Gluten Gabeko Dieta: Euskal Autonomia Erkidegoko pertsona zeliakoen dieta ohiturak

IDOIA LARRETXI LAMELAS

Doktorego Tesia. Vitoria-Gasteiz

2019

eman ta zabal zazu

universidad del país vasco

euskal herriko unibertsitatea

Dieta Sin Gluten: Hábitos dietéticos de la personas celíacas de la Comunidad Autónoma Vasca

IDOIA LARRETXI LAMELAS

Tesis Doctoral. Vitoria-Gasteiz

2019

eman ta zabal zazu

universidad del país vasco
euskal herriko unibertsitatea

Gluten Free Diet: Dietary habits of celiac people from the Basque Country

eman ta zabal zazu

universidad del país vasco
euskal herriko unibertsitatea

IDOIA LARRETXI LAMELAS

Doctoral Thesis. Vitoria-Gasteiz

2019

Badira bide batzuk derrigorrez ibili behar

ditugunak, inora ez doazela jakiteko

Joseba Sarrionaindia

Lehenik eta behin nire zuzendariei, Edurne Simón eta Itziar Txurrukari, urte guzti hauetan zehar emandako laguntza guztiagatik eskerrak eman nahi nizkieke. Lehenengo egunetik nire alboan egon zarete eta eredu izan zarete, alderdi zientifikoan bai, baina baita pertsonalean ere. Talde ezinhobea eratzen duzue, hortaz ez dut imaginatzen zuzendari hobeak tesi hau burutzeko! Mila esker!!

Bigarrenik GLUTENSSS taldeari: Arrate Lasa, Jonatan Miranda, Virginia Navarro, Marian Bustamante, Pily Fernández, Olaia Martinez, Itziar Txurruka, Edurne Simón eta Jesus Salmeróni. Tesi hau zuena ere bada, nirea bezain beste, eta hori da hain zuzen ere gure taldean gehien baloratzen dudana. Zuekin laguntasunaren eta lantaldearen benetako esanahia ikasi dut. Zuek gabe tesi hau ezinezkoa litzateke. Bihotzez, mila-mila esker denoi!!

A Noemí Arias, Saioa Gómez, Itziar Eseberri, Ana Gracia, Andrea Mosqueda y Leixuri Aguirre por todos los "pintxopotes", cenas, congresos, "jóvenes investigadores" y demás eventos. Sin duda vosotras habéis hecho que estos años sean mucho más divertidos e inolvidables. Eskerrik asko!

También quiero agradecer a todas las personas del área de Nutrición y Bromatología su ayuda durante todos estos años y el haber conseguido que desde el primer día que pisé el laboratorio me sintiera como en casa. Sin duda vuestro ejemplo ha hecho que las ganas de investigar crezcan en mí. Eskerrik asko!!

Euskal Herriko Unibertsitatea ere aipatu behar dut, bereziki “Euskararen eta Etengabeko Prestakuntzaren arloko Errektoreordetza”, Tesi hau burutzeko diru laguntza eman baitzidan. Bigarren urtean zehar “Ikerketaren Arloko Errektoreordetzako” laguntza ere jaso nuen Tesi honetan Finlandian egindako egonaldia burutu ahal izateko.

For all the people from Tampere. Thank you first to Katri Kaukinen, for accepting me in your research group. Thank you Kalle Kurppa and Katri Lindfords for helping me with my work during my stay in Finland. Thank you to all the people from the lab and office for making my days in the office easy and teaching me all those interesting things about the celiac disease. Especially to Heidi Kontro who was my first contact with Tampere. You helped me a lot from the first moment till my last day in Tampere. You don’t know how good is to arrive somewhere you don’t know and find someone like you. Thanks a lot, truly!!! Finally to my lovely “celigirls”: Laura Airaksinen, Minna Hietikko, Suvi Kaillokoski and Juliana Cerqueira. You were a wonderful discovery for me and you made me feeling like at home during my stay in Tampere. Thank you girls!!

Tampoco puedo dejar de agradecer su colaboración a tod@s l@s participantes del estudio llevado a cabo en esta tesis, ya que sin ell@s este trabajo no hubiese sido posible. Eskerrik asko guztioi!

Y finalmente y más importante… a mi familia y amigos:

A mis padres; ama, aita, ESKERRIK ASKO!! No tengo palabras para agradecer todo lo que habéis hecho y seguís haciendo por mí. Gracias por vuestro apoyo incondicional, por quererme y cuidarme todos los días de mi *vida. También quiero dar las gracias de manera especial a mi tía Marisol, ya que seguramente sin su apoyo y ánimo esta tesis no hubiese existido ¡muchas gracias tía! A Javi, Mikel, Miren y Ángel, por estar siempre ahí. Y por supuesto a todo el resto de la familia (a l@s que están y a l@s que no) por todo vuestro cariño. ¡Os quiero mucho!*

Kuadrilako lagunei, eskerrik asko. Bidaiengatik, asteburuengatik, bazkariengatik, parrandengatik, beti hor egoteagatik… zuek zalantzarik gabe egin duzue bide hau askoz atseginagoa. Zuekin alboan, edozein gauza hobea da!! Miriam eta Ido, kilometroek pertsonak banatzen ez dituztela erakutsi didazuelako …Eskerrik asko!!

Eta azkenik, Mikeli… Mila esker, nire bizitzan agertzeagatik, zure pazientziagatik, ulermenagatik eta nola ez…tesi honen inguruko "txapa" guztiak aguantatzeagatik! Jiji! Laburbilduz, bide honetan beti nire ondoan egoteagatik. Mila esker bihotzez!

Idoia

AURKIBIDEA

ARTIKULUAK / PAPERS

Doktorego-Tesi hau datozen bost ikerketa-lanetan oinarritzen da **(1-5 eskuizkribuak**):

This Thesis is based on the following five research papers (**manuscripts 1-5**):

1. Churruca I, Miranda J, Lasa A, Bustamante M, Larretxi I, Simon E. Analysis of Body Composition and Food Habits of Spanish Celiac Women. Nutrients. 2015;7(7):5515-31.

2. Larretxi I, Simon E, Benjumea L, Miranda J, Bustamante MA, Lasa A, et al. Gluten-free-rendered products contribute to imbalanced diets in children and adolescents with celiac disease. Eur J Nutr. 2019 Mar;58(2):775-783.

3. González T, Larretxi I, Vitoria JC, Castaño L, Simón E, Churruca I, et al. Celiac Male's Gluten-Free Diet Profile: Comparison to that of the Control Population and Celiac Women. Nutrients. 2018 Nov 8;10(11).

4. Idoia Larretxi, Itziar Churruca, Virginia Navarro, Jonatan Miranda, Arrate Lasa, Maria Ángeles Bustamante, Edurne Simon. Impact of analytically measured fiber and resistant starch from gluten free products on celiac people´s diet. Manuscript .

5. Larretxi I, Churruca I, Navarro V, Lasa A, Bustamante MA, Simon E, Miranda J Micronutrient analysis of gluten-free products: their low content is not involved on gluten-free diet misbalance. Manuscript.

SARRERA

1. AURREKARI HISTORIKOAK

Gaixotasun zeliakoaren (GZaren) lehen definizioa k.a II. mendean agertu zen Aretaeus de Capadocia medikuaren eskutik. “Afekzio zeliakoa” zeritzon jatorrizko testuan Aretaeusek GZaren kausa somatzen zuela antzeman daiteke. Are gehiago, *“Diatesi zeliakoa*” kapituluan lehen aldiz azaltzen dira zeliakian agertzen diren hainbat manifestazio kliniko, hala nola, beherakoa, desnutrizioa, zurbiltasuna, pisu galera, etab. Testu honen beste kapitulu batean gaitz hau jasaten duten pertsonei lehenengo aldiz “zeliako” izena ematen zaie: *“urdailak ez baditu elikagaiak gordetzen eta bere barnetik pasatzen badira liseritu gabe eta nutrienterik asimilatu gabe, pertsona horiek zeliako izendatuko ditugu”.* Aretaeusek pertsona hauek deskribatzen zituenean “*koliakos*” hitz grekarra erabiltzen zuen, bere esanahia “hesteetako gaitza duen pertsona” izanik.

Bestalde, Samuel Geek 1888an “afekzio zeliakoa” izendatu zuen sindromea deskribatu zuen, eta aipagarria da Geek lehena izan zela gaixotasuna eta dietaren arteko erlazioa ikusten. Bere idazkietan honakoa irakur daiteke: “*Elikadura tratamenduaren alde garrantzitsuena da. Irin-jatorrizko elikagai proportzioak minimoa izan behar du…, gaixoa sendatzen bada dietari esker izango da*” (1, 2).

XX. mendearen lehen hamarkadetan medikuek umeengan jarri zuten arreta guztia. Izan ere, hauek ziren eta dira tratamendu dietetikoaren aurrean erantzun onenak eta azkarrenak dituztenak.

1908. urtean Herter pediatrak liburu bat argitaratu zuen haur zeliakiaren inguruan (3). Hain goraipatua izan zen bere lana gaixotasuna izenez aldatu zuela, "Gee-Herter gaixotasuna" deitzera pasatuz. Pediatraren ekarpen nabariena organismoak gantzak karbohidratoak baino hobeto onartzen zituela baieztatzea izan zen.

1921ean Frederik Still pediatrak hurrengoa esan zuen: "*Ogia almidoi mota bat da zeliakiaren sintomak larritzen erantzukizun berezia duena, tamalez ez dut ezagutzen elikagai horretarako egokia den ordezkorik*". Urte berean John Howlandek ume zeliakoekin izandako esperientziak deskribatu zituen bere idazkietan, eta karbohidrato gabeko dieta eramatearen beharra adierazi zuen ere bai. Alabaina, ez zuen GZaren terminoa erabili, garai hartan gaixotasunak izen mordoa baitzituen. Mediku horren arabera, karbohidratorik gabeko dieta jarraitzean pazienteek dietaren beste mantenugaiak askoz hobeto onartzen bazituzten ere, gantzen xurgapena ez zen inoiz paziente osasuntsuetan bezain ona. Howlandek gomendatzen zuen dieta hiru faseetan banatzen zen, eta ogia, zerealak eta patatak pixkanaka sartzea baimentzen zuen azken fasean.

Hiru urte geroago, 1924an, Haas medikua zeliakoen dieta aberasten saiatu zen ondo onartzen zuten karbohidrato bat bilatuz. Ordura arte zeuzkaten ezagupenetan oinarrituta, platano helduetan oinarritutako dietarekin entsegu klinikoak egiten hasi zen. Horrela, Hassek ume zeliakoek fruta eta barazkietatik zetozen karbohidratoak ondo onartzen zituztela demostratu zuen eta, hortaz, dieta aberatsagoa eta erakargarriagoa jarraituz bere osasuna hobetu zezaketela eta gantz eta proteinetan soilik oinarritzen zen dieta ekidin zezaketela ere. Ondorengo urteetan Hass eta bere semeak seiehun paziente baino gehiago tratatu zituzten, dieta horrekin gaixotasunaren pronostikoa izugarri hobetuz (4).

GZaren hainbat ezaugarri ezagutzen baziren ere, 40. hamarkadara arte ez zen gaitza egoki definitu. Dicke pediatrak bere doktorego-tesian (5) funtsezko aurkikuntza egin zuen, bai haur nahiz heldu zeliakoentzat. Aipagarri da II. Mundu Gudan Holandan jasandako gosetearen ondorioz garatu zuela bere ikerketa, nahiz eta aurretik susmoak izan, garai hartan gaixoen sintomak murrizten ikusi baitzuen. Dickek demostratu zuen ume zeliakoek dietatik gari-, zekale- eta olo-irinak baztertzen zituztenean nabari hobetzen zutela gaitza (6). Horrela, elikagai horiek hartu ordez gari- edota arto-almidoiak nahiz arto- edota arroz-irinak kontsumituz pazienteek gosea berreskuratzen zuten eta beherakoa desagertzen zitzaien (7, 8).

Geroago, Charlotte Andersonek eta bere taldeak hori berretsi zuten almidoia eta beste konposatu batzuk gari-irinetik erauzi zituztenean eta glutena deritzon zatiarekin gelditu zirenean, zereal horren alde toxiko bezala definituz. Hortaz, 1950. urtetik aurrera gluten gabeko dieta (GGD) izan da zeliakoen tratamenduaren oinarria (9).
Beranduago, beste aurkikuntza garrantzitsu bat etorri zen J.W. Paulley doktorearen eskutik. Pertsona zeliako bati egindako ebakuntzan jabetu baitzen heste meharreko mintzean hantura zuela. Horrez gain, Paulleyk heste biloen galera ere deskribatu zuen, nutrienteen xurgapena gertatzeko beharrezkoak direnak (10). Ondoren, 1960an Andersonek ikusi zuen heste meharreko mukosa guztiz osa zitekeela GGD jarraitu ezker.

2. GAIXOTASUN ZELIAKOA

Azken hamarkadetan GZa diagnostikatzeko eta tratatzeko jarraibideak mantendu dira. Alabaina, urteak pasa ahala ikerketa berriei esker gaixotasunaren inguruan dagoen ezagutza handituz joan da eta, honekin batera, zeliakiarekiko dugun ikuspuntua moldatu ere. 2012. urtean ESPGHANek (*European Society for Pediatric Gastroenterology, Hepatology and Nutrition* erakundeak) hurrengo definizioa sortu zuen: *Zeliakia aurre-joera genetikoa duten pertsonengan glutena edo/eta erlazionatutako prolaminak hartzeak eragiten duen asaldura autoimmune eta multiorganikoa da. Honako aldagai hauen araberakoa da: gluten ingesta, enteropatia, GZan espezifikoak diren antigorputzak eta HLA-DQ2 eta HLA-DQ8 haplotipoak agertzea, adierazpen klinikoak* (11).

Definizio berri hori baliagarria izan zen aintzinatik zegoen GZren ideia alde batera usteko (soilik umeetan agertzen den malabsortzio sindrome arraroa izatearena alegia). Horrela, gaitza adin guztietan ager daitekeen eta adierazpen kliniko anitzak dituen patologia arrunta eta zabaldua izatera pasatzen da (12).

2.1 Epidemiologia

Azken urteotan GZaren epidemiologia eta sintomatologia aldatu dira. Historikoki, zeliakiak gehienetan biztanleria kaukasikoan izan du eragina eta, ohiko sintoma gastrointestinalen agerreran oinarrituz, bizitzaren lehen urteetan diagnostikatu izan da, heste meharreko biopsiaren bitartez (13). Egun, badakigu GZa adin guztietako pertsonetan ager daitekeela eta emakumeetan nagusiagoa dela, ratioa 2:1 izanik. Bibliografiak adierazten duen moduan, GZaren prebalentzia era esanguratsuan handitu da azken 20 urteetan. Are gehiago, zeliakia heredagarria eta kronikoa den hesteko gaitz ohikoena bilakatu da mundu mailan (14). Hori gertatu da, alde batetik, egungo osasun sistemak diagnostiko hobeak egitea ahalbidetzen duelako eta, bestetik, gaixotasuna eta bere sintomen ezagutza sakonagoa delako osasun profesionalen aldetik.

1996an Catassi et al.-ek frogatu zuten GZa arruntagoa zela garai hartan uste zena baino, 17.000 ikasle osasuntsuri egindako screening serologiko batean oinarritu ziren eta kasu atipikoen bilaketa egin ezean diagnostikatu gabe geldituko zirela aipatu zuten. Horrela, GZaren epidemiologiak icerbergaren ezaugarriak dituela ohartu ziren: sintomatikoak diren pertsonak (sintomak gastrointestinalak zein estraintestinalak izanik) icebergaren tontorrean kokatzen dira eta zeliakia isila edo atipikoa duten pertsonak eta diagnostikatu gabe daudenak, berriz, behealdean. Ideia horrek ahalbidetu du orain arte susmagaitzak ziren ez-ohiko kasu klinikoak diagnostikatzea eta beraien maiztasun altua erakutsi eta prebalentzia erreala ebaluatzea.

Nahiz eta zeliakiaren prebalentzia guztiz finkatuta ez dagoen, oro har, mendebaldeko herrialde gehienetan biztanleriaren %1-2an kokatzen da. Hala ere, alde handia behatu da herrialde ezberdinen artean (13, 15). Adibidez, Peña eta Rodrigok desberdintasun esanguratsuak aurkitu zituzten Ipar Europako eta Mediterraneoko herrialdeen artean, neurtutako prebalentziak %0,016 eta % 2,4 izanik hurrenez hurren (15). Beste ikerketa batzuetan ikusi den moduan, batezbesteko prebalentzia herrialde garatuetan (Estatu

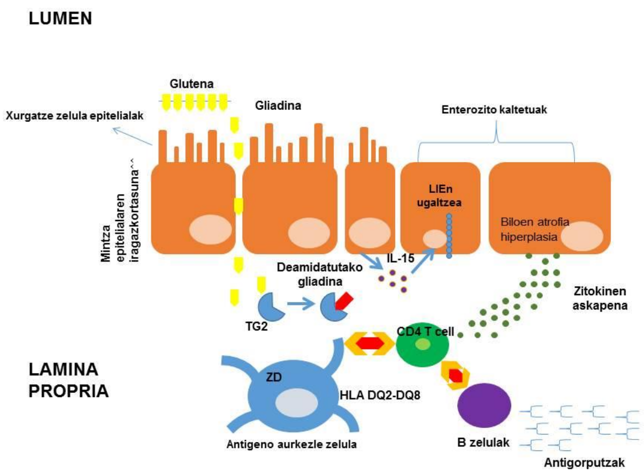
Batuetan, Australian edo Nueva Zelandan esaterako) Europan aurkitzen denaren antzekoa da (13, 15). Bestalde, 1998an Catassi *et al.*-ek %5,6ko prebalentzia neurtu zuten Mendebaldeko Saharako biztanlerian (16). Gertakari hori justifikatzeko bi arrazoi aipa daitezke: gluten kontsumo handiagoa eta HLA-DQ2 eta HLA-DQ8 haplotipoen agerrera. Ekialdeko lurraldeetan ostera, agertzen den prebalentzia anekdotikoa da, seguraski gluten kontsumo eskasagatik eta gaixotasunerako aurre joera sorrarazten duten geneen gabezia edo prebalentzia baxua dela eta (9, 13, 15, 17).

GZa garatzeko probabilitatea zenbait arrisku taldetan handiagoa da: zeliakoen lehen mailako senideetan, A immunoglobulinaren gutxiegitasun selektiboan eta asaldura genetiko jakin batzuk pairatzen dituzten pazienteetan (adibidez, Down, Turner edo William sindromeetan). Halaber, aipagarria da, zeliakiaren prebalentzia areagotua dagoela gaixotasun autoimmuneak dituzten pertsonen artean, esaterako, 1. motako diabetesa, tiroidearen gaixotasun autoimmunea, gibel-gaixotasun autoimmune kronikoa, gazteen artritis kronikoa, Sjögren sindromea, giltzurrun-gaixotasuna edo kardiomiopatia dilatatu idiopatikoa. Beraz, arrisku talde horiek definitzea eta ezagutzea oso garrantzitsua da eta, nahiz eta genetikoki GZarekin erlazionatuta ez egon, agertzen duten zeliakia prebalentzia altuak beharrezkoa egiten du analisi serologikoen bitartez berauek kontrolatzea (18).

2.2 Patogenesia eta aurkezpen klinikoa

Zeliakiaren garapenean faktore genetiko, immunologiko eta ingurugiroko aldagaien arteko elkarrekintza ezinbestekoa da.

Glutena, bi zati proteiko dituena (prolaminak eta glutelinak), elikagaien bidez irensten da eta bere digestioaren ondorioz (1. irudia) peptido txiki bihurtzen da. Ondoren, garia adibidetzat hartuz, gariaren prolaminak, gliadina deritzona, enterozitoen arteko mintz epiteliala gurutzatzen du, zeliakoengan iragazkortasuna handiagoa baita. Horrela, peptidoa lamina propriara ailegatuko da, bertan transglutaminasak (tTG2k) peptidoa deamidatuko du HLA DQ-2k edota DQ8k ezagutzea ahalbidetuz. Antigeno-aurkezle zelulek deamidatutako gliadina CD4 zelulei aurkeztuko diete. Zelula immunologiko horiek, alde batetik, hantura eragiten duten zitokinak jariatzen dituzte enterozitoa minduz (heste-biloaren atrofia eta kripten hiperplasia gertatuz), baina, bestalde, B zelulak aktibatzen dituzte, gliadina edota “toxikoak” diren beste prolaminen kontra joateko beharrezkoak diren antigorputzak sortuz. Era berean, deamidatutako gliadinak enterozitoek IL15 ekoiztea eragiten du. Eta azken horrek, linfozito intraepitelialak (LIE) ugaltzea sustatuko du.



1. irudia. GZaren patogenesia (Moscoso *et al*.-etik moldatuta). LIE:linfozito intraepitelialak, DC:zelula dendritikoa, TG2: transglutaminasa.

GZaren garapenean eragile izan daitezkeen arrisku faktore desberdinak aztertu izan dira, adibidez, bizitzaren lehen urteko elikadura mota. Hala ere, ez dago ebidentzia nahikorik edoskitze naturalak GZa garatzea saihesten duela baieztatzeko, hortaz, ezin da edoskitzerako gomendiorik zabaldu (ez iraupenari ez beikostarekin bateratzeari dagokionez) (19, 20). Glutenaren sarreraren eta gaixotasunaren garapenaren arteko erlazioari dagokionez, emaitza desberdinak proposatu dira (19). Nolanahi ere, ESPGHANek gomendatzen du gluten ingestio goiztiarra (< 4 hilabete) zein berantiarra (>7 hilabete) ekiditea eta, ahal den heinean, glutena umearen elikaduran sartzea edoskitzeak diraun bitartean. Aipagarri da glutendun dietak genetikoki aurrejoera duten umeentzat bakarrik direla arriskutsuak, halere, gomendio hori ume guztientzat luzatu egiten da. Hasieran gluten kopuru txikiak hartzea komeni da eta horiek poliki-poliki handituz joatea. Dena den, ez da oraindik zehaztu optimoa den gluten zenbatekoa (20).

Bestalde, GZaren eta infekzio gastrointestinalen arteko erlazioa ere aztertu da. Badirudi infekzioek GZaren autoimmunitatea handitzen dutela gaixotasunerako genetikoki sentikorrak diren pertsonetan (21) eta rotavirusen kontrako txertoa dutenetan, berriz, txikitu. *Helicobacter pylori*ren kasuan proposatu izan da infekzio gastrikoak zeliakiaren kontrako efektu babeslea eduki dezakeela (22). Laburbilduz, gaixotasun honen etiologian bibliografiak balizko elkarrekintzak deskribatu ditu, infekzio, faktore genetiko eta dietaren artekoak hain zuzen ere.

Aurkezpen kliniko zabala dauka zeliakiak, diagnostiko egokia eta azkarra zailtzen duena. Kasu batzuetan ia ez da sintomarik antzemateen (14), eta agertzen direnean sintoma gastrointestinal eta estragastrointestinalak bereizten dira (1. taula). Gainera, seinaleak nabari aldatzen dira adinaren arabera; umeen kasuan ohikoagoa izanik gastrointestinalak agertzea eta helduetan, aldiz, beste motatako sintomak. Izan ere, heldu diagnostikatuen %25 baino gutxiagok aurkezten du aparatu digestiboko klinika (23).

1. taula. GZ sintomatikoaren aurkezpen klinikoa.

Sintoma	**Adin taldea**	
Digestiboak	Umeak	Apetitu falta Beherako kronikoa Idorreria Sabeleko mina Goitikoa Distentsio abdominala
	Helduak	Heste suminkorraren sindromea Beherako kronikoa Beherako malabsortiboa Idorreria Sabeleko mina Distentsio abdominala
Ez digestiboak	Umeak	Artritisa Keratosi folikularra Suminkortasuna Menstruazio irregularrak Menarkia atzeratua Hazkuntza atzeratua Osteopenia Dermatitis herpetiformea Buruko mina Ezpaineko hotzikara Giharren hipotrofia Malnutrizioa/anemia
	Helduak	Anemia

		Menopausia goiztiarra Osteoporosia/artritisa Parestesia Ataxia/ epilepsia/ neuropatia periferikoa Suminkortasuna Dermatitis herpetiformea Edema periferikoa Ezpaineko hotzikara

Esan bezala, GZ klasikoa paziente guztien portzentaje txiki bati dagokio eta bere ezaugarri nagusiak biloen atrofia eta xurgapen txarraren eraginez sortutako sintoma gastrointestinalak dira eta 6-24 hilabete bitarteko umeetan agertzen da. Bestalde, GZ atipikoa ume nagusiago edo helduen kasuan gertatzen da eta, kasu hauetan, malabsortzio-ezaugarriak arraroagoak dira. Test serologikoak agertu zirenetik GZ isila (itxuraz asintomatikoa dena) gehiago diagnostikatu egin da, arrisku taldeei egiten zaizkien aldizkako *screening*en ondorioz; hala nola, lehen mailako ahaidei edota gaixotasun autoimmune (1. motako diabetesa) edo genetikoak dituzten pertsonei (13). Esan bezala, sintomatikoak diren kasuek gaixotasunaren portzentaje baxua osatzen dute. Berez, ikerketa epidemiologiko askok diotenez, klasikoak ez diren sintomak dituzten pertsonak sintomatologia klasikoa pairatzen duten pertsonak baino gehiago dira (24).

2.3 Diagnostikoa

Zeliakia diagnostikoa jasotzen duen pertsona kopurua handitzen ari den arren, oraindik ere gaitz hau oharkabean pasatzen da askotan. Gaixotasunaren diagnostiko kliniko tradizionala honakoetan oinarritu izan da: sintomak, erlazionatutako gaixotasunak edota arrisku faktoreak agertzea. Alabaina, zeliakiaren inguruko ezagutzak erakutsi duenez, hori ez da nahikoa gaixotasunaren diagnostiko egokia egiteko (25). Hortaz, ESPGHANek gaixotasuna diagnostikatzeko irizpideak berrikusi zituen, gaur eguneko diagnostiko-test immunologikoen zehaztasun eta aurrerapenei esker eta arrisku taldeek zeliakia pairatzeko duten aurrejoeraren inguruko ezagutzari esker (11). Horrela, ESPGHANek zehaztu zuen GZa diagnostikatzeko test serologikoak, azterketa genetikoak eta heste-biopsia behar direla (2. taula) eta horien eta sintomen arabera zeliakia mota erabakiko dela.

2. taula. GZ mota desberdinak.

GZ motak	Genetika (HLA DQ2 edo DQ8)	Anatomia patologikoa	Antigorputzak (anti-tTG, MA edo anti-DPG)	Sintomak
Sintomatikoa	Bateragarria	+	+	+
Isila	Bateragarria	+	+	-
Latentea	Bateragarria	-	±	±
Potentziala	Bateragarria	-	+	±

GZ *isilean* gaixoek zeliakiarako espezifikoak diren antigorputz positiboak daude, HLA bateragarria eta enteropatia aurkeztuko dute, zeliakia *klasikoan* bezala, baina ez dira sintoma nahikoa agertzen diagnostikoa bermatzeko. Kasu horiek normalean arrisku taldeetan dauden pertsonetan agertzen dira, lehenengo mailako ahaideetan adibidez, odol analisiak egiterakoan. GZ *latentean* HLA bateragarria dago baina, aitzitik, enteropatia ez da agertzen. Zeliako horiek sintomak eta gaixotasunaren antigorputzak eduki ditzakete ala ez. GZ *potentziala* antigorputz positiboen presentziagatik eta HLA bateragarriagatik bereizten da, baina heste-biopsian ez da anormaltasunik agertzen (nahiz eta etorkizunean ager daitezkeen). Batzuetan sintomak agertuko dira eta besteetan ez.

Beraz praktika klinikoan, GZaren susmoa dagoenean bai sintomak daudelako, bai pazientea arrisku talderen batean aurkitzen delako, diagnostikoak bide desberdinak jarraituko ditu:

1. <u>Azterketa serologikoa</u>:

Azterketa honek garrantzi handia dauka GZaren diagnostikoan, eta oso erabilia izan da azken 25 urteetan; alde batetik, arrisku altuko taldeetan *screnning*ak egiteko erabiltzen delako eta, bestetik, jadanik enteropatia detektatu den gaixoetan diagnostikoa berresteko balio duelako (26, 27). Analitikak GZaren antigorputz espezifikoak erabiltzen ditu adierazle bezala. Biztanleriaren gehiengoan erabiltzen diren testak ehun-transglutaminasaren kontra doazen IgA autoantigorputzak (anti-tTG) dira. Horiez gain, IgA motatako endomisio kontrako (EMA) antigorputzak ere erabiltzen dira. Bi antigorputz horiek oso sentikortasun eta espezifizitate altua dute diagnostikorako eta, beraz, gehien erabiltzen diren biomarkatzaileak dira. Hori dela eta, bi test horiek ordezkatu dute gliadinaren aurkako antigorputzan (anti-AGAn) oinarritzen zen testa, espezifizitate gutxiago zuena.

Azterketa hauek egin aurretik garrantzitsua da gazuran dagoen IgA kopurua neurtzea; izan ere, zeliakoen %2-3k IgA urritasun hautakorra aurkezten du eta pertsona horiek, bereziki helduek, negatibo faltsuak jaso ditzakete antigorputz-probetan. Egoera horretan IgG antigorputzetan oinarritzen diren testak erabiliko dira, hala nola, anti-tTGa edo deamidatua dagoen gliadina peptidoaren aurkako (anti-DGP) antigorputzak. Izan ere, IgG motatako anti-DGP antigorputzen espezifizitate eta sentikortasuna IgA motatako anti-tTG antigorputzen antzerakoa da (28). Azken bi azterketa horiek 2 urte baino gutxiagoko umeetan ere erabiltzen dira, haiengan IgA antigorputzetan oinarritutakoetan negatibo faltsu asko gertatzen baitira (29).

Aipagarri da, behatu denez, azterketa serologikoak erabilgarriak izan daitezkeela ere GGD zorrotza jarraitzen duten pazienteetan heste-mintzen kaltea eta eboluzioa monitorizatzeko (30).

Dena den, azpimarratzekoa da analisi hauen emaitza negatiboek ez dutela beti GZren absentzia ziurtatzen eta sarritan, gaixotasunaren susmoa oso altua denean, beste azkerketa zuzenagoen beharra dagoela. Esate baterako, sintomatikoa den eta zeliako baten ahaide hurbila den pertsonak, nahiz eta azterketa serologikoan negatiboa jaso, biopsiaren beharra edukiko du.

Azterketa serologikoak egiten direnean pazienteak glutendun dieta jarraitzea beharrezkoa da, emaitzak faltsututa ez daudela bermatzeko. Diagnostikorako gluten ingestio kopuru minimoa 6 asteetan zehar 10g/egun (4 ogi xerra) izan bada ere, azken ikerketek diote denbora tarte laburragoan gluten kopuru txikiagoa nahikoa dela aldaketa serologikoak eragiteko zeliakoengan. Horrela, 2 asteetan zehar gutxienez 3g gluten erabiltzea proposatu da (31, 32).

2. <u>Azterketa genetikoa</u>:

Zeliako gehienak (%95 inguru) HLA-DQ2 positibo dira eta gainerakoak (ia %5), berriz, HLA-DQ8 positibo. Biztanleria orokorrean, aldiz, haplotipo horiek bakarrik %20-30ak adierazten ditu. Hortaz, HLA-DQ2 edo HLA-DQ8 ez edukitzeak diagnostikoa bazter dezake kasu gehienetan (nahiz eta HLA-DQ8 eta HLA-DQ2 negatiboak diren zeliakia kasuak ere detektatu diren) (33).

3. <u>Heste-biopsietako analisi histologikoa</u>:

GZaren behin-betiko diagnostikoa egiteko duodeno proximalaren edo jeiunoaren biopsia erabiltzen da. Azterketa honek hestearen aldaketa estrukturalak edo zelulen anormaltasunak identifikatzen ditu. ESPGHANen azken jarraibideen arabera, gutxienez bost lagin desberdin hartzea gomendatzen da (1 erraboiletik eta beste 4 duodenoaren bigarren eta hirugarren ataletatik) hesteetako lesioa sakabanatuta aurkeztu daitekeelako (11). Azterketa hau ere pazientea glutendun dieta jarraitzen ari denean egin behar da, edo gluten karga bat eragin ondoren behintzat (31).

Biopsiaren azterketan patologoak kontuan hartuko ditu: biopsiaren orientazioa, heste-biloen egoera, kripten elongazio eta atrofia maila, bilo/kripta ratioa, linfozito intraepitelialen (IELs) kopurua eta Marsh-Oberhuber sailkapenaren araberako interpretazioa, praktika klinikoan oso hedatua dagoena. (24, 25, 28, 30, 31, 34, 35).

Heste meharrak duen kaltea neurtzeko Marsh sailkapena erabiltzen da. 4 maila bereizten ditu: 0 maila mintza osasuntsua izanik eta III mailak mintzaren atrofia handiena adieraziz. Zeliako gehienek diagnostikoaren momentuan Marsh III balioa erakusten dute. IV mailak lesio hipoplasikoa erakusten du, atrofia osoa eta kripten hipoplasia bilduz. Hala ere, azken maila hau ez da Gastroenterologiaren Mundu Erakundearen zeliakiarako jarraibideetan agertzen (36).

Kalte histologikoa bateragarria da GZaren diagnostikoarekin baina ez da gaitzaren berariazko ezaugarri klinikoa. Beraz, diagnostikoa berresteko azterketa histologikoa eta azterketa serologikoa uztartu behar dira.

Nahiz eta biopsia gaur egun zeliakia diagnostikatzeko oinarrizkoa den, analisi hori ekidin eta diagnostiko zuzena egiteko aukera aztertzen ari da. 2012an ESPGHANek idatzitako jarraibideetan iradoki zen umeen diagnostikoan biopsia ekidin daitekeela hurrengo baldintzak betetzen direnean: GZaren klinika arrunta, IgA antigorputzen (IgA anti-tTg) balioak ohikoa baino 10 aldiz altuagoak izatea, bigarren lagin batean anti-endomisio (EMA) antigorputzekin aurrekoa egiaztatzea eta test genetiko positiboa, DQ2 edo DQ8 adierazleak aurkeztea alegia (11).

GZa urteetan zehar isil edo latente mantendu daitekenez, kontrol jarraitua eta sakona behar du, gehien bat zeliakoen gertuko ahaideetan edo arrisku taldeetan dauden pertsonetan.

2.4 Tratamendua

Gaur egun zeliakiarako eraginkorra den eta onartuta dagoen tratamendu bakarra GGD jarraitzea da (12). Hala ere, zientzialariak GZaren mekanismo fisiopatologikoen ikasketarekin batera tratamendu alternatibo desberdinak ikertzen ari dira, GGDrekin bateratzeko edo/eta berau ordezkatzeko botikaren bila (37).

Farmakoak GZa pizten duten mekanismoen fase ezberdinak inhibitzeko diseinatu dira, honako helburu espezifiko ezberdinez: entzimen bidezko peptido toxikoen inaktibazioa heste lumenean, gliadinaren bahiketa bere heste mukosarako sarrera ekidin edo murrizteko, jauzi immunologikoan gertatzen den iragazkortasuna edo/eta antigeno aurkezpena modulatzea hantura erantzuna erregulatuz (tTG2ren blokeoa, HLA-DQ eta T zelulen arteko loturen blokeoa),CD4+ gluten espezifikoak diren T zelulentzako epitopoen txertoa, ea. (38).

Halaber, probiotikoek joka dezaketen papera ere ikertu da. Organismoaren mikrobiotaren aniztasuna eta oreka berreskuratzen laguntzen dute eta horrek zeliakian gerta daitekeen hantura-prozesua arindu eta heste epitelioan defentsa mekanismoa hobetu baititzake (24, 39).

Dena dela, bizitza osorako GGD zorrotza, epe luzera mantenduz gero zeliakoen bizi kalitatea hobetu egiten du, baita asintomatikoak diren zeliakoetan ere (40, 41). Alderantziz, tratatu gabeko zeliakoek, glutena hartzen dutenek, alegia, hainbat gaixotasunak garatzeko arrisku handiagotua daukate: hala nola, minbizia, linfomak, heste meharreko neoplasia, tumore orofaringeoak, ugalkortasun asaldurak, osteoporosia, hezurren hausketa, etab.

Aurreko guztia dela eta, behin diagnostikoa egin eta GGD ezarri denean gaixoaren jarraipena egitea gomendatzen da, sintomen arintzea ebaluatzeko, markagailu serologikoak aztertzeko eta dietarekiko atxikimendua ziurtatzeko (42).

2.5 Glutenarekin erlazionatzen diren gainerako gaixotasunak

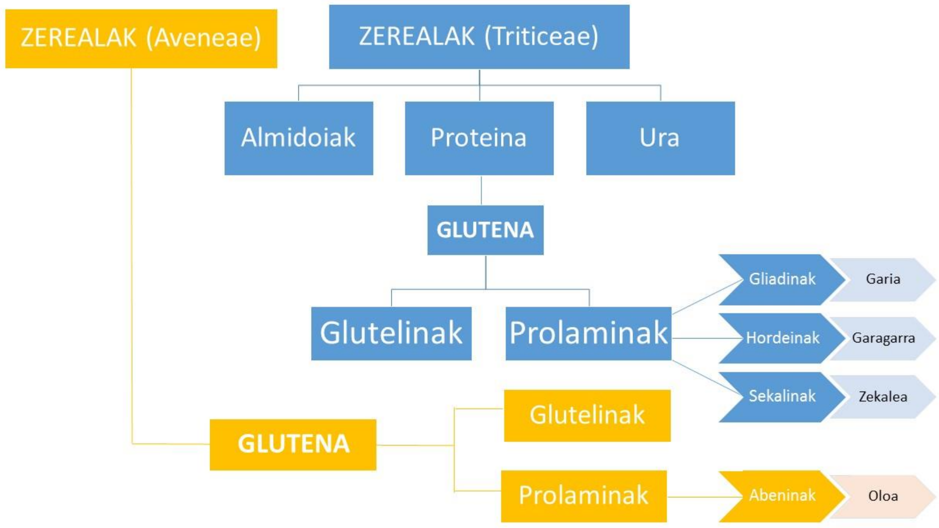
GZa eragiteaz gain, gluten proteinak beste hainbat gaixotasun sorrarazle izatea ere esleitu zaizkio, horien artean aipagarri dira gariarekiko alergia (GA), dermatitis herpetiformea eta glutenak eragindako ataxia edo neuropatia periferikoa (43).

GZak oinarri autoimmunea duten bitartean, GA E-Igen bidezko erantzun immunearen asaldura da. Espozizio mota eta horrek pizten dituen mekanismoen arabera sailkatzen da, honako GAk bereitzen dira: elikagaiekiko alergia, jarduera fisikoak eragindako gari-ingestaren menpeko anafilaxia, asma eta rinitisa eta ukipenezko urtikaria. Forma ohikoena arnas-aparatuan eragina duen alergia da, hots, “okinaren asma ”(44).

Azken urteetan “glutenariko sentikortasun ez-zelikoa” (GSEZ) deritzon gaixotasunarekiko interesa piztu da ikerlari eta medikuen artean. GGD beste hainbat gaixotasunen sintomak lasaitzeko eraginkorra dela behatu da, eta horrela, proposatu da GSEZk glutenarekin erlazionatzen diren hainbat gaitz ezberdin bateratu ditzakeela. Hala ere, bere jatorria ezezaguna da oraindik (44) eta, uste denez, glutena ez omen da gaixotasunaren eragile bakarra. Hala, eztabaida handia dago beste molekula batzuek, hala nola FODMAPSek (oligo-di-mono-sakarido eta poliol hartzigarriek), sindrome honetan duten paperaren inguruan (43, 45). Hortaz, autore batzuek proposatzen dute GSEZ izena aldatzea eta bere ordez “gariarekiko intoleratzia sindrome” erabiltzea (43, 46). Dena den, momentuz GSEZ terminoa erabiltzen da, kasu hauetan: pazienteak, GZa edo GA diagnostikatzeko irizpideak bete gabe, gluten ingestioaren aurrean sintoma gastrointestinalak edo/eta estragastrointestinalak dituenenan eta GGD zorrotza jarraituta desagertzen zaizkionean (43). Laburbilduz, gaixotasun horiek guztiek tratamendua dute komunean, GGD zorrotza, hain zuzen ere.

3. GLUTENA

Glutena proteina-konplexu bat da, *Triticeae* familiako zenbait zerealen hazietan dagoena, hala nola, garian (*Triticum aestivum L.)*, garagarrean (*Hordeum vulgare L.*) eta zekalean (*Secale cereale L.*). Proteinok komunak diren peptido-talde homologoak dituzte. Oloa zerealen subfamilia (*Pooideae*) berdinekoa izanda ere *Aveneae* familiatik dator. Egun eztabaida handia dago oloaren toxikotasunaren inguruan, murritzagoa dirudiena; izan ere, bere prolaminek ezaugarri desberdinak aurkezten dituzte *Triticeae* familiakoekin konparatuz.



2. irudia. Zerealetan agertzen diren prolaminen sailkapena.

Glutenak zereal-aleetako proteinen %70-80 osatzen du. Biltegi funtzioa du, haziaren endospermoan nitrogenoa, karbonoa eta sulfuroaren metaketaz arduratuz (47). Glutena bi proteina multzok osatzen dute (2. irudia): glutelinek (soluzio azidoetan solugarriak direnak) eta prolaminek (alkoholean solugarriak direnak). Nahiz eta proportzio ezberdinetan agertzea zerealetan, prolamin: glutelin ratioa 1:1 dela arbitrariotasuna eskuarki onartzen da. Pertsona zeliakoentzako peptido toxikoak prolaminak dira. Nolanahi ere, ikerketa batzuetan glutelina proteinen sekuentzietan ere peptido immunogeniko edo toxikoak aurkitu dira.

Prolamina bakoitzak izen desberdina hartzen du jatorrizko zerealaren arabera, horrela gariaren prolaminak gliadinak dira; garagarrenak, hordeinak; zekalearenak, sekalinak eta, azkenik, oloarenak abeninak (2. irudia). Prolaminak eta glutelinak zerealetan monomero, oligomero edo polimero gisa ager daitezke, disulfuro loturen bitartez elkartuta. “Prolamina” eta “glutelina” terminoak proteina hauetan dagoen prolina (Pro) eta glutamina (Gln) aminoazido kopuru altuari sor zaizkio, hurrenez hurren. Prolaminek, prolinatan aberatsak izateagatik hain zuzen, digestio-entzimek eragindako degradazio proteolitikoa gainditzen dute, baita pertsona osasuntsuetan ere (48, 49). Hortaz, hestean tamaina handiko peptidoak gelditzen dira liseritu gabe. Horiek endozitosiz edo enterozitoen arteko loturen bidez lamina propriara sartuko dira eta aurrejoera duten pertsonetan sistema immunologikoa martxan jarriko dute GZa piztuz.

Proteina hauek glutendun irinek oreari ematen dizkioten ezaugarrien arduradunak dira. Prolaminek biskositatea esleitzen duten bitartean, glutelinek indarra eta elastikotasuna ematen diote oreari; ondorioz, gasaren atxikimendua areagotu egiten da eta produktuak bukaerako estruktura mantentzen du (50). Hori dela eta, gluten proteina erauzi zaien zerealekin edota eratorriekin ekoizten diren produktuen palatabilitatea ez da glutendun homologoena bezain atsegina, galtzen dituztelako glutenak berez ematen dituen propietate organoleptikoak (kohesioa, tinkotasuna, hezetasuna eta uniformetasuna). Halere, azken urteotan gluten gabeko produktuen (GGPen) merkatua handitu denez, elikadura-industriak aurrerapen handiak egin ditu ekoizpen formula berriak garatuz eta, horrela, GGPen propietate organoleptikoak hobetuz. Elikagai espezifiko horiek ekoizteko, adibidez, era naturalean glutenik ez duten osagaiak erabiltzen dira, nagusiki arroza, artoa edota pseudozerealak (kinoa, artobeltza, amarantoa). Kasu horietan, glutendun zerealak dituzten propietate organoleptikoak imitatzeko beste osagai batzuk gaineratzen dira: hidrokoloideak, emultsionatzaileak, isolatutako proteinak (arrautza eta esnekienak) edo entzimak besteak beste (51).

Hala eta guztiz ere, glutenak elikadura-industriako osagai erabilienetakoa izaten jarraitzen du, lehen aipatutako ezaugarri organoleptikoei eta duen balio nutrizionalari esker (18). Zeliakoek ezinbestekoa dute jakitea zein elikagaik duten edo izan dezaketen glutena eta zeintzuk ez. Izan ere, proteina hau hartzea, kopuru txikian bada ere, kaltegarria da zeliakia duten pertsonentzat. Horrela, elikagaiak gluten edukiaren arabera sailkatzen dira, arriskutsuak izan daitezkeenak erraz detekatzeko asmoz:

1. <u>Glutena duten elikagaiak:</u> Zeliakoen osasunerako kaltegarritzat hartzen dira aipatutako prolaminak dituzten zerealekin, edo haietatik eratorritako irinekin, egindako elikagai edo produktu guztiak. Zerrenda honetatik kanpo geratzen dira garia, garagarra edo zekalearen almidoiekin zeliakoentzat egindako GGP bereziak, baldin eta etiketan

berariaz adierazia agertzen bada ezaugarri hori (ikur edo adierazpen baten bitartez).

2. Glutenik gabeko elikagaiak: Glutenik gabeko dietarako egoki eta seguruak diren bakarrak dira. Talde honen barne bere osakeran *per se* glutenik ez duten zerealak daude (arroza, kinoa, artoa, artatxikia, artobeltza, etab.) eta zerealen eratorriak ez diren beste elikagai taldeak, hala nola, fruta, fruitu lehorrak, barazkiak, lekaleak, arraina, haragia eta esnekiak.

3. Glutena izan dezaketen elikagaiak: Elikagaigintzan gari-irina asko erabiltzen denez, kontuz ibili behar da berez konposizioan glutenik izan behar ez luketen arren elaborazio prozesuan kutsatzeko arriskua izan duten produktuekin edo bere osagaien artean glutena gehitu den produktuekin.

3. taula. Elikagaien sailkapena eta hainbat adibide glutena agertzeko arriskuari dagokionez.

GLUTENDUN ELIKAGAIAK	1. Garia, oloa*, garagarra, zekalea, espelta, tritikalea, basagaria, kamuta, espelta berdea, bulgurra, gari semola. 2. Barazkiak ogi-arrailetan edo irinetan pasatuak. 3. Fruitu lehor irineztatuak (adibidez, pikupasak). 4. Jogurta zerealekin eta jogurta gailetekin. 5. Ogi-arrailetan edo irinetan pasatutako haragia edo arraina, edo saltsa glutendunak dituztenak. 6. Bexamel saltsa. 7. Txokolatea zereal glutendunekin, opilak, pastelak, tartak, gailetak, bizkotxoak eta pastelgintzako beste produktuak. 8. Garagardoa (glutena kentzeko tratamendurik gabea),

	kafe-suzedaneo garagar edo maltadunak, edariak zereal glutendunekin.
GLUTENIK GABEKO ELIKAGAIAK	1. Artoa, arroza, artatxikia, artobeltza edo suge-ilarra, kinoa, amarantoa, algarroboa, tapioka, manioka,batata, azenarioa eta patata. Zerealen eratorriak diren GGPak. 2. Barazki eta ortuari natural guztiak. 3. Fruta guztiak eta fruitu lehor gordinak. 4. Esnea eta esnekiak: gazta, gaztanbera esne-gaina, jogurt naturalak eta mamia. 5. Era guztietako haragi eta errai freskoak, zezina, urdaiazpiko ondua. Arraina eta itsaskia: freskoa eta izoztua (arrautza-irinetan pasatu gabea), kontserban, naturalean edo oliotan. Arrautzak, lekale naturalak. 6. Landare-olioak, gurina, margarina, txerri-gantza, ozpina, espezia puruak. 7. Eztia eta azukrea. Freskagarriak, kafea, tea, fruta- eta nektar-zuku naturalak, cava, ardoak, glutenik gabeko garagardo espezifikoak, muztioa.
GLUTENA IZAN DEZAKETEN ELIKAGAIAK	1. Produktu prestatuak (patata frijituak, patata-purea…), arto- edo arroz-irina (oharkabeko kutsaduragatik). 2. Aurrez prestatutako platerak (barazkipurea, menestra…). 3. Fruitu lehorrak eta txigortu gaziak. 4. Kremak, budinak eta flanak, esne-irabiaki prestatuak, esnez egindako azkenburukoak, zaporedun jogurtak eta fruta zatitxoak dituztenak, zorroko mamia, urtzeko gazta, ataletakoa, igurztekoa eta zaporeduna.

	5. Hestebeteak, haragi- eta arrain kontserba saltsadunak, askotariko pateak, urdaiazpiko egosia. 6. Saltsa prestatuak, soja-saltsak, espezia-prestakinak, salda-laukitxoak, legami deshidratatua hautsean edo pikortatua eta legamia-estraktuak, piperrautsa. 7. Txokolatea, gozokiak eta gomazko gozokiak, kakaoa, izozkiak. Txokolate- eta kafe-suzedaneoak, edari konbinatuak, likoreak.

*Oloa zerealaren kasua berezia da; lehen esan bezala, zereal honek ezaugarri desberdinak aurkezten ditu *Triticeae* familiako zerealekin konparatzerakoan. Bibliografian agertzen denez, olo barietate batzuk GZa pairatzen duten pertsona gehienentzat jasangarriak dira, baina ez guztientzat. Gainera, oloaren immunogenizitatea aldakorra da laborantza batetik bestera (52-55). Hortaz, autore batzuk ziurtatzen dute olo barietate batzuk erantzun immunogenikoa sor dezaketela zeliakoak diren pertsonetan (56), beste olo batzuk ziurrak diren arren. Edonola ere, gaixo zeliakoaren ikuspuntutik zereal honetan arreta berezia jartzea beharrezkoa da; glutendun zerealekin batera hazi, gorde, prozesatu eta garraiatu ohi denez, oloaren kutsadura ohikoa baita. Kutsadura gurutzatuaren adibide garbia Thompson eta kolaboratzaileen argitalpenean ikus daiteke (54), non aztertutako gluten gabeko laginen %32 prolamina toxikoan positiboak ziren.

Kontrara, oloarekin egindako okindegi-produktuak organoleptikoki nahiko atseginak dira, beste gluten gabeko zerealekin egindakoetan gertatzen ez den bezala (57), eta zeliakoek, beraz, gehienetan harrera ona egiten diete. Hori guztia kontuan hartuz, Elikadura Kodearen arabera (58), olo jatorrizko produktuetan gluten gabeko ikurra baimentzea ala ez lurralde bakoitzeko erabakia eta ardura da. Espainia mailan indarrean dagoen araudiaren arabera (828 zbk/2014 Araudia) oloa glutena duten zerealen taldean kokatuko da, kutsatuta egoteko arrisku oso altua baitu.

Azpimarratzekoa da, zerealak izan ezik ia gainerako elikagai fresko guztiak gluten gabeak direla. Aldiz, produktu prozesatu ugarik glutena dute edo izan dezakete bere osagaien artean (3. taula). Horren arrazoi nagusiak dira, lehen esan bezala, elikagadura-industrian gari-irinak oso maiz erabiltzen direla eta ekoizpen prozesuan gerta daitekeen kutsadura gurutzatua. Hortaz, jatorriz glutenik ez duten produktu batzuek, hestebeteek adibidez, amaierako produktu prozesatuan eduki dezakete. Horrela, glutenaren nahigabeko kontsumoa ekiditeko ontziratutako produktuen etiketak ondo irakurri behar dira, osagai zerrendari eta glutenaren gabezia ziurtatzen duten ikurrei arreta berezia jarriz.

Hala ere, GZan gluten gabeko elikagaiak kontsumitzeko beharra guztiz onartuta egon arren, ez dago argi hartutako gluten kantitatearen eta sintomen garapenaren eta asaldura histologikoen artean dagoen erlazioa. Hortaz, pertsona zeliakoentzat segurua den ebaki puntua non kokatzen den finkatzea zaila da (59). Gainera glutena dietatik guztiz baztertzea ez da erraza; alde batetik, GGP berezietan kutsadura ekiditea nahiko zaila delako, eta, bestetik, glutendun zerealetan glutena kentzeak zailtasun tekniko eta gastu ekonomiko nabariak dakartzalako. Ikerketa batzuen aburuz, zeliakoen gehiengoak 10 mg/eguneko ingesta onartzen du kalte histologikorik jasan gabe. Beste autore batzuek diote 50 mg/eguneko ingestek heste-mukosaren kaltea eragiten dutela (59, 60).

GZaren eragilea glutena dela XX. mendetik jakin arren, harrigarria da XXI. mendera arte ez dela berrikusi elikagaien segurtasuna eta legeria. Hots, honen guztiaren kudeaketa eta kontrola lehen mailako gaia bihurtu da azken urteotan. Kasu baterako, duela gutxira arte GGPak etiketatzeko erabiltzen ziren jarraibideak Elikadura Kodearen Batzordeak 1979an ezarritakoak ziren. Garai hartan Kodeak ezartzen zuen elikagaiak *"gluten gabeak"* bezala etiketatu ahal izateko, haien gluten edukia ezin zela 200mg/Kg edo ppm baino handiagoa izan (58). 2008an Kodeko txostena berrikusi zen eta Europar Batzordeko elikagaien konposizioa eta etiketatuaren jarraibideetan oinarrituz *"gluten gabea"* (elikagaiak <20ppm/kg duenean) edo *"gluten gutxikoa"* (elikagaiak 21-100 ppm/ kg duenean) adierazpenak ezarri ziren elikagaien gluten maila aitortzeko (41zk./2009ko urtarrilaren 20a Araudia) (61).

Egun, (EB) 41zk./2009ko urtarrilaren 20a Araudia baliogabetua izan da eta glutenaren inguruko adierazpenak (EB) 828 zbk/2014ko uztailaren 30a Araudiak biltzen ditu (62). Arau bilduma horrek glutenarekiko sentikortasun maila pertsona batetik bestera aldatzen dela azaltzen du eta, hortaz, elikagaietarako gluten kopuru baxuko etiketatu desberdinak posible direla (betiere segurtasunaren barne). Horrela, arau berri horrek baimentzen du elikagaien etiketan hurrengo adierazpenak jartzea: *"egokia glutenarekiko intolerantzia duten pertsonentzat"* edo "*egokia zeliakoentzat"* eta baita *"gluten gabea"* (elikagaiak <20ppm/kg duenean) edo *"gluten oso gutxi"* (elikagaiak 21-100 ppm/ kg duenean) ere. Halere, kontuan hartuz bakoitzaren glutenarekiko sentikortasun maila jakitea ez dela ohikoena, gomendio orokorra da gluten gabeko ikurra duten produktuak kontsumitzea (63). Aurrekoez gain, (EB) 1169 zbk/2011ko urriaren 25a Araudia ere aipagarri da (64), bertan elikagaiei buruz kontsumitzaileari ailegatu behar zaion informazioa ezartzen baita, bai ontziratutak dauden produktuetan bai ontziratu gabe saltzen direnetan.

Egun, elikagaien gluten edukia adierazteko merkatuan ikur desberdinak uztartzen dira, batzuk erakunde ofizialek ezarritakoak dira, hala nola, Espainiako Zeliako Elkarteen Federazioko (FACE) ikurra edo Europako Zeliako Elkarteen Federazioko (AOECS) ikurra (3. irudia), eta beste batzuk elikagaien markek sortutakoak. Azken horiek ez dute inongo berme ofizialarik. FACE eta AOECSeko ikurrak lege espezifikoetan biltzen ez badira ere, kalitate bermeen barnean aurkitzen dira, beren araudi propioak izanik. AOECSeko ikurrak elikagaiak <20ppm/kg gluten edukia duela adierazten du (legeriak finkatzen duena gluten gabeko elikagaia dela adierazteko hain zuzen, (62) eta FACEkoak, berriz, <10ppm/kg. Halere, azken ikur hori 2019ko abenduaren 31an desagertuko da, izan ere 2020. urtetik aurrera ikur guztien esanahia bateratuko da.

3. irudia. GGPetan gluten gabezia bermatzeko ikurrak.

Oro har, merkatuan dauden eta pertsona zeliakoentzat bereziak diren GGPek sarritan gluten hondarrak dituzte (62). Are gehiago, “gluten gabeko” adierazpenak gluten proteinaren gabezia totalari erreferentzia egin beharrean, pertsona zeliakoen osasunarentzat kaltegarria ez den gluten mailari (nahiz eta produktu horren ingesta mugagabea izan) dagokio (59). Izan ere, prolamina toxikoa elikagaitik guztiz ezabatzea ez da lan erraza. Dena den, azken urteetan industriak asko aurreratu du arlo honetan; Bustamante *et al*.-ek ikusi zuten 1998 eta 2016 urteen bitartean aztertutako GGPen 3.141 zereal laginetan gluten edukia murrizteko tendentzia bazegoela eta, are gehiago, Europear Araudiak eraginkorrak zirela ondorioztatu zuten (63).

3.1 Glutenaren detekzioa elikagaietan:

Elikagaietan gluten presentzia antzemateko laborategi-teknika ezberdinak saiatu dira urteetan zehar. Azpimarratu behar da glutenarekiko sentikorrak diren pertsonak babestu ahal izateko, ezinbestekoa dela proteina detektatu eta kuantifikatzeko erabiliko diren teknikak zehatzak eta fidagarriak izatea. HPLCa, Malditoff-a edota PCRa erabiltzen badira ere, teknika immunokimikoak garatu dira gehien; horren isla Codex Alimentarius-ek (Kodea-k) horiek gomendatzen dituela da. 80. hamarkadatik aurrera prolaminen edota glutelinen kontrako antigorputz mono eta poliklonaletan oinarritutako analisi-teknikak ugaritu izan dira.

Horrela, gaur egun Kodearen lehen aukeran gomendatzen duen metodo analitikoa teknika immulogikoa da, R5 antigorputzean oinarritutako ELISA (entzimei lotutako immunoxurgatze entsegua) hain zuzen ere. Horrek, gariko gliadina, zekaleko sekalina eta garagarreko hordeina aldi berean detektatzea ahalmentzen du. Egun, antigorputz monoklonalak ohikoenak dira, hau da, aminoazido sekuentzia bakar bat ezagutzeko gai direnak. Nahiz eta antigorputz asko garatu diren, ELISA teknikarako gutxi batzuk bakarrik arrakastatsu probatu eta balioetsi izan dira AOAC jarraibideen arabera.

Hala, aipatutako R5eaz aparte, badira beste antigorputz batzuk, Nazioarteko AOACk eskatzen dituen betebeharrak asetzen dituztenak (4.taula), besteak beste: *Skerritt*, G12 eta A1 antigorputzak.

4. taula. ELISA (Sandwich eta lehiakorra) teknikarako erabiltzen diren antigorputzak.

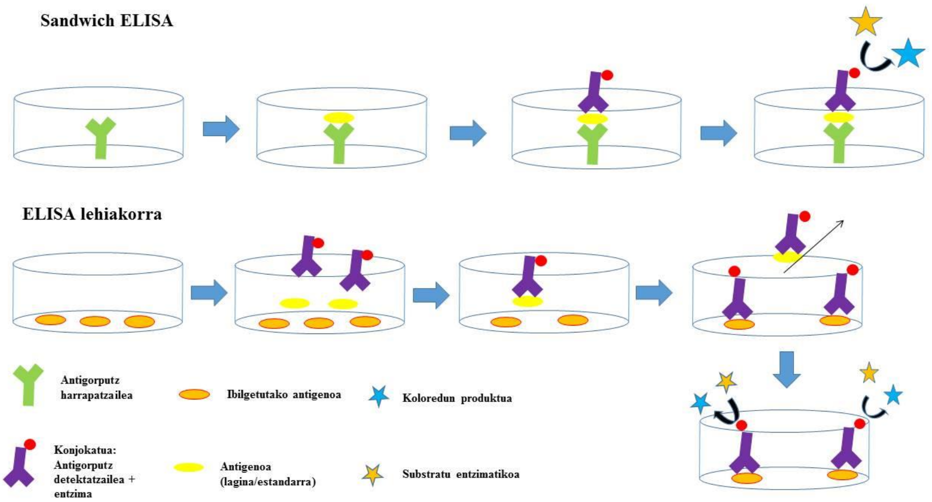
	Frakzio peptidiko ezaguna	**Nazioarteko AOACko Analisi metodo ofiziala**	**Bibliografia**
401.201 Antigorputza edo *Skerrit*	*HMW- GS* (ω-gliadinak)	991.19 Sandwich	***Skerrit & Hill, AOAC 1991***
R5 Antigorputza	QQPFP (ω-sekalinak)	2012.01 Sandwich 2015.05 Lehiakorra 2015.16 Tira imunokromatikoak	***Méndez et al, Eur J Gastroenterol Hepatol 2005***
G12 eta A1 Antigorputza	QPQLPY & QLPYPQP (α2-gliadinak)	2014.03 G12 Sandwich	***Morón et al, Plos One 2008***

Teknika immunologikoak prolaminen zati toxikoa (petido immunogenikoa) hauteman eta kuantifikatzeko diseñatuta daude eta, zentzu horretan, ezinbestekoa da jakitea zein epitopo edo aminoazido-sekuentziaren kontra jotzen duen erabilitako antigorputzak (4. taula). Kontuan hartu behar da industriak elikagaiak ekoizpenean batzuetan zerealen proteinak eraldatzen dituela, hainbat prozesuen bidez, esaterako, deamidazioa, transamidazioa edota hidrolisia eraginez. Ondorioz, aminoazido-sekuentzia horien edukia, kopurua edo egitura alda daiteke, glutenaren detekzioa zailduz (65).

Lehen esan bezala, ELISA aukerazko detekzio metodoa da; alde batetik ezaugarri ezinhobeak dituelako horretarako, zehaztasuna, sentikortasuna eta errepikakortasuna, alegia, eta, bestetik, beste erreferentzia metodo independienterik ez dagoelako. Gainera, emaitzak azkar ematen ditu eta, beste teknika molekularrekin konparatuz, merkea da (entsegu hau aurrera eramateko laborategiko ekipamendu sinplea izatea nahikoa da).

ELISA teknika bi era nagusitan gara daiteke laborategian: sandwich metodoa edo metodo lehiakorra jarraituz. Sandwich ELISAk (4.irudia) prolaminak, teknikarako antigeno direnak, bi antigorputz geruzen artean kuantifikatzen ditu. “Harrapatzaile” diren antigorputzak mikroplakaren zorura atxikituta daude eta, horrela, prolaminaren frakzio toxikoak lotzen zaienean antigenoa atxikituta mantenduko dute. Geroago, entzima batera konjokatuta dagoen bigarren antigorputza, “detektatzailea” dena, gaineratzen da kikaretan. Hori, prolaminaren beste gune bat ezagututa, antigorputz harrapatzaile-antigeno konplexura lotzen da. Hortaz, teknika honen bidez prolamina kopurua neurtu ahal izateko, proteinak gutxienez bi epitopo antigeniko oso eduki behar ditu (era berean bi antigorputzetara, harrapatzailera eta detektatzailera, lotu ahal izateko). Normalean konjokatuta erabiltzen diren entzimak errefau-peroxidasa eta fosfatasa alkalinoa dira, beraien helburua amaieran erantsiko den kromogenoarekin erreakzionatu (substratua) eta kolorea sortzea da (produktua), metodo espektrofotometrikoen bitartez neurtuko dena. Hortaz, absorbantzia lagineko prolamina kantitaterekiko proportzionala izango da.

ELISA lehiakorra (4. irudia) berriz, lagineko prolaminaren eta metodoan eransten den prolamina estandarren artean gertatzen den lehian oinarritzen da. Lagina (detekatu nahi den prolamina izan dezakeena) gliadinarekin tapizatutako kikareetan erantsi egiten da. Ostean, konjokatua gaineratu egiten zaio, R5 antigorputz monoklonalari konjokatutako peroxidasa alegia. Momentu horretan lagineko prolamina librea eta kikaran ibilgetutako gliadina lehian sartzen dira, antigorputzaren lotura-guneetan lotzeko. Gero, kikaran lotu ez den konjokatua garbitu ondoren, kromogenoa gaineratuko da, entzima-konjokatuak erreakziona dezan eta produktu koloredun bat sor dadin, espektrofomotria bidez neurtuko dena. Hortaz, absorbantzia lagineko prolamina kantitaterekiko alderantziz proportzionala izango da. Metodo honek antigorputz bakarra darabil, hots, epitopo bakarra detektatuz dihardu. Horrek ahalbidetzen dio metodoari egitura osoa mantentzen ez duten prolaminen detekzioa, kasu horietan aukerazko teknika izanik, adibidez, proteina hidrolizatuen peptido txikiak antzematea. Hala ere, baliteke sandwich ELISAk bezain besteko espezifitatea ez izatea, detekziorako antigeno-antigorputz lotura puntu bakarrean gertatzen baita.



4. irudia. ELISA teknika immunologikoaren oinarria.

ELISA metodoak gehienetan gliadinak edo hordeinak darabiltza erreferentzia estandar gisa, beraz, ez ditu aintzat hartuko glutelina erako balizko proteina toxikoak (66). Are gehiago, glutendun zerealen proteinan gliadina eta glutelina proportzioa 1:1 dela kontuan hartuz, ELISA teknikan prolaminatik abiatuz elikagaian dagoen gluten eduki totala estimatzeko x2 bihurtze faktorea erabiltzea onartzen ohi da (67).

4. GLUTEN GABEKO DIETA

GGDk naturalki glutenik ez duten elikagaiek edo/eta espezifikoki ekoiztutako GGPek osatuko dute. Dietak gehienetan heste meharreko lesioen hobekuntza esanguratsua, serologiaren normalizazioa eta sintomen desagerpena dakar, pertsona zeliakoen ongizatea bermatuz. Baina, GGD zorrotza eta fidagarria mantentzea erronka handia da pertsona zeliakoentzat (68), glutena elikagai askotan agertzen baita, 3. taulako sailkapenean behatzen den moduan. Hasteko, berez glutenik ez duten zerealetan (arroza, artoa, artatxikia, artobeltza eta abarretan adibidez) kutsadura gurutzatua gerta daiteke, bai laborantza momentuan, bai elikagaiak prozesatzerakoan. Gainera, ez da arraroa gluten iturri ezkutuak diren elikagaiak ohiko elikaduraren parte izatea; izan ere, elikadura-industrian proteina honen erabilera oso hedatuta baitago, askotan gehigarri gisa erabiltzen baita (lodigarri, zapore-indartzaile, emultsionatzaile gisa esaterako) (63). Bestalde, ez da ahaztu behar GGDk glutenaren absentzia ziurtatzeaz gain, nutrizionalki orekatua izan behar duela, beharrizan energetiko eta nutrizionalak betetzeko. Hori dela eta, ikerlariek urteak daramatzate GGDk pertsona zeliakoetan epe luzera eduki dezakeen eragina ikasten, behatu denez batzuetan nutrieeten desoreka edota gabeziak sor baititzake (68).

Pertsona zeliakoak GGDri guztiz atxikitzea ezinbestekoa da; ez bakarrik arestian aipatutako heste-mintza osatzeko, sintoma klinikoak arintzeko eta horrek dakarren bizi-kalitatea hobetzeko (69), baita etorkizuneko osasun-zailtasunak prebenitzeko ere, besteak beste anemia, osteoporosia, heste meharreko linfoma, urritasun nutrizionalak eta asaldura autoimmuneen garapena, diabetes mellitusa edo hipotiroidismoa adibidez. (70-72).

Atxikimendu egokia lortzekotan, bi asteetako GGD jarraitu ondoren gehienetan pazientearen sintoma klinikoak arintzen dira (73, 74) eta antigorputzen odol-maila esanguratsuki jaisten da. Are gehiago, antigorputzen maila ez normaltzea transgresio dietetikoak detektatzeko markatzaile da askotan (36). Kalte histologikoak sendatzeko, berriz, denbora gehiago behar da. GGDri egoki atxikitzen zaizkion heldu zeliakoen %65ak bi urte behar ditu heste-meharraren mintza guztiz osatzeko (75-77). Umeen kasuan GGDren eragina azkarragoa izan ohi da eta %88-96ak kalte histologikoa bi urteko epealdian guztiz berreskuratzen du (77, 78), eta bost urteko epealdian %100ak (77).

4.1 GGD heldu zeliakoetan: nutrizio-egoera eta ohitura dietetikoak.

4.1.1 Diagnostikoaren uneko nutrizio-egoera

Gaixotasunaren diagnostikoa egiten den momentuan, zeliakoak malnutrizioa pairatzeko arriskuan daude, heste meharrean dagoen kaltearen eraginez mikronutrienteen malabsortzioa gertatzen baita. Zenbait ikerketen aburuz, tratatu gabeko gaixoetan urritasun nutrizionalak arruntak dira (bitamina eta mineral maila plasmatiko baxuak ikusi dira). Burdina eta kobrea, A, D, E eta B taldeko bitaminekin batera, eskasia gehien erakusten duten mikronutrienteak dira diagnostikatu berri diren pertsonetan (79, 80). Hortaz, diagnostikoa egiten den momentuan garrantzitsua da pertsonaren nutrizio-egoera aztertu eta ebaluatzea, gabezi nutrizionalen ondorioz gerta daitezkeen patologiak zaintzeko: erretinopatia (A bitaminaren gabezia), neuropatia periferiko sistemikoa (B_{12} eta E bitaminen gabezia), haurdunaldirako zailtasunak (burdina eta azido folikoaren gabezia), hortzeriaren asaldurak, osteopenia edo osteoporosia (D bitaminaren gabezia). B_{12} bitaminaren gabezia ohikoa bada ere, anemia ferropenikoa da zeliakoengan arruntena, hestearen egitura berreskuratu bitartean behintzat (79, 81). Gainera, GZa laktasa entzimaren urritasunarekin erlazionatu izan da, eta, horren ondorioz, pertsona zeliakoek laktosarekiko intolerantzia izaten dute maiz diagnostikoaren unean (81). Era berean, diagnostikatu berri diren zeliakoen egoera nutrizionala neurri antropometrikoen bitartez aztertu denean, heldu zeliakoek, heldu osasuntsuek baino pisu, gorputz masa indizea (GMI), gantz-masa (GM) eta ihar-masa (IM) baxuagoak erakutsi dituzte (82-86).

4.1.2 GGD jarraitu bitartean

Diagnostikatu berri diren zeliakoen malnutrizioa ondo ikasi bada ere, zalantzan dago zer nolako eboluzioa duten haien nutrizio-egoerak eta gorputz-osaerak GGD epe luzez jarraituz gero (87). Pentsa daiteke diagnostikoaren aurretik gertatzen den hesteetako atrofia GGD eginez sendatuko dela eta, horrekin batera, bitamina eta mineralen absortzioa hobetuko dela. Izan ere, ikusi da diagnostiko ostean tratamenduarekin hasten diren pertsonek sintomen hobekuntza nabaritzen dutela. Hala ere, berriki egindako ikerketek adierazten dutenez, tratamendu aurreko gabezia nutrizionalak ez dira guztiz zuzentzen GGD ezarritakoan (82, 83, 86-88). Pertsona bakoitzaren osatze maila hainbat faktoreren menpe dago: adina diagnostikoaren momentuan, GZ diagnostikatu gabe egon den denbora tartea eta beste osasun faktore batzuk, besteak beste. Horregatik, GGD jarraitzen duten pertsonen nutrizio-gabezien identifikazio goiztiarra funtsezkoa izango da etorkizuneko balizko arazoak prebenitzeko eta zeliakoen bizi-kalitatea hobetzeko (81).

Esate baterako, Capristo *et al*-ek GGD jarraitu ostean mintz duodenalean hobekuntza zuten zeliakoen *vs* kontrolen aldagai metabolikoak konparatu zituzten. Ikusi zutenez, hemoglobina, hematokritoa, burdina eta transferrina gazur-kontzentrazioak baxuagoak ziren pertsona zeliako guztietan (bai diagnostikatu berri ziren pertsonetan eta baita tratamenduan zeudenetan ere) (88). Bere aldetik, Hallert *et al*.-ek (89) homozisteina plasmatiko totala, folatoa, piridoxal-6-fosfatoa (B_{6}- bitaminaren forma aktiboa) eta B_{12} bitamina neurtu zituzten 8-12 urte bitarteko GGD jarraitzen zuten 30 heldu zeliakoetan, kontrolekin aldaratuz. Zeliakoek, homozisteina plasmatiko maila altuagoa erakutsi zuten, egoera bitaminiko kaskarra adierazi, eta haietatik %47ak B_6 bitamina edota folato maila baxuak aurkeztu zituen; B_{12} bitaminaren gabezia, aldiz, ez zen agertu. Beraz, pertsona zeliakoen jarraipenean bitaminen oreka maiz berrikusi behar dela ondorioztatu zuten. Hala ere, eztabaida dago gabezi nutrizionalak zuzentzearen inguruan; izan ere, badira beste ikerlari batzuk honekin bat ez datozela (71, 90) eta mikronurienteen gabeziak GGDren hasturekin justifikatzen dituztenak.

Era berean, neurri antropometrikoetan, aipagarria da autore guztiek ez dutela adostasunik lortzen GGDren ondorengo nutrizio-egoeraren hobekuntzari dagokionez. Batzuk GGD zorrotza jarraitzen duten zeliakoek GMI, IM eta GM portzentaje egokiak berreskuratzen dutela baieztatzen duten bitartean (37, 91, 92), beste batzuk behatu dute GGD dieta ez-orekatua ezarteak pisu irabazi osasungaitzera eraman ditzakela. GGD errespetatuz heste-mintza osatzen da eta, ondorioz, nutrienteen xurgapena areagotzen da, GGDren desorekak pisu irabazte ez desiragarria ekarriz (93-95).

Zeliakoen dietaren nutrizio-konposaketaz ari garenean, ikusi da GGD ez dela orekatua energia ingesta, makronutrienteen banaketa eta mikronutrienteen beharrei dagokienez (96-98). Oro har, energia ingesta altua, energiaren gantz eta proteina portzentaje handiegia eta karbohidrato portzentaje baxuegia antzematen dira. Ikerketa askotan frogatu den moduan, GGD zuntzatan eskasa da glutendun dietarekin konparatzen denean (89-90).

Mikronutriente ingestari dagokionez antzera gertatzen da; ikerketa gehienetan bitamina eta mineralen gabeziak aurkitu dira GGD glutendun dietarekin erkatzerakoan. Zehazki, honako mikronutrienteen gabeziak deskribatu dira: D bitamina, E bitamina, Azido folikoa, B_1 bitamina, B_2 bitamina, B_6 bitamina, Mg, Fe, Ca, I, Se (79, 80).

Heldu zeliakoen ohitura dietetikoei dagokienez, SENCeko elikagaien taldeko kontsumo gomendioei atxikidura urria ikusten da. Askotan GGD jarraitzeak erronka dietetikoa bihurtzen da eta glutena ekiditeko asmoz zeliakoek bere dietan zerealen taldeko elikagai gutxi hartzen dituzte. Bestalde, horiek ordezkatzeko animalia jatorriko elikagaien kontsumoa areagotzen da, gantz aseen kontsumoa handituz. Biztanleri orokorrean gertatzen den bezala kolektibo honetan ere fruta eta barazkien kontsumoa baxua da eta ez da gomendatutako ingesta betetzen.

4.2. GGD ume eta nerabe zeliakoetan: nutrizio-egoera eta ohitura dietetikoak

Umeen artean GZa da elikagaiekin erlazionatuta dagoen gaixotasun kroniko arruntena. Europar umeen artean zeliakiaren intzidentzia %1 inguruan kokatzen da (cita). Gainera, kopurua etengabe handitzen ari da eta oraindik diagnostikatu gabe dagoen portzentajea esanguratsua da. Suediar *screening* ikerketa batek aipatu zuen prebalentzia %3 dela 12-14 urteko nerabeen artean (99).

Lehen esan bezala, nabarmendu beharra dago zuzena den GGDk segurua izan ez ezik orekatua ere izan behar duela, beharrizan energetiko eta nutrizionalak betetzeko. Betekizun horrek haurren eta nerabeen kasuan are garrantzia handiagoa dauka, dietak hazkuntza prozesu osasuntsua bermatu beharko baitu, etorkizuneko urritasunak ekidinez eta bizitza osasuntsua sustatuz. Sarritan ume zeliakoek pisu, altuera eta GMI baxuagoa dute bere adin eta sexu berdineko umeek baino. Besteetan berriz, autore batzuk deskribatzen dute GGD eramateak desiragarria ez den pisua irabaztera eramaten dituela, dieta honen desoreka nutrizionala dela eta (68, 93, 95).

Helduen antzera, ikerketa ugarik ume zeliakoen ingesta dietetikoa desorekatua dela behatu dute: energi ingesta murriztua (96), karbohidrato konplexu, gantz asegabe eta zuntz kontsumo txikia (93,97,98) eta lipido (gantz total eta ase) eta proteina ingesta altua (96,98,100). Halaber, ume eta nerabe zeliakoek jarraitzen duten GGD biztanleri orokorraren dietarekin aldaratzerakoan, bien kasuan patroi dietetikoa desorekatua egon arren, desorekaren jatorria desberdina dela (101,102).

Umeen GGDk mikronutriente batzuen urritasuna ere ekari dezake, hala nola, D, A, K, E eta B12 bitaminak, azido folikoa, zinka eta burdina nutrienteena (100-104). Gabezia larrienak diagnostikatu gabeko kasuekin erlazionatzen da, GGD hasi aurretik, heste-biloen atrofiaren eraginez zeliakoek pairatzen duten malabsortzioagatik. Baina, autore batzuen iritsiz, ume eta heldu zeliakoen mikronutrienteen urritasunak GGD jarraituta ere dirau (68, 97, 105). Hortaz, GGD ezarrita, balizko gabezia horiek zorrotz zaindu behar dira haur hauengan.

Ume eta nerabe zeliakoen ohitura dietetikoei dagokienez, elikagai-taldekako kontsumoan atxikidura eskasa ikusten zaie Mediterranear piramidearen jarraibideei. Zeliakoek zerealen taldeko elikagai gutxi hartzen dituzte glutena ekiditeko. Babiok eta bere taldeak ikusi zuten zeliakoek kontsumitzen zutela almidoi gutxiago (pasta, ogia, opilak) eta proteinadun elikagai gehiago (haragia, arraina, arrautza) biztanleri orokorrak baino. Gainera, ume zeliakoek ez dute betetzen zuntz iturriak diren elikagaien kontsumoaren gomendioa, esate baterako, lekale, barazki edo frutaren gomendioa.

5.taula. Ohiko akats nutrizionalak GGD jarritzen duten heldu, ume eta nerabeetan. (68, 88, 93, 100-102, 104-111)

	Helduak		**Umeak eta nerabeak**	
	Gabeziak	**Gehiegizko ingesta**	**Gabeziak**	**Gehiegizko ingesta**
Energia	Energia		Energia	
Makronutrienteak	KH konplexuak	Proteinak Gantz totalak	KH konplexuak GAAG	KH sinpleak Gantz totalak GAA
Beste osagaiak	Zuntza		Zuntza	
Bitaminak	D, E, Azido folikoa, B1, B2, B6		D, C, Tiamina	
Mineralak	Mg, Fe, Ca, I, Se		Mg, Fe, Ca, Se	

Notak: KH: karbohidratoak, GAAG: gantz azido asegabeak, GAA: gantz azido aseak, Mg: magnesioa, Fe: burdina, Ca: kaltzioa, Se: selenioa

4.3. GGDren gabeziak: zergatia?

Oro har, GGD jarraitzen duten zeliakoen nutrizio-egoeraren eta ohitura dietetikoen inguruan dagoen bibliografiak adierazten duenez, zeliakoek ez dute beti lortzen segurua, askotarikoa eta orekatua den elikadura eta ohitura dietetiko egokiak ezartzea. Horrek beraien egoera nutrizionala, osasuna eta bizitza-kalitatea kolokan jar dezake.

GGD aztertzean adostasuna dago makronutrienteen proportzioan desoreka dagoela eta zenbait mikronutrienteren gabezia agertzen dela baieztatzean. Hori kausa desberdinen ondorioa izan daiteke. Alde batetik, GGD zorrotza jarraitzen duten pertsonek glutendun zerealak baztertu behar dituzte beren elikaduratik, baita horietatik eratorritako elikagai guztiak ere: ogia, pasta, irinak, etab. Horrek dieta orekatua jarraitzea zaildu egiten du, izan ere, elikadura piramideko lehen mailan kokatutako elikagai taldea da, maiz kontsumitu beharrekoa. Gainera, elikagai horiek gure dietan energia eta zuntza iturri garrantzitsuak izateaz aparte, gariak, zekaleak eta garagarrak ume osasuntsuen zuntza hartualdiaren %35 eskaintzen dute (112) eta burdina, zinc, magnesioa eta B bitaminen taldean aberatsak dira (113, 114).

Beste aldetik, pertsona zeliakoek GGP finduak kontsumitzera jotzen dute. Produktu horien nutrizio-osaera aztertzen dituzten ikerketek diferentzia esanguratsuak aurkitu dituzte GGPak haien glutendun homologoekin konparatzean, besteak beste, kaloria kopuruan, makronutriente edukian, dota zuntza, sodioa, gantz eta kolesterol kantitatean (115). Lehen esan bezala, glutenak ezaugarri berezi eta organoleptikoki aproposak ematen dizkie glutendun zerealekin egindako produktuei. Beraz GGPen formulazio berrien helburu nagusietako bat bukaerako produktuaren ezaugarriak hobetzea izango da; horretarako egiten diren aldaketek produktuaren nutrizio-osaera okertu badezakete ere

Horren adibidea da sarritan GGPei gantzetan aberatsak diren osagaiak, hala nola, animalia- edo landare-olioak edo/eta emultsionatzaileak (gantz azidoen mono eta diglizeridoak) gaineratzen zaizkiela beraien palatabilitatea eta ehundura hobetzeko asmoz. Gainera, produktu horien eduki proteikoa txikiagoa izan ohi da almidoien, proteina baxuko zerealen edo goma eta entzimen erabilera dela eta (116). Bere aldetik, zuntzaren kontsumoaren inguruan dauden datuek erakusten dute zeliakoen artean zuntzaren gomendioak betetzen ez direla (117). Hori bi arrazoik azaltzen dute. Lehengoa, glutendun zerealek gomendatutako zuntza osoaren heren bat eskaintzen dutela (112) eta horiek ekiditean zuntza iturri bat galtzen dela (68). Bigarrena, GGP finduen konposizioa dela eta, zuntza kopuru gutxiago dute glutendun eta findu gabeko analogoek baino, ekoizpen prozesuan, haziaren azala erauzten delako (zuntzatan aberatsa dena) (68, 115).

Bitamina eta mineralei dagokienez, ikerketek diotenez GGDk mikronutrienteetan gabeziak aurkeztu ditzazke (9, 97, 105, 118). Halaber, maiz zeliakoek diagnostikoa jasotzen duten momentuan mikronutrienteen gomendio nutrizionalak ez dituzte betetzen, heste-mintza eta xurgapena minduta izatearen ondorioz. Gabezia horiei guztiei aurre egiteko, oso garrantzitsua da GGPen mikronutriente edukia ezagutzea. Apagarri da, alde batetik, produktuok ez dutela zertan informazio hau etiketan agertu, eta bestetik, GGPetan ez dela gluten gabeko elikagaiak gotortzeko politika ohitura berdina jarraitzen. Horrek guztiak agerian jartzen du elikagai horiekin lan asko egiteke dagoela, ez bakarrik ikerketa alorrean, baita elikadura-industria eta administrazio mailan ere.

Historikoki pertsona zeliakoei emandako nutrizio-aholkua gluten absentzira bideratu izan da, dietaren oreka nutrizionala ahaztu egin delarik. Aitzitik, kontuan hartu behar da GGPen kalitate nutrizionala egokia izatea oso garrantzitsua dela(115), are gehiago zenbait kolektibotan egun osoko energiaren %36,6 GGPetatik datorrela ikusi denean (101). Hala ere, zeliakoen tratamenduan naturalki glutenik ez duten elikagaien kontsumoa sustatu behar da. Ildo horretatik, gluten gabeko zerealen eta pseudozerealen kontsumoa areagotzen ari da azkenaldi honetan, karbohidrato eta proteina iturri ona izateaz gain bitamina eta mineralena ere badirelako (97); gainera nahiz eta pseudozerealen gantz edukia altua izan, glutendun zerealekin aldaratzen dugunean duten gantz azido mota hobea da, asegabeak izanik (97). Oloak eztabaida sortzen duen arren, autore batzuen aburuz zereale hau zeliakoen dietan sartzeak ez du eragin kaltegarririk, ez helduetan (119, 120) ezta umeetan ere (121, 122). Are gehiago, pertsona zeliakoei galdetu zaienean, bere dietan oloa hartzea aukeratu dute (122). Izan ere, onuragarriak diren hainbat ezaugarri esleitu zaizkio zereal honi, hala nola, kaltzio, fosforo, potasio eta zink eduki altuak eta gantz azido asegabeen, folatoen eta tiamina bitaminen ekarpen handia. Azpimarratzekoa da ere duen zuntza kopuru handia, beta-glukanoak nagusiki.

Egun, jakina da populazio orokorrak ere ohitura dietetiko desegokiak jarraitzen dituela eta horrek dietaren desoreken agerraldia eragiten duela. Hortaz, pentsa daiteke, pertsona zeliakoek printzipioz egoera konplexuago batean aurkitzen direnak ere dieta desorekatua jarraituko dutela. Egoera ikusita, berebiziko garrantzia du zeliakoek jarraitzen duten GGD aztertzeak. Agertzen diren akatsak identifikatu eta horien jatorria zehaztu behar dira, horrela bakarrik ahalko baitzaie aurre egiten, konponbide espezifiko eta eraginkorrak bilatuz. Hala, hizpide dugun kolektibo honen nutrizio-egoera hobetzearekin batera, beren osasuna eta bizitza-kaliteta hobetu ahalko litzateke.

4.4. GGDrekiko atxikimendua:

Lehen esan bezala, GGDren jarraipen zorrotza aukera bakarra da zeliakoaren osasun egoera hobetzeko eta etorkizunean ager dakizkioken albo ondorioak ekiditeko. Baina, tamalez, pertsona zeliakoentzat ez da erreza izaten GGD betetzea eta aukera dietetiko egokiak egitea (68). Aurreko ohitura okerrak baztertzea eta bizitza osorako ohitura dietetiko berriak ezartzea, zorrotzak direnak, eta mantentzea pertsona askorentzat erronka handia da (90). Are gehiago, badira prozesuan eragina duten beste hainbat faktore: GGPen prezio altuak (123), aisialdian glutenik gabeko espazio seguruak aurkitzea, nahigabeko transgresioak, estigma, etab. Aurreko guztiak sarritan eragiten du dietaren jarraipena desegokia izatea (124) eta hori sahiesteko ezinbestekoa da zeliakoek eta bere ingurukoek GGDri buruzko hezkuntza jasotzea, baita dieta ez jarraitzearen ondorioak ondo ezagutzea ere, bere garrantziaz jabetzeko.

Praktika klinikoan jasotako datuen arabera, paziente zeliakoen %20-80 partzialki atxikitzen zaio GGDri. Nabaria da bereziki *screening*en bitartez diagnostikatu izan diren pertsonen kasua (90, 125), non gehienek, asintomatikoak izanik, askotan ez dituzten GGD jarraitzearen abantailak ikusten (126). Beste autore batzuk, berriz, atxikimendu maila altuagoak aurkitu dituzte; adibidez Kurppa *et al*.-ek %88ko atxikimendua behatu zuten, batezbeste 7 urtez GGD jarraitu zuen ume eta heldu zeliako talde baten ohitura dietetikoak aztertzean. Ikerketa horretan dietarekiko atxikimendu egokia galdetegi dietetiko eta azterketa serologikoen bitartez neurtu zuten. Gertatu ohi den moduan, ez zuten aurkitu GGDrekiko atxikidurarekin erlazionatzen den faktorerik (generoa (127, 128), aurkezpen klinikoa (129, 130), komorbilitateen presentzia (129), egoera psikologikoa (129), familia-historia, etab.). Aitzitik, bai ohartu ziren aldizkako urratzeak egiten zituzten pertsonen adina, edo/eta diagnostiko momentuan zuten adina, baxuagoa zela; eta hortaz, talde horretako pertsonak sintoma kliniko gehien mantentzen zituztenak ziren (131). Halaber, Fabiani *et al*.-ek (125) ikusi zuten nerabezaroan dietarekiko atxikimendu eskasa zela eta Ciacci *et al*.ek (132) 20 urte baino gehiagorekin diagnostikatuak izan ziren zeliakoek hobeto jarraitzen zutela GGD. Horren arrazoia heldutasun falta izan daiteke, izan ere nerabezaroa aldaketa askoko denboraldia da (90) eta horrek GGD egokia jarraitzea eragotzi dezake. Halaber, behatu egin da ume helduek sintoma gastrointestinal gutxiago aurkezten dituztela ume txikiagoek baino, eta, hortaz pentsa daiteke deserosotasunik sentitzen ez dutenez garrantzia gutxiago ematen diotela tratamenduari (133). Hala eta guztiz ere, esan bezala, ez dago argi zeintzuk diren dietarekiko atxikimendu onarekin erlazionatzen diren faktoreak ezta zeintzuk izan daitezkeen aurrera eraman daitezkeen interbentzioak zeliakoek beren tratamendua hobeto doitzeko (134).

Zeliakoek GGDri egoki atxikitu ahal izateko azpimarratzeko beste faktore bat da elikagaiek jasan dezaketen kutsadura gurutzatua. Arestian aipatu den moduan, askotan gertatzen da produktuen osagaiak gluten gabeak izanda ere, bukaerako produktuek gluten kopuru ez ziurra izatea, etiketan adieraziko ez dena beti eta, jakina, urratze dietetikoak areagotuko dituena. Halaber, lehen esan bezala, araudiak dio GGPek 20 ppm-ra arteko gluten edukia izan dezaketela; beraz, oso zaila da banakoarentzat egunean zehar hartzen duen gluten kopuru totala kontrolatzea, GGPen kontsumoaren araberakoa izango da.

4.5 GGD jarraitzen duen pertsonaren egoera psikosoziala

Aipagarria da pertsona zeliakoek denbora asko itxaron behar dutela gaixotasunaren diagnostikoa lortzen duten arte. Pelegrí C. *et al*-ek 98 helduekin egindako ikerketan sintomak agertu zitzaizkienetik diagnostikoa jaso zutenera arte 9 urte pasa zirela behatu zuten (135). Ildo berean, Cranney *et al*.-ek (136) eta Rodrigo *et al*.-ek (137) egindako ikerketetan 11,7 eta 10 urte pasatu ziren hurrenez hurren. Egoera horrek pazienteen frustrazioa dakar eta, lehen aipatu den moduan, beste patologia batzuen garapena areagotu daitezke, hala nola, osteoporosia, anemia, depresioa, etab.

Pertsona zeliakoek diagnostikoa jasotzen dutenean ezberdin erantzuten dute. Batzuek lasaitasunez hartzen dute, izan ere, bere egonezinaren kausa eta hortaz konponbidea topatu baitute (138, 139). Beste batzuk, berriz, gainezka sentitzen dira (130). Alde batetik, hasieran zailtasunak topatzen dituzte GGDren jarraibideei egoki atxikitzeko; eta bestetik, askotan gizarteak jartzen dien traba ugariei aurre egin behar die, gogorra egiten zaiena.

Zeliakoek gainditu behar duten arazo nagusietako bat etxetik kanpo jatea da. Izan ere, taberna edo/eta jatetxeetan dagoen GGPen eskaintza ez da oso zabala eta jaten dutena glutenarekin kutsatuta egoteko beldur dira (140). Egoera horrek sozialki baztertutak sentitzera eramaten ditu. Zarkadas *et al*-en ikerketan Canadiar Zeliakoen Elkarteko kideen %80k jatetxeetara joaten ez zela adierazi zuen (141), balizko kutsadura ekiditeko. Ikertalde berdinak beste ikerketa batean ostalaritzak eskaintzen duen GGPen eskaintzari buruz galdetu zuen, parte-hartzaileen %70ak adierazi zuen aukera eskasegien aurrean inpotentzia sentitzen zuela (138).

Etxean bazkaltzen dutenean ere zenbait arazo topatzen dituzte gluten gabeko elikagaiak hautatzeko. Pelegrí C *et al*.-ek (142) egindako ikerketan parte-hartzaileen %93ak elikagaien etiketak ulertzea zaila zela adierazi zuen, baita glutendun ala gluten gabeko elikagaiak ziren zehaztea ere. Ildo berean, Casellas *et al*-ek (143) behatu zuten beren ikerketako parte-hartzaileen %88,5ek aurkitzen zuela nolabahiteko zailtasuna etiketak interpretatzerako orduan.

Faktore horiek guztiak direla eta, autore askok ondorioztatu dute GGD jarraitzeak zeliakoaren bizi-kalitatea murrizten duela, (138, 144, 145) nahiko paradoxikoa dena. Hots, tratamenduak osasuna sustatuko du, eta horrek teorikoki pertsona horien bizitza-kalitatea hobetu beharko luke; baina, GGD bera erronka gogorra da pertsona hauentzat, haien sozializazioa oztopatzen baitu eta, hartara, bizitza-kalitatea, maila horretan, kaltetzen baitu.

HELBURUA ETA DISEINU ESPERIMENTALA

HELBURUA ETA DISEINU ESPERIMENTALA

GZ oinarri immunologikoa duen patologia da, gluten ingestak sortzen du aurrejoera genetikoa duten pertsonengan eta iraunkorra da, hots, bizitza osoan zehar mantentzen da.. Europa mailan hesteetako gaixotasun kroniko ohikoenetako bat da. Egun dagoen tratamendu bakarra GGD zorrotza jarraitzea izanik, proteinaren absentzia bermatzea zeliakoentzat ezinbestekoa da, bai ekoizten diren produktu bereziak kontrolatuz eta bai ohiko elikagaien balizko kutsadura gurutzatua ekidinez. Izan ere, gluten ingestio txikienek ere kalte egin diezaiekete; adibidez, Catassi *et al*.-ek (60) erakutsi zuten hiru hilabetez 50mg/egun gluten hartzeak pertsona zeliakoaren heste mukosa kaltetu dezakela. Testuinguru honetan, 20 mg/kg-ko muga gainditzen ez duen GGDk hartualdi ziurra bermatuko du. Are gehiago, GGD segurua izateaz gain orekatua ere izan behar da, hots, norbanakoaren beharrizan energetiko eta nutrizionalak bete behar ditu gabeziak saihesteko.

Gure ikertaldeak lantzen duen ildo nagusi bat populazio zeliakoaren nutrizio egoerari buruzkoa da hain zuzen ere, bere ohitura dietetikoak aztertu eta nutrizio-ebaluazioa egitea alegia. Autore ugarik ikusi dutenez, zeliakoek glutena ekiditeko asmoz dietan zerealen kontsumoa gehiegi murriztu ohi dute, eta ondorioz, karbohidrato gutxiegiko dieta jarraitzeko arriskua dute (68, 106, 107). Gainera, GGD mikronutriente batzuetan urria izan daiteke (9, 97, 105, 118). Halaber, azken urteotan GGPen merkatua esponentzialki handitzen ari da eta egun enpresa asko ari dira produktu hauek ekoizten. Aitzitik, zerealetatik datozen produktuetatik glutena erauzterakoan zailtasun teknikoak aurkitzen dituzte. Glutenak oreari ematen dizkion antzeko ezaugarri erreologikoak lortzeko asmoz, beste osagai batzuk gehitzen zaizkie produktuei, hala nola, gomak, hidrokoloideak, gantzak edo entzimak (51). GGPetan gehien erabiltzen den osagaia almidoia da (18), oreei biskoelastikotasuna ematen die, aire burbuilak harrapatzen dituelako, gari-oreen antzeko masa lortuz. Halere, ikusi da produktuen birformulazioak GGPen nutrizio-osaera eralda dezakeela (18). Horrela, ikerketa batzuek, gureek barne, erakutsi dutenez, zerealen taldeko GGP espezifikoak eta beren glutendun homologoak nutrizionalki desberdinak dira, bada lehengoek gantz-azido ase gehiago eta zuntz eta proteina gutxiago dute (115, 146, 147), baita mikronutriente eduki txikiagoa ere (tiamina, erriboflabina, niazina, folatoa, D bitamina, kaltzioa edota burdin gutxiago bibliografiaren arabera) (97, 148). Azpimarratzekoa da gantz-ase gehiegiko eta zuntz baxuko dieta gaixotasun kardiobaskularren prebalentzia altuagoarekin erlazionatzen dela. Era berean, zeliakoengan ohikoak dira azido folikoaren edo/eta burdinaren gabeziarekin lotutako anemiak eta kaltzioaren eta D bitaminaren eskasiarekin erlazionatzen den osteoporosia. Ondorioz, ezinbestekoa da, batetik, populazio honen dieta zaintzea, bai atxikimenduari (gluten absentziari) bai nutrizio-kalitateari dagokienez eta, bestetik, GGD osatzen duten elikagaiak eta produktuak (GGP) aztertzea, nutrizio-osaera egokia dutela bermatzeko.

Aurreko guztia kontuan hartuz lan honen helburuak honako hauek izan dira:

A. Euskal Autonomia Erkidegoko (EAEko) populazio zeliakoaren nutrizio-egoera eta elikadura ohiturak aztertzea:

1) EAEko ume eta nerabe zeliakoen GGDren kalitate dietetikoa aztertzea; nutrizio-egoera hobeezina lortzeko dieta orekatua jarraitzea oztopatzen duten faktore nagusiak zehaztea. (**2. eskuizkribua**).

2) EAEko emakume zeliakoen nutrizio-egoera eta jarraitzen duten GGDren nutrizio-osaera aztertzea; nazioarteko dieta orekatuaren gomendioekin aldaratu, desberdintasun kuantitatibo eta kualitatiboak detektatzeko, eta, aldi berean, populazio orokorreko emakumeen datuekin aldaratu (**1go eskuizkribua**).

3) EAEko gizon zeliakoen nutrizio-egoera eta jarraitzen duten GGDren nutrizio-osaera aztertzea; nazioarteko dieta orekatuaren gomendioekin aldaratu, desberdintasun kuantitatibo eta kualitatiboak detektatzeko, eta, aldi berean, populazio orokorreko gizonen datuekin aldaratu (**3. eskuizkribua**).

4) EAEko gizon eta emakume zeliakoen nutrizio-egoera eta jarraitzen duten GGDren nutrizio-osaera konparatzea (**3. eskuizkribua**).

B. GGPen nutrizio-osaera aztertzea eta horiek GGDn duten eragina ebaluatzea.

5) GGP batzuen eta beraien glutendun homologoen zuntz dietetiko eta almidoi iraunkor (AI) edukia laborategian neurtzea eta aldaratzea **(4. eskuizkribua).**

6) GGP batzuen eta beraien glutendun homologoen mikronutriente edukia laborategian neurtzea eta aldaratzea **(5. eskuizkribua).**

7) EAEko umeek eta nerabeek jarraitzen duten GGDren nutrizio-osaeran, balizko desorekan, GGPek duten erantzukizuna aztertzea; baita hauek zuntza eta mikronutriente ahorakin totalari egiten dioten ekarpena analizatzea ere. (**2., 4. eta 5 eskuizkribuak**).

C. Finlandiako populazio zeliakoaren GGDrekiko atxikimendua aztertzea eta honek sintoma klinikoen osatzean duen eragina.

8) Finlandiako heldu zeliakoen ohitura dietetikoak eta horiekin erlazionatzen diren ezaugarri klinikoak aztertzea.

A. EAEko populazio zeliakoaren nutrizio-egoera eta elikadura ohiturak:

1. Parte-hartzaileak eta prozedura:

Pertsona zeliakoen elikadura-ohiturei dagokienez, EAEn burututako ikerketa prospektiboa aurkezten da. Parte-hartzaileak adin-talde guztietakoak izan ziren (6. taulan lagina deskribatzen da) eta 2011 eta 2013 urteen artean bildu ziren EAEko hiru lurraldeetan.

6. Taula. Parte-hartzaileen banaketa.

Taldea	**Adina (urte)** **Tartea (BB±DE)**	**n**
Emakumeak	≥18 (37,1±11,7)	47
Gizonak *	≥18 (34±14,9)	37
Umeak (neskak/mutilak)	3-17 (9,4 ± 4,0)	83 (52/31)
Guztira		167

Notak: BB: Bataz bestekoa; DE: desbiderapen estandarra

Parte-hartzaileak EZErekin eta Donostiako Unibertsitate-Ospitaleko Gastroenterologia zerbitzuarekin elkarlanean errekrutatu ziren. EZEk diagnostikatu berri diren kide zeliakoekin harremana du, egoera berriari aurre egiten laguntzeko, baina baita aspaldiko kide zeliakoak, haientzako zerbitzu eta ekintza desberdinak antolatzen dituztelako. Parte-hartzea borondatezkoa bazen ere, hiru lurraldeetako (Araba, Bizkaia eta Gipuzkoa) pertsonak hautatzea izan zen helburua, betiere bakoitzaren proportzioa mantenduz. Horretarako, EZEk proiektuaren berri zabaldu zuen bere web eta aldizkariaren bitartez. Gizonen kohortea handitzeko, aurrekoez gain Gurutzetako Unibertsitate-Ospitalean egindako ikerketa bateko datuak ere jaso ziren.

Azterketan parte hartzeko bete beharreko baldintza izan zen urte batez gutxienez GGD jarraitu izana. Baztertze irizpideak, aldiz, hauek izan ziren: gaixotasun kronikoen historia klinikoa aurkeztea (hala nola gaixotasun kardiobaskularrak, diabetesa, hipertiroidismo/hipotiroidismoa, hiperkolesterolemia, hipertriglizeridemia, hipertentsio arteriala) edo/eta murrizketa dietetiko espezifikoak behar dituzten beste patologia digestiboak nozitzea.

Proiektu hau Euskal Herriko Unibertsitateko, UPV/EHUko, Etika Batzordeak onartu zuen (CEISH/76/2011 eta CEISH/194M/2013), baita Gurutzeta Unibertsitate-Ospitaleko Etika Batzordeak ere gizonei dagokienez (CEIC E-08/66). Parte-hartzaileen, eta umeen kasuan haien tutore legalen, idatzizko baimen informatua eskuratu zen, proiektuaren eta osatu beharreko galdetegien inguruan informazioa eman ostean.

Partaide guztien (n=167) neurri antropometrikoak jaso ziren eta galdetegi dietetiko desberdinak bete zituzten. Horrela haien nutrizio-egoera eta ohitura dietetikoak aztertu ziren.

Datuak jasotzen eta aztertzen bukatu ondoren, parte-hartzaile bakoitzak banakako txosten bat jaso zuen norbere emaitzen balorazioarekin: nutrizio-egoera, dietaren ezaugarriak, egindako akatsak eta horiek zuzentzeko hainbat aholku dietetikoak.

2. Neurri antropometrikoak:

Neurri antropometrikoak biltzen esperientzia zuten ospitaleetako eta EZEko nutrizionistak gurekin batera aritu ziren. Gorputz-pisua (±10g) baskula bat erabiliz neurtu zen (SECA 760), altuera (±5mm) estadiometro baten bidez (SECA 220) eta banakoen GMIa (kg/m^2) kalkulatu zen aurreko datuak erabiliz. GMI balioak Obesitatea Ikasten eta Ikertzen duen Erakundeak (SEEDOk) ezarritako irizpideen arabera sailkatu ziren: gutxiegiko pisua (<18,5 kg/m^2), normopisua (18,5-24,9 kg/m^2), gainpisua (25-29,9 kg/m^2) eta obesitatea (>30 kg/m^2). Umeen kasuan, neska eta mutilentzako GMIrako desbiderazio estandarrak (DE) kalkulatu ziren Z-balioak erabiliz. Horiek, era berean, Cole *et al*.-en LMS metodoarekin kalkulatu ziren (erref.6). Gutxiegizko edo gehiegizko pisua definitu ziren, adina eta sexuaren arabera garatutako GMI ebaki-puntuen arabera. Aipagarri da ebaki-puntuak kalkulatzeko oinarria nazio mailako eta nazioarteko datu baseak erabili izan zirela (149, 150).

3. Gorputz-konposaketa eta energia-gastua:

Gorputz-konposaketa, hots, gantz-masa eta masa iharra, segmentukako frekuentzia anitzeko inpedantzia bioelektriko zuzenaren bidez neurtu zen (Inbody 230; Biospace, Seoul, Korea). Horretarako, bina azal-elektrodo kokatu ziren parte hartzaileen oinetan eta eskuetan. Prozedura estandarraren arabera, metodo analitiko honekin gorputzak korronte elektrikoari eragiten dion erresistentzia neurtzen da. Proba hau egiteko ezinbestekoa zen baldintza hauek betetzea: parte-hartzaileek ura ez edatea, gutxienez proba egin baino ordu eta erdi lehenago, eta neurketa egin aurretik komunera joatea gernu-maskuria husteko. Pertsona guztien gantz-masa eta ihar-masa kalkulatu ziren gorputz osoko inpedantziatik abiatuta (Z). Gastu energetikoa kalkulatzeko, hasteko oinarrizko metabolismoa kalkulatu zen. Harris-Benedict formula erabili zen parte-hartzailearen pisu, altuera eta adinaren datuetan oinarrituz. Horri jarduera fisikoren gastua erantsi zitzaion, eta kasu guztietan 1,5 zuzenketa-faktorea aplikatu zen, neurrizko jarduera fisikoari dagokiona. Jarduera fisiko maila sailkatzeko OMEk ematen dituen gomendioak erabili ziren oinarritzat (>60 minutuko neurrizko edo trinko jarduera fisiko/egun) eta gantz masaren portzentajeari dagokionez Gallagher *et al*.-en jarraibideak erabili ziren erreferentzia moduan (151).

4. Dieta:

Ahorakin dietetikoa galdetegien bitartez aztertu zen, elikagaien kontsumo-maiztasunari buruzko galdera-sorta bat eta hiru 24 orduko oroitzapen (2 lan-egunekoak eta bat asteburukoa) osatu ziren hain zuzen ere.

24 orduko oroitzapenaren bitartez pertsonak egun oso batean zehar kontsumitutako janari eta edari guztiak jasotzen dira. Kontsumo maiztasunari buruzko galdetegia berriz elikagai talde desberdineko aleen zerrenda bat da eta denboraldi luzeago batean parte-hartzaileak elikagai horiek zer maiztasunez kontsumitu dituen jakitea da bere helburua. Ikerketa honetarako erabilitako bi galdetegiek berezitasun bat daukate: GGPen kontsumoa jasotzeko aukera ematen dutela hain zuzen ere, beste batzuek ez bezala (1go eta 2.eranskinak)

Galdetegiak nutrizionisten laguntzarekin bete zituzten partehartzaileek eta elikagai eta anoen tamainak argazkien bitartez zehaztu ziren (152). Dieta erregistroetatik abiatuta, energia eta nutrienteen ekarpena *“Alimentación y Salud”* programaren bitartez kalkulatu zen (AyS, Software, Tandem Innovam, Inc., Huesca, Spain), zeinak elikagaien nutrizio-osaeraren datu basean oinarrituz eta jasotako elikagai mota eta kantitateak zenbatuz dietaren nutriente ekarpena kalkulatzea ahalbidetzen duen. Aitzitik, dieten ebaluazioa egiteko erabilitako softwareak ez zituen GGPak bere elikagaien datu basean. Hortaz, azpimarratzekoa da parte-hartzaileek kontsumitzen zuten GGP guztien nutrizio-osaera banan-banan bildu zela eta jatorrizko softwarearen datu basera gehitu zela dieta guztiak ebaluatu aurretik. Halaber, GGPak ez dutenez mikronutrienteen edukia (bitaminak eta mineralak) etiketan adierazten, glutendun homologoen datuak erabili ziren. Umeen kasuan, Missbach *et al*.-ek sortutako GGPen mikronutriente-edukia biltzen zuen datu basea erabili zen (153); horretarako, GGPen mikronutriente-osaera berriz gehitu zen "Alimentación y Salud" softwareko jatorrizko datu basera, hartara umeen dieten azterketa nutrizionala egoki egin ahal izan zen. .*Espainiako Nutrizio*, *Elikadura eta Dietetika Elkarteen Federazioa*k (FESNADek) 2010ean ezarritako nutrienteen ingestaren erreferentzia-balioak erabili ziren 24 orduko galdetegien datuak baloratzeko (154). Elikagaien kontsumo-maiztasunari buruzko galdetegiak interpretatzeko, ordea, *Espainiako Komunitate-Nutrizioaren Elkarteak* (SENCek) ezarritako gomendioak hartu ziren kontuan (155).

GGDren emaitzak dieta orotarikoa zeramaten eta populazio orokorraren datuak kontroltzat hartu ziren.. Horrela, kontrol helduen datuak Estatu mailan egindako ENIDE nutrizio-inkestatik hartu ziren. ENIDE azterlana estatu mailako populazio helduaren erakusgarria da, izan ere 18-65 urte bitartean zeuden 3.323 pertsonen erantzunetan oinarrituta dago. Nabarmentzekoa da ENIDE gure ikerketaren garai berdinean egin zela eta dieta neurtzeko zerabiltzan tresnak, hots, galdetegi dietetikoak, guk erabilitakoen berdinak direla, 24 orduko hiru oroitzapen eta elikagaien kontsumo-maiztasunari buruzko galdetegi bat alegia (156). Umeen datu antropometriko eta dietetikoak konparatzeko Eusko Jaurlaritzak 2005. urtean argitaratutako Nutrizio Inkesta erabili zen. Itaunketa horren helburua zen EAEko 4 eta 18 urte bitarteko biztanleriaren (n=1178) nutrizio-osasunaren egoera eta determinatzaileak ezagutzea eta nutrizio-arriskuko talde nagusiak identifikatzea (157).

GGDan glutendun zerealetatik eratorritako hainbat elikagai GGPek ordezkatzen dituztenez baina horien konposizioa ezberdina dela kontuan hartuz zeliakoen nutriente ahorakina alda daiteke. GGPek dietaren nutrizio-osaeran duten eragina neurtzeko asmoz, parte-hartzaile zeliakoen “glutendun dieta” simulatu zen, GGDrekin konparatzeko. Horretarako, galdetegien bitartez jasotako GGDk bikoiztu ziren, GGPak, eta dagokien nutrizio-osaera, ezabatu eta horien ordez glutendun homologoak sartu ziren, beren nutrizio-osaerarekin. GGPak glutendun produktuekin ordezkatzeko erabilitako irizpidea antzekotasuna izan zen, osagai-zerrenda antzekoenak dituzten produktuak bilatzea alegia. Horrela, GGDtik abiatuz glutendun dieta simulatu zen, non glutendun produktuek GGPak ordezkatzen zituzten, besterik ez. Ondoren, bi dieten nutrizio-osaerak aldaratu ziren.

Dietaren kalitatea ere ebaluatu zen umeen taldean, KIDMED indizearen bitartez Mediterranear Dietarekiko (MD) atxikimendua neurtu zen (eranskina 3). Indize hori sarritan erabiltzen da MDren ohitura dietetiko osasuntsuen adierazle moduan eta, izan ere, korrelazio egokia du kontrol glizemikoarekin eta osasun kardiobaskularrarekin. Galdetegi bakun bat da, non dietaren puntuazioa 0tik (kalitate baxuko dieta) 9ra arte (kalitate altuko dieta) baloratzen den. Horrela, pertsona zeliakoek fruta, fruitu lehor, barazki, tuberkulu, lekale, arrain edo zerealen ingestio egokia dutenean puntu bana jasotzen dute, elikagai talde bakoitzeko, baita gantz monoasegabeen eta aseen erlazioa egokia denean ere. Kontsumoa urriegia denean, berriz, ez dute punturik jasotzen. Haragia eta bere eratorrien kasuan kontrakoa gertatzen da, kontsumoak baxua izan behar du puntu bat jasotzeko eta gehiegizko kontsumoa egiteak du punturik lortarazten.

GGDren azterketan nutrizio-osaera eta gomendioen betetzea aztertzeaz gain, dietaren balizko gluten edukia analizatzea garrantzitsua da, transgresioak alegia, dakarten arriskua dela eta. Transgresioak aztertzeko normalean pazienteei galdetu ohi zaie, baina haien erantzuna zintzoa ez izateko beldurra eta nahigabeko transgresioak ere badirela kontuan hartuz, horiek detektatzeko bioadierazgailuak bilatu nahian dabil komunitate zientifikoa. Aurreko guztia dela eta, *University of Tampere*ko *Celiac Disease Research Center*en egonaldia egin zen, Katri Kaukinen katedradunaren zuzendaritzapean, transgresioen gaia jorratzeko asmoz.

B. GGPen nutrizio-osaera aztertzea eta horiek GGDn duten eragina ebaluatzea:

1. Makronutrienteen edukia:

Laborategiko analitika hauek egiteko MDren piramidearen oinarrian dauden 37 GGP aukeratu ziren: 13 gosaltzeko zereal, 12 ogi eta 12 pasta. Elikagaiak giro tenperaturan edo izoztuta biltegiratu ziren (ekoizleen jarraibideen eta lagin motaren arabera).

Elikagaien proteina eta gantza edukia Kjeldahl eta Soxhlet erauzketa metodoen bitartez neurtu zen hurrenez hurren (158) (AOAC2005).

Kjeldahl metodoak lagin batean dagoen nitrogeno kopurua neurtzen du, proteina edukia kalkulatzeko abiapuntu dena. Metodoa 3 fasetan banatzen da: laginaren liseriketa, destilazioa eta balorazioa. Liseriketan laginean dagoen nitrogeno organikoa amonio ioi (NH_4^+) bihurtuko da, horretarako azido sulfurikoa (H_2SO_4) eta kobre II sulfato ($CuSO_4$) katalizatzailea erabiliko dira. Ondoren, destilazioan, amonioa ioia amoniako (NH_3) bihurtuko da sodio hidroxidoa (NaOH) adizioaren bitartez eta gas hori azido borikoak [$B(OH)_3$] jasoko du. Azkenik, boriko soluzio horren destilatua azido klorhidrikoarekin (HCl) baloratuko da nitrogeno edukia kalkulatzeko.

Soxhlet metodoren bidez aurrez hidrolizatua eta lehortuta dagoen lagin baten gantzaren erauzketa egiten da. Horretarako, iragaz-paperean dagoen lagina soxhlet aparatuan dagoen kartutxoan edo kukurutxoan sartuko da eta hexano edo petroleozko eterra gaineratuko zaio, gantza disolbatu eta erauzteko. Ostean, soxhlet aparatua piztu eta irakiten jarriko da disolbatzaile guztia lurrundu arte. Karbohidratoen edukia, elikagaien hezetasuna eta errautsak neurtu ostean, diferentziaz estimatuko da.

Emaitzak glutendun produktu baliokideen datu analitikoekin aldaratu ziren, GGPekin batera eta era berean analizatu zirenak, eta baita GGPen etiketetako datuekin.

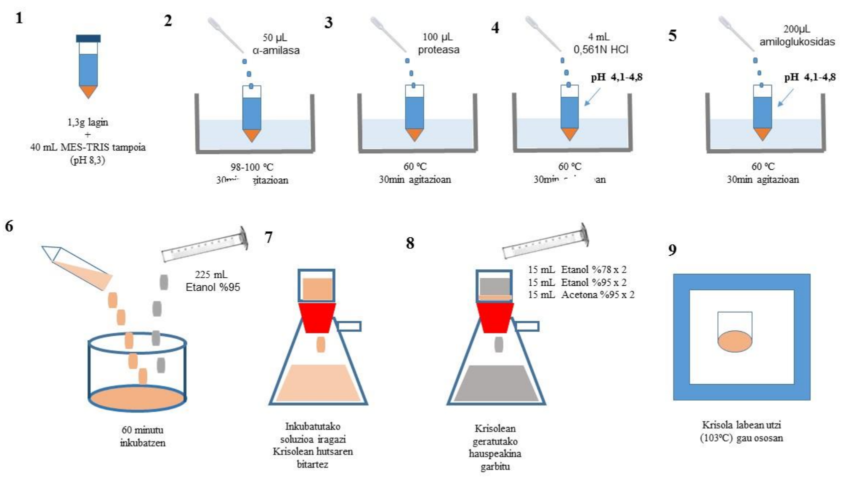
2. Zuntza eta almidoi iraunkorra (AI) edukia:

Zerealen taldeko glutendun produktuak (n=55) eta haien gluten gabeko homologoak (GGP) (n=55) hautatu eta erosi ziren. Aukera gure kohorte zeliakoaren ohituretan oinarritu zen, gehien kontsumitzen dituen GGPak hautatuz. Elikagai-laginak 7 talde desberdinetan sailkatu ziren GGPak eta glutendun homologoak sartuz talde bakoitzean: 1. ogiak (n=11/11), 2. okintzako produktuak eta tartak (n=14/14), 3. Opilak (n=11/10), 4. gosaltzeko zerealak (n=3/3), 5. umeen zereale-formulak (n=3/3), 6. pasta eta irinak (n=6/7) eta 7. oreak (n=7/7). Behin lagin guztiak laborategian zeudela, giro tenperaturan edo izoztuta biltegiratu ziren (ekoizleen jarraibideen eta lagin motaren arabera). Analisia hasi aurretik, beharrezkoa zen kasuetan lagina txikitu egin zen, eta ondoren, lagin guztiak deshidratatu ziren 105ºCtan pisu konstantera heldu arte. Ondoren, zuntz dietetiko totalaren (ZDT) eta almidoi iraunkorraren (AI) analisia egin zen.

2.1 Zuntz dietetiko totalaren analisia:

ZDTren analisia egiteko *Total Dietary Fiber* kit komertziala (Megazyme International, *Wicklow, Ireland)* erabili zen, AOAC 991.43 (159) metodoan oinarritzen dena (5. irudia). Horrez gain, etanol, azido hidrokloriko edo sodio azetato bezalako erreaktibo orokorrak Panreac Química SLU (Bartzelona, Espainia) enpresak hornitu zituen. ZDTren neurketa gantzarik gabeko eta deshidratatuak dauden laginetan egiten da. Metodoak liseriketa fisiologikoan gertatzen diren pausuak imitatzea dauka oinarritzat. Elikagai-lagin (1,3 g) 40 mL MES-TRIS indargetzaile soluziorekin (pH 8,2) batera irabiatu ziren, *Falcon* hodietan 30 segundoz. Gero, 50µL α-amilasa soluzio gehitu ziren eta hodiak aluminiozko paperarekin estali ziren. Hodiak 98-100ºCtako bainuan sartu ziren 30 minutuz etengabeko agitazioan. Horrela, almidoiaren gelatinizazioa, hidrolisia eta depolimerizazioa eragiten da. Ondoren, bainutik atera eta beren tenperatura 60 ºC-tara jaitsarazi zen. Hodien hormetan edota hondoan lagin apurrak geratu ezkero espatula baten laguntzaz askatu eta 9 mL ur distilatu gaineratu zitzaien ondo garbitzeko. Momentu horretan 100 µL proteasa soluzio gaineratu, proteinen solubilizazioa eta depolimerizazioa eragiteko eta hodiak berriz estali eta inkubatu ziren, 30 minutuz 60ºC-tan etengabeko agitazioarekin. Bukatutakoan, 0,561 N HCl 4 mL gehitu zitzaien pH 4,1-4,8 tartea lortzeko. Horrela, 200 µL amiloglukosidasa erantsi almidoi zatiak hidrolizatzeko eta hodiak berriz ere inkubazioan jarri ziren, 60 ºC-tan etengabeko agitazioan. Ostean, 60º C tenperatura berdinean zegoen %95eko etanol 4 bolumen erantsiz (225 mL inguru) zuntza dietetikoa hauspeatu zen, giro tenperaturan 60 minuturen buruan. Prezipitatua jasotzeko, soluzioa iragazi zen Celite geruza bat erantsita zuten krisol-iragazkien bidez. Azken horiek aldez aurretik prestatu ziren, krisolean 1g Celite (± 0,1 mg) pisatuz eta 130 ºC-tara lehortuz, pisu konstantea lortu arte (3 ordu gutxi gorabehera).

Iragazi ondoren, hondakinak garbitu ziren, 15mLko bi bolumen %78 etanol, beste bi bolumen %95 etanol eta, azkenik, bi bolumen azetona huts-bomba erabiliz iragaziz. Ostean, laginak lehor zitezen gau osoan utzi ziren labean 103 ºC-tan. Biharamunean, labetik atera eta lehorgailuan sartu ziren giro-tenperaturara eramateko. Orduan hondarra pisatu zen, prozesuko azken krisol osoa pisatuz eta horri aldez aurretik prestatutako Celitedun krisolaren pisua kenduz. Azkenik, bina aztertutako laginen krisolak banatuko dira; baten hondarra proteina neurtzeko erabiliko da, Kjeldahl metodoaren bitartez (AOAC, 960.52A), eta bestea muflan 525ºCtan sartuko da errautsa lortzeko (AOAC, 923.03). Izan ere, datuok zuntza totalaren edukia kalkulatzeko erabiltzen baitira.

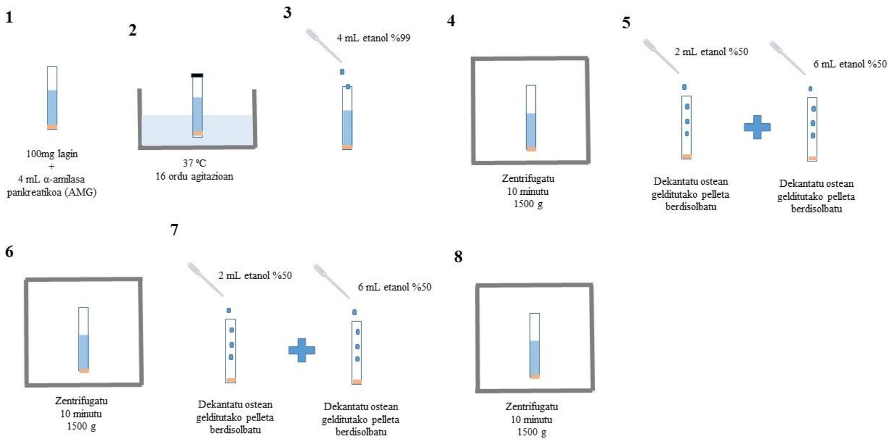


5. Irudia. Zuntza totalaren edukia analitikoki aztertzeko jarraitzen diren pausuak

2.2 Almidoi iraunkorraren analisia:

AI *Megazyme Resistant Starch* kita (Megazyme International, Wicklow, Ireland) erabiliz neurtu zen, McCleary and Monaghan-ek proposatu bezala eta nazioarteko AOACk onartu bezala (2002.02 metodoa) (160). Metodo entzimatiko honek *in vivo* digestio prozesua eta bere baldintzak imitatzen ditu.

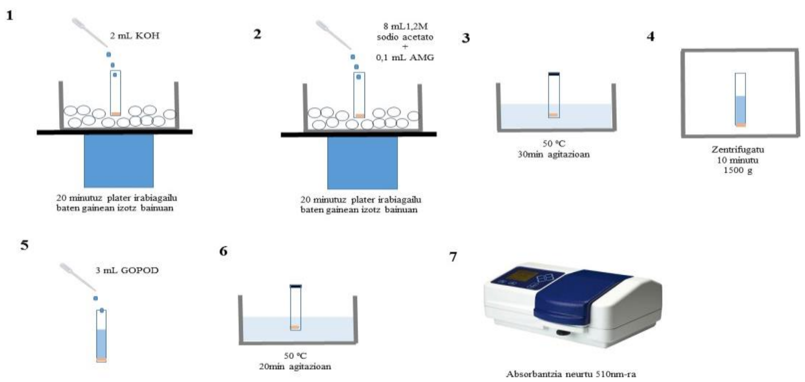
Lehenengo pausuan almidoi ez-erresistentea hidrolizatu eta disolbatu egiten da. Horretarako, kristalezko saiodi batean 100 ± 5 mg lagin pisatu ziren (aurretik lehortuta eta pisu konstantea izanik) eta 4 mL α-amilasa pankreatiko (amiloglukosidasa entzima zuena (AMG, 3U/mL)) gaineratu zitzaien. Tapoia jarri eta saiodiak 37ºC-tan inkubatu ziren, 16 orduz etengabeko irabiaketan (100 rpm). Gero, 4 mL %99 etanol gaineratu zen saiodi bakoitzean, borteatu eta zentrifugatu ziren, 1500 g-ra 10 minutuz. Ostean, gainjalkina dekantatu zen eta hauspeakina, AI duena, berdisolbatu zen 2 mL %50 etanolean bortizki irabiatuz. Horrela, lagina bitan garbitu zen, beste 6 mL %50 etanol gaineratuz, irabiatuz eta berriz ere berdin zentrifugatuz (6. irudia).



6. irudia. AIren edukia analitikoki aztertzeko jarraitzen diren pausuak (1. zatia)

Analisiaren bigarren pausuan AIa neurtu egiten da. Horretarako, kristalezko saiodiak izotz bainuan kokatu ziren, 2 mL KOH gehitu zitzaien eta 20 minutuz eragin ziren prezipitatuak disolba zitezen. Ondoren, 8 mL 1,2M sodio azetato indargetzaile gaineratu, eragin eta, berehala, 0,1 mL AMG (3300 U/mL) gaineratu zitzaien. Ondo irabiatu ondoren laginak inkubatu ziren, 30 minutuz 50ºC-tan zegoen bainuan. Horrela, almidoi iraunkorra apurtu eta glukosa molekulak askatzen dira. Amaierako AI edukia %10 baino txikiagoa izatea espero zenez, laginak ez ziren gehiago diluitu baizik eta zuzenean zentrifugatu ziren, 10 minutuz 1500g-ra.

Amaitzeko, glukosa edukia kuantifikatzeko,, gainjalkin 0,5 mL-ri 3 mL glukosa oxidasa plus peroxidasa eta 4-aminoantipirina (GOPOD erreaktiboa) gaineratu zitzaien eta azkenengo inkubazioa egin zen, 20 minutuz 50ºC-tan. Azken produktua espektrofotometriaz neurtzen da, absorbantzia 510 nm-tan neurtuz (7. irudia). Datu horretatik, AI kantitatea kalkulatzen da, amaieran emaitza lagin osoarekiko ehuneko bezala adieraziz.



7. irudia. Almidoi iraukorraren edukia aztertzeko jarraitzen diren pausuak (2. zatia)

3. Mikronutrienteen edukia:

Mikronutrienteen azterketa egiteko, aukeratutako laginak makronutrienteen azterketan erabilitako berdinak izan ziren, hots, dieta orekatuan zerealen taldeko hiru elikagai nagusiak: gosarirako zerealak (n=13), pasta (n=12) eta ogia (n=12). / GFP from the three main contributor cereal food-types to a balanced diet, such as flakes (n=13), pasta (n=12) and bread (n=12), were selected.

Mineralak analizatzeko mikrouhinek lagundutako digestioa burutu zen Mars 5 mikrouhin aparatu itxian (CEM, Vertex, Barcelona, Spain), zeinak 8-24 tefloi-ontzi eta tenperatura kontrolagailuak zituen. Selenio, manganeso eta kobrearen analisi kuantitatiboak egiteko erabili ziren ICP-MSa (7700x, Agilent Technologies, Palo Alto, USA) eta MicroMist mikro-xurgatze beirazko langargailu zentrokidea (Glass Expansion, West Melbourne, Victoria, Australia). Kaltzio, sodio, zink eta burdinaren kasuan ICP-OESa (Horiba Jobin Yvon, Activa) erabili zen, kuartzozko Meinhard langargailu zentrokidearekin, Scott motako esprai-ganberarekin eta ohiko koartzozko konexio-zorroarekin esprai-ganberaren eta lastargiaren artean.

Bitaminen analisia egiteko (biotina, folatoa, niazina, piridoxin, erriboflavina, tiamina, B5 eta B12) kromatografia likidoa (HPLCa) erabili zen, kuadrupolo hirukoitz teknologiadun masa-espektrometria detekzio sistemarekin batera. Bitaminen identifikazioa lortu zen elektrospraiaren inoizazio positiboaren eta erreakzio anizkoitzen monitorizazioaren bidez. Bitaminak kuantifikatu aurretik laginak likido-likido erauzketaz jaso ziren, nahasketa urtsu azido bat erabiliz, zentrifugatuz eta iragaziz. E bitamina analizatzeko, aldez aurretik laginak saponifikatu egin ziren, likido-likido erauzketa eta purifikazioa burutzeko. Ondoren, errendimendu handiko kromatografia likidoa, fluoreszentzia detektagailuarekin, erabili zen erauzietan bitamina azterzeko.

Azkenik, laborategian aztertutako GGPen mikronutriente edukia Espainiako Elikagaien Konposizioa biltzen duen Datu Baseko (BEDCA) (161) glutendun ogi (n=19), gosari-zereal (n=18) eta pasta (n=8) homologoekin erkatu zen. Biotina edukia aztertutako elikagai guztientzat eta kobrearena zerealen taldeko elikagaientzat McCance and Widdowson’s elikagaien konposizio tauletatik (162) hartu zen. Zerealen kasuan ez zen manganeso edukiaren daturik aurkitu ez Espainia ezta Erresuma Batuko, Estatu Batuetako eta Australiako datu baseetan ere (161-164).

Lan esperimentalari dagokionez, makronutrienteen, zuntzaren eta AIren edukiaren azterketa analitikoa UPV/EHUko Nutrizio eta Bromatologia arloan burutu izan da eta mikronutrienteen azterketa UPV/EHUko Ikerkuntzarako Zerbitzu Orokorrak (SGIker), mineralentzat, eta kanpoko laborategi baten zerbitzuak, bitaminentzat, kontratatu ziren.

Analisi estatistikoa:

Analisi estatistikoa IBMren SPSS programa estatistikoaren 23. bertsioa erabiliz burutu zen (IBM Inc., Armonk, NY, USA). $P< 0,005$ balioak onartu ziren esanguratsu bezala. Emaitzak aurkezteko batezbesteko aritmetikoa ± desbiderazio estandarra (DE) erabili da.

Datuen banaketaren normaltasuna *Kolmogorov-Smirnov Test*aren bitartez aztertu zen eta homogeneizitatea *Levene Test*aren bitartez.

Talde zeliakoaren elikadura ohiturei dagozkien emaitzak populazio kontrolaren datuekin konparatzeko erabilitako test estatistikoa neurri laburtuen *t Student´s Test*a izan zen; gizon eta emakume zeliakoen arteko konparaketak egiteko *Chi-karratu test*a jarraitu zen.

Erregresio linealaren analisia erabili zen KIDMED indizearen emaitzak (kalitate baxua, ertaina edo altua, alegia) beste parametro batzuekin erlazionatzeko, hala nola, sexua, adina, jarduera fisikoa, GGDn egondako denbora eta gorputz-pisu egokia. Aipagarria da distribuzio ez-normala zuten aldagaiak logaritmikoki eraldatu zirela distribuzio normala lortzeko.

Zeliakoen gluten gabeko eta glutendun bihurtutako dietak aldaratzerakoan, hots, binakako batezbesteko balioen analisi estatistikoa egiteko, *Wilcoxon Test* ez-jarraia erabili zen.

Elikagai talde bakoitzaren zuntz ekarpena dieta osoaren zuntza edukiarekin erlazionatzeko erregresio linealaren analisia erabili zen. Berriz ere, distribuzio ez-normala zuten aldagaiak logaritmikoki eraldatu ziren distribuzio normala lortzeko.

Zeliakoen zuntza kontsumoa kontrolen ahorakinarekin erkatzeko neurri laburtuen T Student´s testa erabili zen. Mann-Whitney U Testa erabili zen desberdintasun esanguratsuak ezartzeko dieta orokorrarekin eta GGDren zuntz dietetiko eta almidoi iraunkor kopuruei dagokienez. Baita laborategian lortutako datu analitikoak eta etiketen zuntzaren datuak erkatzeko ere.

Elikagaien makronutriente laborategiko analisien datuak aztertzeko binakako T Student proba erabili zen. Mikronutrienteen kasuan emaitzek (bai datu analitikoek baita datu baseenek ere) banaketa ez-jarraia zutenez Mann-Whitney U Testa erabili zen.

Doktorego-Tesi lan hau garatzeko, doktoregaia UPV/EHUko Euskara Errektoreordetzako Doktorego-Tesia Euskaraz Prestatzeko, Idazteko eta Defendatzeko Beka baten onuradun izan da. Tampereko egonaldirako ere diru-laguntza jaso izan zuen, UPV/EHUko Ikerketarako Errektoreordetzak emandako Mugikortasuna Sustatzeko eta Ikerketaren Emaitzak Dibulgatzeko Laguntza bat hain zuzen ere.

EMAITZAK ETA EZTABAIDA

RESULTS AND DISCUSSION

Manuscript 1

Itziar Churruca, Jonatan Miranda, Arrate Lasa, María Á. Bustamante, **Idoia Larretxi** and Edurne Simón

Analysis of Body Composition and Food Habits of Spanish Celiac

Nutrients 2015, 7, 5515-5531

Nutrients **2015**, *7*, 5515-5531; doi:10.3390/nu7075234

OPEN ACCESS

nutrients

ISSN 2072-6643

www.mdpi.com/journal/nutrients

Article

Analysis of Body Composition and Food Habits of Spanish Celiac Women

Itziar Churruca, Jonatan Miranda, Arrate Lasa, María Á. Bustamante, Idoia Larretxi and Edurne Simon *

Gluten Analysis Laboratory of University of the Basque Country, Department of Nutrition and Food Science, University of the Basque Country (UPV/EHU), Vitoria-Gasteiz 01006, Spain;
E-Mails: itziar.txurruka@ehu.es (I.C.); jonatan.miranda@ehu.es (J.M.); arrate.lasa@ehu.es (A.L.); marian.bustamante@ehu.es (M.A.B.); idoia.larrechi@ehu.es (I.L.)

* Author to whom correspondence should be addressed; E-Mail: edurne.simon@ehu.es;
Tel.: +34-945-013-069; Fax: +34-945-013-014.

Received: 28 May 2015 / Accepted: 1 July 2015 / Published: 8 July 2015

Abstract: The purpose of the present work was both to analyze composition of Spanish celiac women and to study the food habits and gluten-free diet of these celiac patients, in order to determine whether they achieve a balanced and healthy diet as well as to highlight nutritional qualitative and/or quantitative differences. 54 adult celiac women (34 ± 13 years) took part in the six-month study. Height, weight and body composition were measured. An analysis of energy consumption and of the macronutrient distribution of their diet was carried out. Their fulfillment of micronutrient intake recommendations was verified. Participants showed a Body Mass Index of 21.6 ± 2.4 kg/m^2. Energy Intake was slightly lower than the Dietary Reference Intakes. Excessive protein apart from over-consumption of fat was observed. More than three quarters of participants consumed meat in excess. Carbohydrate consumption along with that of fiber was below recommended levels. Vitamin D, iron, and iodine had a low percentage of recommendation compliance. In general, participants followed the recommendations of dairy products and fruit intake whereas vegetable consumption was not enough for the vast majority. We conclude that although the diet of celiac women does not differ much from the diet of general population, some considerations, such as reducing fat and protein consumption and increasing fiber intake, must be taken into account.

Keywords: celiac disease; gluten-free diet; diary recommended intake; food habit; body composition

1. Introduction

Celiac disease (CD) is one of the most common chronic intestinal diseases in Europe and is defined as a permanent intolerance to gluten proteins. This intolerance is maintained throughout a lifetime and is presented in genetically predisposed subjects. Its aetiology is unknown but genetic, environmental (gluten) and immunological factors contribute to its development [1]. Its estimated prevalence in Europeans and their descendants is 1%, this being more frequent in women in a 2:1 ratio [2]. Moreover, there is a significant percentage of patients who remain undiagnosed [2].

The only effective treatment for celiac disease is a strict lifelong gluten-free diet (GFD). To meet this challenge, it is essential to control the production of foods and dishes for celiac people, in order to guarantee the absence of these proteins in them. Catassi *et al.* [3] demonstrated that the ingestion of small amounts of gluten, as little as 50 mg of gluten per day over three months, can cause important damage in the intestinal mucosa. In this context a GFD with the threshold of 20 mg/kg ensures an intake of less than 50 mg/day of gluten and provides a sufficient safety margin [4,5].

Apart from gluten control, the correct suitable GFD must be nutritionally balanced too; it must fulfil all the energy and nutrient requirements and be sufficient to meet the nutritional needs of each person and prevent deficiencies. The possibility that excessively restricted cereal consumption as a solution to avoid gluten intake becomes a low carbohydrate (CHO) intake with an excess of fat and protein cannot be discarded. Additionally, the American Dietetic Association mentions in its guide for celiac patients [6] the possible consequences of complying with a GFD. According to this guide, following this diet could imply a low intake of carbohydrates, iron, folate, niacin, zinc, and fiber. Other authors also indicate that consumption of refined grains, processed as gluten-free products, entails a lower intake of vitamin B group, vitamin D, and calcium [7].

It must be taken into account that the prevalence of some diseases such as cardiovascular diseases is associated with low fiber and high saturated fat intake. Similarly, anaemia, related to a lack of iron and folic acid, and osteoporosis, associated with a lack of calcium and vitamin D, are closely linked with common symptoms of celiac patients [8,9].

As far as we know no data exist concerning the nutritional adequacy of a GFD for celiac women in Spain. In this context, the main aim of the present work was to evaluate body composition and the nutritional composition of the GFD followed by adult celiac women, as well as to compare it with the international recommendations, in order to highlight qualitative and quantitative nutritional differences. Furthermore, we found it relevant to describe the food intake and habits of celiac women in comparison with Spanish women in the general population.

2. Experimental Section

2.1. Participants and Procedure

Gluten and Food Safety is a prospective SUSFOOD study conducted in the Basque Country (Spain). The present study makes use of data from the celiac women cohort recruited in 2011 from three regions in the north of Spain (Alava, Gipuzkoa, and Bizkaia). All celiac patients were members of the Basque Country Celiac Society, with a confirmation of CD diagnosis (intestinal biopsy and/or serological test). Participants had all been compliant with the GFD for at least one year (mean length in years ± SD: 23 ± 11) and were followed up on for this analysis until April 2012 (for six months). 54 celiac women and older than 16 years took part in the study (mean age ± SD: 34 ± 13). All women claimed to be in remission from clinical symptoms. Exclusion criteria included a history of cardiovascular disease or diabetes, pregnancy, hyperthyroidism/hypothyroidism, total cholesterol levels > 300 mg/dL, levels of triglyceride > 300 mg/dL and blood pressure level > 140/90 mm Hg. All participants received verbal and written information about the nature and purpose of the survey, and all gave their written consent for their involvement in the study. This study was approved by the Ethical Committee of the University of The Basque Country (CEISH/76/2011).

Each subject underwent anthropometric parameters and dietary habits record for nutritional status assessment.

2.2. Anthropometric Measurements

Body weight (±10 g) was measured after voiding using a digital integrating scale (SECA 760, SECA, Hamburg, Germany). Height was measured to the nearest 5 mm using a stadiometer (SECA 220, SECA, Hamburg, Germany).

For each subject body mass index (BMI) was calculated as weight (kg)/height (m)^2. The BMI values were categorized according to the World Health Organization (WHO) criteria as follows: Below 18.5 kg/m^2 as underweight, 18.5–24.9 kg/m^2 as normal weight, 25–29.9 kg/m^2 as overweight and $\geqslant$30 kg/m^2 as obese [10].

2.3. Body Composition and Energy Expenditure

Body composition (fat mass and fat-free mass) was estimated with a direct segmental multiple-frequency bioelectrical impedance analysis method (Inbody 230; Biospace, Seoul, Korea). Two skin electrodes were placed on the feet and two electrodes on the hands. According to the standard procedure, whole-body resistance and reactance were measured. For all subjects, fat mass and fat-free mass were evaluated from total-body impedance (Z). Energy expenditure was calculated using Harris-Benedict formula and applying 1.5 factor for mild/light physical activity.

Regarding the percentage of body fat, the guidelines of Gallagher *et al.* were used as reference [11].

2.4. Dietary Assessment

24 h food recall (24HR) of three days and a food frequency questionnaire (FFQ), previously described [12], were kept for each patient with food portions and amounts determined by using photographs of rations and sizes described in Photo Album food [13]. Trained nutritionist-dieticians carried out the 24HRs, two on weekdays and one at the weekend. Nutrient intake was calculated by using a computerized program system (AyS, Software, Tandem Innova, Inc., Huesca, Spain). The nutrient content data of the specific gluten-free products manufactured for celiac people were collected from the manufacturers and added into the food composition database of the program before calculations. As

gluten-free product labels did not indicate micronutrient content (vitamins and minerals), an estimation with homologous gluten-containing products was carried out [12]. Dietary reference intakes (DRI) for Spanish population issued by the Spanish Societies of Nutrition, Feeding and Dietetics (FESNAD) in 2010 were taken as references for the interpretation of the 24HR [14]. Other Recommended Dietary Values such as Institute of Medicine (IoM) were taken into consideration [15].

In the case of FFQ, Spanish Society of Community Nutrition (SENC) recommendations were used for the correct interpretation of the results [16]. The energy, nutrient and food intakes of celiac women were compared to nutritional data obtained from a Spanish reference women population in ENIDE [17], a nutritional survey carried out in 2011, at the same period of time as the present work.

ENIDE study is representative at national level of the adult population. It was based on a random selection of more than 3323 individuals, providing a level of confidence of 95% and an accuracy of $\pm 1.8\%$. The survey was conducted on 1589 men and 1734 women aged between 18 and 65 years old. The methodology used 24 h recall, daily food over three random days, and a food frequency questionnaire [17].

2.5. Mediterranean-Diet Score

Adherence to the Mediterranean diet was measured by the Mediterranean-Diet Score (MDS) [18]. The diet score varied from 0 (low quality diet) to 9 (high quality diet). With regard to fruits and nuts, vegetables/potatoes, legumes, fish, and cereals as well as the component ratio between monounsaturated fatty acids and saturated fatty acids, a value of 1 was assigned to celiac women whose consumption was equal to or higher than the median value, and 0 to the others. For meat and meat products and dairy products, a value 1 was assigned to celiac women whose consumption was less than the median, and 0 to the others. Although seven participants were under 18 years, taking into account that they consume alcohol, these criteria were also computed for MDS.

2.6. Statistical Analysis

Statistical analyses of our results were performed by using the IBM SPSS statistical program 19 (IBM Inc., Armonk, NY, USA). The results for continuous variables are given as the arithmetic mean ± SD and the range. The results for non-continuous variables are given as the frequency and the percentage. Statistical analyses were performed with Student's or Welch's t test and F-Snedecor test. p values < 0.05 were accepted as significant.

3. Results

3.1. Anthropometric and Body Composition Measurements

Main anagraphic data and anthropometric/body composition measurements of CD women are shown in Table 1. The BMI were within normal in 81.5% of cases, and there were only six people with low weight and four cases of overweight women. None of them were obese. Accordingly, the majority of adult women had a normal fat percentage [11].

Table 1. Characteristic of celiac participants included in survey.

Characteristic	Women
n	54
Age (year)	34.4 ± 12.9
Mean duration of GFD (year)	10.9 ± 8.5
Height (cm)	164 ± 6
Weight (kg)	57.9 ± 7.3
Body Fat %	27.1 ± 6.9
Body-mass index	
Mean (kg/m^2)	21.4 ± 27
Underweight <18.5—no. (%)	6 (11.1)
Normal 18.5–24.9—no. (%)	44 (81.5)
Overweight 25–30—no. (%)	4 (7.4)

Notes: Values are means ± SD; SD, standard deviation; no, number of subjects; GFD, gluten free diet.

3.2. Dietary Intakes

3.2.1. Energy, Macronutrients and Fiber

The average energy intake of celiac women (Table 2) was in good accordance with their estimated energy expenditure (1904 ± 161 kcal) but below that of the DRI's [14]. Although this was also observed in women from the ENIDE study, the energy intake of celiac patients was significantly lower (Table 2). When comparing energy sources among groups, similar macronutrient intakes were found between celiac and Spanish women—control women groups (Figure 1).

Regarding proteins, this nutrient intake represented 17.3% of total energy intake, which was similar to the data observed in the Spanish survey (Figure 1), therefore it was consumed in excess in both celiac and general women—control women populations. Animal protein intakes were the main contributor (69%) to the total protein consumption. Dairy products provided nearly 20% of total protein intake whereas meat and meat products, fish and eggs provided nearly 50%.

Carbohydrate consumption was enough to cover the minimum established as DRI (130 g/day) by FESNAD (Table 2). With regard to lipids, the percentage of this macronutrient markedly exceeded the recommendations (Figure 1). In fact, almost all the celiac women reached a fat consumption which represented over 30% of total energy. In order to evaluate the dietary fat quality, the lipid profile was calculated. In general terms, saturated and unsaturated fatty acids related ratios were reached, as was the case in the ENIDE survey (Table 2) [19].

Celiac women consume small amounts of dietary fiber. Indeed, 43% of celiac women did not reach 15 grams per day (Table 2). Compared to Spanish women—control women from the ENIDE survey, the celiac group consumed a significantly lower daily amount of fiber (Table 2). Low fiber intake was more frequent in young adult celiac women; the mean fiber consumption in the 16–44 years celiac women (n = 41) was 15.6 ± 5.5 g/day, women older than 45 years (n = 13) consumed on average 19.0 ± 5.1 g/day (p = 0.005).

Table 2. Energy and nutrient distribution in celiac and Spanish women.

	Celiac Women (*n* = 54)	Spanish Women—Control Women (*n* = 1734)	*p*
Energy (kcal)	1847 ± 362	2038 ± 655	0.003
Protein (g)	79.2 ± 16.6	88.0 ± 37.8	< 0.001
Carbohydrate (g)	192.3 ± 40.7	199.7 ± 75.9	0.002
Fat (g)	84.6 ± 23.0	93.2 ± 35.6	0.022
(PUFA+MUFA)/SFA	2.1	2.0	-
PUFA/SFA	0.49	0.49	-
Cholesterol (mg)	324 ± 137	336 ± 151	0.544
Fiber (g)	16.4 ± 5.6	18.9 ± 10.1	0.002

Notes: Values are means ± SD; Spanish Adult women data were taken from the Spanish dietary nutritional assessment (ENIDE study, representative of the adult population at national level); SD, standard deviation; PUFA, polyunsaturated fatty acids; MUFA, monounsaturated fatty acids; SFA, saturated fatty acids.

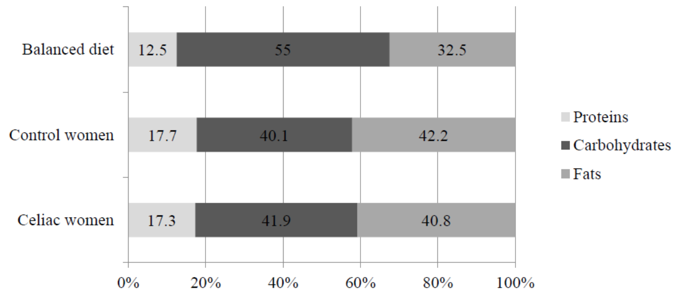


Figure 1. Energy percentage from each macronutrient in celiac (*n* = 54) and Spanish women—control women (*n* = 1734) (ENIDE study, representative at national level of the adult population) compared to the recommended percentage in a balanced diet proposed by the Federation of Spanish Societies of Nutrition and Dietetics (FESNAD).

3.2.2. Micronutrients

Of the 19 micronutrients analyzed, vitamin D, vitamin E, folate, iodine, iron, calcium, and selenium had an especially marked low compliance and these deficiencies were also similar to those found in general population (Table 3). None of the celiac women showed low intakes of vitamin B12 (Table 3). According to FESNAD recommendations, iodine, vitamin D, and vitamin E deficiencies were highly presented because 80%, 48% and 39%, respectively, of the celiac women did not accomplish 2/3 of DRI of these nutrients (Figure 2). Also, nearly 1 in 3 women did not achieve 2/3 of the iron and selenium DRIs (Figure 2). With respect to folate, calcium and vitamin A, around 18%, 13% and 11% of celiac were below 2/3 of recommended levels (Figure 2).

Comparing these with the Spanish general population survey—control women, micronutrient deficiencies follow a very similar scheme/ pattern (Table 3). Statistical differences were found in some of them, such as vitamin E and niacin as well as magnesium and selenium minerals, which were lower in celiacs (Table 3). On the other hand, riboflavin, vitamin B6, and folate were noticeably higher in the celiac group (Table 3).

Table 3. Micronutrient intake in celiac and Spanish women compared to the FESNAD' and IoM' recommendations.

	Celiac Women (*n* = 54)	**Spanish Women—Control Women (*n* = 1734)**	***p***	**DRI FESNAD (2010) [14]**	**DRI: RDA and AI IoM (2011) [15]**
Vitamin A (µg)	819 ± 556	723 ± 323	0.001	600	700
Thiamin (mg)	2.1 ± 3.5	1.8 ± 4.9	0.056	1	1.1 [k]
Riboflavin (mg)	2.1 ± 1.5	1.4 ± 3.2	0.001	1.3 [a]	1.1 [k]
Vitamin B6 (mg)	2.1 ± 0.8	1.7 ± 3.7	< 0.001	1.2 [b]	1.3 [b]
Vitamin B12 (µg)	8.1 ± 5.6	6.1 ± 4.9	0.230	2	2.4
Vitamin C (mg)	153 ± 65	133 ± 80	0.180	60 [c]	75
Vitamin D (µg)	4.9 ± 4.0	3.7 ± 3.7	0.331	5	15
Vitamin E (mg)	11.2 ± 3.8	13.4 ± 7.0	0.003	15	15
Niacin (mg)	26.4 ± 11.4	39.4 ± 39.7	< 0.001	14	14
Folate (µg)	373 ± 556	266 ± 113	< 0.001	300	400 [j]
Calcium (mg)	897 ± 264	835 ± 293	0.274	900 [d]	1000 [d]
Iron (mg)	14.5 ± 5.0	13.7 ± 6.2	0.162	18 [e]	18 [e]
Magnesium (mg)	297 ± 92	354 ± 126	0.011	300 [f]	320 [f]
Iodine (µg)	78.7 ± 38.7	84.8 ± 47.3	0.187	150	150
Phosphorus (mg)	1223 ± 314	1295 ± 380	0.191	700 [g]	700 [g]
Zinc (mg)	9.2 ± 4.0	8.7 ± 3.3	0.156	7 [g]	8
Sodium (mg)	1916 ± 802	2349 ± 810	0.505	1500	1500 [l]
Potassium (mg)	2950 ± 806	2858 ± 827	0.485	3100 [h]	4700
Selenium (µg)	48.2 ± 19.3	53.7 ± 28.9	0.031	55 [i]	55

Notes: Values are means ± SD; Spanish Adult women data were taken from the Spanish dietary nutritional assessment (ENIDE study, representative of the adult population at national level); SD, standard deviation; DRI, dietary reference intake; RDA, recommended dietary allowances; AI, adequate intake; FESNAD, Federation of Spanish Societies of Nutrition and Dietetics; IoM, Institute of Medicine; [a] Riboflavin, 1.2 mg for 16–19 range and >60 years women; [b] Vitamin B6, 1.3 mg for 16–19 years old women (FESNAD), 1.2 mg for 16–18 years and 1.5 mg for >50 (IoM); [c] Vitamin C, 65 mg for 16–18 years (IoM) and 70 mg for >60 years (FESNAD); [d] Calcium 1000 mg (FESNAD) or 1300 mg (IoM) for 16–19 years range and 1000 mg >50 years women (FESNAD) or 1200 mg (ioM) for >50 years; [e] Iron, 15 mg for 16–19 years, 50–59 years and 10 mg for >60 years (FESNAD) or15 mg for 16–18 years and 8 mg for >51 years (IoM); [f] Magnesium 360 mg for 16–18 years, 310 mg for 18–30 years, 320 mg for >60 years (IoM); [g] Phosphorus and Zinc, 800 and 8 for 16–19years (FESNAD) or 1250 mg and 9 for 16–18 years (IoM); [h] Sodium, 1300 mg for >50 years women; [i] Selenium, 45 µg for 16–19 years (FESNAD); [k] Thiamin and Riboflavin: 1.0 mg for 16–18 years; [l] Sodium, 1300 mg for 51–70 years and 1200 mg for >70 years (IoM).

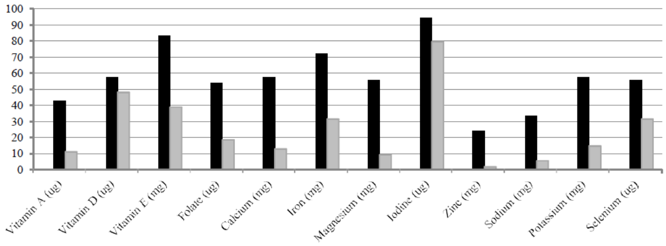


Figure 2. Percentage of celiac women who do not achieve dietary reference intake of the vitamins and minerals proposed by the Federation of Spanish Societies of Nutrition and Dietetics (FESNAD). ■ (black) Do not fulfilled completely (100%) dietary reference intakes; ■ (grey) with a fulfillment of dietary reference intakes in the range of 99%–67%.

3.3. Food Consumption Frequency

Main food group consumption is summarized in Figure 3. The cereal consumption data indicated that only one out of ten celiac people ate the minimum of four recommended servings per day. Moreover, nearly half the celiac women (48%) consumed a very small amount of cereals (fewer than two servings) per day. Nevertheless, three day 24HR questionnaires showed that grains and cereal derivatives provided 23% of total energy intake and that these were the main source of CHO (37.9%). Cereal derivatives consumed usually took the form of naturally gluten-free grains and the incorporation of gluten free rendered cereals (GFP) into GFD was quite low. Specifically gluten-free products formed only 3% of total energy intake and contained only about 11 g of the 73 carbohydrate g provided by the cereal group.

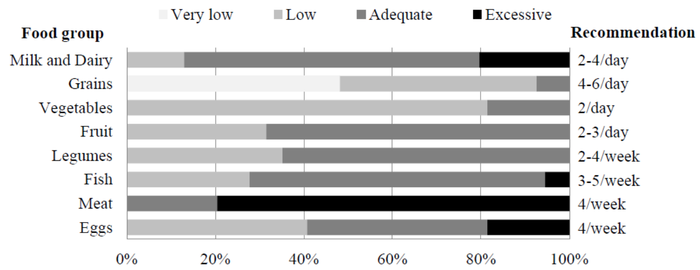


Figure 3. Compliance of food frequency consumption of celiac women by servings per day or week, according to Spanish Society of Community Nutrition (SENC).

One third of celiac women did not consume the minimum recommended two servings of fruit daily. In relation to the intake of vegetables, only 18.5% of celiacs consumed the daily minimum of two servings. Even weekly, the vast majority of participants, 70%, did not reach vegetable consumption recommendations (10 portions/week) [16].

As far as food of animal origin is concerned, 87% of celiac patients consumed two or more serving daily of milk or dairy products. Almost 40% of celiac patients reached the weekly recommended egg consumption by SENC. More than three quarters of participants had an excessive consumption of meat and meat products. Additionally, half this amount was fatty meat/red meat (pork, beef, lamb for instance). In the case of fish, its consumption was sufficient in two out of three adult celiacs and very few consumed it in excess. This food group (meat, fish and eggs) supplied 18% of total energy, and nearly 50% of total protein intake, whereas milk derivatives contributed 14% of total energy and 20% of total protein.

65% of celiac women usually included two or more portions of legumes and, on average, daily intake was about 20 g. Chickpeas, beans and lentils were the preferred pulses, making up 3% of the total daily energy intake.

With respect to other energy sources such as vegetable oils, sugar, chocolate, alcohol and/or pastries, most participants consumed them fairly correctly (data not shown). In general, this population chose olive oil as its fat source, a positive aspect that contributed to maintaining the high proportion of monounsaturated fatty acid in the diet.

3.4. Score for Adherence to Mediterranean Diet

The MDS showed that diet followed by celiac women was intermediate in quality, in terms of adherence to the Mediterranean pattern. The mean MDS for all studied participants was 4.6 ± 1.8.

4. Discussion

Dietary treatment of celiac disease involves certain dietary restrictions that can limit the nutritional status of celiac people and make the implementation of the recommendations for a balanced diet difficult. In a previous study carried out by our research group, females and males were collected in order to analyze their GFD and to carry out a nutritional comparison between gluten-free diets and diets containing equivalent products with gluten [14]. Surprisingly, women and men showed different patterns of eating habits. Following a GF diet in women resulted in a lower protein intake and in a higher fat intake when comparing the same diet with equivalent products containing gluten. However in men these differences were not observed. Taking these results into account, a more in-depth study should be carried out in order to analyze the nutritional status of celiac women following a GFD. Thus, in the present research, we evaluated this situation in 54 celiac women from the Basque Country (from Alava, Gipuzkoa, and Bizkaia regions in the north of Spain), in order to point out any differences between celiac women and women in the general population.

The anthropometric study conducted with celiac women indicated that only less than 6.5% were overweight and that there were no cases of obesity. In good accordance with these results, Bardella *et al.* found a lower BMI index in celiac people than in the general population [20]. Our results were also lower than those corresponding to Spanish women—control women overall [21]. There are several reasons that could justify this fact: (1) the celiac disease pathological situation could provoke a low bioavailability of nutrients as was reported by others in newly diagnosed celiacs [22]; (2) these patients were more concerned about their dietary habits than the general population and therefore there is a lower percentage or incidence of overweight and obesity.

With regard to the first issue, it is hard to believe that malabsorption related to celiac disease could be an explanation, considering that participants in our study had followed a long-term GFD (10.9 years). However, it is true that in the case of the adult celiac population complete normalization of duodenal lesions is rare [23], leading to additional nutrient deficiencies. One of the weaknesses of the present research is that no intestinal biopsy was performed prior to the study, therefore there was no possibility of ensuring the histological remission of celiac disease, which makes it difficult to discount if any of the celiacs studied might have more deficiencies or not. Nevertheless, the purpose of the study was to define whether GFD itself could provoke nutrient deficiencies or not, regardless of histological remission. Despite its limitations, the information extracted from the present research could answer why celiac patients in biopsy-proven remission and who adhered to a strict gluten-free diet for years were prone to the development of various vitamin deficiency states [24].

The second reason itself could also be considered as one possible limitation of this work, due to the fact that research study participants usually show more interest in the topic of nutrition. Taking into account that the present study was voluntary and was not done randomly, the self-selection bias was inevitable. However, it is noteworthy that our results were consistent with those obtained in a study conducted with an American celiac population. Zipser *et al.* [25] found a prevalence of overweight and obesity of 14% and 4% respectively in American Celiac population, percentages which were a long way from the prevalence of overweight and obesity in the United States. Other recent research carried out in Finland also showed that although the mean BMI is significantly increasing in Western countries and a similar trend is described for celiac patients, a more favourable BMI [26] is found in the latter population.

Another important aspect in assessing the nutritional status of celiac was the dietary intake evaluation. Nowadays there are no databases that include specific composition of GFP. Furthermore, gluten-free product labels do not indicate micronutrient (vitamin and mineral) content. As stated before, in the present study an estimation using homologous gluten-containing products was carried out. This strategy has also been used by other authors, publishing some results in good accordance to ours [27].

According to data obtained from the three day records, the macronutrient imbalance in the celiac population was quite predictable considering that it also exists in the general population. In fact, the national survey of dietary intake of Spain (ENIDE) showed that a similar percentage of energy intake comes from protein, and a slightly larger one from fats (42.2%) in the Spanish population—control women [17]. These similarities between GFD energy distribution and that of the general population have been reported in other studies conducted in Europe [28,29].

It is important to remark that the low consumption of CHO detected in celiac women by our study, was accentuated in general Spanish population—control women. Furthermore, it must be considered that the percentage contribution of carbohydrates to total energy has steadily decreased in recent years. This fact is due at least in part to reductions in cereal and grain intake. The ENIDE survey indicated that 39% of total energy from CHO came from cereal, and that this food group contributed 17% (394 kcal) of total energy, which is comparable to the data reached by our celiac group (37,9% of total carbohydrate and 23% of total energy; 417 kcal) [17]. Both sets of data, the ENIDE survey and our study, showed cereal and grain consumption below Spanish data registered in 2006 and in previous years [30].

With the outcomes obtained from celiac women *vs.* general women population—control women it could be hypothesized that the dietary habits of celiac women were healthier than those of the general population. Zuccotti *et al.* [29] compared dietary intake of celiac children to a control group, suggesting a similar conclusion. Food frequency questionnaires of that study revealed a difference in eating habits. Indeed, celiac children consumed more bread and rice, accompanied by greater gluten-free product consumption than controls [29]. Nevertheless, it is necessary to point out that celiac women from our research did not consume such a large amount of commercially available gluten-free foods as those children did.

As suggested by Subar *et al.* [31], in the general United States population grain foods contribute a large percentage to the adult daily intake of several nutrients including thiamine, riboflavin, niacin, folate, iron, and fiber [31]. Therefore, non-consumption of the recommended amount of whole grain products could have important implications for dietary intakes of B-vitamins, iron, and fiber.

With regard to our results, the intake of dietary fiber in a GFD was a long way from the recommended amount and it was even much lower than Spanish women's consumption—control women. This fact was not surprising, taking into account that GFD and GFP are often formulated and produced in low fiber forms to avoid the risk of gluten cross-contamination [32]. Unlike our results, in a recent study conducted with the German celiac population, dietary fiber intake was very similar to that of the general German population [33], which suggests that perhaps commercial gluten-free food contains added fiber in that country. However, another study conducted with Spanish families [34] indicated that the average fiber consumption is about 16.4 g/day, which would be a similar amount to that found in the celiac women population. Therefore it could be postulated that adult celiac women share nutritional goals with the rest of the Spanish population, at least for this component of the diet.

As micronutrients are concerned, if we compare the results of the celiac population with Spanish general population—control women, the data are similar to those reported in the ENIDE study [17]. Vitamin D and iodine represent the most important deficiencies in the celiac women of this study [14].

The compliance rate for recommended vitamin D consumption, both for the celiac as well as for the general population, is around 70%–80% of RDI. The lack of vitamin D can alter the normal metabolism of the bone, which can lead to rickets in children or osteoporosis or osteomalacia in adults [35]. In fact, numerous studies indicate that a suitable intake of calcium is crucial in the celiac group [36,37]. In this sense, the data showed that 57% of celiacs do not obey the FESNAD DRI for calcium [14]. A review of the topic suggested that up to 75% of celiac people exhibit low bone density and increased risk of fractures [38]. What is more, a study of persistent mucosal damage and the risk of fracture in celiac disease concluded that the association between persistent villous atrophy and hip fractures implies thinner subcutaneous tissue [39]. Taking into account that our result showed that 6% of participants were underweight and deficient in vitamin D intake, the risk of fractures in our cohort is more than probable.

Similarly, as in the Spanish general population—control women, there was a deficiency of iodine in the celiac participants (94% of them demonstrated iodine deficiency according to FESNAD's recommendations) [14]. In good accordance with the result obtained, remarkably low iodine intake was also found in Europe in 2010 for the general population [40]. Nevertheless, it is important to point out that celiac disease is accompanied by other pathologies related to thyroid function, where adequate iodine consumption is a limiting factor to avoid clinical consequences [41].

In the case of vitamin A, this micronutrient had a low rate of compliance in celiac women, even though the average intake was greater than in the Spanish general population—control women (ENIDE study) [17]. For the rest of the minerals and vitamins analyzed, recommended intakes were reached (Table 3) [14]. Although, riboflavin results show that the celiac population had doubled RDI [14], there is no evidence of adverse effects due to excessive intake of riboflavin, possibly due to a limited extent of absorption in the intestine and its rapid excretion in urine.

Vitamin 6, folate, and vitamin B12 deficiencies commonly lead to moderate elevations in total plasma homocysteine levels which, in turn, may increase the tendency to develop occlusive venous and arterial disease in both celiac people and the general population [42]. In our research, the recommendations for vitamin B6, folate, and vitamin B12 were amply fulfilled by celiac women. Taking into account that it has been demonstrated that most bread, pastas, and cold cereals are not fortified with folate for instance [43], the intake of this vitamin, like that of B12 and B6, must have been compensated by other sources.

The frequency consumption questionnaire conducted revealed the daily or weekly consumed rations of different food groups. Although macronutrient distribution of the diet was similar to the rest of the Spanish population [16], this questionnaire highlighted that the food source of these nutrients was different. CHO in the general population should come from cereals, fruits and pulses/legumes, respectively. As represented in Figure 3, celiac women consumed a very low amount of cereals, consequently in the case of celiac people the pulses and legumes group had greater importance as sources of CHO. Thus, if we compare the consumption of legumes of celiac people in our research with Spanish general population [30], it is possible to point out the great difference between groups (19.3 g/day *vs.* 11.9 g/day). The strategy followed by celiac people consists in focussing on avoiding gluten in the diet, promoting the consumption of legumes instead of cereals [44].

In relation to the intake of vegetables, although only 19% of celiac adults consume the daily minimum of two servings these data are in good accordance with the rest of the Spanish adult population—control women [17]. More than three quarters of participants reported excessive (more than three servings) consumption of meat and meat products per week. Additionally, rather than celiac people consuming one serving per week of fatty meat/red meat, the data obtained revealed that half of their meat consumption was pork, beef, or lamb. Nevertheless, the daily consumption of meat by celiac patients was lower than that of the general Spanish population (110 g/day *vs.* 179 g/day) [30].

The PREDIMED trial among others, has demonstrated that following a Mediterranean Diet can be considered a sustainable ideal model for cardiovascular disease prevention [45]. According to data published in the ENIDE study, celiac women demonstrated better adhesion to the Mediterranean diet than did the general Spanish women population—control women. The percentage of celiac women with low adherence to the Mediterranean dietary pattern (less than index 4) was 30%, while in the Spanish women population—control women this reached 62%. Consequently, celiac women showed higher percentages of intermediate (indexes 4–6) and high (>6) Mediterranean-Diet Score; 56 *vs.* 31 and 13 *vs.* 7 respectively. In good accordance with the results obtained in the present work, a recent study conducted with Spanish women—control women established a relationship between higher adhesion to the Mediterranean diet and decreased risk of overweight or obesity [46].

In summary, as far as our result is concerned there are marked differences between the body composition of adult celiac and non-celiac women. Furthermore it could be postulated that energy and macronutrient intake of celiac women follows trends similar to those found in Spanish women—control women as a whole, with an imbalanced distribution of macronutrients and an inadequate consumption of certain micronutrients and fiber (Table 4).

Table 4. Summary of Spanish societies recommendations' compliance concerning energy intake, obese and overweight percentage, nutrients, fiber and food consumption frequency by celiac women and Spanish women.

<table>
<tr><th rowspan="2"></th><th colspan="2">Compliance/Suggestion</th></tr>
<tr><th>Celiac Women</th><th>Spanish Women—Control Women</th></tr>
<tr><td>Energy intake (kcal)</td><td colspan="2">√</td></tr>
<tr><td>Obese and overweight (%)</td><td>√</td><td>×/↓</td></tr>
<tr><td>Macronutrient distribution</td><td></td><td></td></tr>
<tr><td>Protein (%)</td><td colspan="2">×/↓</td></tr>
<tr><td>Carbohydrate (%)</td><td colspan="2">×/↑</td></tr>
<tr><td>Fat (%)</td><td colspan="2">×/↓</td></tr>
<tr><td>Fiber (g)</td><td>×/↑↑</td><td>×/↑</td></tr>
<tr><td>Micronutrient intake</td><td></td><td></td></tr>
<tr><td>Vitamin D (μg)</td><td colspan="2">×/↑</td></tr>
<tr><td>Vitamin E (mg)</td><td colspan="2">×/↑</td></tr>
<tr><td>Folate (μg)</td><td colspan="2">×/↑</td></tr>
<tr><td>Calcium (mg)</td><td colspan="2">×/↑</td></tr>
<tr><td>Iron (mg)</td><td colspan="2">×/↑</td></tr>
<tr><td>Iodine (μg)</td><td colspan="2">×/↑</td></tr>
<tr><td>Selenium (μg)</td><td colspan="2">×/↑</td></tr>
<tr><td>Food consumption frequency</td><td></td><td></td></tr>
<tr><td>Grains</td><td>×/↑</td><td>√/=</td></tr>
<tr><td>Legumes</td><td>√/=</td><td>×/↑</td></tr>
<tr><td>Vegetables</td><td colspan="2">×/↑</td></tr>
</table>

Notes: ×, values do not achieve Spanish societies (FESNAD and SENC) recommendations; √ values do achieve Spanish societies (FESNAD and SENC) recommendations; ↑ it is suggested an intake increase; ↓ it is suggested an intake reduction; FESNAD, Federation of Spanish Societies of Nutrition and Dietetics; SENC, Spanish Society of Community Nutrition.

5. Conclusions

Even though specific nutritional education is not necessary for celiac people some specific consideration must be provided in order to improve eating habits and nutritional status among adult celiac women.

Acknowledgments

This research was supported by a grant from the University of the Basque Country, Vitoria-Gasteiz, Spain (UPV/EHU) (University-Society US12/10). We thank the collaboration of "Federación Española de Asociaciones de Celiacos (FACE), Madrid, Spain", specifically that of Mireia Apraiz, and the efforts of dieticians Beatriz Zulueta, Nerea Segura and Estibaliz Olabarri.

Author Contributions

Itziar Churruca and Edurne Simon conceived and designed the experiments; Arrate Lasa and Idoia Larretxi performed the experiments; Jonatan Miranda analyzed the data; María Á. Bustamante contributed materials/analysis tools; Itziar Churruca wrote the paper.

Conflicts of Interest

The authors declare no conflict of interest.

References

1. Ludvigsson, J.F.; Green, P.H. Clinical management of coeliac disease. *J. Int. Med.* **2011**, *269*, 560–571. [CrossRef] [PubMed]
2. Catassi, C.; Gatti, S.; Fasano, A. The new epidemiology of celiac disease. *J Pediatr. Gastroenterol. Nutr.* **2014**, *59*, S7–S9. [CrossRef] [PubMed]
3. Catassi, C.; Fabiani, E.; Iacono, G.; D'Agate, C.; Francavilla, R.; Biagi, F.; Volta, U.; Accomando, S.; Picarelli, A.; de Vitis, I.; *et al.* A prospective, double-blind, placebo-controlled trial to establish a safe gluten threshold for patients with celiac disease. *Am. J. Clin. Nutr.* **2007**, *85*, 160–166. [PubMed]
4. Rajpoot, P.; Makharia, G.K. Problems and challenges to adaptation of gluten free diet by Indian patients with celiac disease. *Nutrients* **2013**, *5*, 4869–4879. [CrossRef] [PubMed]
5. Gibert, A.; Kruizinga, A.G.; Neuhold, S.; Houben, G.F.; Canela, M.A.; Fasano, A.; Catassi, C. Might gluten traces in wheat substitutes pose a risk in patients with celiac disease? A population-based probabilistic approach to risk estimation. *Am. J. Clin. Nutr.* **2013**, *97*, 109–116. [CrossRef] [PubMed]
6. American-Dietetic-Association. Celiac disease. In *Evidence Based Nutrition Practice Guideline*; American-Dietetic-Association: Chicago, IL, USA, 2009.
7. Saturni, L.; Ferretti, G.; Bacchetti, T. The gluten-free diet: Safety and nutritional quality. *Nutrients* **2010**, *2*, 16–34. [CrossRef] [PubMed]
8. Pietzak, M.M. Follow-up of patients with celiac disease: Achieving compliance with treatment. *Gastroenterology* **2005**, *128*, S135–S141. [CrossRef] [PubMed]
9. Valdimarsson, T.; Toss, G.; Löfman, O.; Ström, M. Three years' follow-up of bone density in adult coeliac disease: Significance of secondary hyperparathyroidism. *Scand. J. Gastroenterol.* **2000**, *35*, 274–280. [CrossRef] [PubMed]

10. World Health Organization (WHO). Obesity: Preventing and managing the global epidemic. In *Report of a WHO Consultation on Obesity*; World Health Organization: Geneva, Switzerland, 1997; p. 276.
11. Gallagher, D.; Heymsfield, S.B.; Heo, M.; Jebb, S.A.; Murgatroyd, P.R.; Sakamoto, Y. Healthy percentage body fat ranges: An approach for developing guidelines based on body mass index. *Am. J. Clin. Nutr.* **2000**, *72*, 694–701. [PubMed]
12. Miranda, J.; Lasa, A.; Bustamante, M.A.; Churruca, I.; Simon, E. Nutritional differences between a gluten-free diet and a diet containing equivalent products with gluten. *Plant. Foods. Hum. Nutr.* **2014**, *69*, 182–187. [CrossRef] [PubMed]
13. Russolillo, G.; Marques, I. *Food Portion Sizes Album*; Imagen Comunicación Multimedia: Madrid, Spain, 2008; p. 186.
14. Federation of Spanish Societies of Nutrition and Dietetics (FESNAD). Dietary reference intakes (DRI) for spanish population, 2010. *Act Diet.* **2010**, *14*, 196–197.
15. Ross, C.; Taylor, C.; Yaktine, A.; del Valle, H. *Dietary Reference Intakes for Calcium and Vitamin D*; The National Academies Press: Washington, DC, USA, 2011; p. 1132.
16. Aranceta, J.; Serra-Majem, L. Dietary guidelines for the Spanish population. *Public. Health. Nutr.* **2001**, *4*, 1403–1408. [CrossRef] [PubMed]
17. Nutritional Assessment Studies: Spanish Population Dietary Intakes. National Health Survey. ENIDE. Available online: http://aesan.msssi.gob.es/AESAN/web/evaluacion_riesgos/seccion/estudios_evaluacion_nutricional.shtml (accessed on 25 May 2015).
18. Trichopoulou, A.; Costacou, T.; Bamia, C.; Trichopoulos, D. Adherence to a Mediterranean diet and survival in a Greek population. *N. Engl. J. Med.* **2003**, *348*, 2599–2608. [CrossRef] [PubMed]
19. Serra-Majem, L.; Aranceta, J. Nutritional objectives for the Spanish population: Consensus from the Spanish Society of Community Nutrition. *Public Health Nutr.* **2001**, *4*, 1409–1413. [CrossRef] [PubMed]
20. Bardella, M.T.; Fredella, C.; Prampolini, L.; Molteni, N.; Giunta, A.M.; Bianchi, P.A. Body composition and dietary intakes in adult celiac disease patients consuming a strict gluten-free diet. *Am. J. Clin. Nutr.* **2000**, *72*, 937–939. [PubMed]
21. Pedrera-Zamorano, J.D.; Roncero-Martin, R.; Lavado-Garcia, J.M.; Calderon-Garcia, J.F.; Rey-Sanchez, P.; Vera, V.; Martinez, M.; Moran, J.M. Segmental fat-free and fat mass measurements by bioelectrical impedance analysis in 2224 healthy Spanish women aged 18–85 years. *Am. J. Hum. Biol.* **2014**. [CrossRef]
22. Wierdsma, N.J.; van Bokhorst-de van der Schueren, M.A.; Berkenpas, M.; Mulder, C.J.; van Bodegraven, A.A. Vitamin and mineral deficiencies are highly prevalent in newly diagnosed celiac disease patients. *Nutrients* **2013**, *5*, 3975–3992. [CrossRef] [PubMed]
23. Lanzini, A.; Lanzarotto, F.; Villanacci, V.; Mora, A.; Bertolazzi, S.; Turini, D.; Carella, G.; Malagoli, A.; Ferrante, G.; Cesana, B.M.; *et al.* Complete recovery of intestinal mucosa occurs very rarely in adult coeliac patients despite adherence to gluten-free diet. *Aliment. Pharmacol. Ther.* **2009**, *29*, 1299–1308. [CrossRef] [PubMed]

24. Hallert, C.; Grant, C.; Grehn, S.; Grännö, C.; Hultén, S.; Midhagen, G.; Ström, M.; Svensson, H.; Valdimarsson, T. Evidence of poor vitamin status in coeliac patients on a gluten-free diet for 10 years. *Aliment. Pharmacol. Ther.* **2002**, *16*, 1333–1339. [CrossRef] [PubMed]
25. Zipser, R.D.; Patel, S.; Yahya, K.Z.; Baisch, D.W.; Monarch, E. Presentations of adult celiac disease in a nationwide patient support group. *Dig. Dis. Sci.* **2003**, *48*, 761–764. [CrossRef] [PubMed]
26. Ukkola, A.; Mäki, M.; Kurppa, K.; Collin, P.; Huhtala, H.; Kekkonen, L.; Kaukinen, K. Changes in body mass index on a gluten-free diet in coeliac disease: A nationwide study. *Eur. J. Int. Med.* **2012**, *23*, 384–388. [CrossRef] [PubMed]
27. Pellegrini, N.; Agostoni, C. Nutritional aspects of gluten-free products. *J. Sci Food. Agric.* **2015**. in press.
28. Martin, J.; Geisel, T.; Maresch, C.; Krieger, K.; Stein, J. Inadequate nutrient intake in patients with celiac disease: Results from a German dietary survey. *Digestion* **2013**, *87*, 240–246. [CrossRef] [PubMed]
29. Zuccotti, G.; Fabiano, V.; Dilillo, D.; Picca, M.; Cravidi, C.; Brambilla, P. Intakes of nutrients in Italian children with celiac disease and the role of commercially available gluten-free products. *J. Hum. Nutr. Diet.* **2013**, *26*, 436–444. [CrossRef] [PubMed]
30. Varela-Moreiras, G.; Avila, J.M.; Cuadrado, C.; del Pozo, S.; Ruiz, E.; Moreiras, O. Evaluation of food consumption and dietary patterns in Spain by the Food Consumption Survey: Updated information. *Eur. J. Clin. Nutr.* **2010**, *64*, S37–S43. [CrossRef] [PubMed]
31. Subar, A.F.; Krebs-Smith, S.M.; Cook, A.; Kahle, L.L. Dietary sources of nutrients among US adults, 1989 to 1991. *J. Am. Diet. Assoc.* **1998**, *98*, 537–547. [CrossRef]
32. Gallagher, E.; Gormley, T.R.; Arendt, E.K. Recent advances in the formulation of gluten-free cereal-based products. *Trends Food. Sci. Technol.* **2004**, *15*, 143–152. [CrossRef]
33. Martin, U.; Mercer, S.W. A comparison of general practitioners prescribing of gluten-free foods for the treatment of coeliac disease with national prescribing guidelines. *J. Hum. Nutr. Diet.* **2014**, *27*, 96–104. [CrossRef] [PubMed]
34. Ruiz-Roso Calvo de Mora, B.; Perez-Olleros Conde, L. Preliminary results on dietary fibre intake in Spain and health benefits associated to soluble fiber intake. *Res. Esp Nutr. Comunitaria* **2010**, *16*, 147–153. [CrossRef]
35. Yoshida, T.; Stern, P.H. How vitamin D works on bone. *Endocrinol. Metab. Clin. North. Am.* **2012**, *41*, 557–569. [CrossRef] [PubMed]
36. Cellier, C.; Flobert, C.; Cormier, C.; Roux, C.; Schmitz, J. Severe osteopenia in symptom-free adults with a childhood diagnosis of coeliac disease. *Lancet* **2000**. [CrossRef]
37. Kalayci, A.G.; Kansu, A.; Girgin, N.; Kucuk, O.; Aras, G. Bone mineral density and importance of a gluten-free diet in patients with celiac disease in childhood. *Pediatrics* **2001**, *108*, E89. [CrossRef] [PubMed]
38. Lucendo, A.J.; García-Manzanares, A. Bone mineral density in adult coeliac disease: An updated review. *Rev. Esp. Enferm. Dig.* **2013**, *105*, 154–162. [CrossRef] [PubMed]
39. Lebwohl, B.; Michaëlsson, K.; Green, P.H.; Ludvigsson, J.F. Persistent mucosal damage and risk of fracture in celiac disease. *J. Clin. Endocrinol. Metab.* **2014**, *99*, 609–616. [CrossRef] [PubMed]

40. Zimmermann, M.B.; Andersson, M. Prevalence of iodine deficiency in Europe in 2010. *Ann. Endocrinol.* **2011**, *72*, 164–166. [CrossRef] [PubMed]
41. Kalyoncu, D.; Urganci, N. Antithyroid antibodies and thyroid function in pediatric patients with celiac disease. *Int. J. Endocrinol.* **2015**. [CrossRef] [PubMed]
42. Hadithi, M.; Mulder, C.J.; Stam, F.; Azizi, J.; Crusius, J.B.; Peña, A.S.; Stehouwer, C.D.; Smulders, Y.M. Effect of B vitamin supplementation on plasma homocysteine levels in celiac disease. *World. J. Gastroenterol.* **2009**, *15*, 955–960. [CrossRef] [PubMed]
43. Thompson, T. Folate, iron, and dietary fiber contents of the gluten-free diet. *J. Am. Diet. Assoc.* **2000**, *100*, 1389–1396. [CrossRef]
44. Mlynekov, Z.; Chrenkov, Mr.; Formelov, Z. Cereals and Legumes in Nutrition of People with Celiac Disease. *Int. J. Celiac Dis.* **2014**, *2*, 105–109.
45. Martínez-González, M.A.; Salas-Salvadó, J.; Estruch, R.; Corella, D.; Fitó, M.; Ros, E. Benefits of the Mediterranean Diet: Insights from the PREDIMED Study. *Prog. Cardiovasc. Dis.* **2015**, *58*, 50–60. [CrossRef] [PubMed]
46. Sayón-Orea, C.; Santiago, S.; Cuervo, M.; Martínez-González, M.A.; Garcia, A.; Martínez, J.A. Adherence to Mediterranean dietary pattern and menopausal symptoms in relation to overweight/obesity in Spanish perimenopausal and postmenopausal women. *Menopause* **2014**, *22*, 750–757. [CrossRef] [PubMed]

Manuscript 2

I. Larretxi, E. Simón, L. Benjumea, J. Miranda, M.A Bustamante, A. Lasa and F.J Eizaguirre

Gluten-free-rendered products contribute to imbalanced diets in children and adolescents with celiac disease

***European Journal of Nutrition* 2019 Mar; 58(2):775-783**

Gluten-free rendered products contribute to imbalanced diets in children and adolescents with celiac disease

I. Larretxi, E. Simon, L. Benjumea, J. Miranda, M.A. Bustamante, A. Lasa*, F. J. Eizaguirre, I. Churruca

1 Gluten Analysis Laboratory of the University of the Basque Country, Department of Nutrition and Food Science, University of the Basque Country (UPV/EHU), Vitoria, Spain
2 Department of Pediatric Gastroenterology, Hospital Universitario Donostia, San Sebastián, Spain.

Running title: Gluten-free rendered products and diet

*Corresponding author/Request for reprints
A. Lasa PhD. Department of Nutrition and Food Science, Faculty of Pharmacy, University of the Basque Country (UPV/EHU), Paseo de la Universidad, 7, 01006 Vitoria-Gasteiz (Spain).
Telephone number: +34-945-014070/ Fax number: +34-945-013014
e-mail address: arrate.lasa@ehu.eus/ ORCID code: 0000-0003-2399-4979

Acknowledgements
This research was supported by grants from the University of the Basque Country (University-Society US12/10 and US15/06) and a grant from the Basque Government (Proyectos de Investigación Focalizada Agricultura PA15/01). Idoia Larretxi is a PhD scholar of the University of the Basque Country, UPV/EHU (Euskararen eta Etengabeko Prestakuntzaren arloko Errektoreordetza). We thank the collaboration of Hospital Universitario

Donostia and "Federación Española de Asociaciones de Celiacos" (FACE), specifically that of Mireia Apraiz (EZE).

ABSTRACT

Purpose

As well as adhering to the safe limit for gluten intake, a suitable gluten-free (GF) diet must also be nutritionally balanced. However, malnutrition has been observed in the population with celiac disease (CD). This is even more important in the case of children and adolescents, whose GF diet must also ensure their proper growth. The aim of the present study was to assess the diet quality of children and adolescents with CD in order to attain optimal nutritional status, determining the most relevant factors that affect a balanced diet.

Methods

Eighty three children and adolescents with CD (9.2±3.8 years) took part in the study. Height, weight and body composition were measured. An analysis of energy consumption and of the macronutrient distribution of their diet was carried out. Adherence to Mediterranean diet by KIDMED index was analyzed, and energy and nutrients intake.

Results

The diet of participants was not balanced, containing more fat and less carbohydrate than recommended. Most children and adolescents revealed adequate Body Mass Index and suitable body fat percentage. Two-thirds of them showed moderate or poor KIDMED index, the case of girls being remarkable. When the GF diet, containing GF rendered foodstuffs, was compared to a similar type of diet but substituting GF products with their analogues containing gluten, important nutritional differences were revealed.

Conclusions

Even though celiac children and adolescents' diet is unhealthy due to its inappropriate dietary pattern, following a diet based on GF products raises extra difficulty in complying with nutritional recommendations.

Keywords: gluten free products, celiac disease, dietary unbalance, KIDMED index, children, adolescents

INTRODUCTION

Celiac disease(CD) is a permanent, autoimmune disorder that causes a reaction to gluten, which is found in wheat, barley, rye and oats [1].CD has become one of the most common food-related chronic intestinal diseases among children in Europe and the USA [2]. Its estimated prevalence in European children is 1% (1/71 in Spanish children) and it is more frequent among girls [3,4].

At present, the gluten-free (GF) diet is the only treatment available for CD which achieves a resolution of symptoms for the majority of patients. For instance, CD pathogenesis leads to a malabsorption syndrome which implies some nutritional deficiencies [5-7], and following the GF diet could correct these deficiencies in order to guarantee good nutritional health. Apart from maintaining the safe limit of gluten intake, a suitable GF diet must also be nutritionally balanced; it must cover all energy and nutrients requirements to prevent deficiencies and ensure a healthy life. This is even more the case for children and/or adolescents, whose GF diet must also allow appropriate growth [8,9].

An inadequate dietary pattern is a common problem in Western countries that also affects patients with CD. Nutritional deficiencies have been observed in all age-ranges of the population with CD [10]. Patients with CD restricted to a GF diet have to exclude gluten-containing grains and foods such as bread, flour, pasta, breakfast cereals and bakery products from their diet. These products are a major source of energy, protein and carbohydrates but also contain micronutrients, such as iron, zinc, calcium, magnesium and group B vitamins [10-12].

However, all age-range patients with CD tend to consume refined GF cereal products, which do not have the same nutritional composition as their unrefined analogues [13]. Previously, we reported differences in energy, macronutrient, sodium, salt and cholesterol content between some GF rendered and gluten-containing foodstuffs [14]. Moreover, wheat flour products are usually enriched, and thus, GF cereal products often do not contain the same levels of micronutrients, such as thiamine, riboflavin, niacin, folate, vitamin D calcium and iron [10,15,16].

In dietary assessment studies carried out in children following a GF diet, imbalanced dietary intakes have been observed, due to a reduced energy intake with a decreased intake of complex carbohydrate and subsequent higher protein and fat consumption [17-21]. Furthermore, low intake of micronutrients has been also described [18,20,22-24]. Consequently, children with CD can commonly develop complications such as anaemia, related to an insufficient intake of iron and folic acid, and disorders affecting the bone health, associated to a lack of calcium and vitamin D [8,25,26]. Nevertheless, it remains worthwhile to evaluate the possible contribution of specific GF foodstuffs to nutrient balance in children and adolescents with CD following a GF diet.

In this context, the aim of the present study was to assess the dietary quality of children and adolescents with CD in order to seek an optimal nutritional status by determining the most relevant factor that affects their balanced diet. Here, we report the influence of GF rendered products on the achievement of a balance diet in children and adolescents with CD.

MATERIALS AND METHODS

Participants and procedure

"Gluten free-diet and Food Safety" is a prospective SUSFOOD (SUStainable FOOD production and consumption) study conducted in the Basque Country (Spain). Eighty three patients with CD (♀: n=53; ♂: n=30) from 3 to 18 years of age (mean age ± SD: 9.2± 3.8) took part in the present study. They were recruited during 2011 and 2013 from 3 regions of the Basque Country in the north of Spain (Araba, 16.9% of participants; Gipuzkoa, 55.4%; and Bizkaia, 27.7%) in collaboration with the Association of Basque patients with CD. All participants followed a GF diet for at least one year and claimed to be in remission from clinical symptoms. Exclusion criteria included history of chronic diseases such as cardiovascular disease, diabetes, hyperthyroidism/hypothyroidism, hypercholesterolemia, hypertriglyceridemia or high blood pressure levels, and other digestive pathologies which need specific dietetic advice as well as a lack of motivation to participate in the study. Written informed consent was obtained from legal tutors, after receiving information about the survey, for all participants. This study was approved by the Ethical Committee of the University of The Basque Country (CEISH/76/2011 and CEISH/194M/2013).

Anthropometric measurements

Anthropometric measurements were collected by trained nutritionists. Body weight (±10 g) was measured after voiding using a digital integrating scale (SECA 760). Height was measured to the nearest 5 mm using a stadiometer (SECA 220). Body Mass Index (BMI) was calculated from weight and height (kg/m2). Sex- and age- independent BMI standard deviation scores were calculated using z-values for BMI, calculated via the LMS method by Cole *et al* (2000) [27]. Thinness and overweight were defined according to age- and sex-standardized BMI cut-off points based on national and international databases [27-30].

Fat mass, energy expenditure and physical activity

For all subjects, fat mass percentage was estimated with a direct segmental multiple-frequency bioelectrical impedance analysis method (Inbody 230; Biospace, Seoul, Korea). Participants were controlled to void urinary bladder after a fast > 1.5 h to the experiment. Two skin electrodes were placed on the feet and two electrodes on the hands. According to the standard procedure, whole-body resistance and reactance were measured.

Weight, height and age were used to calculate individual energy expenditure according to the Harris-Benedict formula. Standard activity levels value was applied.

Physical activity (PA) fulfilling was assessed by World Health Organization recommendation (>60 min/day of moderate to vigorous activities) [31].

Dietary assessment

Dietary intake was assessed using 3 days 24-hour food recalls (24HR), two on weekdays and one at the weekend, and a 136-item food frequency questionnaire (FFQ). The FFQ was specifically developed for people with CD from validated FFQ for general population [32-34]. Participants were requested to include the name and brand of the consumed GF foodstuffs.

Trained nutritionist-dieticians kept the records for participants, who were accompanied by their parents. Food portions and amounts were determined by the three-pass method, which included a quick list, a detailed description review, and visual aids. In this last case, printed food models, plates, glasses, spoons as wells as household measures or albums of portion photographs were used [35]. Children and adolescents over 12 years self-reported the FFQ and 24HR whereas parents' assistance was needed for younger children. Energy and nutrient intakes were calculated by means of the nutritional software program "Alimentación y Salud" (AyS, Software, Tandem Innova, Inc.). Dietary reference intakes (DRI) for Spanish population issued by the Spanish Societies of Nutrition, Feeding and Dietetics (FESNAD) in 2010 were taken as references for the interpretation of the 24HR [36]. Macronutrient energy distribution of patients with CD was compared to non-celiac children and adolescents of the same region as well as to the recommended percentage in a balanced diet [37,38]. In the case of FFQ, Spanish Society of Community Nutrition (SENC) recommendations were used for the correct interpretation of the results [39].

The composition of specific GF products for patients with CD was retrieved from the product labels and added into the food composition database of the program before calculations, as in previous studies [14]. As GF product labels did not indicate micronutrient content, Missbach *et al* GF product database was used for vitamin and mineral estimation [40].

A simulation of a gluten-containing diet was performed by duplicating recorded GF diets and replacing specific GF foodstuffs by their equivalent gluten-containing foods. Then a nutritional comparison between both diets was carried out. The GF products' brands were those specifically reported by patients with CD. In the case of gluten-containing products, these were available on the same shops as their GF counterparts and had the same name as well as the most similar appearance.

Diet quality was evaluated as adherence to the Mediterranean diet (MeD) by the KIDMED index (Mediterranean Diet Quality Index in children and adolescents). MeD has been widely used as an indicator of healthy dietary habits, due to its good correlation with improved glycemic control and cardiovascular health [41-43].The KIDMED index was based on principles sustaining Mediterranean dietary patterns, as well as those that could potentially impair the MeD, described elsewhere [44]. Levels of adherence were classified into three groups: poor (0–3), moderate (4–7), and high adherence (8–12) to the MeD.

Statistical analysis

Statistical analyses of results were performed by using the IBM SPSS statistical program, version 23 (IBM Inc., Armonk, NY, USA). P values <0.05 were accepted as significant.

The results for continuous variables are given as the arithmetical mean ± standard deviation (SD) and the range. The results for non-continuous variables are given as the frequency and the percentage. Normality in the distribution was assessed by Kolmogorov-Smirnov test and the homogeneity by Levene test.

Linear regression analysis was used to examine the association of adherence to the KIDMED index (poor, moderate and high) with sex, age interval, physical activity fulfilling, province, time following GF diet and adequate weight. Variables with skewed distribution were logarithmically transformed to obtain a more symmetrical distribution.

Statistical analyses of paired means for GF diet and gluten-containing diet was performed with non-parametrical Wilcoxon's test.

RESULTS

Anthropometric measurements

Anthropometric characteristics of the children and adolescents, stratified by age, group and gender, are shown in Table 1. Nearly 70% of the participants presented an adequate BMI, 20% were thin and there were only 9 cases (11%) of overweight. None was obese. Although greater weights were observed in boys, accompanied by a tendency to higher heights, not differences in BMI values were found. Accordingly, body fat percentage was adequate in most cases. There were no differences between genders from 3 to 13 years, but at age of 14-18 body-fat percentages were higher in girls than in boys, as expected.

Healthy behaviour

Healthy behaviour is summarized in Table 2. A low percentage of the population studied (19%) reached physical activity recommendations. By contrast, a small number of girls and boys with CD (less than 5%) followed unhealthy practices such as skipping breakfast, eating commercially baked goods or pastries at this meal or eating more than once a week in fast food restaurants.

Although almost all children and adolescents with CD introduced cereal products into their breakfast, only 33% of the participants ate the minimum of four recommended servings per day of cereals (recommendation: 4-6 portion/day). Along similar lines, around 21% of them ate pasta or rice almost daily. Moreover, two out of ten patients with CD consumed a very small amount of cereals (fewer than two servings). Notwithstanding, grains and cereal derivatives provided 27% of energy intake, GF rendered cereals being the main contributors (24% of total energy intake) (Table 2).

The vast majority of the participants, 86%, did not reach vegetable consumption recommendations (2 portions/day). While the consumption of only one fruit or fruit juice daily was extended among patients with CD, nearly half of them ate the second serving of fruit daily (recommendation: 2-3 portion/day).

With respect to animal origin food, 73% of participants included two or more servings daily of milk or dairy products (recommendation: 2-4 portion/day). However, two out of ten children and adolescents with CD consumed more than four servings/day. Meat and meat product consumption was excessive in 64% of participants (recommendation: 4 portion/week), whereas almost 50% of them reached the weekly recommended amount of fish (3-5 portions/week) and 42% that of eggs (4 portions/week). Animal origin food provided 32% of total energy intake: 14% from milk and dairy products and 18% from meat and meat products, fish and eggs.

Eight out of ten participants included two or more portions of pulses per week (recommendation: 2-4 portion/week). By contrast, regular nut consumption was covered by merely 28% of the sample studied. Regarding other energy sources such as vegetables oils, sugar, chocolate and pastries, most participants consumed them correctly.

KIDMED index

The average of diet quality index, KIDMED index, was 6.8, and was higher for boys than girls (Table 2). While 47% of boys with CD demonstrated high adherence to MeD, only 25% of girls reached this goal. Table 3 represents linear regression statistics, showing the positive association of KIDMED interval (poor, moderate and high) with sex and adequate weight.

Dietary Intakes

The energy intake of children with CD was under their estimated energy expenditure (supplementary Figure 1). They reached, on average, around 80% of recommended energy.

As in the case of non-celiac children and adolescents, the diet of patients with CD was not balanced, containing more fat (39.6%) and less carbohydrate (45.4%) than recommended by the European Food Safety Authority (EFSA) (supplementary Figure 2) [38]. Protein intake was around 17%. As far as micronutrients were concerned, a low percentage (<10%) of participants showed insufficient intakes of thiamin, riboflavin, vitamin B6 and B12, as well as niacin, phosphorus, and magnesium (supplementary Figure 3). By contrast, more than 2/3 of participants did not reach recommendations for vitamin D, and over half of them did not fulfil the recommendations for folic acid and calcium, nor about 1/4 those for iron (supplementary Figure 3).

In the present research, the amount of calories coming from GF rendered foods supposed 24% of the total energy intake. Therefore, it was very important to analyze the potential impact of these products on dietary imbalance. For this purpose, diets containing GF rendered foodstuffs (the ones reported by the participants) were compared to similar diets but substituting those foodstuffs with analogue gluten-containing products. Diet composition was significantly changed, providing more protein and carbohydrate and less fat content in the gluten-containing one (Table 4). With regard to fatty acids, higher contents in polyunsaturated fatty acids (PUFA) and monounsaturated fatty acids (MUFA) and lower saturated fatty acids (SFA) were observed (Table 4). The micronutrient comparison of GF diet against gluten-containing diet indicated higher levels of vitamin E, zinc, potassium and calcium consumption, but lower of vitamin A, thiamine, riboflavin, niacin, vitamin B6, vitamin B12, folate, biotin, vitamin D, pantothenate, magnesium, sodium, iron, cupper, iodine, chlorine, manganese and selenium intake (Table 5).

DISCUSSION

It has been previously demonstrated that diets followed by adults with CD can lead to imbalances in their macronutrient intake and to vitamin and mineral deficiencies [5,45-47]. Therefore, it could be suggested that patients with CD tend to develop nutrition-related pathologies such as hypercholesterolemia, osteoporosis or anemia [25,26,46]. This matter gains even more importance in children or adolescents with CD, who often have suboptimal nutritional status at the time of diagnosis [8] and should meet all nutrient intake recommendations in order to ensure their growth and development.

With similar data to those observed in adult celiac studies, children and adolescents from the present study followed an unbalanced diet (excessive fat, insufficient carbohydrate). However, most participants revealed adequate BMI and body fat percentage. Furthermore, a lower BMI than those reported for Spanish children and adolescents was observed. National research studies in adolescents found a prevalence of overweight, including obesity, of 25.7% in boys and 19.1% in girls [48] whereas its prevalence in children reached 26.7% in boys and 25.7% in girls [49]. By contrast, 11% of young people with CD were overweight with no differences in gender. Even though authors such as Babio *et al*. did not find differences in BMI values between celiac and non-celiac adolescents, other studies have indicated that children on a GF diet have lower weight, height and BMI compared to age- and sex-matched controls [50,51].

Dietary pattern is one of the main factors that could explain the observed unbalanced intakes of macronutrients, as well as deficiencies in mineral and vitamins. For this reason, we assessed the adherence to the MeD by KIDMED index in order to evaluate the adequacy of the healthy behaviour - dietary and lifestyle- of children and adolescents with CD [42,52].

Most of the participants showed a moderate KIDMED index. With regard to dietary pattern, cereal consumption as well as fruit and vegetables was low in children and adolescents with CD, highlighting that the majority of participants did not fulfil recommendations (almost 50% for fruit and 86% for vegetables). By contrast, foodstuffs of animal origin, especially meat, were eaten to excess, representing on average 32% of the total calorie consumption. These data are in good accordance with those published by other authors, who observed that adolescents with CD consumed less starch and more animal products (meat, fish and eggs) than children without CD [51]. Taken together, these results could suggest that celiac children and adolescents tend to replace cereals and, in general, plant origin foods in the diet with animal products.

Data concerning PA showed that only almost 20% of participants fulfilled the WHO`s recommendations (>60 min/day of moderate to vigorous activities) [31]. Similarly, Babio *et al.* observed that 32% of their celiac patients were active or very active (very active being the equivalent to fulfilling WHO recommendations)[51]. As in the present study, other studies carried out among Spanish adolescents pointed out the gender difference related to PA, with significantly more boys being active than girls (32% and 17%, respectively) [53].

After introducing the GF diet, nutritional deficiencies arising from the malabsorption syndrome of CD could be corrected [8]. Nevertheless, it has been demonstrated that diets followed by patients with CD can lead to imbalances in macronutrient intake and to vitamin and mineral deficiencies [5,45-47]. Although dietary pattern could be one explanation for malnutrition, other factors could play a role. Taking into account that several studies have revealed higher amounts of fat and lower quantities of protein, carbohydrate and several vitamins and minerals in GF products [14,40,54,55], the influence of GF product composition cannot be neglected.

In the present research, GF rendered foods provided almost a quarter of the total calorie intake, which is close to data published by others (36%) [50]. This result indicates that, first of all, GF rendered foodstuffs play a major role in the diet of children and adolescents with CD, and secondly, that the impact of these products on nutritional balance compliance should be taken into account.

GF diets resulted in higher fat ingestion (especially due to saturated fat) and lower protein and carbohydrate consumption. Micronutrient intake also proved to be lower when comparing GF diets with their analogues containing gluten. Specifically, vitamin A, thiamine, riboflavin, niacin, vitamin B6, vitamin B12, folate, biotin, vitamin D, pantothenate, magnesium, sodium, iron, cupper, iodine, chlorine, manganese and selenium intakes were lower in GF diets. Other studies among children following a GF diet have suggested unhealthy diets, characterised by low intakes of complex carbohydrate and subsequent higher protein and fat intakes [17-21] as well as lower intake of folic acid, zinc, iron and magnesium [51]. It is important to point out that all the research reported evaluated the diet followed by patients with CD, and not the influence of GF rendered products. As stated before, in the case of the present study the dietary pattern followed by young patients with CD is inappropriate. However, taken as a whole, data from the present study indicate that GF rendered foodstuffs contribute to a higher degree of unbalance among those children and adolescents with CD.

This study has an important limitation. Since there are no food composition tables containing GF rendered foodstuffs, information regarding their energy and macronutrient content was based on our previous research, where data provided on the GF product labels were collected in a database and used for the comparison of these products versus the ones containing gluten [14]. Unfortunately, micronutrient data are not provided in the product labels so the database provided by Missbach *et al*. was used for this purpose [40]. They included GF products from another European market (Germany), therefore the composition could vary slightly from ours (the Spanish market). However, we believe that it is essential to include the specific composition of GF products, in order to assess the real micronutrient intake of GF diet followed by children and adolescents with CD. Otherwise, it would be inaccurate to claim that a GF diet promotes inadequate micronutrient intake, as has been concluded in other studies [5,45-47,51].

In conclusion, the present study revealed that a relevant participant sample of children and adolescents with CD do not meet energy and nutrient recommendations and showed, on average, moderate adherence to the Mediterranean diet. Moreover, the consumption of GF rendered foods increases the imbalances observed in energy and nutrient intakes. Thus, although GF products could guarantee the absence of gluten in the diet, following a diet based on them adds to the difficulty of reaching a nutritional balance for patients with CD, which means that specific nutritional guidelines should be put forward for this collective.

AUTHORSHIP STATEMENT

I. Larretxi and M.A. Bustamante performed the anthropometric measurements and collected questionnaires. Eizaguirre and L. Benjumea recruited the patients, performed anthropometric measurements and collected questionnaires. I. Churruca and E. Simon carried out the experimental design and analyzed data. J. Miranda and A. Lasa wrote the manuscript.

CONFLICTS OF INTEREST

Authors declare no conflict of interest

REFERENCES

1. Bai JC, Ciacci C (2017) World Gastroenterology Organisation Global Guidelines: Celiac Disease February 2017. J Clin Gastroenterol 51 (9):755-768. doi:10.1097/MCG.0000000000000919
2. Ludvigsson JF, Leffler DA, Bai JC, Biagi F, Fasano A, Green PH, Hadjivassiliou M, Kaukinen K, Kelly CP, Leonard JN, Lundin KE, Murray JA, Sanders DS, Walker MM, Zingone F, Ciacci C (2013) The Oslo definitions for coeliac disease and related terms. Gut 62 (1):43-52. doi:10.1136/gutjnl-2011-301346
3. Catassi C, Gatti S, Fasano A (2014) The new epidemiology of celiac disease. J Pediatr Gastroenterol Nutr 59 Suppl 1:S7-9. doi:10.1097/01.mpg.0000450393.23156.59
4. Mariné M, Farre C, Alsina M, Vilar P, Cortijo M, Salas A, Fernández-Bañares F, Rosinach M, Santaolalla R, Loras C, Marquès T, Cusí V, Hernández MI, Carrasco A, Ribes J, Viver JM, Esteve M (2011) The prevalence of coeliac disease is significantly higher in children compared with adults. Aliment Pharmacol Ther 33 (4):477-486. doi:10.1111/j.1365-2036.2010.04543.x
5. Capristo E, Malandrino N, Farnetti S, Mingrone G, Leggio L, Addolorato G, Gasbarrini G (2009) Increased serum high-density lipoprotein-cholesterol concentration in celiac disease after gluten-free diet treatment correlates with body fat stores. J Clin Gastroenterol 43 (10):946-949. doi:10.1097/MCG.0b013e3181978e4d

6. Wierdsma NJ, van Bokhorst-de van der Schueren MA, Berkenpas M, Mulder CJ, van Bodegraven AA (2013) Vitamin and mineral deficiencies are highly prevalent in newly diagnosed celiac disease patients. Nutrients 5 (10):3975-3992. doi:10.3390/nu5103975
7. Dickey W, Kearney N (2006) Overweight in celiac disease: prevalence, clinical characteristics, and effect of a gluten-free diet. Am J Gastroenterol 101 (10):2356-2359. doi:10.1111/j.1572-0241.2006.00750.x
8. Snyder J, Butzner JD, DeFelice AR, Fasano A, Guandalini S, Liu E, Newton KP (2016) Evidence-Informed Expert Recommendations for the Management of Celiac Disease in Children. Pediatrics 138 (3). doi:10.1542/peds.2015-3147
9. Valitutti F, Trovato CM, Montuori M, Cucchiara S (2017) Pediatric Celiac Disease: Follow-Up in the Spotlight. Adv Nutr 8 (2):356-361. doi:10.3945/an.116.013292
10. Wild D, Robins GG, Burley VJ, Howdle PD (2010) Evidence of high sugar intake, and low fibre and mineral intake, in the gluten-free diet. Aliment Pharmacol Ther 32 (4):573-581. doi:10.1111/j.1365-2036.2010.04386.x
11. Whitton C, Nicholson SK, Roberts C, Prynne CJ, Pot GK, Olson A, Fitt E, Cole D, Teucher B, Bates B, Henderson H, Pigott S, Deverill C, Swan G, Stephen AM (2011) National Diet and Nutrition Survey: UK food consumption and nutrient intakes from the first year of the rolling programme and comparisons with previous surveys. Br J Nutr 106 (12):1899-1914. doi:10.1017/S0007114511002340
12. Størsrud S, Hulthén LR, Lenner RA (2003) Beneficial effects of oats in the gluten-free diet of adults with special reference to nutrient status, symptoms and subjective experiences. Br J Nutr 90 (1):101-107. doi: 10.1079/BJN2003872
13. Thompson T, Dennis M, Higgins LA, Lee AR, Sharrett MK (2005) Gluten-free diet survey: are Americans with coeliac disease consuming recommended amounts of fibre, iron, calcium and grain foods? J Hum Nutr Diet 18 (3):163-169. doi:10.1111/j.1365-277X.2005.00607.x
14. Miranda J, Lasa A, Bustamante MA, Churruca I, Simon E (2014) Nutritional differences between a gluten-free diet and a diet containing equivalent products with gluten. Plant Foods Hum Nutr 69 (2):182-187. doi:10.1007/s11130-014-0410-4
15. Thompson T (1999) Thiamin, riboflavin, and niacin contents of the gluten-free diet: is there cause for concern? J Am Diet Assoc 99 (7):858-862. doi:10.1016/S0002-8223(99)00205-9
16. Saturni L, Ferretti G, Bacchetti T (2010) The gluten-free diet: safety and nutritional quality. Nutrients 2 (1):16-34. doi:10.3390/nu20100016

17. Dell'Olio D, Palma L, Malorgio E, Ansaldi Balocco N (1995) What do celiac children eat? Dietary analysis of a group of children with celiac disease on a diet. Minerva Gastroenterol Dietol 41 (4):269-273
18. Ohlund K, Olsson C, Hernell O, Ohlund I (2010) Dietary shortcomings in children on a gluten-free diet. J Hum Nutr Diet 23 (3):294-300. doi:10.1111/j.1365-277X.2010.01060.x
19. Mariani P, Viti MG, Montuori M, La Vecchia A, Cipolletta E, Calvani L, Bonamico M (1998) The gluten-free diet: a nutritional risk factor for adolescents with celiac disease? J Pediatr Gastroenterol Nutr 27 (5):519-523. doi:10.1097/00005176-199811000-00004
20. Hopman EG, le Cessie S, von Blomberg BM, Mearin ML (2006) Nutritional management of the gluten-free diet in young people with celiac disease in The Netherlands. J Pediatr Gastroenterol Nutr 43 (1):102-108. doi:10.1097/01.mpg.0000228102.89454.eb
21. Ferrara P, Cicala M, Tiberi E, Spadaccio C, Marcella L, Gatto A, Calzolari P, Castellucci G (2009) High fat consumption in children with celiac disease. Acta Gastroenterol Belg 72 (3):296-300
22. Rujner J, Socha J, Syczewska M, Wojtasik A, Kunachowicz H, Stolarczyk A (2004) Magnesium status in children and adolescents with coeliac disease without malabsorption symptoms. Clin Nutr 23 (5):1074-1079. doi:10.1016/j.clnu.2003.10.018
23. Blazina S, Bratanic N, Campa AS, Blagus R, Orel R (2010) Bone mineral density and importance of strict gluten-free diet in children and adolescents with celiac disease. Bone 47 (3):598-603. doi:10.1016/j.bone.2010.06.008
24. Kautto E, Ivarsson A, Norström F, Högberg L, Carlsson A, Hörnell A (2014) Nutrient intake in adolescent girls and boys diagnosed with coeliac disease at an early age is mostly comparable to their non-coeliac contemporaries. J Hum Nutr Diet 27 (1):41-53. doi:10.1111/jhn.12125
25. Pietzak MM (2005) Follow-up of patients with celiac disease: achieving compliance with treatment. Gastroenterology 128 (4 Suppl 1):S135-141. doi:10.1053/j.gastro.2005.02.025
26. Valdimarsson T, Toss G, Löfman O, Ström M (2000) Three years' follow-up of bone density in adult coeliac disease: significance of secondary hyperparathyroidism. Scand J Gastroenterol 35 (3):274-280. doi: 10.1080/003655200750024146
27. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH (2000) Establishing a standard definition for child overweight and obesity worldwide: international survey. BMJ 320 (7244):1240-1243. doi:10.1136/bmj.320.7244.1240

28. Cole TJ, Lobstein T (2012) Extended international (IOTF) body mass index cut-offs for thinness, overweight and obesity. Pediatr Obes 7 (4):284-294. doi:10.1111/j.2047-6310.2012.00064.x
29. Cole TJ, Flegal KM, Nicholls D, Jackson AA (2007) Body mass index cut offs to define thinness in children and adolescents: international survey. BMJ 335 (7612):194. doi:10.1136/bmj.39238.399444.55
30. Fernández C, Lorenzo H, Vrotsou K, Aresti U, Rica I, Sánchez E (2011) Curvas y tablas de crecimiento (Estudios Longitudinal y Transversal). Fundación Faustino Orbegozo Eizaguirre, Bilbao
31. World Health Organization (2010) Global recommendations on physical activity for health. WHO, Geneve
32. Bel-Serrat S, Mouratidou T, Pala V, Huybrechts I, Börnhorst C, Fernández-Alvira JM, Hadjigeorgiou C, Eiben G, Hebestreit A, Lissner L, Molnár D, Siani A, Veidebaum T, Krogh V, Moreno LA (2014) Relative validity of the Children's Eating Habits Questionnaire-food frequency section among young European children: the IDEFICS Study. Public Health Nutr 17 (2):266-276. doi:10.1017/S1368980012005368
33. Rodríguez VM, Elbusto-Cabello A, Alberdi-Albeniz M, De la Presa-Donado A, Gómez-Pérez de Mendiola F, Portillo-Baquedano MP, Churruca-Ortega I (2014) Validación de un nuevo registro alimentario precodificado. Revista Española de Nutrición Humana y Dietética; Vol 18, Núm 3 (2014): Revista Española de Nutrición Humana y Dietética. doi:10.14306/renhyd.18.3.84
34. Rodríguez IT, Ballart JF, Pastor GC, Jordà EB, Val VA (2008) Validation of a short questionnaire on frequency of dietary intake: reproducibility and validity. Nutr Hosp 23 (3):242-252
35. Russolillo G, Marques I (2008) Food portion sizes album. Imagen Comunicación Multimedia, Madrid
36. FESNAD (2010) Dietary reference intakes (DRI) for spanish population, 2010. Actividad Dietética 14 (4):196-197. doi:10.1016/S1138-0322(10)70039-0
37. Gobierno Vasco (2005) Encuesta de nutrición 2005. Departamento de Sanidad del Gobierno Vasco, Vitoria-Gasteiz
38. European Food Safety Authority (2017) Dietary Reference Values for nutrients Summary report. EFSA Supporting Publications 14 (12):e15121E-n/a. doi:10.2903/sp.efsa.2017.e15121
39. Aranceta J, Serra-Majem L (2001) Dietary guidelines for the Spanish population. Public Health Nutr 4 (6A):1403-1408
40. Missbach B, Schwingshackl L, Billmann A, Mystek A, Hickelsberger M, Bauer G, König J (2015) Gluten-free food database: the nutritional

quality and cost of packaged gluten-free foods. PeerJ 3:e1337. doi:10.7717/peerj.1337
41. Zhong VW, Lamichhane AP, Crandell JL, Couch SC, Liese AD, The NS, Tzeel BA, Dabelea D, Lawrence JM, Marcovina SM, Kim G, Mayer-Davis EJ (2016) Association of adherence to a Mediterranean diet with glycemic control and cardiovascular risk factors in youth with type I diabetes: the SEARCH Nutrition Ancillary Study. Eur J Clin Nutr 70 (7):802-807. doi:10.1038/ejcn.2016.8
42. Esteban-Cornejo I, Izquierdo-Gomez R, Gómez-Martínez S, Padilla-Moledo C, Castro-Piñero J, Marcos A, Veiga OL (2016) Adherence to the Mediterranean diet and academic performance in youth: the UP&DOWN study. Eur J Nutr 55 (3):1133-1140. doi:10.1007/s00394-015-0927-9
43. Martin-Calvo N, Chavarro JE, Falbe J, Hu FB, Field AE (2016) Adherence to the Mediterranean dietary pattern and BMI change among US adolescents. Int J Obes (Lond) 40 (7):1103-1108. doi:10.1038/ijo.2016.59
44. Serra-Majem L, Ribas L, Ngo J, Ortega RM, García A, Pérez-Rodrigo C, Aranceta J (2004) Food, youth and the Mediterranean diet in Spain. Development of KIDMED, Mediterranean Diet Quality Index in children and adolescents. Public Health Nutr 7 (7):931-935. doi:10.1079/PHN2004556
45. Bardella MT, Fredella C, Prampolini L, Molteni N, Giunta AM, Bianchi PA (2000) Body composition and dietary intakes in adult celiac disease patients consuming a strict gluten-free diet. Am J Clin Nutr 72 (4):937-939
46. Hallert C, Grant C, Grehn S, Grännö C, Hultén S, Midhagen G, Ström M, Svensson H, Valdimarsson T (2002) Evidence of poor vitamin status in coeliac patients on a gluten-free diet for 10 years. Aliment Pharmacol Ther 16 (7):1333-1339. doi: 10.1046/j.1365-2036.2002.01283.x
47. Martin J, Geisel T, Maresch C, Krieger K, Stein J (2013) Inadequate nutrient intake in patients with celiac disease: results from a german dietary survey. Digestion 87 (4):240-246. doi:10.1159/000348850
48. Moreno LA, Mesana MI, Fleta J, Ruiz JR, González-Gross M, Sarría A, Marcos A, Bueno M, Group AS (2005) Overweight, obesity and body fat composition in spanish adolescents. The AVENA Study. Ann Nutr Metab 49 (2):71-76. doi:10.1159/000084738
49. Pérez-Farinós N, López-Sobaler AM, Dal Re M, Villar C, Labrado E, Robledo T, Ortega RM (2013) The ALADINO study: a national study of prevalence of overweight and obesity in Spanish children in 2011. Biomed Res Int 2013:163687. doi:10.1155/2013/163687
50. Zuccotti G, Fabiano V, Dilillo D, Picca M, Cravidi C, Brambilla P (2013) Intakes of nutrients in Italian children with celiac disease and the role

of commercially available gluten-free products. J Hum Nutr Diet 26 (5):436-444. doi:10.1111/jhn.12026
51. Babio N, Alcázar M, Castillejo G, Recasens M, Martínez-Cerezo F, Gutiérrez-Pensado V, Masip G, Vaqué C, Vila-Martí A, Torres-Moreno M, Sánchez E, Salas-Salvadó J (2017) Patients With Celiac Disease Reported Higher Consumption of Added Sugar and Total Fat Than Healthy Individuals. J Pediatr Gastroenterol Nutr 64 (1):63-69. doi:10.1097/MPG.0000000000001251
52. Papadaki S, Mavrikaki E (2015) Greek adolescents and the Mediterranean diet: factors affecting quality and adherence. Nutrition 31 (2):345-349. doi:10.1016/j.nut.2014.09.003
53. Inchley J, Currie D, Young T, Samdal O, Torsheim T, Augustson L, Mathison F, Aleman-Diaz A, Molcho M, Weber M, Barnekow V (2016) Growing up unequal. HBSC 2016 study (2013/2014 survey).
54. Kulai T, Rashid M (2014) Assessment of Nutritional Adequacy of Packaged Gluten-free Food Products. Can J Diet Pract Res 75 (4):186-190. doi:10.3148/cjdpr-2014-022
55. Wu JH, Neal B, Trevena H, Crino M, Stuart-Smith W, Faulkner-Hogg K, Yu Louie JC, Dunford E (2015) Are gluten-free foods healthier than non-gluten-free foods? An evaluation of supermarket products in Australia. Br J Nutr 114 (3):448-454. doi:10.1017/S0007114515002056

Table 1. Anthropometric results of celiac boys and girls aged 3-18 years, stratified by age group and gender.

	3-8 years		9-13 years		14-18 years	
Characteristic	Girls	Boys	Girls	Boys	Girls	Boys
n	20	14	20	11	13	5
Age (year)	5.1 ± 1.5	5.7 ± 1.4	10.2 ± 1.2	10.9 ± 1.2	15.8 ± 1.2	15 ± 0.7
Height (cm)	113 ± 11.3	118 ± 10.5	140 ± 11.1	144 ± 8	160 ± 6.0	174 ± 8.9
Weight (kg)	21.1 ± 4.5	23.0 ± 5.1	37.8 ± 9.9	37.4 ± 7.6	57.9 ± 8.2	62.7 ± 12.6*
Body Fat %	16.4 ± 1.5	16.6 ± 1.4	22.5 ± 7.5	19.8 ± 10.8	25.3± 5.8	13.6 ± 9.2**
Body -mass index:						
Mean (kg/m²)	16.4 ± 1.5	16.6 ± 1.4	18.5 ± 2.6	17.7 ± 2.6	21.2 ± 2.5	20.7 ± 3.7
Z-score	1.0 ± 0.1	1.0 ± 0.1	1.0 ± 0.2	1.0 ± 0.2	1.0 ± 0.2	1.0 ± 0.2
Thinness - no. (%)	3 (15)	2 (14)	5 (25)	4 (36)	2 (13)	1 (20)
Adequate weight - no. (%)	15 (75)	10 (72)	13 (65)	7 (64)	9 (74)	3 (60)
Overweight- no. (%)	2 (10)	2 (14)	2 (10)	0 (0)	2 (13)	1 (20)

Notes: Values are means + Standard Deviation (SD); n: number of subjects. Asterisks represent significant difference between boys and girls: * *P*<0.05; ** *P*<0.01.

Table 2. Dietary habits of study sample according to the KIDMED index.

	All	Girls	Boys
n	83	53	30
Healthy behaviour following			
PA recommendation (%)	19.3	17.0	23.0
Skips breakfast (%)	3.6	3.8	3.3
Dairy product for breakfast (%)	95.2	98.1	90.0
Two yogurts and/or some cheese daily (%)	30.1	32.1	26.7
Cereal or cereal product for breakfast (%)	92.8	92.5	93.3
Pasta or rice almost daily (≥5/wk) (%)	20.5	9.6	10.8
Fresh or cooked vegetables daily (%)	25.3	20.8	33.3
Fresh or cooked vegetables daily-no. (%)	1.2	1.9	0
Fruit or fruit juice daily (%)	86.8	90.0	88.0
Second serving of fruit daily (%)	50.6	50.9	50.0
Regular fish consumption (≥2–3/wk) (%)	84.3	83.0	86.7
More than 1/wk fast food (hamburger) restaurant (%)	3.6	5.7	0.0
Pulses >1/wk (%)	94	90.6	100
Regular nut consumption ((≥2–3/wk) (%)	27.7	28.3	26.7
Use of olive oil at home (%)	90.4	92.5	86.7
Sweets and candy several times a day (%)	7.2	7.5	6.7
Commercially baked goods or pastries for breakfast(%)	1.2	0.0	3.3
KIDMED index (0–16)	6.8 ± 1.6	6.6 ± 1.6	7.0 ± 1.5
KIDMED index (%)			
Poor (≤3)	2.4	3.8	0.0
Moderate (4–7)	63.9	44.6	19.3
High (≥8)	33.7	26.4	46.7
Gluten-free habits			
Time following GF diet (years)	6.43 ± 4.18	6.56 ± 4.53	6.20 ± 3.54
Energy intake from GF rendered foods (Kcal)	478 ±186	487 ± 161	463 ± 227
Percentage of daily total energy intake	24.3%	23.8%	24.3%

Notes: Values are mean ± SD or percentages; GF. Gluten-free; PA. physical activity.

Table 3. Lineal regression coefficients (β) and standard errors (SE) considering the associations of KIDMED index interval with sex, age interval, physical activity fulfilling, province, time following gluten-free diet and adequate weight.

	β	**SE**	***P***
Sex	0.31	0.13	0.02
Age interval (3-8, 9-13, 14-18)	0.13	0.12	NS
PA fulfilling	-0.03	0.16	NS
Province	-0.05	0.08	NS
Adequate weight	0.38	0.13	<0.01
Time following GF diet [a]	-0.26	0.24	NS

Notes: Normal weigh consideration was determined by Orbegozo cut-offs values and physical activity (PA) fulfilling by World Health Organization recommendation[30,31]
[a] Analysis was performed with log-transformed data.

Table 4. Energy and macronutrient intake comparison between following gluten free and gluten containing diet.

	Gluten free diet		Gluten containing diet		
	Mean	**SD**	**Mean**	**SD**	***P***
Energy (Kcal/d)	1952	344	1979	438	NS
Protein (g/d)	*79.8*	*17.0*	*83.5*	*17.2*	*< 0.001*
%	*16.7*	*2.6*	*17.4*	*2.4*	*< 0.001*
Fat (g/d)	*86.2*	*19.6*	*83.2*	*19.5*	*< 0.001*
%	*40.1*	*5.9*	*38.9*	*5.0*	*< 0.001*
MUFA (g/d)	*31.1*	*8.9*	*33.1*	*9.1*	*< 0.001*
%	*46.9*	*6.0*	*48.3*	*5.3*	*< 0.001*
PUFA (g/d)	*8.2*	*4.0*	*9.4*	*4.3*	*< 0.001*
%	*12.2*	*4.2*	*13.6*	*4.2*	*< 0.001*
SFA (g/d)	*27.1*	*7.9*	*25.9*	*7.4*	*0.002*
%	*40.9*	*6.6*	*37.9*	*5.9*	*< 0.001*
Carbohydrate (g/d)	*219.0*	*47.1*	*223.2*	*42.2*	*< 0.001*
%	*42.6*	*6.4*	*43.8*	*5.4*	*< 0.001*

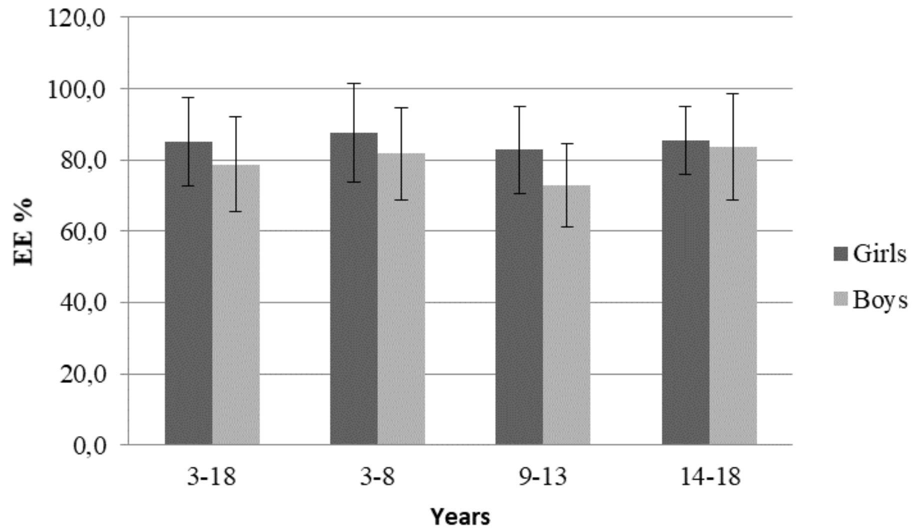
Notes: in italic, nutrients with significance level $P < 0.05$; Kcal/d, kilocalories/day; g/d. grams/day; MUFA, monounsaturated fatty acids; NS, not significance; PUFA, polyunsaturated fatty acids; SD, standard deviation; SFA, saturated fatty acids.

Table 5. Micronutrient intake comparison between following gluten free and gluten containing diet.

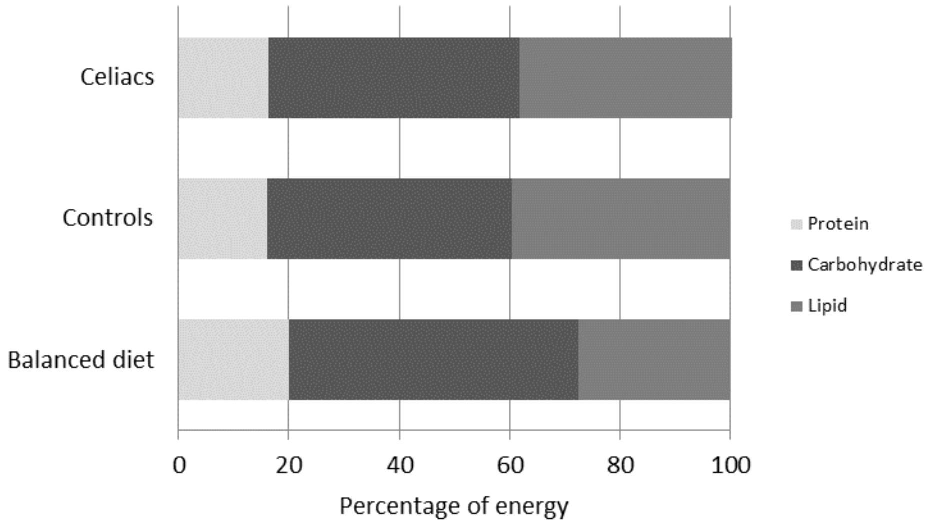
	Gluten free diet		Gluten containing diet		
	Mean	**SD**	**Mean**	**SD**	***P***
Vitamin A (µg/d)	*555*	*224*	*602*	*231*	*< 0.001*
Thiamin (mg/d)	*1.3*	*0.4*	*1.4*	*0.5*	*< 0.001*
Riboflavin (mg/d)	*1.6*	*0.4*	*1.8*	*0.5*	*< 0.001*
Vitamin B6 (mg/d)	*1.6*	*0.4*	*1.9*	*0.5*	*< 0.001*
Vitamin B12 (µg/d)	*5.9*	*2.9*	*6.8*	*3.1*	*< 0.001*
Vitamin C (mg/d)	89.6	46.0	92.2	48.1	0.9
Vitamin D (µg/d)	*3.3*	*3.5*	*3.8*	*3.3*	*< 0.001*
Vitamin E (mg/d)	*10.4*	*3.9*	*9.6*	*3.4*	*< 0.001*
Niacin (mg/d)	*21.2*	*5.6*	*23.7*	*6.2*	*< 0.001*
Pantothenate (mg/d)	*4.1*	*1.1*	*4.6*	*1.4*	*< 0.001*
Biotin (µg/d)	*4.2*	*2.3*	*4.3*	*2.3*	*< 0.001*
Folate (µg/d)	*186*	*76*	*233*	*88*	*< 0.001*
Sodium (mg/d)	*1819*	*529*	*1932*	*533*	*< 0.001*
Potassium (mg/d)	*2788*	*585*	*2747*	*565*	*0.001*
Calcium (mg/d)	*900*	*217*	*887*	*219*	*0.007*
Phosphorus (mg/d)	1297	287	1298	285	0.7
Magnesium (mg/d)	242	66	279	66	< 0.001
Iron (mg/d)	12.2	3.3	13.9	4.1	< 0.001
Zinc (mg/d)	15.1	13.3	8.7	2.6	< 0.001
Iodine (µg/d)	63.0	28.6	66.8	29.3	< 0.001
Copper (mg/d)	0.5	0.3	0.7	0.3	< 0.001
Chlorine (mg/d)	697	322	1118	518	< 0.001
Manganese (mg/d)	76.3	67.4	77.1	67.1	< 0.001
Selenium (µg/d)	36.9	17.6	52.0	20.2	< 0.001

DRI, FESNAD: A vitamin (µg/d): 400 for 3-5 y, 450 for 6-9 y, 600 for 10-13 y, 600 for 14-18 y ♀, 800 for 14-18 y ♂. **Thiamin (mg/d):** 0.7 for 4-5 y, 0.8 for 6-9 y, 0.9 for 10-13 y ♀, 1 for 10-13 y ♂, 1 for 14-18 y ♀, 1.2 for 14-18 y ♂. **Riboflavin (mg/d):** 0.9 for 4-5 y, 1.1 for 6-9 y, 1.2 for 10-18 ♀, 1.3 for 10-13 y ♂, 1.5 for 14-18 y ♂. **Vitamin B6 (mg/d):** 0.9 for 4-5 y, 1 for 6-9 y, 1.1 for 10-13 y ♀, 1.2 for 10-13 y ♂, 1.3 for 14-18 y ♀, 1.4 for 14-18 y ♂. **Vitamin B12 (µg/d):** 1.1 for 4-5 y, 1.2 for 6-9 y, 1.8 for 10-13 y, 2 for 14-18 y. **Vitamin C (mg/d):** 45 for 4-9 y, 50 for 10-13 y, 60 for 14-18 y. **Vitamin D (µg/d):** 5 for 4-18 y. **Vitamin E (mg/d):** 7 for 4-9 y, 11 for 10-13 y, 15 for 14-18 y. **Niacin (mg/d):** 11 for 4-5 y, 12 for 6-9 y, 13 for 10-13 y ♀, 14 for 14-18 y ♀, 15 for 10-18 y ♂. **Pantothenate (mg/d):** 3 for 4-9 y, 4 for 10-13 y, 5 for 14-18 y. **Biotin (µg/d):** 12 for 4-9 y, 20 for 10-13 y, 25 for 14-18 y. **Folic Acid (µg/d):** 150 for 4-5 y, 200 for 6-9 y, 250 for 10-13 y, 300 for 14-18 y. **Sodium (mg/d):** 1200 for 4-9 y, 1500 for 10-18 y. **Potassium (mg/d):** 1100 for 4-5 y, 2000 for 6-9 y, 2900 for 10-13 y♀, 3100 for 14-18 y♀, 3100 for 10-18 y♂. **Calcium (mg/d):** 700 for 4-5 y, 800 for 6-9 y, 1100 for 10-13 y, 1000 for 14-18 y. **Phosphorus (mg/d):** 500 for 4-5 y, 600 for 6-9 y, 900 for 10-13 y, 800 for 14-18 y. **Magnesium (mg/d):** 120 for 4-5 y, 170 for 6-9 y, 250 for 10-13 y ♀, 280 for 10-13 y ♂, 300 for 14-18 y ♀, 350 for 14-18 y ♂. **Iron (mg/d):** 8 for 3-5 y, 9 for 6-9 y, 12 for 10-13 y ♂, 11 for 14-18 y ♂, 15 for 10-18 y ♀. **Zinc (mg/d):** 6 for 4-5 y, 6.5 for 6-9 y, 8 for 10-13 y, 8 for 14-18 y ♀, 11 for 14-18 y ♂. **Iodine (µg/d):** 90 for 4-5 y, 120 for 6-9 y, 130 for 10-13 y ♀, 135 for 10-13 y ♂, 150 for 14-18 y. **Copper (mg/d):** 0.6 for 4-5 y, 0.7 for 6-9 y, 1 for 10-18 y. **Chlorine (mg/d):** 1900 for 4-9 y, 2300 for 10-18 y. **Manganese (mg/d):** 1.5 for 4-9 y, 1.6 for 10-13 y ♀, 1.9 for 10-13 y ♂, 1.67 for 14-18 y♀, 2.2 for 14-18 y ♂. **Selenium (µg/d):** 20 for 3-5 y, 25 for 6-9 y, 35 for 10-13 y, 45 for 14-18 y ♀, 50 for 14-18 y ♂.

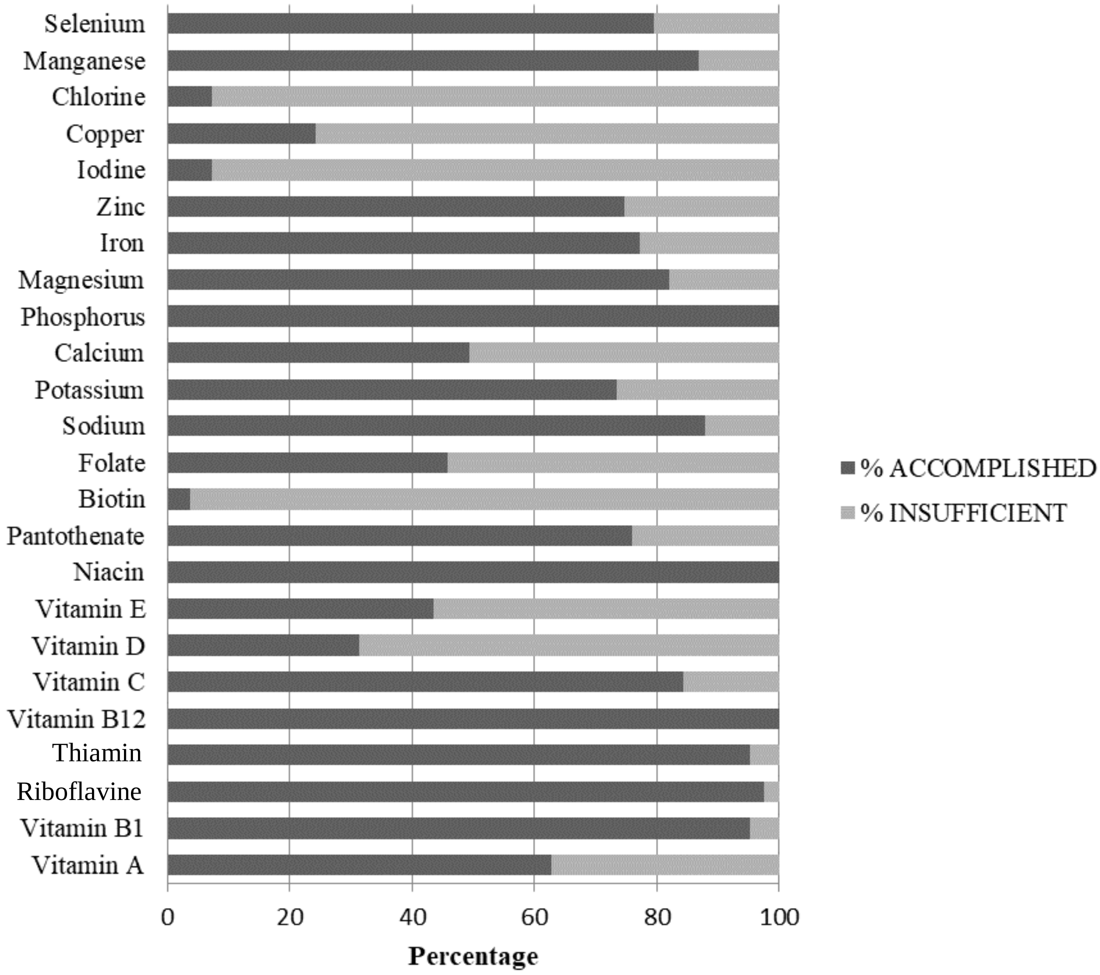
Notes: in italic, nutrients with significance level $P < 0.05$; NS. not significant; SD. standard deviation .DRI, dietary reference intake; FESNAD, Federation of Spanish Societies of Nutrition and Dietetics; mg/d, miligrams/day; µg/d, micrograms/day; y, years; ♀, girls; ♂, boys.



Supplementary Figure 1. Energy intake of celiac children and adolescents represented as the percentage of estimated energy expenditure. EE: Energy Expenditure.



Supplementary Figure 2. Macronutrient energy distribution of celiac and non-celiac children and adolescents' diet compared to the recommended percentage in a balanced diet (proposed by the European Food Safety Authority, EFSA). Kcal, kilocalories; SD, Standard Deviation.



Supplementary Figure 3. Percentage of celiac children and adolescents who accomplished or did not achieve dietary reference intake of the vitamins and minerals (proposed by the Federation of Spanish Societies of Nutrition and Dietetics, FESNAD).

Manuscript 3

Teba González, **Idoia Larretxi**, Juan Carlos Vitoria, Luis Castaño, Edurne Simón, Itziar Churruca, Virginia Simón and Arrate Lasa.

Celiac Male's Gluten-free Diet Profile: Comparison to that of the Control Population and Celiac Women

Nutrients. 2018 Nov 8;10 (11)

nutrients

MDPI

Article

Celiac Male's Gluten-Free Diet Profile: Comparison to that of the Control Population and Celiac Women

Teba González [1,†], Idoia Larretxi [2,†], Juan Carlos Vitoria [1,3], Luis Castaño [1], Edurne Simón [2], Itziar Churruca [2,*], Virginia Navarro [2,*] and Arrate Lasa [2]

1 Instituto de Investigación Sanitaria Biocruces Bizkaia, Hospital Universitario Cruces, UPV/EHU, CIBERDEM, CIBERER, 48903 Barakaldo, Spain; teba.gonzalez@gmail.com (T.G.); jcvitoria45@gmail.com (J.C.V.); lcastano@osakidetza.eus (L.C.)

2 Gluten Analysis Laboratory of the University of the Basque Country, Department of Nutrition and Food Science, Faculty of Pharmacy, University of the Basque Country (UPV/EHU), Paseo de la Universidad, 7, 01006 Vitoria-Gasteiz, Spain; ilarretxi@hotmail.com (I.L.); edurne.simon@ehu.eus (E.S.); arrate.lasa@ehu.eus (A.L.)

3 Servicio de Gastroenterología Pediátrica, Hospital Universitario Cruces, UPV/EHU, 48903 Barakaldo, Spain

* Correspondence: itziar.txurruka@ehu.eus (I.C.); virginia.navarros@ehu.eus (V.N.); Tel.: +34-945-013071 (I.C.); +34-945-013923 (V.N.); Fax: +34-945-013014 (I.C. & V.N.)

† González and Larretxi contributed equally and should be considered as first authors.

Received: 16 October 2018; Accepted: 5 November 2018; Published: 8 November 2018

check for updates

Abstract: The aim of the present work was to analyze the body composition and dietary profile of Spanish celiac men and to compare them to control men and celiac women from our previous studies. Forty-two celiac men (31.5 ± 11.9 years) were recruited and anthropometric measurements were taken. Analysis of energy consumption, macro- and micronutrient intake and food frequency consumption was carried out. Celiac men were more overweight and obese than celiac women, but less than the control population, reporting the same energy intake and macronutrient distribution. Most micronutrient deficiencies in celiac men were not directly related to a gluten free diet; these were also observed for the entire population. The least adherence to Dietary Reference Intakes in women was reported for iron, iodine, potassium and selenium, whereas magnesium intake was higher than in men. Among celiac participants (both genders), cereal, vegetable and legume consumption was poor and meat intake was contrastingly excessive. In conclusion, the dietary profile of celiac men is as unbalanced as that of control men but slightly more than that of celiac women. General nutritional education should be given to both general and celiac populations, and specific advices to celiac men, in order to decrease the risk of celiac disease-related pathologies.

Keywords: celiac disease; gluten-free diet; diary recommended intake; food habit; body composition

1. Introduction

Celiac disease (CD) is described as a permanent intolerance to gluten and is the most common chronic intestinal disease in Europe. Its estimated prevalence in Europe is around 1%, and this only refers to those that are diagnosed, since a significant number of patients have not been diagnosed yet [1,2].

This intestinal disease is more frequent in women than in men in a 2:1 ratio [3] and thus, in the vast majority of studies carried out in celiac people, it is common to find a larger number of female participants than males. Moreover, it must be pointed out that, in general, the male population is less likely to take part in health promotion programs than women are. Different reasons for this have been put forward, such as their low recruitment, delayed help-seeking behavior, and less interest and knowledge about health-related topics and habits [4,5]. As a result, this population cohort is often undervalued and does not always receive gender-specific healthcare.

Nutrients **2018**, *10*, 1713; doi:10.3390/nu10111713 www.mdpi.com/journal/nutrients

The only effective treatment for celiac disease is a strict lifelong gluten-free diet (GFD). In fact, small amounts of gluten ingestion can cause important damage-causing disorders in the intestinal mucosa. Apart from gluten absence, a GFD must guarantee nutritional balance and so prevent deficiencies. However, when the nutritional composition of GFD of celiac people has been assessed, imbalanced proportions of macronutrients and several deficiencies in vitamin and mineral content have been observed [6–11]. Specifically, a GFD is usually accompanied by a low intake of carbohydrates, iron, calcium, folate, niacin, zinc and fiber and excess of saturated fats.

Some aspects of the GFD profile could be linked to a higher risk of several diseases. For instance, the relationship between low fiber, high saturated fat intake and cardiovascular diseases and obesity has been widely described [12,13]. Micronutrient deficiencies are related to comorbidities such as ferropenic anemia and osteopenia. This fact enhances the importance of complying with not only the intake recommendations of some key nutrients such as iron, calcium, vitamin D, but also those of other important molecules that regulate the immune system and help metabolic status to be balanced (zinc, magnesium, selenium, folate and so on), either in women or in men. In fact, it has been described that these deficiencies persist in some of these patients, even if they follow a strict GFD [8,10,14,15].

Bearing in mind all the above mentioned, the aim of the present work was to evaluate the body composition of adult celiac men from a region in Spain and the nutritional composition of the GFD they followed, as well as compare it with international recommendations, and with dietary habits of the general population (Spanish adult men). As a second objective, energy and nutrient intake and dietary habits of celiac men were compared to those reported in our previous studies for celiac women from the same region.

2. Materials and Methods

2.1. Participants and Procedure

The present study used data from a celiac men cohort recruited between 2007 and 2013 from three regions of the Basque Country (Araba, Gipuzkoa, and Bizkaia), in the north of Spain, as part of a prospective SUSFOOD study conducted in collaboration with the Gastroenterology and Endocrinology Units of Cruces University Hospital and Coeliac Association from the Basque Country. Forty-two celiac men took part in the study (mean age ± SD: 31.5 ± 11.9); all participants were diagnosed with celiac disease according to ESPGHAN guidelines and followed a GFD for at least one year. Exclusion criteria included a history of chronic diseases such as cardiovascular disease, diabetes, hyperthyroidism/hypothyroidism, hypercholesterolemia, hypertriglyceridemia or high blood pressure levels, other digestive pathologies that need specific dietary advice, and lack of motivation to participate in the study. Written informed consent was obtained from all participants, after receiving information about the survey. This study was approved by the Ethical Committee of Cruces Hospital (CEIC E-08/66) and the Ethical Committee of the University of The Basque Country (CEISH/76/2011).

Celiac women were recruited simultaneously and their data were collected in the same way as results from men; in fact, the woman cohort is part of the SUSFOOD study mentioned, which has already been published [6]. Control men data were obtained from the ENIDE nutritional survey carried out in Spain, based on 1589 adult men and conducted over the same period of time as the present work (ENIDE).

2.2. Anthropometric Measurements

Anthropometric measurements were collected by trained personnel. Body weight (±10 g) was measured after voiding using a digital integrating scale (SECA 760). Height was determined to the nearest 5 mm using a stadiometer (SECA 220). Body Mass Index (BMI) was calculated from weight and height (kg/m^2). The BMI values were categorized according to the World Health Organization (WHO) criteria as follows: Below 18.5 kg/m^2 considered as underweight, 18.5–24.9 kg/m^2 as normal weight, 25–29.9 kg/m^2 as overweight and >30 kg/m^2 as obese (WHO).

2.3. Body Composition and Energy Expenditure

Fat mass was estimated by a direct segmental multiple-frequency bioelectrical impedance analysis method (Inbody 230; Biospace, Seoul, Korea). Two skin electrodes were placed on the feet and two on the hands. Following the standard procedure, whole-body resistance and reactance were measured. Fat mass was evaluated from total-body impedance (Z). The guidelines of Gallagher et al. were used as reference for body fat mass [16].

Weight, height and age were used to calculate individual energy expenditure according to the Harris-Benedict formula. Standard activity level value was applied.

2.4. Dietary Assessment

Dietary intake was assessed using 3-day 24-h food recalls (24 HR), two on weekdays and one at the weekend. Sixteen participants filled out a food frequency questionnaire (FFQ). Trained nutritionist-dieticians recorded the answers of participants. Food portions and amounts were determined by using photographs of rations and sizes described in the Photo Album, as per the author of [17]. Energy and nutrient intakes were calculated by the nutritional software program "Alimentación y Salud" (AyS, Software, Tandem Innova, Inc., Huesca, Spain). The composition of specific gluten-free products for celiac people consumed by the participants was collected from the manufacturers and added into the food composition database of the program before calculations. As gluten-free product labels did not indicate micronutrient content (vitamins and minerals), an estimation with homologous gluten-containing products was carried out.

Dietary reference intakes (DRI) for Spanish population issued by the Spanish Societies of Nutrition, Feeding and Dietetics (FESNAD) in 2010 were taken as references for the interpretation of the 24 HR [18]. In the case of FFQ, Spanish Society of Community Nutrition (SENC) recommendations were used for the correct interpretation of the results [19].

Moreover, the results were compared to energy, nutrient and food intake of celiac women [6] and those of the mentioned Spanish reference population (ENIDE) [20].

2.5. Statistical Analysis

Statistical analyses of results were performed by using the IBM SPSS statistical program, version 23 (IBM Inc., Armonk, NY, USA). Normality in the distribution was assessed by the Kolmogorov-Smirnov test, and homogeneity by Levene's test. Statistical analyses were performed in order to calculate differences between celiac men and control population were performed with summary *t* Student's test, and those between celiac men and celiac women with Chi-square test. p values < 0.05 were accepted as significant.

3. Results

3.1. Anthropometric Measurements

Anthropometric data of celiac men from the present study are shown in Table 1.

57.1% of the participants showed normal BMI values and only 4.8% were underweight. In contrast, 38.1% of them were above normal BMI values, 26.2% were overweight, and 11.9% obese. Fat mass measurements indicated that 52% of the participants had excessive adiposity and 41% were between normal values. Only three participants (7%) had very low fat mass values.

Table 1. Characteristic of celiac participants included in the survey.

Characteristic	Men
N	42
Age (year)	31.5 ± 11.9
Mean duration of GFD (year)	20.9 ± 11.9
Height (cm)	176.2 ± 6.2
Weight (kg)	75.8 ± 13.9
Fat mass (%)	24.1 ± 8.1
Body-Mass Index	
Mean (kg/m^2)	24.7 ± 4.1
Underweight < 18.5—no. (%)	4.8
Normal 18.5–24.9—no. (%)	57.1
Overweight 25–29.9—no. (%)	26.2
Obese > 30—no. (%)	11.9

Notes: Values are mean ± SD or percentages; SD, standard deviation; *N*: sample size; no, number of subjects; GFD, gluten-free diet.

3.2. Dietary Intakes

3.2.1. Energy, Macronutrients, Fiber and Cholesterol Intake in Celiac Men

Daily energy intake was comparable to that observed in control men in ENIDE study. Energy distribution among macronutrients was not balanced in celiac men. To be specific, proteins and fats were consumed in excess (17% and 41% respectively) accompanied by a small amount of carbohydrates (42%) (Figure 1). When these data were compared to those of the ENIDE study, no significant differences were observed in energy and macronutrient consumption.

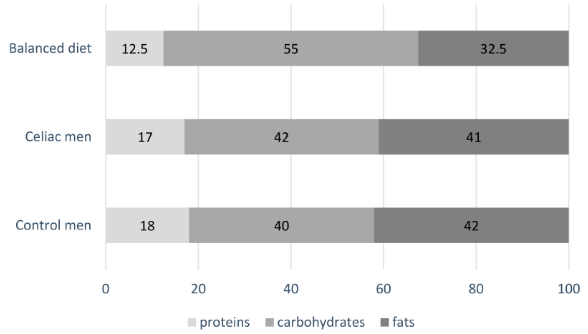


Figure 1. Mean contribution of macronutrients to energy in Spanish celiac (n = 42) and control men (n = 1589) (ENIDE study, representative at national level of the adult population) compared to the recommended contribution in a balanced diet proposed by the Federation of Spanish Societies of Nutrition and Dietetics (FESNAD).

In order to evaluate fat sources, dietary lipid profile was calculated. While saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) consumption was similar in celiac men and control men, polyunsaturated fatty acid (PUFA) consumption of celiac men was lower. In general terms, saturated and unsaturated fatty acids ratios were reached, as in the ENIDE survey (Table 2) [21]. Cholesterol ingestion was also similar in both groups, higher than that recommended [18].

Table 2. Energy and nutrient distribution in celiac and Spanish control men and results from recent studies.

Characteristic	Celiac Men (n = 42)	Spanish Men (ENIDE Study) (n = 1589)	p Value	Sheperd 2013 (11 Men)	Martin 2013 (18 Men)	Wild 2010 (31 Men)
Energy (kcal)	2355.4 ± 659.0	2546.8 ± 860.9	NS	2697 ± 445	2401 ± 592	2500 ± 717
Protein (g)	100.2 ± 30.1	109.4 ± 47.7	NS	98.8 ± 22.83	92.4 ± 17.5	92.7 ± 24.9
Carbohydrate (g)	220.7 ± 84.7	242.7 ± 101.8	NS	294 ± 72.3	267 ± 95	315.6 ± 98.5
Fat (g)	114.0 ± 32.2	114.9 ± 46.0	NS	67.9 ± 50.58	97.4 ± 29.7	93.2 ± 36.6
MUFA (g)	52.4 ± 16.1	48.29 ± 22.2	NS	-	-	-
PUFA (g)	13.2 ± 5.8	15.5 ± 8.5	<0.05	-	-	-
SFA (g)	34.5 ± 11.2	32.71 ± 15.55	NS	34.7 ± 7.7	-	-
(PUFA + MUFA)/SFA	1.93	1.95	-	-	-	-
PUFA/SFA	0.38	0.47	-	-	-	-
Cholesterol (mg)	421.2 ± 132.8	423.82 ± 181.25	NS	-	-	-
Fiber (g)	20.3 ± 7.1	20.94 ± 11.38	NS	30.2 ± 7.7	22.3 ± 6.0	13.7 ± 5.3

Notes: Values are means ± SD; Spanish adult men data were taken from the Spanish dietary nutritional assessment (ENIDE study, representative of the adult population at national level); SD, standard deviation; NS: non-significant; PUFA, polyunsaturated fatty acids; MUFA, monounsaturated fatty acids; SFA, saturated fatty acids.

Regarding dietary fiber consumption, celiac men were below recommendations (25–35 g/day). In fact, fiber consumption of 28% of participants was below 15 g per day and that of 43% between 15.1 and 25 g per day. A similar intake of fiber was also reported in control men, taken from the ENIDE survey (Table 2).

A comparison of energy intake between celiac men from the present study and celiac women from our previous studies [6] revealed that while the majority of women (65%) consumed the correct amounts of calories in their diet, less than half the men (46%) did so (Table 3). Moreover, only 6% of women consumed calories in excess, which, by contrast, 14% of men did. However, macronutrient distribution, similar in both genders, was higher than recommendations for proteins and fat intakes, and lower in the case of carbohydrates.

Table 3. Percentage of celiac men (n = 42) and women (n = 54) that fulfilled energy and macronutrient recommendation and their comparison.

	Recommended Intake *	Celiac Men			Celiac Women [6]			p Value
		Low	Adequate	Excessive	Low	Adequate	Excessive	
Energy intake	±20% of EE	40	46	14	30	65	6	<0.001
Protein	10–15%	0	19	81	0	20	81	NS
Carbohydrate	50–60%	98	2	0	91	9	0	NS
Total Fat	30–35%	0	5	95	4	11	85	NS
SFA	<10%	0	17	83	0	33	67	<0.05
MUFA	15–20%	14	31	55	31	37	28	NS

Notes: * Recommended energy and nutrient intake in a balanced diet proposed by the Federation of Spanish Societies (FESNAD). EE: energy expenditure; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; NS: non-significant. p value corresponds to differences between celiac males' and females' suitable intakes.

Dietary lipid profiles were similar between celiac men and women. In both groups, the highest percentage of participants consumed excessive SFA and cholesterol, though even more so in men.

Although both groups contained a high proportion of subjects with low fiber intakes (71% in men vs. 96% in women), there were more men that achieved adequate fiber consumption (26% in men vs. 4% in women).

3.2.2. Micronutrients Intake in Celiac Men

When vitamin and mineral mean intake of celiac participants was compared to that of control men, differences in eight micronutrient mean consumption were observed (Table 4). Celiac men consumed lower amounts of vitamin E, niacin and magnesium than the control group. By contrast, their riboflavin, B6, zinc, potassium and selenium mean intake was higher.

Table 4. Micronutrients mean intake in celiac and Spanish men.

	Celiac Men (n = 42)	Control Men (ENIDE Study) (n = 1589)	DRI: FESNAD (2010)	p Celiac Men vs. Spanish Men (ENIDE Study)
Vitamin A (ug)	802 ± 340	748 ± 338	700 [a]	NS
Thiamin (mg)	2.0 ± 1.4	2.1 ± 6.7	1.2 [b]	NS
Riboflavin (mg)	2.2 ± 1.1	1.5 ± 0.8	1.6	<0.001
Vitamin B6 (mg)	2.7 ± 0.9	2.0 ± 0.9	1.5 [c]	<0.001
Vitamin B12 (ug)	8.1 ± 5.6	7.9 ± 6.1	2	NS
Vitamin C (mg)	143 ± 82	131 ± 81	60 [d]	NS
Vitamin D (ug)	4.4 ± 4.5	4.3 ± 4.5	5 [e]	NS
Vitamin E (mg)	10.8 ± 5.1	14.9 ± 8.4	15	<0.001
Niacin (mg)	38.5 ± 13.6	45.7 ± 39.5	18 [f]	<0.01
Folate (ug)	302 ± 115	296 ± 121	300	NS
Calcium (mg)	939 ± 295	886 ± 345	900 [g]	NS
Iron (mg)	16.5 ± 5.1	16.1 ± 6.5	9 [h]	NS
Magnesium (mg)	323 ± 107	396 ± 139	350	<0.001
Iodine (ug)	117 ± 88	100 ± 50.5	150	NS
Phosphorus (mg)	1580 ± 442	1535 ± 471	700 [i]	NS
Zinc (mg)	12 ± 4.0	10.5 ± 3.7	9.5 [j]	<0.01
Potassium (mg)	3481 ± 980	3045 ± 917	3100	<0.01
Selenium (ug)	93.8 ± 53.5	63.5 ± 35.1	55	<0.001

Notes: Values are means ± SD. Spanish adult men data were taken from the Spanish dietary nutritional assessment (ENIDE study, representative at national level of the adult population). SD, standard deviation; DRI, dietary reference intake; FESNAD, Federation of Spanish Societies of Nutrition and Dietetics; NS: non-significant. [a] Vitamin A, 800 mg for 16–19 year range men; [b] Thiamin, 1.1 mg for >60 years old men; [c] Vitamin B6, 1.4 mg for 16–18 years old and 1.6 mg for >60 years old men; [d] Vitamin C, 70 mg for >60 years old men; [e] Vitamin D, 7.5 mg for >60 years old men; [f] Niacin, 17 mg for 50–69 years old men; [g] Calcium, 1000 mg 16–19 years old and for >60 years old men; [h] Iron, 10 mg for >60 years old men; [i] Phosphorus, 800 mg for 16–19 years old men; [j] Zinc: 10 mg for >60 years old men.

A 67% (2/3) DRI cutoff value for deficient micronutrient intake was set as reported by the literature [22,23]. According to this cutoff, vitamin A, D and E, iodine, folate and magnesium were deficient, as a small amount (41–81%) of participants accomplished it (Table 5). In the case of calcium, zinc, selenium and vitamin C, most participants (around 90% of them) fulfilled 2/3 of the DRIs. None of the celiac men showed low intakes of vitamin B6, B12, niacin, riboflavin, thiamine, phosphorus and iron.

Table 5. Percentage of celiac men (n = 42) and women (n = 54) that fulfilled at least 2/3 of DRI (FESNAD, 2010) and their comparison.

	Celiac Men	Celiac Women [6]	p Value
	% of Participants that Fulfilled at Least 67% of Recommendations	% of Participants that Fulfilled at Least 67% of Recommendations	
Vitamin A (ug)	81	89	NS
Thiamin (mg)	98	100	NS
Riboflavin (mg)	98	98	NS
Vitamin B6 (mg)	100	100	NS
Vitamin B12 (ug)	100	100	NS
Vitamin C (mg)	93	96	NS
Vitamin D (ug)	45	52	NS
Vitamin E (mg)	48	61	NS

Table 5. *Cont.*

	Celiac Men	Celiac Women [6]	*p* Value
	% of Participants that Fulfilled at Least 67% of Recommendations	% of Participants that Fulfilled at Least 67% of Recommendations	
Niacin (mg)	100	100	NS
Folate (ug)	76	82	NS
Calcium (mg)	86	87	NS
Iron (mg)	100	69	<0.001
Magnesium (mg)	71	91	<0.05
Iodine (ug)	50	20	<0.01
Phosphorus (mg)	100	100	NS
Zinc (mg)	91	98	0.093
Potassium (mg)	98	85	<0.05
Selenium (ug)	93	69	<0.01

Notes: Recommended intake of vitamins and minerals proposed by the Federation of Spanish Societies (FESNAD). NS: non-significant. *p* value correspond to differences between celiac men and celiac women's appropriate intakes.

Several differences were found in mineral intake accomplishment, but not in vitamins, when comparing celiac men and women habits (Table 5). Celiac women fulfilled magnesium requirements better than celiac men and, by contrast, iron, iodine, potassium, and selenium DRIs were better complied by celiac men.

3.3. Food Consumption Frequency of Celiac Men

Figure 2 summarizes main food group consumption frequency. Cereal consumption data indicated that only 13% of celiac men fulfilled these food group recommendations, which means at least four servings per day (Figure 2). Moreover, almost half of them (44%) consumed a small or a very small amount of cereals (fewer than two servings) per day.

The vast majority of participants (84%) did not reach vegetable consumption recommendations (10 portions/week) [19]. One-third of celiac men did not consume the minimum recommended two servings of fruit daily. Furthermore, legume consumption was also low in 42% of participants, which means that they consumed less than two portions of pulses per week.

With regard to animal origin food consumption, almost half of the participants fulfilled the recommendations of dairy products, 2–3 servings per day, whereas 25% reported an excessive consumption, which means more than 4 dairy servings per day. Only 19% of celiac men achieved egg consumption recommended by the SENC, while 38% had an excessive consumption. The ingestion of fish and derivatives was sufficient in nearly half of celiac men and almost 20% of participants consumed it in excess. By contrast, meat consumption was excessive in the vast majority of the subjects (88%), which means that they ate more than 4–5 servings of meat per week.

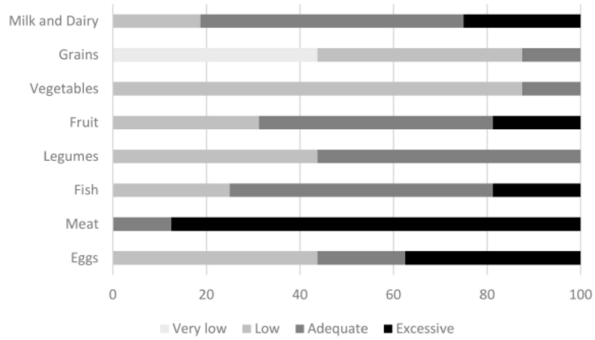


Figure 2. Compliance of food frequency consumption in celiac men by servings per day or week, according to the Spanish Society of Community Nutrition (SENC).

In relation to the foods considered for occasional consumption, it must be pointed out that celiac men followed the recommendations (data not shown). Participants used olive oil, which is rich in monounsaturated fatty acids, as their fat source, and discarded other lipid sources such as margarine or butter as their main lipid source for cooking.

Finally, the analysis of food frequency questionnaires in celiac men and celiac women revealed similar results. No differences were found in dairy products, grains, vegetables, fruits and meat consumption. However, when fish and egg consumption were evaluated, a tendency toward higher percentage of celiac men with excessive consumption of these foods was observed, compared to celiac women. Concretely, 19% of men consumed fish in excess vs. 6% of women ($p = 0.09$). Moreover, 37% of men consumed eggs in excess, whereas 17% of women did so ($p = 0.08$).

4. Discussion

Male participation in health promotion programs has usually been lower than that of women, probably due to their lower interest in health-related topics, among other reasons [4,5]. The studies in the literature comparing men and women's dietary habits provide different outcomes, attributable to differences in methodologies, population sizes, regional habits, and recommendations. Some studies described different patterns of deficiencies either in macro- or micronutrients between genders [24,25]. In the case of celiac disease, even though clinical trials have been published with both female and male participants, the recruited number of men is usually small. Thus, evidence about celiac men's body composition, diet quality, and eating pattern is rather scarce. The present study presents detailed energy and nutrient intake and dietary habits of a meaningful celiac male group, comparing them to a male control population and to celiac women from the same geographical region.

Obesity and overweight are rising among celiac patients, which is increasingly of concern to clinicians [11,26]. In this study, anthropometric data revealed that celiac men showed an alarming 38.1% of overweight and obesity, while 52% of participants showed an excessive fat mass. Nevertheless, this prevalence was lower than that observed in the control population [27,28]. These results were consistent with other studies, where celiac men registered similar BMI index values [8,29,30] which were also lower than those of the control populations [6,7,31,32].

Comparing these results with data from women (only 7.4% of overweight in celiac women), major gender differences were found. Accordingly, the prevalence of obesity and overweight in general population is greater for men [33–35]. These differences could, as stated before, be due to the lesser interest that men pay to health status and its care [4,5]. Nevertheless, it must be pointed out that in the case of celiac patients, most studies do not find gender differences in this parameter [8,29]. However, Tucker et al. (2012) [36] found that females were more likely to be obese, when BMI reaches values of 30 or above.

Even though energy intake of celiac men was similar to that of the control and to celiac men from other studies [8,20,29,30], our data revealed that 14% of celiac men consumed calories in excess, more than 120% of their energy expenditure, and 24% of them exceeded 110% of requirements. These results were not found in women, whose energy intake was suitable in 65% of the participants. All of the above explains the high percentage of obesity found among male participants from the present study. By contrast, 40% of the participants were under 80% of their energy expenditure, reflecting a possible underreporting, as found by others [29,37].

Celiac men showed imbalanced energy distribution, similarly to previous studies, but it must be pointed out that participants in this study consumed even fewer carbohydrates and more fat than those of earlier studies [29,30]. However, the macronutrient consumption of Spanish celiac women and the control population was similar to that observed in the male celiac group studied. Thus, it seems that imbalanced energy distribution is not GFD-related but could be associated to geographical dietary habits. Of these, low cereal and vegetable consumption and excessive meat intake, observed in both men and women [6], as well as in control men [38], could be the main contributors to the macronutrient imbalance.

With regard to dietary fat sources, SFA, MUFA and cholesterol were consumed in excess, although adequate fatty acid ratios were reached. These results were similar to those observed in control population, reinforcing the idea of this not being a GFD-related imbalance. The excessive consumption of meat (88% participants) and eggs (37%) could justify the results observed. However, the proportion of men eating excessive SFA and cholesterol was significantly greater than that of women. Although meat consumption did not differ between men and women, the excessive consumption of eggs among celiac men could impair these parameters. By contrast, intake of PUFAs was slightly lower in the male celiac population than in control men. However, 90% of celiac men participating showed adequate PUFA intake, more than women did. The higher fish consumption observed in men could be on the basis of this difference.

As far as fiber is concerned, celiac male consumption was low, similar to other studies conducted with celiac men and control population [8,20,29,30]. The low consumption of vegetables, legumes and grains could explain, at least in part, this outcome. However, fiber recommendations were more obeyed by celiac men than by celiac women, due to higher general food intake and thus greater amounts of plant origin foods.

The celiac population has been associated with an increased cardiovascular risk [39,40]. Although factors influencing the association between CD and cardiovascular disease could be related to the pathophysiology of CD, there is no doubt that the body composition and dietary pattern of celiac men described, with high intake of deleterious components, such as SFA, and low intake of protective ones, such as fiber, play an essential role.

As stated previously, micronutrients have also been a matter of concern for the celiac population. In our study, some differences in micronutrient mean intake arose between celiac men and their control. Vitamin E, niacin and magnesium was lower in celiac men than in control men and, by contrast, that of riboflavin, B6, zinc, potassium and selenium was higher, pointing to a different dietary pattern of this collective. While mean intakes provided some important information, it was necessary to analyze to what extent this population was below micronutrient recommendations, in order to identify possible deficiencies and health-risks.

The percentage of celiac men who complied with micronutrient DRI varied among nutrients, and also between genders. The most alarming deficiencies reported in this work for celiac men were those of vitamin D, vitamin E, and iodine, all of them with less than 50% of individuals fulfilling two thirds of DRI.

Wild et al. (2010) also reported vitamin D deficient intake in celiac men (no more than 20% of patients fulfilled the DRI) [8,29,30]. These data were also in accordance with the general population [8,20,29,30]. Vitamin E and iodine mean intakes were also low for both celiac and control men. In particular, vitamin E was even lower in celiacs, where only 48% reached the 67% of DRI. Iodine and vitamin E are not usually analyzed in GFD assessments, but considering their extremely low intake, specific nutritional advice, and probably supplementation, should be considered for the celiac male population.

In addition to the aforementioned important deficiencies, other micronutrients, such as folate and magnesium, were also below recommendations in celiac men (less than 80% achieved two-thirds of DRI). These data were in agreement with previous studies from the literature [8,30,37].

The comparison between celiac men and women in their adherence to vitamin and mineral recommendations revealed differences only in some mineral intakes. Less fulfilment of DRIs in women was reported for iron, iodine, potassium, and selenium, whereas magnesium intake was higher than that of men. This could be due to the lower energy intake of women [6], which could hinder the achievement of some mineral intakes. Iron was a special case, because men fulfilled totally the DRIs, but only 69% of women achieved 2/3, due to more demanding requirements of this mineral at some life stages for them. Considering the high prevalence of anemia in patients with CD, this is an important issue to be addressed in women.

In general, it can be stated that micronutrient deficiencies in celiac men were not directly related to a GFD, and were concerning for the entire population. Only magnesium and vitamin E intake appeared to be lower than the control, and below recommendations for both groups in the case of vitamin E. Folate, iodine and vitamin D intakes were the most worrying, giving the anemia, thyroid disease and osteopenia prevalence of this collective [41,42].

One of the most cited reasons for micronutrient deficiency in a GFD is gluten-free products (GFP) composition. In fact, gluten-containing grains and foods are a major source of micronutrients [43,44], and often GFP do not contain the same levels of micronutrients as their counterparts—e.g., thiamine, riboflavin, niacin, folate, vitamin D, calcium, or iron [10,45–48]. Instead of those GFP, higher gluten-free whole cereals (amaranth, sorghum, millet, etc.) and pseudocereals (quinoa, buckwheat, etc.) are more interesting, as they contain the fiber and micronutrients necessary to balance [11,49]. Nevertheless, fortification of gluten-free flours and GFP could be also of interest, but more research is necessary in order to correctly identify which nutrients are suitable for general fortification and which for personal supplementation.

For this purpose, all micronutrients should be addressed in the studies, and the micronutrient composition of GFP, which needs to be studied in depth, should be incorporated into the databases. Selenium, iodine and vitamin E intakes are scarcely analyzed in the literature, and they are still important to ensure the health status of celiac people.

The main limitation of the present study was that micronutrient intake coming from GFP was calculated according to the micronutrient content of their gluten-containing counterparts. Thus, data from the present work could represent an overestimation of the real consumption of vitamins and minerals. Moreover, people participating in this kind of studies are more concerned about health issues and self-caring, which could also have overestimated the results. Furthermore, the experimental design did not include a control group of men, and people with comorbidities were excluded from the study. Nevertheless, this is the first study where dietary habits of a cohort of Spanish celiac men were compared to those of the control population and to those of Spanish celiac women. It is worth noting that the sample size was higher than those used in other studies with celiac men.

In summary, inadequacies in terms of both, macro- and micronutrients, in celiac male diets are found. Some of them also appear in the diet of the control population, pointing to unsuitable habits of the entire population, but other changes are gender-specific and fewer GFD-dependent. Effective general recommendations for the global male population should be given, as well as specific advice for celiac people, principally regarding micronutrient intake. Greater consumption of gluten-free cereals or pseudocereals, vegetables, and legumes and less of that of meats should be recommended. Personalized dietary advice and long-term follow-up for celiac people, in particular for celiac men, could contribute towards preventing nutrient deficiencies related to dietary imbalance and to obtaining a better health status, lowering the risk of CD-related pathologies.

Author Contributions: T.G. and I.L. recruited the patients, performed anthropometric measurements and collected questionnaires. J.C.V. and L.C. designed the experiment. I.C. and A.L. performed the analysis of diet and food habits. V.N. and E.S. analyzed all data and contributed to statistical analysis. E.S., I.C., V.N. and A.L. wrote the manuscript.

Acknowledgments: Idoia Larretxi is a fellowship of the University of the Basque Country, UPV/EHU (Euskararen eta Etengabeko Prestakuntzaren arloko Errektoreordetza). This research was funded by University of the Basque Country (UPV/EHU) and Basque Government, grant numbers US12/10 and GV2008111042, respectively. We thank the participation of all patients, and the collaboration of "Asociación de Celiacos de Euskadi (EZE)", specifically that of Mireia Apraiz, Beatriz Zulueta, Nerea Segura, Estíbaliz Olabarri Elena Pérez-Estevez, Lissette Delgado, Aranzazu Sainz-Espiga, Sagrario Martínez and Lorea Martínez-Indart.

Conflicts of Interest: The authors declare that they have no conflict of interest.

References

1. Lionetti, E.; Catassi, C. New clues in celiac disease epidemiology, pathogenesis, clinical manifestations, and treatment. *Int. Rev. Immunol.* **2011**, *30*, 219–231. [CrossRef] [PubMed]
2. Peña, A.; Rodrigo, L. Epidemiology of celiac disease and non-celiac gluten-related disorders. In *Advances in the Understanding of Gluten Related Pathology and the Evolution of Gluten-Free Foods*; Arranz, E., Fernández-Bañares, F., Rosell, C., Rodrigo, L., Peña, A., Eds.; OmniaScience: Barcelona, Spain, 2015; pp. 27–73.
3. Catassi, C.; Gatti, S.; Fasano, A. The new epidemiology of celiac disease. *J. Pediatr. Gastroenterol. Nutr.* **2014**, *59 (Suppl. 1)* (Suppl. 1), S7–S9. [CrossRef]
4. Sharp, P.; Bottorff, J.L.; Hunt, K.; Oliffe, J.L.; Johnson, S.T.; Dudley, L.; Caperchione, C.M. Men's Perspectives of a Gender-Sensitized Health Promotion Program Targeting Healthy Eating, Active Living, and Social Connectedness. *Am. J. Men's Health* **2018**, 2157–2166. [CrossRef] [PubMed]
5. Galdas, P.M.; Cheater, F.; Marshall, P. Men and health help-seeking behaviour: Literature review. *J. Adv. Nurs.* **2005**, *49*, 616–623. [CrossRef] [PubMed]
6. Churruca, I.; Miranda, J.; Lasa, A.; Bustamante, M.; Larretxi, I.; Simon, E. Analysis of Body Composition and Food Habits of Spanish Celiac Women. *Nutrients* **2015**, *7*, 5515–5531. [CrossRef] [PubMed]
7. Bardella, M.T.; Fredella, C.; Prampolini, L.; Molteni, N.; Giunta, A.M.; Bianchi, P.A. Body composition and dietary intakes in adult celiac disease patients consuming a strict gluten-free diet. *Am. J. Clin. Nutr.* **2000**, *72*, 937–939. [CrossRef] [PubMed]
8. Martin, J.; Geisel, T.; Maresch, C.; Krieger, K.; Stein, J. Inadequate nutrient intake in patients with celiac disease: Results from a german dietary survey. *Digestion* **2013**, *87*, 240–246. [CrossRef] [PubMed]
9. Capristo, E.; Malandrino, N.; Farnetti, S.; Mingrone, G.; Leggio, L.; Addolorato, G.; Gasbarrini, G. Increased serum high-density lipoprotein-cholesterol concentration in celiac disease after gluten-free diet treatment correlates with body fat stores. *J. Clin. Gastroenterol.* **2009**, *43*, 946–949. [CrossRef] [PubMed]
10. Vici, G.; Belli, L.; Biondi, M.; Polzonetti, V. Gluten free diet and nutrient deficiencies: A review. *Clin. Nutr.* **2016**. [CrossRef] [PubMed]
11. Theethira, T.G.; Dennis, M. Celiac disease and the gluten-free diet: Consequences and recommendations for improvement. *Dig. Dis.* **2015**, *33*, 175–182. [CrossRef] [PubMed]
12. Phillips, C.M.; Kesse-Guyot, E.; McManus, R.; Hercberg, S.; Lairon, D.; Planells, R.; Roche, H.M. High dietary saturated fat intake accentuates obesity risk associated with the fat mass and obesity-associated gene in adults. *J. Nutr.* **2012**, *142*, 824–831. [CrossRef] [PubMed]
13. Slavin, J.L. Dietary fiber and body weight. *Nutrition* **2005**, *21*, 411–418. [CrossRef] [PubMed]
14. Capristo, E.; Mingrone, G.; Addolorato, G.; Greco, A.V.; Corazza, G.R.; Gasbarrini, G. Differences in metabolic variables between adult coeliac patients at diagnosis and patients on a gluten-free diet. *Scand. J. Gastroenterol.* **1997**, *32*, 1222–1229. [CrossRef] [PubMed]

15. Capristo, E.; Addolorato, G.; Mingrone, G.; De Gaetano, A.; Greco, A.V.; Tataranni, P.A.; Gasbarrini, G. Changes in body composition, substrate oxidation, and resting metabolic rate in adult celiac disease patients after a 1-y gluten-free diet treatment. *Am. J. Clin. Nutr.* **2000**, *72*, 76–81. [CrossRef] [PubMed]
16. Gallagher, D.; Heymsfield, S.B.; Heo, M.; Jebb, S.A.; Murgatroyd, P.R.; Sakamoto, Y. Healthy percentage body fat ranges: An approach for developing guidelines based on body mass index. *Am. J. Clin. Nutr.* **2000**, *72*, 694–701. [CrossRef] [PubMed]
17. Russolillo, G.; Marques, I. *Food Portion Sizes Album*; Imagen Comunicación Multimedia: Madrid, Spain, 2008; 186p.
18. FESNAD. Dietary reference intakes (DRI) for spanish population, 2010. *Actividad Dietética* **2010**, *14*, 196–197. [CrossRef]
19. Aranceta, J.; Serra-Majem, L. Dietary guidelines for the Spanish population. *Public Health Nutr.* **2001**, *4*, 1403–1408. [CrossRef] [PubMed]
20. AECOSAN, Agencia Española de Consumo, Seguridad Alimentaria y Nutrición. Spanish Population Dietary Intakes. National Health Survey (ENIDE). Available online: http://aesan.msssi.gob.es/AESAN/docs/docs/evaluacion_riesgos/estudios_evaluacion_nutricional/valoracion_nutricional_enide_macronutrientes.pdf (accessed on 8 June 2017).
21. SENC. Nutritional objectives for the Spanish population: Consensus from the Spanish Society of Community Nutrition. *Rev. Esp. Nutr. Com.* **2011**, *17*, 178–199.
22. Marshall, T.A.; Stumbo, P.J.; Warren, J.J.; Xie, X.J. Inadequate nutrient intakes are common and are associated with low diet variety in rural, community-dwelling elderly. *J. Nutr.* **2001**, *131*, 2192–2196. [CrossRef] [PubMed]
23. Roman-Vinas, B.; Ribas Barba, L.; Ngo, J.; Martinez-Gonzalez, M.A.; Wijnhoven, T.M.; Serra-Majem, L. Validity of dietary patterns to assess nutrient intake adequacy. *Br. J. Nutr.* **2009**, *101* (Suppl. 2), S12–S20. [CrossRef]
24. Arganini, C.; Saba, A.; Comitato, R.; Virgili, F.; Turrini, A. Gender Differences in Food Choice and Dietary Intake in Modern Western Societies. In *Public Health-Social and Behavioral Health*; Maddock, J., Ed.; InTech: Hampshire, UK, 2012; ISBN 978-953-51-0620-3. Available online: http://www.intechopen.com/books/public-health-social-and-behavioral-health/gender-differences-in-foodchoice-and-dietary-intake-in-modern-western-societies (accessed on 15 October 2018).
25. Rippin, H.L.; Hutchinson, J.; Jewell, J.; Breda, J.J.; Cade, J.E. Adult Nutrient Intakes from Current National Dietary Surveys of European Populations. *Nutrients* **2017**, *9*, 1288. [CrossRef] [PubMed]
26. Valletta, E.; Fornaro, M.; Cipolli, M.; Conte, S.; Bissolo, F.; Danchielli, C. Celiac disease and obesity: Need for nutritional follow-up after diagnosis. *Eur. J. Clin. Nutr.* **2010**, *64*, 1371–1372. [CrossRef] [PubMed]
27. Aranceta Bartrina, J.; Pérez Rodrigo, C. Inequality, health and nutrition in Spain: A regional view of the body mass index. *Nutr. Hosp.* **2018**, *35*, 142–149. [PubMed]
28. López-Sobaler, A.M.; Aparicio, A.; Aranceta-Bartrina, J.; Gil, Á.; González-Gross, M.; Serra-Majem, L.; Varela-Moreiras, G.; Ortega, R.M. Overweight and General and Abdominal Obesity in a Representative Sample of Spanish Adults: Findings from the ANIBES Study. *Biomed. Res. Int.* **2016**, *2016*, 8341487. [CrossRef] [PubMed]
29. Wild, D.; Robins, G.G.; Burley, V.J.; Howdle, P.D. Evidence of high sugar intake, and low fibre and mineral intake, in the gluten-free diet. *Aliment. Pharmacol. Ther.* **2010**, *32*, 573–581. [CrossRef] [PubMed]
30. Shepherd, S.J.; Gibson, P.R. Nutritional inadequacies of the gluten-free diet in both recently-diagnosed and long-term patients with coeliac disease. *J. Hum. Nutr. Diet.* **2013**, *26*, 349–358. [CrossRef] [PubMed]
31. Brambilla, P.; Picca, M.; Dilillo, D.; Meneghin, F.; Cravidi, C.; Tischer, M.C.; Vivaldo, T.; Bedogni, G.; Zuccotti, G.V. Changes of body mass index in celiac children on a gluten-free diet. *Nutr. Metab. Cardiovasc. Dis.* **2013**, *23*, 177–182. [CrossRef] [PubMed]
32. Larretxi, I.; Simon, E.; Benjumea, L.; Miranda, J.; Bustamante, M.A.; Lasa, A.; Eizaguirre, F.J.; Churruca, I. Gluten-free-rendered products contribute to imbalanced diets in children and adolescents with celiac disease. *Eur. J. Nutr.* **2018**. [CrossRef] [PubMed]
33. Peralta, M.; Ramos, M.; Lipert, A.; Martins, J.; Marques, A. Prevalence and trends of overweight and obesity in older adults from 10 European countries from 2005 to 2013. *Scand. J. Public Health* **2018**, *46*, 522–529. [CrossRef] [PubMed]
34. Rodríguez-Rodríguez, E.; López-Plaza, B.; López-Sobaler, A.M.; Ortega, R.M. Overweight and obesity among Spanish adults. *Nutr. Hosp.* **2011**, *26*, 355–363. [CrossRef] [PubMed]

35. Gutiérrez-Fisac, J.L.; Banegas Banegas, J.R.; Artalejo, F.R.; Regidor, E. Increasing prevalence of overweight and obesity among Spanish adults, 1987–1997. *Int. J. Obes. Relat. Metab. Disord.* **2000**, *24*, 1677–1682. [CrossRef] [PubMed]
36. Tucker, E.; Rostami, K.; Prabhakaran, S.; Al Dulaimi, D. Patients with coeliac disease are increasingly overweight or obese on presentation. *J. Gastrointest. Liver Dis.* **2012**, *21*, 11–15.
37. Hallert, C.; Grant, C.; Grehn, S.; Grännö, C.; Hultén, S.; Midhagen, G.; Ström, M.; Svensson, H.; Valdimarsson, T. Evidence of poor vitamin status in coeliac patients on a gluten-free diet for 10 years. *Aliment. Pharmacol. Ther.* **2002**, *16*, 1333–1339. [CrossRef] [PubMed]
38. De Batlle, J.; Gracia-Lavedan, E.; Romaguera, D.; Mendez, M.; Castaño-Vinyals, G.; Martín, V.; Aragonés, N.; Gómez-Acebo, I.; Olmedo-Requena, R.; Jimenez-Moleon, J.J.; et al. Meat intake, cooking methods and doneness and risk of colorectal tumours in the Spanish multicase-control study (MCC-Spain). *Eur. J. Nutr.* **2018**, *57*, 643–653. [CrossRef] [PubMed]
39. Emilsson, L.; Lebwohl, B.; Sundström, J.; Ludvigsson, J.F. Cardiovascular disease in patients with coeliac disease: A systematic review and meta-analysis. *Dig. Liver Dis.* **2015**, *47*, 847–852. [CrossRef] [PubMed]
40. Ciaccio, E.J.; Lewis, S.K.; Biviano, A.B.; Iyer, V.; Garan, H.; Green, P.H. Cardiovascular involvement in celiac disease. *World J. Cardiol.* **2017**, *9*, 652–666. [CrossRef] [PubMed]
41. Lucendo, A.J.; García-Manzanares, A. Bone mineral density in adult coeliac disease: An updated review. *Rev. Esp. Enferm. Dig.* **2013**, *105*, 154–162. [CrossRef] [PubMed]
42. Nijhawan, S.; Katiyar, P.; Nagaich, N.; Saradava, V.; Nijhawan, M.; Gupta, G.; Mathur, A.; Sharma, R.; Nepalia, S. Prevalence of associated disorders in Indian patients with celiac disease. *Indian J. Gastroenterol.* **2013**, *32*, 330–334. [CrossRef] [PubMed]
43. Whitton, C.; Nicholson, S.K.; Roberts, C.; Prynne, C.J.; Pot, G.K.; Olson, A.; Fitt, E.; Cole, D.; Teucher, B.; Bates, B.; et al. National Diet and Nutrition Survey: UK food consumption and nutrient intakes from the first year of the rolling programme and comparisons with previous surveys. *Br. J. Nutr.* **2011**, *106*, 1899–1914. [CrossRef] [PubMed]
44. Størsrud, S.; Hulthén, L.R.; Lenner, R.A. Beneficial effects of oats in the gluten-free diet of adults with special reference to nutrient status, symptoms and subjective experiences. *Br. J. Nutr.* **2003**, *90*, 101–107. [CrossRef] [PubMed]
45. Thompson, T. Thiamin, riboflavin, and niacin contents of the gluten-free diet: Is there cause for concern? *J. Am. Diet. Assoc.* **1999**, *99*, 858–862. [CrossRef]
46. Penagini, F.; Dilillo, D.; Meneghin, F.; Mameli, C.; Fabiano, V.; Zuccotti, G.V. Gluten-free diet in children: An approach to a nutritionally adequate and balanced diet. *Nutrients* **2013**, *5*, 4553–4565. [CrossRef] [PubMed]
47. Cornicelli, M.; Saba, M.; Machello, N.; Silano, M.; Neuhold, S. Nutritional composition of gluten-free food versus regular food sold in the Italian market. *Dig. Liver Dis.* **2018**. [CrossRef] [PubMed]
48. Kulai, T.; Rashid, M. Assessment of Nutritional Adequacy of Packaged Gluten-free Food Products. *Can. J. Diet. Pract Res.* **2014**, *75*, 186–190. [CrossRef] [PubMed]
49. Lee, A.R.; Ng, D.L.; Dave, E.; Ciaccio, E.J.; Green, P.H. The effect of substituting alternative grains in the diet on the nutritional profile of the gluten-free diet. *J. Hum. Nutr. Diet.* **2009**, *22*, 359–363. [CrossRef] [PubMed]

Manuscript 4

Idoia Larretxi, Itziar Churruca, Virginia Navarro, Jonatan Miranda,
Arrate Lasa, Maria Ángeles Bustamante and Edurne Simon

Impact of analytically measured fiber and resistant starch from gluten free products on celiac people´s diet

Manuscript

Impact of analytically measured fiber and resistant starch from gluten free products on celiac people´s diet

Running head: Fiber and celiac´s diet

Idoia Larretxi[1], Itziar Churruca[1], Virginia Navarro[1], Jonatan Miranda[1], Arrate Lasa[1], Maria Ángeles Bustamante*[1], Edurne Simon[1].

[1]Gluten Analysis Laboratory of the University of the Basque Country, Department of Nutrition and Food Science, University of the Basque Country (UPV/EHU), Vitoria-Gasteiz, Spain

*Corresponding author: Maria Ángeles Bustamante, Dpt. Nutrición y Bromatología, Facultad de Farmacia, Paseo de la Universidad 7, 01006 Vitoria-Gasteiz (Spain). Phone +34-945-01 4086, e-mail marian.bustamante@ehu.eus

Abstract

Objective: The aim of the present work was to assess fiber and resistant starch (RS) content of several Gluten Free Products (GFP), compare them with their gluten containing analogues (GCP) analytically and with label data, and to measure the contribution of these foodstuffs to the fiber content of a gluten free diet.

Methods: Total dietary fiber and RS content in a large amount of GFP (n=55) and their counterparts (n=55) was measured by analytical techniques based on AOAC methods. Dietary assessment was carried out with a sample of 141 individuals (pediatric and adult celiac patients) submitted to a food frequency questionnaire and a 24-h recall survey (in three different days).

Results: In general terms, GFP and GCP showed similar fiber and RS content, with the exception of breads, which revealed higher percentage of both in GFP. This food group was the main contributor to fiber intake adult and pediatric celiacs. Even if GFP were blamed for being nutritionally poorer than GCP, it is not the case of fiber.

Conclusions: It is noteworthy that label information can underestimate fiber consumption among celiac people, and it must be considered for future database revision. In addition, we demonstrated a high impact on fiber intake of children and adults due to GFP, and especially to bread, a widely consumed basic product.

Keywords: total dietary fiber; resistant starch; gluten-free diet; gluten-free products

1. Introduction

The gluten-free products (GFP) market has grown exponentially during recent decades. Nowadays many food companies are involved in the production of these foodstuffs. In fact, their popularity and consumption has not only increased among gluten-sensitive persons, but also in people not suffering from cardiovascular diseases (CVD) [1,2]. Whatever the reason for their use is, food industries have to deal with technical problems when gluten is removed from cereal-based foodstuffs.

Gluten is a complex mixture of proteins found in cereals from the Triticeae family. Wheat prolamin and glutelin are responsible for the rheological characteristics of dough; while gliadin brings viscosity, glutenin provides elasticity and strength [3]. To obtain the same rheological properties, other ingredients such as gums, hydrocolloids, fats or enzymes are used in GFP manufacture. However, it has been shown that they could modify the nutritional composition of GFP [4]. One of the raw material most used to improve the texture of GFP are starches [5,6], which can trap air bubbles, forming a viscoelastic mass close to wheat dough [7].

GFP are a valuable alternative for celiac patients, whose restrictive diet can be difficult to follow, but represents the only treatment with proven benefits. In recent years, negative impacts on health have been described for this restrictive diet, blaming the nutritional characteristics of GFP [8-10]. For instance, it has been stated that patients with CVD who follow a GFD do not always consume healthy amount of fats, carbohydrates and fiber [11]. It is important to note that gluten-containing grains, such as wheat, barley and rye, are an important source of complex carbohydrates, fiber and B-group vitamins; so the removal or substitution of those grains in diet and in GFP by other ingredients could adversely affect nutrient intake. Specifically, fiber plays a special role in the nutritional improvement of food products, due to its health promoting effects and its technological properties [12]. Many researchers who found that the GFD of celiac people is usually low in fiber content [13,14] and lower than that of the general population [15-17].

Starch represents an interesting ingredient for GFP. Concretely, a small proportion of it, called resistant starch (RS). The physiological behavior of RS, which is similar to that of the soluble, fermentable fiber, has placed the compound under the spotlight. Its positive effects range from the generation of short chain fatty acids because of the prebiotic effect in beneficial bacteria, to the decrease of postprandial glycaemia and insulinaemia as well as serum lipemia [18,19].

It must be noted that fiber analysis in foodstuffs is a complex process and methods for its quantification differ among laboratories. This results in widely variable fiber contents reported in databases and thus in inaccuracy, not only of fiber intake determination among celiac and non-celiac people, but also of the theoretical calculation of fiber content in labels. Moreover, poor data concerning the amount of different fiber components, such as RS, in addition to the total dietary fiber (TDF) content, is available.

The aim of the present study was to assess the fiber and RS content of some representative GFP and compare them with their gluten-containing counterparts. Moreover, the obtained analytical data were compared to those reported on the label by the food manufacturers. To our knowledge, this is the first research to analyze fiber and RS content in a large amount of GFP and their counterparts, using analytical techniques based on AOAC methods.

This study provides novelty by assessing the contribution of GFP to the total dietary fiber content of a GFD.

2. Material and Methods

2.1. Food samples

Conventional cereal derived products (n=55) and gluten-free cereal products (n=55) were purchased in local supermarkets. A “GFP” was considered a cereal-based foodstuff where the usual gluten-containing cereal with its inherent bread-making or baking characteristic was replaced by non-gluten-containing ingredients. Once in the laboratory, samples were maintained until analysis at room temperature or frozen at -18 ºC, according to the instructions of the manufacturer. Before analysis, samples were milled if necessary and all samples were dried at 105 ºC to reach a constant weigh for the analysis of TDF and RS.

2.2. Total dietary fiber analysis

TDF analysis was performed using the commercial kit Total Dietary Fiber (Megazyme International, Wicklow, Ireland) based on the AOAC method 991.43 [20] with the following modifications. A sample of around 1.3 g was mixed in a beaker with 40 mL of MES-TRIS blend buffer solution, pH 8.2, by vortexing for 30 seconds. Then, 50 µL of α-amylase solution were added to each beaker and incubated at 98-100 ºC for 30 minutes and then, cooled to 60 ºC. The beaker was rinsed with 9 mL of distilled water and samples were incubated with 100 µL of protease solution at 60 ºC for 30 minutes. After incubation, 4 mL of 0.561 N HCl and 200 µL of amyloglucosidase solution were added, and the beakers were incubated again at the same conditions. TDF was precipitated at room temperature by adding pre-heated 95% ethanol. The precipitate was filtered through a Celite® bed (Megazyme International, Wicklow, Ireland). After filtration, the residue was successively washed using vacuum with two 15 mL portions of 78% ethanol, 95% ethanol and acetone. Residue was dried overnight at 103 ºC, and the next day, it was cooled in a desiccator. Finally, it was weighed subtracting the weight of the crucible and Celite®. Ash (AOAC, 923.03) and protein (AOAC, 960.52A) content were determined in a duplicate residue in order to calculate the percentage of total dietary fiber [20].

2.3. Resistant starch analysis

RS was measured according to McCleary and Monaghan [21] using the commercial kit Megazyme Resistant Starch (Megazyme International, Wicklow, Ireland), approved by AOAC International (method 2002.02) [20]. It consists in an enzymatic method that mimics the in vivo conditions of the digestive system.

Briefly, the first step was hydrolysis and solubilization of non-resistant starch, which was done as follows: 4.0 mL of pancreatic α-amylase (10 mg/mL) containing amyloglucosidase enzyme (AMG, 3 U/mL) were added to 100 mg of dried sample weighed in a screw cap tube. In the second step, the RS was measured: the tubes with the pellet were placed in an ice bath and the pellets were re-suspended by adding 2 mL of 2 M KOH and stirred during 20 minutes. Then, 8 mL of 1.2 M sodium acetate buffer (pH 3.8) were added to each tube, stirred, and, immediately, 0.1 mL of AMG (3,300 U/mL) was added. The tubes were mixed again and incubated at 50°C for 30 minutes. Samples were centrifuged at 1,500 g for 10 min, 3.0 mL of GOPOD reagent were added to 0.5 mL of the supernatant and samples were incubated at 50°C for 20 min. Finally, the product's absorbance was measured at 510 nm.

Quantification of the RS was carried out following the calculations proposed by McCleary and Monaghan [21]. Results were expressed as percentages in the tested sample, also related to total starch.

2.4.Dietary assessment

Dietary assessment was carried out in 68 pediatric celiacs (42/26 female/male ratio) from 3 to 14 years old with a mean Body Mass Index (BMI) of 17.5 kg/m2 and 73 celiac patients (57/16 female/male ratio) older than 14 years old with a mean BMI of 21.9 kg/m2. For total fiber intake and food group consumption analysis in pediatric age, participants were divided into three groups aged between 3-6, 7-10 and 11-14 years old. All subjects gave their informed consent for inclusion before they participated in the study. They belonged to a cohort of patients from two previous studies which had been conducted in accordance with the Helsinki Declaration, and the protocol was approved by the Ethics Committee of the University of The Basque Country, UPV/EHU, (CEISH/76/2011 and CEISH/194M/2013) [22,23].

The sample of 141 individuals was submitted to a food frequency questionnaire and a 24-h recall survey, which was repeated three times on different days [17]. Printed food models, plates, glasses and spoons were used as visual aids to improve information recovery. In order to determine TDF intake of celiac people, recall information was processed by a computerized program system (AyS, Software, Tandem Innova, Inc.). The fiber content data (analytical or from the label, when available) of the specific manufactured GFP for celiac people were added to the program database. Dietary reference intakes (DRI) for Spanish population issued by the Spanish Societies of Nutrition, Feeding and Dietetics (FESNAD) in 2010 were taken as references for the interpretation of fiber intake [24].

2.5. Data treatment and statistical analysis

IBM SPSS Statistics software version 23.0 (New York, USA) was used for statistical analysis. The results were given as the arithmetic mean (standard deviation). Linear regression analysis was used to examine the association of fiber content of each food group with that of total diet (variables with skewed distribution were logarithmically transformed to obtain a more symmetrical distribution). Mann-Whitney U test was applied to establish significant differences in the TDF and RS content between conventional and GFP and between analytical results and label information. In all cases, significance level was set at $P \leq 0.05$.

3. Results

3.1. Comparison between analytical fiber content and fiber data in label between gluten-containing and gluten-free rendered foodstuffs

Table 1 illustrates analytical and label fiber content of some representative GFP compared to their gluten-containing counterparts. With the exception of breads, the rest of assayed cereal-based products did not show differences fiber content calculated by analytical methods. In the case of breads, higher fiber content was found in GFP than in gluten-containing breads. No modification in fiber data label was observed between 55-paired samples. By contrast, analytical and label fiber content in the cereal-based products of the study revealed statistical differences in the case of bread, and bakery and cakes food-groups.

3.2. Analytical resistant starch assessment in gluten-containing and gluten-free rendered foodstuffs

When comparison between GFP and their counterpart was performed, no signifficant differences were found in bakery and cakes, biscuits, breakfast cereals, pasta and flours and dough food-groups (Table 2). Analyzed gluten-free bread showed higher values of RS and RS/Total starch ratio (RS/TS) than gluten-containing breads (P=0.002 and P=0.011, respectively). In the case of cereal infant formulas, the measurements revealed opposite trends, with lower values of RS and RS/TS in GFP than in their counterparts (P=0.043 and P=0.05, respectively).

3.3. Fiber intake in adult and pediatric celiacs

Taking into account label fiber content, fiber intake of celiac participants was calculated, by age group and gender (Supplementary table). In the case of adult celiac participants, the observed fiber intake was 16.6 (5.8) g/d for women and 20.2 (6.8) for men. These results did not follow dietary reference intakes for Spanish people. Similarly, fiber intake of celiac children was below recommendations (14.4 g/d, for girls, and 15.1 g/d, for boys).

3.4. Contribution of gluten-free products to the total dietary fiber content in GFDs

The intake of cereals as well as vegetables was below the recommendations for adult and pediatric celiacs. In the case of legumes, celiac children and adolescents achieved the intake recommendation, but not celiac adults. On the contrary, celiac adults as well as children in the range of 3-10 years old consumed enough grams per days of fruits, while celiac adolescents did not reached the intake recommendation. Linear regression for the influence of each food groups revealed that the main contributors to the fiber intake in adult and pediatric celiac following a gluten-free diet, are GFP (P<0.001) (Table 3).

4. Discussion

In the comparison of GFP with their GC counterparts, data revealed that, generally, fiber content of cereal-based GF foodstuffs was similar or even higher than that of their homologous GC partner. This was especially remarkable in breads, whose analytical data revealed a higher fiber content in GFP than in gluten-containing breads (9.8% and 3.9%, respectively), as reported elsewhere [25]. Surprisingly, these analytical data were not always in line with the corresponding label (when available), whose displayed fiber content was significantly lower, being nearly halved in GF breads. It must to be highlighted that all studies in the literature (ours included) have always calculated the GFP fiber composition based on label information. Even though the theoretical calculation of nutritional information of food products is less accurate, the European Union normative [26] allows it. Fiber content information is not mandatory, so it is not always available for consumers. This is especially important for specific food products such as GFP, whose composition can be different from their counterparts and does not usually appear in general databases.

Although some studies in the past found lower fiber content in GFP [27], recently published works [10,28], did not find differences in fiber content between GFP and their gluten-containing counterparts, which is in accordance with data from the present study. This positive evolution in fiber content could be due to the profound innovations made by GFP over the last few years [29]. GFP have evolved to be not only safer from the gluten contamination perspective [30], but also more nutritionally balanced in terms of gluten content. Focusing on fiber, the use of new ingredients has improved, at least in part, the nutritional profile of these products.

As explained in the introduction section, RS takes the role of the fermentable fiber and thus exerts beneficial effects on several pathologies, raising its interest in nutritional amelioration of GFP matrixes. Our analytical results revealed a similar trend of RS content in GFP to that observed in total dietary fiber similarities between TDF and RS. However, GF breads showed more RS content than gluten containing breads, which is in accordance to the higher fiber content observed for this food group, and in accordance with other studies [31,32]. This fact could be justified by the use of some starchy ingredients for texture and structure construction [5], and due to the selection of bread as the election matrix because of its important role in the daily diet. On the other hand, GF cereal infant formula showed lower amount of RS than gluten-containing formula. In this case, the use of different grains could directly alter the final RS content of the product, but also the use of different ingredients (vitamins and fiber) could affect the digestibility of the starch present in the final product.

The average consumption of cereals and vegetables was low in all celiac participants. Comparing these results with those from other studies where the same aged control populations were analyzed [33,34], lower consumption of cereals was observed in both celiac populations from the present work. Similar findings were reported by Babio *et al*. among Spanish children and adolescents with celiac disease [35]. By contrast, the consumption of other fiber rich food groups was higher than that reported for the control population: children consumed more vegetables and more fruits (in 11-14 years old range), and celiac adult ate more legumes and fruit. These results suggested that the strategy used to avoid gluten consisted in reducing grain consumption.

In addition, the contribution of each fiber-rich food group to the TDF consumption was calculated. The main fiber source for celiac population was GFP group, entailing 53% and 50% of the fiber intake in pediatric and adult participants, respectively. Accordingly, linear regression revealed that GFP was a significant food group for dietary fiber intake. The contribution of GFP to total dietary fiber observed was not as low as reported in previous studies [16,17], especially for breads. Different dietary habits have been reported for bread consumption in celiac people, compared to controls. In order to assure a high impact on the global diet of a given population, this information results essential for the designing specific products. In our study, even though the intake of cereal-based foodstuff in celiac group was lower than in control, bread consumption was similar. However, Zuccotti *et al*. [36] found higher bread and rice intake in 4-10 years old celiacs and Valitutti *et al*. [37] reported lower bread intake in adult celiacs. Differences in gastronomy and dietary habits between countries may have brought about this discrepancy between studies, resulting in a different impact on GFD. In the present study, gluten free bread provided 54.4% and 68.4% of the total fiber coming from GFP in children and adults respectively. Thus, it is remarkable that nutritional improvements performed in a basic product, such as bread, have had such a noticeable impact on diet.

Although fiber intake in GFD was only slightly below recommendations (21-38 g/d, depending on age), considering that the celiac population has a higher prevalence of risk factors for CVD, the dietary pattern of celiac people should be balanced with an increase in fiber amount. Moreover, fiber coming not only from GF breads, but also from other natural important fiber sources, such as fruits, vegetables and legumes, which are also rich in other important nutrients such as vitamins and minerals should be enhanced. However, this does not imply that an appropriate offer of balanced processed food could not positively influence their adherence to a GFD, which is sometimes stressful to follow. Nevertheless, to that end, nutrients other than fiber must be also assessed, like saturated fats or sugars, to holistically enhance GFP nutritional value.

In the case of RS, even though some GFP showed higher contents than their counterparts, it has to be noted that in order to benefit from RS's positive physiological effects, a dose of 20 g/d is recommended, whereas worldwide intakes vary from 5 to 40 g/d [19]. In our study, RS intakes coming from GFP were around 4 g/d. Then, although RS intake could be slightly higher after consuming GFP, it remains quite a way from the recommended dose, even considering the RS from other sources (like legumes and fruits), which was not included.

Finally, it must be pointed out that food industries should emphasize this great effort to improve GFP updating fiber analytical content in nutritional labelling. Due to the fact that these are the tools that are used to assess nutrient intake and taking into consideration the differences observed between label and analytical data of GFP and estimated TDF intake. This highlights the importance of updating food composition databases to avoid errors when dietary intakes are analyzed in nutritional studies.

5. Conclusions

Even though GFP have been assumed to be low in fiber and in other important nutrients, here we reported an equivalent fiber and RS content, which was even higher in breads, compared to their homologues. These results showed the improvements carried out over recent years in innovation and research in GFP manufacture in order to enhance their nutritional value. In addition, here we demonstrated a high impact on fiber intake of children and adults due to GFP, and especially to bread -a widely consumed basic product-. This shows the importance of strategically improving food products in order to reach the goal of ameliorating the nutritional balance of consumer’s diet.

Moreover, label information, which can be theoretically calculated, was different from the analytical results in some cases. This highlights the importance of updating and revising food composition databases frequently. In fact, as GFP are not always available on food composition databases, estimation of fiber intake in celiac population might be inaccurate. Here we show that the use of label or analytical information on determining the fiber intake of GFD could completely change the final diagnosis.

Acknowledgments

Idoia Larretxi is a PhD fellowship of the University of the Basque Country, UPV/EHU (Euskararen eta Etengabeko Prestakuntzaren arloko Errektoreordetza). This research was supported by a grant from the UPV/EHU (University-Society US15/06).

We thank the collaboration of "Asociación de Celiacos de Euskadi (EZE)", specifically that of Mireia Apraiz, and the collaboration of Servicio de Gastroenterología y Nutrición Pediátrica, Hospital Universitario Donostia, specifically that of Laura Benjumea in the celiac people recruitment.

References

[1] Kim H, Demyen M, Mathew J, Kothari N, Feurdean M, Ahlawat S. Obesity, Metabolic Syndrome, and Cardiovascular Risk in Gluten-Free Followers Without Celiac Disease in the United States: Results from the National Health and Nutrition Examination Survey 2009-2014. Digestive Diseases and Sciences. 2017; 62: 2440-48.

[2] Kim HS, Patel KG, Orosz E, et al. Time Trends in the Prevalence of Celiac Disease and Gluten-Free Diet in the US Population: Results From the National Health and Nutrition Examination Surveys 2009-2014. JAMA Intern Med. 2016; 176: 1716-17.

[3] E S, I L, I C, et al. Nutritional and analytical approaches of gluten-free diet in celiac disease: Springer, Cham, 2017.

[4] Matos ME, Rosell CM. Understanding gluten-free dough for reaching breads with physical quality and nutritional balance. J Sci Food Agric. 2015; 95: 653-61.

[5] Witczak M, Ziobro R, Juszczak L, Korus J. Starch and starch derivatives in gluten-free systems - A review. Journal of Cereal Science. 2016; 67: 46-57.

[6] Horstmann SW, Lynch KM, Arendt EK. Starch Characteristics Linked to Gluten-Free Products. Foods. 2017; 6.

[7] Woo H-D, We GJ, Kang T-Y, et al. Physicochemical and Gelatinization Properties of Starches Separated from Various Rice Cultivars. Journal of Food Science. 2015; 80: E2208-E16.
[8] Kulai T, Rashid M. Assessment of Nutritional Adequacy of Packaged Gluten-free Food Products. Can J Diet Pract Res. 2014; 75: 186-90.
[9] Lebwohl B, Cao Y, Zong G, et al. Long term gluten consumption in adults without celiac disease and risk of coronary heart disease: prospective cohort study. Bmj-British Medical Journal. 2017; 357.
[10] Missbach B, Schwingshackl L, Billmann A, et al. Gluten-free food database: the nutritional quality and cost of packaged gluten-free foods. PeerJ. 2015; 3: e1337.
[11] Vici G, Belli L, Biondi M, Polzonetti V. Gluten free diet and nutrient deficiencies: A review. Clin Nutr. 2016.
[12] Lockyer S, Spiro A, Stanner S. Dietary fibre and the prevention of chronic disease – should health professionals be doing more to raise awareness? Nutrition Bulletin. 2016; 41: 214-31.
[13] Kautto E, Ivarsson A, Norström F, Högberg L, Carlsson A, Hörnell A. Nutrient intake in adolescent girls and boys diagnosed with coeliac disease at an early age is mostly comparable to their non-coeliac contemporaries. J Hum Nutr Diet. 2014; 27: 41-53.
[14] Salazar Quero JC, Espin Jaime B, Rodriguez Martinez A, et al. [Nutritional assessment of gluten-free diet. Is gluten-free diet deficient in some nutrient?]. An Pediatr (Barc). 2015; 83: 33-9.
[15] Ohlund K, Olsson C, Hernell O, Ohlund I. Dietary shortcomings in children on a gluten-free diet. J Hum Nutr Diet. 2010; 23: 294-300.
[16] Martin J, Geisel T, Maresch C, Krieger K, Stein J. Inadequate nutrient intake in patients with celiac disease: results from a german dietary survey. Digestion. 2013; 87: 240-6.
[17] Miranda J, Lasa A, Bustamante MA, Churruca I, Simon E. Nutritional Differences Between a Gluten-free Diet and a Diet Containing Equivalent Products with Gluten. Plant Foods For Human Nutrition. 2014; 69: 182-87.
[18] Fuentes-Zaragoza E, Riquelme-Navarrete MJ, Sánchez-Zapata E, Pérez-Álvarez JA. Resistant starch as functional ingredient: A review. Food Research International. 2010; 43: 931-42.
[19] Moraes C, Borges N, Mafra D. Resistant starch for modulation of gut microbiota: Promising adjuvant therapy for chronic kidney disease patients? European Journal of Nutrition. 2016; 55: 1813-21.
[20] AOAC International. Official Methods of Analysis of AOAC International, 18th Edition, 2005 (Revision 3, 2010).

[21] McCleary B, Monaghan D. Measurement of resistant starch. Journal of Aoac International. 2002; 85: 665-75.
[22] Churruca I, Miranda J, Lasa A, Bustamante M, Larretxi I, Simon E. Analysis of Body Composition and Food Habits of Spanish Celiac Women. Nutrients. 2015; 7: 5515-31.
[23] Larretxi I, Simon E, Benjumea L, et al. Gluten-free-rendered products contribute to imbalanced diets in children and adolescents with celiac disease. Eur J Nutr. 2018.
[24] FESNAD. Dietary reference intakes (DRI) for spanish population, 2010. Actividad Dietética. 2010; 14: 196-97.
[25] Segura ME, Rosell CM. Chemical composition and starch digestibility of different gluten-free breads. Plant Foods Hum Nutr. 2011; 66: 224-30.
[26] Regulation (EU) No 1169/2011 of the European Parliament and of the Council of 25 October 2011 on the provision of food information to consumers. Official Journal of the European Union, L304, (November 22, 2011).
[27] Thompson T, Dennis M, Higgins LA, Lee AR, Sharrett MK. Gluten-free diet survey: are Americans with coeliac disease consuming recommended amounts of fibre, iron, calcium and grain foods? J Hum Nutr Diet. 2005; 18: 163-9.
[28] Wu JH, Neal B, Trevena H, et al. Are gluten-free foods healthier than non-gluten-free foods? An evaluation of supermarket products in Australia. Br J Nutr. 2015; 114: 448-54.
[29] Stantiall SE, Serventi L. Nutritional and sensory challenges of gluten-free bakery products: a review. Int J Food Sci Nutr. 2017: 1-10.
[30] Bustamante MA, Fernandez-Gil MP, Churruca I, et al. Evolution of Gluten Content in Cereal-Based Gluten-Free Products: An Overview from 1998 to 2016. Nutrients. 2017; 9: 12.
[31] Murphy MM, Douglass JS, Birkett A. Resistant starch intakes in the United States. J Am Diet Assoc. 2008; 108: 67-78.
[32] Alsaffar A. Effect of food processing on the resistant starch content of cereals and cereal products - a review. International Journal of Food Science and Technology. 2011; 46: 455-62.
[33] Gobierno Vasco. Encuesta de nutrición 2005. Vitoria-Gasteiz: Departamento de Sanidad, 2005.
[34] Agencia Española de Seguridad Alimentaria y Nutrición (AESAN), Evaluación Nutricional de la Dieta Española I. Energía y Macronutrientes, sobre datos de la Encuesta Nacional de Ingesta Dietética (ENIDE), 2011.
[35] Babio N, Alcazar M, Castillejo G, et al. Patients With Celiac Disease Reported Higher Consumption of Added Sugar and Total Fat Than Healthy

Individuals. Journal of Pediatric Gastroenterology and Nutrition. 2017; 64: 63-69.
[36] Zuccotti G, Fabiano V, Dilillo D, Picca M, Cravidi C, Brambilla P. Intakes of nutrients in Italian children with celiac disease and the role of commercially available gluten-free products. J Hum Nutr Diet. 2013; 26: 436-44.
[37] Valitutti F, Iorfida D, Anania C, et al. Cereal Consumption among Subjects with Celiac Disease: A Snapshot for Nutritional Considerations. Nutrients. 2017; 9.

Table 1. Analytical and label fiber content in gluten-containing and gluten-free rendered foodstuffs divided by food groups.

	Analytical Fiber (%)				Fiber data in label (%)				Analytical vs. label	
	N (GFP/GCP)	GFP	GCP	P	N (GFP/GCP)	GFP	GCP	P	GFP P	GCP P
Bread	11/11	9.8(4.8)	3.9(1.2)	< 0.001	10/10	5.5(2.7)	4.0(1.7)	NS	< 0.01	NS
Bakery and cakes	14/14	3.4(2.2)	2.4(1.0)	NS	10/11	2.0(1.7)	2.3(1.0)	NS	< 0.01	NS
Biscuits	11/10	3.7(2.4)	3.6(1.5)	NS	10/7	2.9(1.3)	1.8(1.2)	NS	NS	< 0.05
Breakfast cereals	3/3	4.2(2.2)	6.0(1.1)	NS	3/3	3.9(0.9)	5.8(1.6)	NS	NS	NS
Cereal infant formula	3/3	2.0(2.1)	4.1(1.2)	NS	3/3	3.4(1.5)	3.1(0.2)	NS	NS	NS
Pasta and flours	6/7	7.9(2.5)	7.4(2.5)	NS	5/3	4.5(5.0)	2.8(0.3)	NS	NS	NS
Doughs	7/7	6.4(3.7)	3.5(1.9)	NS	2/4	4.2(5.9)	2.2(0.3)	NS	NS	NS

Notes: Values are means (standard deviation). GFP, gluten-free product; GCP, gluten-containing product; *P*, statistical significance; NS, not significant.

Table 2. Analytical resistant starch content in gluten-containing and gluten-free rendered foodstuffs divided by food groups.

		RS (%)			RS/TS (%)		
	N (GFP/GCP)	GFP	GCP	P	GFP	GCP	P
Bread	11/11	3.6 (1.5)	1.3 (1.0)	0.002	6.1 (2.9)	3.5 (4.3)	0.011
Bakery and cakes	14/14	1.7 (1.4)	0.9 (0.5)	NS	5.1 (4.7)	2.4 (0.8)	NS
Biscuits	11/10	1.3 (1.0)	0.6 (0.4)	NS	3.5 (2.3)	2.0 (1.4)	NS
Breakfast cereals	3/3	1.6 (1.8)	0.9 (1.0)	NS	2.5 (2.5)	2.8 (3.6)	NS
Cereal infant formula	3/3	0.1 (0.1)	0.6 (0.2)	0.043	0.1(0.1)	0.9 (0.3)	0.05
Pasta and Flours	6/7	1.0 (1.0)	1.5 (1.8)	NS	1.6 (1.6)	2.3 (2.2)	NS
Doughs	7/7	0.9 (0.4)	1.0 (0.8)	NS	1.9 (0.8)	2.2 (1.8)	NS

Notes: Values are means (standard deviation). GFP, gluten-free product; GCP, gluten-containing product; *P*, statistical significance for the differences between GFP and GCP; NS, not significant; RS, resistant starch; TS, total starch.

Table 3. Fiber intake of gluten-free diet in adult and pediatric celiacs and linear regression for the contribution of each food group

	0-14 years old (N=68)					>14 years old (N=73)				
	g/day	%	ß	SE	P	g/day	%	ß	SE	P
Legumes	1.7 (2.3)	8	0.452	0.018	<0.001	2.0 (2.3)	8	0.228	0.018	<0.001
Fruit & vegetables	4.6 (2.3)	21	0.343	0.031	<0.001	6.8 (3.6)	26	0.275	0.023	<0.001
GFP	11.7 (5.3)	53	0.741	0.046	<0.001	13.2 (7.6)	50	0.657	0.042	<0.001
Rice, corn and derivatives	0.6 (0.8)	3	-	-	NS	0.5 (0.8)	2	-	-	NS
Other sources	3.6 (2.2)	16	0.283	0.023	0.001	4.1 (2.8)	15	0.218	0.031	0.001
Total fiber intake	22.2 (6.8)	100				26.5 (10.6)	100			

Notes: Fiber intake values are means (standard deviation). β Lineal regression coefficients; SE, standard errors; GFP, gluten-free products; N: number of participants; *P*, statistical significance; NS, not significant; %: percentage of total intake. Analysis was performed with log-transformed data.

Supplementary Table. Fiber intake of celiac participants, according to label fiber content, by age group and gender.

Participants	Adult celiac participants			Pediatric celiac participants			3-6 years old			7-10 years old			11-14 years old		
	All	Women	Men	All	Girls	Boys	All	Girls	Boys	All	Girls	Boys	All	Girls	Boys
Fiber (g/d)	17.3±6.1	16.6±5.8	20.2±6.8	14.7±4.0	14.4±4.0	15.1±4.1	14.0±3.7	12.5±3.4	16.3±3.0	15.1±4.3	15.4±3.5	14.4±5.7	15.1±4.2	15.6±4.9	14.4±3.2
Fiber DRI		19-50y: 38 g/d men, 25 g/d women > 51 y: 30 g/d men, 21 g/d women						25 g/d			7-8y: 25g/d 9-10y:31 g/d boys, 26g/d girls			11-13y: 31 g/d boys, 26g/d girls 14y: 38 g/d boys, 26g/d girls	

Notes: Values are means ± SD. SD, standard deviation. DRI, Dietary Reference Intake of fiber (proposed by the Federation of Spanish Societies of Nutrition and Dietetics. FESNAD). g/d, graams/day. Y, years old.

Manuscript 5

Larretxi I, Churruca I, Navarro V, Lasa A, Bustamante MA, Simon E, Miranda J

Micronutrient analysis of gluten-free products: their low content is not involved on gluten-free diet misbalance

Manuscript

Micronutrient analysis of gluten-free products: their low content is not involved in gluten-free diet imbalance

Larretxi I, Churruca I, Navarro V, Lasa A, Bustamante MA, Simon E, Miranda J.

Gluten Analysis Laboratory of the University of the Basque Country, Department of Nutrition and Food Science, University of the Basque Country (UPV/EHU), Vitoria, Spain

Running title: Micronutrient content of gluten-free products

Corresponding authors/Request for reprints

Bustamante M. and Simon E. PhD. Department of Nutrition and Food Science, Faculty of Pharmacy, University of the Basque Country (UPV/EHU), Paseo de la Universidad, 7, 01006 Vitoria-Gasteiz (Spain).

Telephone number: +34-945-014086/ Fax number: +34-945-013014

e-mail address: marian.bustamante@ehu.eus, edurne.simon@ehu.eus

Abstract

Data about nutritional composition of gluten-free products (GFP) are still limited. Most studies are based on ingredient and nutrition information described on the food label. However, analytical determination is considered the gold standard for compositional analysis of food. Macronutrient and micronutrient analytical content differences were observed in a selection of GF breads, flakes and pasta, when compared with their respective gluten-containing counterparts. In general terms, lower iron, piridoxin, riboflavin, thiamin, niacin, folate, manganese and vitamin B5 can be underlined. Variations in biotin and vitamin E content differed among groups. In order to clarify the potential contribution of the GFP to the GFD's micronutrient shortages, analytical data were used to evaluate GFD in a cohort of celiac children and adolescent. Participants did not reach recommendations for vitamin A, vitamin E, folic acid, vitamin D, biotin, iodine, copper and chlorine. It does not seem that the lower micronutrient content of analyzed GFP groups contributed to the micronutrient deficits detected in GFD. Nevertheless, GFP fortification for folate and biotin is proposed to prevent the deficiencies observed in GFD, at least in the case of pediatric celiac.

Keywords Celiac disease; gluten-free diet; gluten-free product, micronutrient, vitamin and minerals, dietary recommendation.

Abbreviations CD: Celiac disease; GFP: Gluten-free product; GCP, gluten-containing product; GFD: Gluten-free Diet

Introduction

Celiac disease (CD) is a chronic immune-mediated inflammatory pathology triggered by the gluten in the diet of genetically predisposed individuals. The need to avoid this protein in the diet of celiac people brought about some years ago the development of specific cereal-based gluten-free products (GFP). Despite the fact that these GFP allowed them to include a wide variety of foods, in recent years researchers have highlighted differences in nutrient composition of GFP with respect to gluten containing counterparts (1, 2), leading to a minor health rating in some food-groups (3, 4).

It is important to note that most of the studies about nutrient composition of the GFP are based on ingredients and nutrition information described on the food label (2-4). Some works, such as that carried out by Mazzeo et al (2015) (5), take advantage of the retention factors for each nutrient, including losses due to heating or other food preparation steps, to improve these data. However, analytical determination is considered the gold standard for composition analysis of food. Accurate analysis could also provide detailed information about vitamins and minerals which is not totally or commonly available on label (6). Therefore, access to micronutrient data is already restricted to hardly any research (7-9).

Furthermore, a gluten-free diet (GFD) often implies some nutritional imbalances, as recognized in the literature (10, 11). Not only have inadequate fat, protein, sugar and fiber consumption been observed in GFD, but also a poor intake of micronutrients such as iron, zinc, magnesium, calcium, folate, vitamin D and B12 (12). Similarly, celiac people seem to have lower blood values for haemoglobin, ferritin, vitamin D, and cooper than the rest of the population (13, 14). There has been speculation about whether the characteristic composition of GFP is responsible for GFD inadequacy. A potential correlation between both facts has been proposed by others (15).

In view of the above, the aim of this study was to assess analytically the macronutrient and micronutrient content of a selection of GF breads, flakes and pasta, and to compare it with their respective gluten-containing counterparts. Additionally, in order to clarify the potential contribution of the GFP to the GFD's micronutrient shortages, vitamins measured and mineral analytical data were used to evaluate GFD in a cohort of celiac children and adolescent.

Materials and Methods

Analytical nutrient content of GF bread, breakfast cereals and pasta

The measured samples were thirty-seven selected GFP signed with the Crossed Grain symbol: 13 breakfast cereals, 12 breads and 12 pasta products. All the food items were purchased from the local market (Vitoria, Spain) and they were stored frozen (–20°C) until analyzed.

Crude protein and fat contents were determined by the Kjeldahl and Soxhlet extraction method (AOAC 2005) (16), respectively. After measuring moisture and ash content, carbohydrates were estimated by difference. These results were compared to those of equivalent gluten-containing products, which were analyzed in the same way and at the same time, and to the data described in the food label of GFPs.

For mineral determination, microwave-assisted digestion was carried out in a closed microwave device Mars 5 (CEM, Vertex, Barcelona, Spain) equipped with 8 - 24 teflon vessels and temperature controllers. The quantitative analysis of selenium, manganese and cooper was performed by using ICP-MS (7700x, Agilent Technologies, Palo Alto, USA) and MicroMist micro-uptake glass concentric nebulizer (Glass Expansion, West Melbourne, Victoria, Australia). ICP-OES (Horiba Jobin Yvon, Activa) was used with a quartz Meinhard concentric nebulizer, a Scott-type spray chamber and a standard quartz sheath connection between the spray chamber and the torch, in the case of calcium, sodium, zinc and iron quantification.

Biotin, Folate, Niacin, Piridoxin, Riboflavin, Tiamin, vitamin B5 and B12 were measured by liquid chromatography (LC) with triple quadrupole mass spectrometry detection. The identification of each vitamin was achieved by positive ionization of the electrospray and multiple reaction monitoring. As a step prior to vitamin quantification, samples were extracted by liquid-liquid extraction using an aqueous acidic mixture, centrifuged and filtrated. Vitamin E determination was carried out by previous saponification of the samples, followed by a liquid-liquid extraction and purification of the extracts. Afterwards, high performance LC with fluorescence detector method was used to analyze vitamin E in each extract.

Analytically determined micronutrient content of GF foodstuffs was compared with the data of equivalents gluten-containing breads (n = 19), breakfast cereals (n = 18), and pasta products (n = 8) obtained from Spanish Food Composition Database - BEDCA database- (17). Data for biotin in all studied food groups and copper in cereals were obtained from McCance and Widdowson’s “composition of foods integrated dataset” from the United

Kingdom (18). No available data were found with regard to manganese content of cereal flakes in food composition databases from the UK, Australia, the USA or Spain (17-20).

Subjects

Eighty-three minor celiac (53 girls and 30 boys) from the Basque Country took part in the study. The age of the participants was selected due to their higher consumption of GFP comparing to adults (21, 22). All participants received oral and written information about the nature and purpose of the survey, and all of them gave written consent for involvement in the study. This study was approved by the Ethical Committee in University of Basque Country (CEISH/76/2011 and CEISH/194M/2013).

Dietary Assessment

The dietary assessment followed in the research was described elsewhere (22). Three days food records (two weekdays and one weekend day) were selected for each patient, 24-hour food recall were filled in by each celiac patient. Micronutrient intake was calculated by a computerized nutrition program system (AyS, Software, Tandem Innova, Inc.). The analytically measured vitamin and mineral content of tested GF products was added into the food composition database of the program before calculations. Dietary reference intakes (DRI) for Spanish population issued by the Spanish Societies of Nutrition, Feeding and Dietetics (FESNAD) in 2010 were taken as references for the interpretation of the 24HR (23).

Statistical analysis

Results are presented as mean ± standard deviation of the mean (SD). Statistical analysis was performed using SPSS 24.0 (SPSS Inc., Chicago, IL, USA). After confirming the normal distribution of lipid, protein and carbohydrate content variables using Shapiro-Wilks normality, paired-samples student's t test was used for comparison. Due to their skewed distribution, micronutrients variables for analytical and database information were analyzed by Mann-Whitney U. The level of significance was set to $p<0.05$.

Results and Discussion

Macronutrient content of GF rendered foods

With the aim of assessing representative products of a GFD, GFP from the three main cereal food-types contributing to a balanced diet, such as flakes, pasta and bread, were selected. Macronutrient content of the three GFP groups analyzed is shown in Table 1. Results were compared to nutritional composition of their gluten-containing counterparts. With regard to breads, lipid content was higher and protein content was lower than that of gluten containing products. Similarly, GF bread has been described as poor in proteins and rich in fat content by others (24). GF pasta provided higher carbohydrate content but lower protein amount; although in the last case the comparison to gluten containing pasta did not reach statistical significance. In general terms, lower protein content in GFP than in their counterpart has been proposed by previous research (2-4). Nevertheless, and in good accordance with our data, Missbach et al did not observe this pattern in the case of flakes (2).

It is important to point out that the comparative study between GFP and their homologues with gluten in the present work was performed as suggested by Staudacher and Gibson (6), by direct analytical methods and in paired form. As stated in the introduction, most of the studies evaluating the differences between both foodstuffs are based on nutrition information taken from the food label. For this reason, the analytical results obtained were compared to those reported in the nutritional panel information and some interesting data were collected. With regard to bread, experimental data reported a lower lipid (23%, $p=0.07$) and higher protein (37%; $p=0.03$) content than that supplied by the label. Similarly, in the case of cereal flakes, measured protein amount was higher (19%; $p=0.04$). No differences were observed between analyzed and labelled information in GF pasta.

In view of Regulation (EU) No 1169/2011 (25), the declared values on labels shall be average values based on a) the manufacturer's analysis of the food; b) a calculation from the known or actual average values of the ingredients used; c) a calculation from generally established and accepted data. It is not possible for us to determine how each manufacturer calculated label information. However, it must be highlighted that nutrient variations observed in bread types are not within the tolerance ranges between label information and our direct food analysis (tolerance ranges: ±1.5g for lipids and ±2g for proteins, when its content in food is <10g per 100 g). This information brings to light that previous studies about bread described in the literature could be reconsidered, and additionally, it validates, in part, others about pasta and cereals.

Micronutrient content of GFP, compared to gluten-containing products

Despite the growing market of the GFP (26), data about their vitamin and mineral contribution remain scarce. Moreover, the data found in the literature are usually calculated from ingredients and their composition databases, which has been proposed to lead to overestimation (5). Table 2 shows analytical micronutrient content of GF bread, flakes and pasta, compared to that of their gluten-containing counterparts. Lower iron, piridoxin, riboflavin and thiamin content was found in the three GFP groups analyzed. Niacin reduction was observed in GF flakes and breads. With regard to iron, similar results were found by Rybicka (8), who described that 273 of 408 GFP analyzed fulfilled less than 10% of recommended nutrient intake per portion and only 23 products were major contributors to daily intake (over 25% of recommendation intake per portion). In research performed with 368 GFP, including flours, breads, pasta and cold cereals, overall it was observed that this kind of products contained lower amounts of thiamin, riboflavin and niacin than the wheat product they were intended to replace (27). These results are in line with the results obtained in the present study.

Folate content was lower in GF flakes and bread types but only reached statistically significance in the latter; manganese amount was lower only in GF pasta, and that of vitamin B5 in GF flakes. Commonly used ingredients for GFP are maize and rice flours as well as a variety of starches (potato, corn), among others. It seems that removal of protein-rich fractions from flours may result in dramatic depletion of folates. Additionally, rice flours are not very rich in this vitamin (9)). In fact, we calculated a reduction of almost 80% of folate content in rice flour with respect to wheat flour (p=0.05) comparing nutrient composition of both flours obtained from food composition databases from the UK, Australia, the USA or Spain 17-20).

Several studies have claimed lower zinc and cooper and higher sodium content for GFP (4, 28). However, no significant differences in those minerals were found in our data.

Finally, biotin content differed widely among groups, being higher in cereal flakes and lower in pasta GFP than in their counterparts. Moreover, we found that some GF cereals were fortified with biotin, thus explaining its higher content in this GF food group. Similarly, although vitamin E contribution from GFP was lower in flakes, no differences were observed in pasta and bread. Moreover, it is worth mentioning that some bread types showed a formulation with sunflower oil, which led to higher vitamin E content in those specific stuffs.

It is important to point out that food technology interventions to improve the shelf life and rheological properties of GFP have influenced their nutritional profile (12). In order to avoid the absence of the mentioned micronutrients without fortifying foodstuffs, different strategies can be proposed: avoiding starch as a major ingredient, sourdough fermentation, and using less popular grain GF flour such as that from pseudocereals (buckwheat, quinoa, amaranth and teff), including wholemeal forms of gluten-free cereals (29, 30).

Micronutrient intake in celiac children and adolescents

It is known that GFD can lead to imbalanced macronutrient distribution. Our previous work (22) reported that celiac children and adolescent consumed more fat and less carbohydrate than recommended and pointed at GF rendered foods as one of the culprits. Thus, taking into account directly analysed micronutrient content, their intake on that pediatric cohort was calculated, stratified by age group and gender, and compared to FESNAD' recommendations (Supplementary Figure 1).

More than 1/4 of participants did not reach recommendations for vitamin A and vitamin E. Four out of ten children and adolescents with CD showed low intake of folic acid, which was even less than 66% of the recommendation for 25% of participants. 60% of participants did not get that for vitamin D, and moreover, about 40% of them did not reach 25% of the recommendation. Most participants showed very low intakes of biotin, iodine, copper and chlorine. Slightly over half the participants did not fulfil 50% of iodine recommendation and more than 40% were no able to achieve 25% of that of biotine. The intake of the rest of micronutrient was appropriate. With the exception of vitamin D, the results obtained differ from those obtained in similar pediatric research on celiac children, where iron, calcium, selenium and magnesium low intake was observed (31-33).

Considering all the above mentioned, it does not seem that GFP groups analyzed contribute to the micronutrient deficits detected in young celiac people's diet. In fact, cereals are not the natural source of these micronutrients. In this scenario, general recommendations to promote healthy GFD should be given, including greater consumption of vegetables and fruits. Moreover, in our previous study (Larretxi et al) we reported unhealthy dietary habits (low cereal, vegetable, fruit, nut, and excessive meat consumption) in this celiac cohort.

It must be pointed out that, in the case of folic acid, we observed a lower content of this vitamin in GFP than in their gluten containing equivalents. In this regard, in Canada and EEUU (34, 35) the fortification of wheat flour with folic acid is mandatory, but not for other alternative flours, such as the ones used in GFP. Taking into account the folate deficiency in GFD observed, its fortification in GFP or ingredients could be of interest for celiac children. Folate fortification measures could also be extended to biotin, whose widespread diet-deficiency in celiac population was alarming. In fact, some of the GF cereals analyzed were supplemented with this vitamin.

It is of interest to point out that some deficiency diseases found in celiac people, such as anaemia, low bone density or zinc depletion (36) are not only justified by nutritional shortages. Other pathological situations such as systemic inflammation or intestinal microbiota alteration appear to contribute to the persistence of those deficiencies in some celiac individuals (12, 37, 38).

It has to be highlighted that this paper presents wide-ranging high-quality nutritional information about GF bread, pasta and cereal micronutrient content. This remains limited in the literature and even more so in food panels or in databases used for GFD design and evaluation, where it is crucial. Moreover, it has assessed not only GFP composition but also its dietetic role, discounting, in general terms, its involvement in micronutrient deficiencies of the GFD of children and adolescents. Nevertheless, extrapolation to celiac adults is limited and needs further research. Finally, as proposed elsewhere (39), the bioavailability of GFP is a matter of concern that should also be taken into account in further studies.

The practical outcomes of the present study are relevant in improving the universal guidelines for food fortification in CD (40, 41). Some individualized supplementation is usually proposed for celiac people based on micronutrient related blood monitoring. Nevertheless, GFP fortification for folate and biotin could contribute to preventing the deficiencies observed in GFD, at least in the case of celiac children and youngsters.

Acknowledgments: Idoia Larretxi is a fellowship of the University of the Basque Country, UPV/EHU. This research was supported by a grant from the Basque Government (Proyectos de Investigación Focalizada Agricultura PA15/01) and a grant from the University of the Basque Country, UPV/EHU, (University-Society US15/06 and US18/15).

Conflict of Interest The authors declare that they have no conflict of interest.

References

1. do Nascimento AB, Fiates GM, Dos Anjos A, Teixeira E. Analysis of ingredient lists of commercially available gluten-free and gluten-containing food products using the text mining technique. Int J Food Sci Nutr. 2013;64(2):217-22.
2. Missbach B, Schwingshackl L, Billmann A, Mystek A, Hickelsberger M, Bauer G, et al. Gluten-free food database: the nutritional quality and cost of packaged gluten-free foods. PeerJ. 2015;3:e1337.

3. Wu JH, Neal B, Trevena H, Crino M, Stuart-Smith W, Faulkner-Hogg K, et al. Are gluten-free foods healthier than non-gluten-free foods? An evaluation of supermarket products in Australia. Br J Nutr. 2015;114(3):448-54.
4. Miranda J, Lasa A, Bustamante MA, Churruca I, Simon E. Nutritional differences between a gluten-free diet and a diet containing equivalent products with gluten. Plant Foods Hum Nutr. 2014;69(2):182-7.
5. Mazzeo T, Cauzzi S, Brighenti F, Pellegrini N. The development of a composition database of gluten-free products. Public Health Nutr. 2015;18(8):1353-7.
6. Staudacher HM, Gibson PR. How healthy is a gluten-free diet? Br J Nutr. 2015;114(10):1539-41.
7. Gliszczyńska-Świgło A, Klimczak I, Rybicka I. Chemometric analysis of minerals in gluten-free products. J Sci Food Agric. 2018;98(8):3041-8.
8. Rybicka I. The Handbook of Minerals on a Gluten-Free Diet. Nutrients. 2018;10(11).
9. Yazynina E, Johansson M, Jägerstad M, Jastrebova J. Low folate content in gluten-free cereal products and their main ingredients. Food Chemistry. 2008;111(1):236-42.
10. Babio N, Alcázar M, Castillejo G, Recasens M, Martínez-Cerezo F, Gutiérrez-Pensado V, et al. Patients With Coeliac Disease Reported Highter Consumption of Added Sugar and Total Fat Than Healthy Individuals. J Pediatr Gastroenterol Nutr. 2016.
11. Barone M, Della Valle N, Rosania R, Facciorusso A, Trotta A, Cantatore FP, et al. A comparison of the nutritional status between adult celiac patients on a long-term, strictly gluten-free diet and healthy subjects. Eur J Clin Nutr. 2016;70(1):23-7.
12. Gobbetti M, Pontonio E, Filannino P, Rizzello CG, De Angelis M, Di Cagno R. How to improve the gluten-free diet: The state of the art from a food science perspective. Food Res Int. 2018;110:22-32.
13. Salazar Quero JC, Espin Jaime B, Rodriguez Martinez A, Arguelles Martin F, Garcia Jimenez R, Rubio Murillo M, et al. [Nutritional assessment of gluten-free diet. Is gluten-free diet deficient in some nutrient?]. An Pediatr (Barc). 2015;83(1):33-9.
14. Botero-López JE, Araya M, Parada A, Méndez MA, Pizarro F, Espinosa N, et al. Micronutrient deficiencies in patients with typical and atypical celiac disease. J Pediatr Gastroenterol Nutr. 2011;53(3):265-70.
15. Pellegrini N, Agostoni C. Nutritional aspects of gluten-free products. J Sci Food Agric. 2015.

16. International A. Methods 968.06 & 992.15. Official Methods of Analysis of AOAC International, 18th
Edition, 2005 (Revision 3, 2010).2005. p. p129.
17. AESAN/BEDCA. Base de datos española de composición de alimentos v 1.0 [Available from: http://www.bedca.net.
18. Roe M, Pinchen H, Church S, Finglas P. McCance and Widdowson's The Composition of Foods Seventh Summary Edition and updated Composition of Foods Integrated Dataset. Nutrition Bulletin. 2015;40(1):36-9.
19. Australian Food Composition Database (Januari 2019). Available from:
http://www.foodstandards.gov.au/science/monitoringnutrients/afcd/Pages/default.aspx
20. United States Department of Agriculture. Agricultural Research Service. USDA Food Composition Databases. Available from: https://ndb.nal.usda.gov/ndb/search/list
21. Zuccotti G, Fabiano V, Dilillo D, Picca M, Cravidi C, Brambilla P. Intakes of nutrients in Italian children with celiac disease and the role of commercially available gluten-free products. J Hum Nutr Diet. 2013;26(5):436-44.
22. Larretxi I, Simon E, Benjumea L, Miranda J, Bustamante MA, Lasa A, et al. Gluten-free-rendered products contribute to imbalanced diets in children and adolescents with celiac disease. Eur J Nutr. 2019;58(2):775-83.
23. FESNAD. Dietary reference intakes (DRI) for spanish population, 2010. Actividad Dietética. 2010;14(4):196-7.
24. Matos ME, Rosell CM. Understanding gluten-free dough for reaching breads with physical quality and nutritional balance. J Sci Food Agric. 2015;95(4):653-61.
25. Regulation (EU) No 1169/2011 of the European Parliament and of the Council of 25 October 2011 on the provision of food information to consumers, amending Regulations (EC) No 1924/2006 and (EC) No 1925/2006 of the European Parliament and of the Council, and repealing Commission Directive 87/250/EEC, Council Directive 90/496/EEC, Commission Directive 1999/10/EC, Directive 2000/13/EC of the European Parliament and of the Council, Commission Directives 2002/67/EC and 2008/5/EC and Commission Regulation (EC) No 608/2004. Official Journal of the European Union, L304; (November 22, 2011).
26. Lee AR, Wolf RL, Lebwohl B, Ciaccio EJ, Green PHR. Persistent Economic Burden of the Gluten Free Diet. Nutrients. 2019;11(2).
27. Thompson T. Thiamin, riboflavin, and niacin contents of the gluten-free diet: is there cause for concern? J Am Diet Assoc. 1999;99(7):858-62.

28. Krupa-Kozak U, Wronkowska M, Soral-ŚMietaNa M. Effect of Buckwheat Flour on Microelements and Proteins Contents in Gluten-Free Bread2011. 103–8 p.
29. Rybicka I, Gliszczyńska-Świgło A. Minerals in grain gluten-free products. The content of calcium, potassium, magnesium, sodium, copper, iron, manganese, and zinc. Journal of Food Composition and Analysis. 2017;59:61-7.
30. Rybicka I, Gliszczynska-Swiglo A. Gluten-Free Flours from Different Raw Materials as the Source of Vitamin B. J Nutr Sci Vitaminol (Tokyo). 2017;63(2):125-32.
31. Mariani P, Viti MG, Montuori M, La Vecchia A, Cipolletta E, Calvani L, et al. The gluten-free diet: a nutritional risk factor for adolescents with celiac disease? J Pediatr Gastroenterol Nutr. 1998;27(5):519-23.
32. Ohlund K, Olsson C, Hernell O, Ohlund I. Dietary shortcomings in children on a gluten-free diet. J Hum Nutr Diet. 2010;23(3):294-300.
33. Babio N, Alcázar M, Castillejo G, Recasens M, Martínez-Cerezo F, Gutiérrez-Pensado V, et al. Patients With Celiac Disease Reported Higher Consumption of Added Sugar and Total Fat Than Healthy Individuals. J Pediatr Gastroenterol Nutr. 2017;64(1):63-9.
34. Government of Canada. 2018. In: Laws J, ed. Food and drug regulations. Ottawa, Ontario, Canada.
35. US Department of Health and Human Services. 2015. Questions and answers on FDA’s fortification policy – guidance for ndustry. In: Food and Drug Aministration. Available at https://www.fda.gov/media/94563/download (accesed on 2019 May 23)
36. Melini V, Melini F. Gluten-Free Diet: Gaps and Needs for a Healthier Diet. Nutrients. 2019;11(1).
37. Harper JW, Holleran SF, Ramakrishnan R, Bhagat G, Green PH. Anemia in celiac disease is multifactorial in etiology. Am J Hematol. 2007;82(11):996-1000.
38. Olechnowicz J, Tinkov A, Skalny A, Suliburska J. Zinc status is associated with inflammation, oxidative stress, lipid, and glucose metabolism. J Physiol Sci. 2018;68(1):19-31.
39. Suliburska J, Krejpcio Z, Reguła J, Grochowicz A. Evaluation of the content and the potential bioavailability of minerals from gluten-free products. Acta Sci Pol Technol Aliment. 2013;12(1):75-9.
40. Caruso R, Pallone F, Stasi E, Romeo S, Monteleone G. Appropriate nutrient supplementation in celiac disease. Ann Med. 2013;45(8):522-31.
41. Dennis M, Lee AR, McCarthy T. Nutritional Considerations of the Gluten-Free Diet. Gastroenterol Clin North Am. 2019;48(1):53-72.

Table 1. Analytical macronutrient content in gluten-free rendered foodstuffs divided by food groups, compared to gluten-containing products, expressed by 100 g of foodstuffs

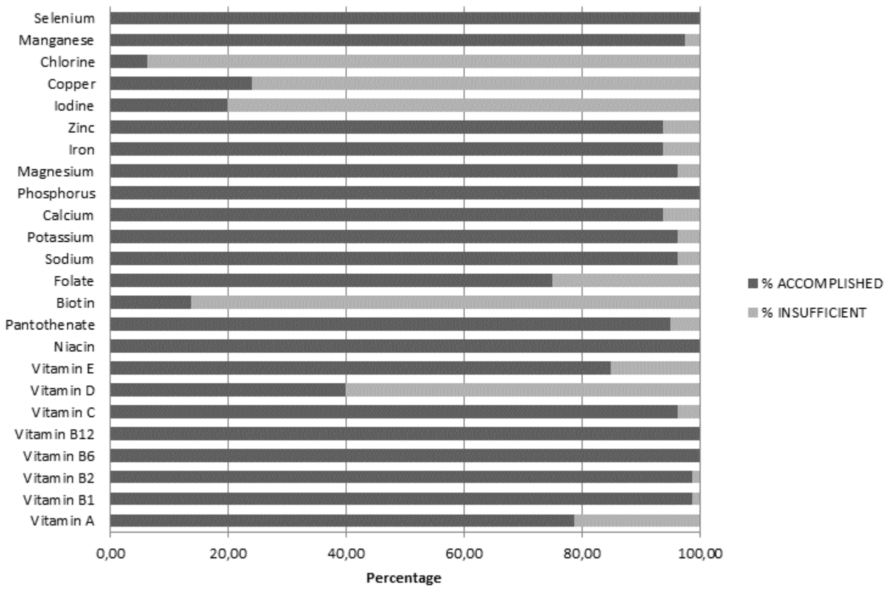
	Cereal flakes			**Bread**			**Pasta**		
	GFP	GCP	P	GFP	GCP	P	GFP	GCP	P
Carbohydrates	77.4 ± 7.0	79.4 ± 10.6	NS	56.8 ± 15.3	49.9 ± 5.7	NS	71.2 ± 16.3	50.3 ± 25.7	< 0.01
Lipids	3.9 ± 5.3	2.6 ± 2.0	NS	5.6 ± 4.2	3.5 ± 4.1	0.05	3.2 ± 4.5	2.2 ± 1.1	NS
Proteins	7.4 ± 0.7	7.8 ± 3.1	NS	2.4 ± 2.0	9.0 ± 1.5*	< 0.001	6.5 ± 1.4	9.8 ± 4.2*	NS

Notes: Values are means ± SD. SD, standard deviation; GFP, gluten-free product; GCP, gluten-containing product; *P*, statistical significance; NS, not significant.

Table 2. Analytical (115) micronutrient content in gluten-free rendered foodstuffs divided by food groups, compared to gluten-containing products, expressed by 100 g of foodstuffs.

	Cereal Flakes					Breads					Pasta				
	GCP		GFP		p	GCP		GFP		p	GCP		GFP		p
	mean	SD	mean	SD		mean	SD	mean	SD		mean	SD	mean	SD	
Calcium (mg)	141	183	22.3	16.8	NS	60.5	34.4	90.8	57.5	NS	22.9	10.0	27.3	27.3	NS
Iron (mg)	9.87	5.19	1.8	1.8	<0.001	6.75	19.99	1.1	0.9	0.009	1.83	0.88	0.7	0.5	0.002
Sodium (mg)	332	332	357.1	313.6	NS	423	280	570.8	248.1	NS	61.0	155	34.8	65.6	NS
Zinc (mg)	5.94	12.9	0.9	0.5	NS	0.84	0.48	0.5	0.4	NS	1.45	1.10	1.1	0.5	NS
Copper (mg)	0.2	0.2	0.3	0.3	NS	0.14	0.11	0.1	0.1	NS	0.31	0.20	0.1	0.1	NS
Manganese (mg)			0.4	0.4		0.58	0.51	0.2	0.3	NS	2.22	1.18	0.5	0.8	0.042
Biotin (ug)	2.3	2.9	40.7	95.7	0.001	12.7	13.6	9.74	21.6	NS	15.8	14.8	1.10	1.00	0.002
Folate (ug)	275	35.4	55.4	88.9	0.062	32.55	14.79	32.8	56.8	NS	19.4	10.3	3.14	3.42	0.003
Niacin (mg)	16.2	8.25	3.02	2.41	<0.001	3.49	2.15	1.02	1.46	0.004	3.75	3.46	3.62	10.58	NS
Piridoxin (mg)	1.80	0.90	1.19	4.15	0.001	0.12	0.10	0.02	0.02	<0.001	0.12	0.07	0.01	0.01	<0.001
Riboflavin (mg)	1.45	0.72	0.12	0.17	<0.001	0.13	0.11	0.04	0.09	0.001	0.07	0.04	0.01	0.02	0.002
Tiamin (mg)	1.32	0.70	0.19	0.28	<0.001	0.20	0.12	0.01	0.01	<0.001	0.19	0.15	0.03	0.04	0.001
B5 (mg)	7.55	3.46	1.03	1.81	0.045	0.39	0.07	0.42	0.57	NS	0.70	0.40	0.29	0.36	0.067
B12 (ug)	0.85	0.52	3.68	4.18	0.03	0.02	0.05	88.2	236	0.026	0.04	0.08	0.73	1.37	NS
Vitamin E (mg)	3.00	6.22	0.14	0.47	0.01	0.30	0.33	1,04	1,2	NS	0.09	0.12	0.2	0.1	NS

Notes: Values are means ± SD. SD, standard deviation; GFP, gluten-free product; GCP, gluten-containing product; *P*, statistical significance; NS, not significant



Supplementary Figure 1. Percentage of celiac children and adolescents who accomplished or did not achieve 2/3 of the dietary reference intake of the vitamins and minerals (proposed by the Federation of Spanish Societies of Nutrition and Dietetics, FESNAD).

LABURBILKETA

SUMMARY

LABURBILKETA

Doktoretza Tesi honetan burututako lana bi zati garrantzitsuetan bana daiteke. Lehen hiru eskuizkribuetan, Euskal Autonomia Erkidegoko zeliako kohorte baten nutrizio-egoeraren eta ohitura dietetikoen inguruko datu berriak nabarmentzen dira. GGD-k oreka lortzeko dituen ohiko akatsak eta berau jarraitzeko oztopoak definitzen dira, adin-tartearen eta generoaren arabera (A helburua). Ondoren, hurrengo bi eskuizkribuetan (4. eta 5. eskuizkribuak), GGPen nutrizio-osaera aztertu egiten da, glutendun elikagaiekiko konparatuta. Halaber, horrek GGD osoan izan dezakeen eragina ondorioztatu da (B helburua).

A. EAEko populazio zeliakoaren nutrizio-egoera eta elikadura ohiturak

1. Neurri antropometrikoak

Zeliakoen neurri antropometrikoak aztertutakoan behatu denez, gehiengoak normopisua du. Parte-hartzaileak taldeka banatzean, emakumeek GMI osasungarriagoa dutela ikusi da gizonekin edo/eta umeekin aldaratuz. Izan ere, gizonen artean gainpisu eta obesitate kasu ugari aurkitu dira, erdiak baino gehiagok gantz-masa portzentaje altua izanik, beste bi taldeetan ez bezala. Kontrara, umeen %20ak pisu baxua du. Emaitzok populazio orokorrean dauden datuekin erkatzean, ondorioztatu daiteke zeliako taldeak gainpisu eta obesitate prebalentzia gutxiago duela; gizonen kohorteko datuak desegokiagoak izan arren, prebalentziak populazio orokorrean dagoena baino baxuagoa izaten jarraitzen du. Bardella *et al.*-ek antzeko emaitzak argitaratu zuten egindako beste ikerketa batean (106); hots, zeliakoek kontrol taldeak baino pisu, gantz-masa, ihar-masa eta hezur-masa baxuagoak zituztela deskribatu zuten.

Ikerketa batzuk agerian jarri duten legez, zeliakoen tratamendu aurreko malnutrizioa ez da batzuetan hobetzen GGD jarraitutakoan (GMIa, gantz-masa eta hezurren mineralizazio baxuak mantenduz) (165-167). Smecuol *et al.*-ek (168) bestelako emaitzak lortu zuten burututako ikerketan non diagnostikatu berri ziren 25 pertsona zeliako bildu ziren. Talde horrek gantz-masa, ihar-masa eta hezur-masa portzentaje txikiegiak zituen hasieran baina 37 asteko GGD tratamendua bete ondoren, neurri antropometrikoak esanguratsuki hobetu zitzaizkien. Ildo beretik jarraituz, Ukkola eta kolaboratzaileek (169) azaldu zuten diagnostiko momentuan pisu gutxi zuten pazienteek urte batez GGD jarraitu ostean pisua berreskuratzen zutela. Gainpisua edo obesitatea zutenek, berriz, pisua galdu zuten, GGD egokia edota orekatuagoa jarraitzeari esker. Bere aldetik, Barone *et al*.-ek (165) 39 pertsona zeliakoren datu antropometrikoak aldaratu zituzten GGD jarraitu aurretik eta ostean. Diagnostiko unean zeliako gehienek (%82k) GMI normala edo gainpisua zuen eta bakarrik talde osoaren %10ak zeukan desnutrizioa. GGDrekin hasi ostean, GMI normala zuten pertsonetan pisu-irabazte esanguratsua ikusi zen, gainpisura heldu gabe, eta desnutrizioa zeukaten artean (n=4) erdiak GMI normala izatea lortu zuten. Beste erdiaren (n=2) hobekuntza eza, bestelako ikerketetan ere agertu den legez, dietaren atxikimendu eskasarekin edota farmako batzuen kontsumoarekin justifikatu liteke, besteak beste (167, 170).

Sexuen arteko ezberdintasunari dagokionez, emaitza ohikoa dela aipatu behar da, populazio orokorrean ere aurkitzen baita (55, 171-173). Horren azalpena honakoa izan daiteke: eskuarki gizonek osasunarekiko duten arreta eta interes falta, emakumeena baino txikiagoa behintzat, ikerketa batzuetan deskribatu izan denez (174, 175).

Oro har, zeliakoek GGD zorrotza jarraitzen dutenean beren neurri antropometrikoak hobetzen dituzte. Izan ere, gehienetan GGD jarraitzeak sintomen arintzea dakar, zeliakoaren nutrizio-egoera aurreratzea eragiten duena. Are gehiago, kolektibo honek elikadurarekiko sentikortasun handia du eta, beraz, baliteke ohitura dietetiko osasuntsuak konplitzeaz gehiago arduratzea. Hortaz, pisu, gantz-masa eta GMI balio egokiagoak dituzte GGDrekin hasi aurretik zituztenekin aldaratuz. Izan ere, adostasun nahikoa dago ondorengoarekin: zenbat eta lehenago hasi GGD, hainbat eta azkarrago normalizatuko dira neurri antropometrikoak (91, 176) Aitzitik, beraien nutrizio-egoera hobetzen bada ere, batzuetan ez da pertsona osasuntsuen mailara ailegatzen (166). Horren azalpena izan daiteke zeliakoek pairatzen duten egoera patologikoak nutrienteen biodisponibilitate baxuagoa eragiten duela edo/eta Kautto *et al*-ek beren ikerketan iradoki zuten moduan nahigabeko dieta transgresioak egiten dituztela (111).

2. Makronutriente kontsumoa

Zeliakiaren tratamendua erronka dietetiko garrantzitsua da diagnostikatu berri den pertsonarentzat. Izan ere, Mediterranear Dieta (MD) orekatuan oinarrizkoak diren elikagai batzuk baztertu behar dira, hala nola, garizko ogi, pasta, zereale eta irinak edo horiek osagai duten gainerako elikagaiak. Kontuan hartu behar da, halaber, zereal taldeko elikagai horiek hainbat makronutriente eta mikronutrienteren iturri nagusi direla eta, hortaz, kolektibo horren nutrizio-egoera baldintzatua egon daitekeela balizko dieta desorekatuaren eta mikronutrienteen gabezien ondorioz (96-98, 113). Horrela, beste lan askorekin ados (93, 100, 108-110), ikerketa honen parte-hartzaileen dieta ebaluatzeak agerian jarri du zeliakoen dieta, orokorrean, desorekatua dela. Erreferentziazko Ingesta Dietetikoekin konparatzen denean (154), honakoa behatzen da: energia, karbohidrato konplexu eta zuntz kopuru gutxi hartzen dutela eta, aldiz, proteina eta gantz, konkretuki gantz ase, gehiegi. Makronutrienteen desoreka nahiko aurresangarria da zeliakoen dietan, zerealen kontsumo urria eta karbohidrato konplexuen beste iturrien ahorakin txikia dela eta. Halaber, populazio orokorrak egiten duen bezala, haragiaren eta animalia jatorriko elikagaien gehiegizko kontsumoa antzeman da .

Ikerketetan parte hartu duten gizonak eta emakumeak aldaratutakoan, gizon portzentaje altuago batek behar duen baino energia gehiago hartzen duela ikusi da, horrek gizon zeliakoetan antzemandako gainpisu eta obesitate maila handiagoa justifika dezake. Makronutrienteen banaketa desoreka, berriz, antzekoa da bi sexu taldeetan, baita gantz dietetikoen profilak ere. Dena den, talde biek gantz ase eta kolesterol gehiegi kontsumitu arren, gizonek emakumeek baino ezegokiago egiten dute.

Zeliakoen eta populazio orokorraren elikadura ohiturak konparatutakoan antzeko emaitzak aurkitzen dira, makronutrienteen desoreka alegia.(156). Beste autore batzuk ere baieztatu dituzte antzeko banaketa energetikoak GGD eta kontrolen dieta erkatzerakoan (101, 107). Barone *et al*.-ek (165) zeliakoen GGDren nutrizio-osaera aztertu zuten talde kontrol batekiko aldaratuz, eta energia, karbohidrato eta proteina kopuru parekoak hartzen zituztela behatu zuten. Hala ere, zeliakoek gantz ingesta altuagoa eta zuntz kontsumo baxuagoa zuten. Aldiz, Bardella *et al*.-ek zeliakoek kontrolek baino energia eta karbohidrato ingesta baxuagoa zutela ikusi zuten, gantz ehunekoa handiagoa izanik (106). Gure ikerketan, emakumeen energia ahorakina kontrolena baino txikiagoa izan zen, baita hiru makronutrienteen kontsumoa ere. Gizonen kasuan ez zen ezberdintasunik behatu. Karbohidratoei dagokienez, bai pertsona zeliakoak bai kontrolak gomendioaren azpitik kokatu ziren. Guztiek gutxieneko Erreferentziazko Ingesta Dietetikoa (130 g/egun) betetzen zuten arren, kopurua ebaluatuz, zeliakoek gramo gutxiago irensten zituzten kontrolek baino. Zeliakoen kasuan, energia osoaren %42 bakarrik eratortzen da karbohidratoetatik, populazio orokorrean datua antzekoa izanik.

Zeliakoetan zuntz kontsumoa kontroletan baino urriagoa izan zen; are gehiago, emakume zeliakoek, gazteenek zehazki, ingesta txikiagoa egiten zuten gizon zeliakoek baino. Hau da, nahiz eta bi sexu taldeetan zuntz ahorakina txikia izan, gizon portzentaje gehiagok bete zuen zuntzaren gomendioa (gizonen %26k betetzen du vs emakumeen %4k).

Beste aurretiko ikerketa batzuek agerian jarri zuten GGD zuntz gutxikoa dela, glutendun dieta baino urriagoa. Wild *et al*.-ek (177) erakutsi zuten emakume zeliakoen zuntz ingesta esanguratsuki txikiagoa zela Erresuma Batuko populazioaren inkesta dietetikoen datuekin konparatzean. Martin *et al*.-ek emaitza berdinak lortu zituzten haien ikerketako gizonen kohortea Alemaniako populazio orokorrarekin aldaratzean (107). Izan ere, zuntz kontsumo murritza GGPen kontsumoarekin erlazionatu daiteke, maiz produktu horien formulazioa zuntzan eskasa baita ekoizpen prozesuan glutenarekin kutsatzeko arriskua ekiditeko (178).

Halere, azken urteotan GGPen gaineko interesa handitu da eta, horrekin batera, beren kalitatea hobetzeko kezka ere. Horrela, elikagai-industriak beraien ezaugarri organoleptikoak eta nutrizio-balioa hobetzeko asmoz osagai berri batzuk erabiltzen hasi da GGPen ekoizpenean (179). Osagaiok iraganean ikusitako konposizio desegokia (115) zuzentzen saiatzeko sartu dira.

Lehengai berri horien artean AIa dago, zuntz iturri dena. AIaren jokaera fisiologikoa zuntza solugarriaren antzekoa da, bere efektu onuragarriak eraginez (180, 181). Gure taldeak azterketa analitikoen bitartez lortutako datuek (**4. eskuizkribua**) diote gluten gabeko ogiek glutendun ogiek baino zuntza eta AI kopuru handiagoa dutela, GGPen hobekuntza nutrizionalaren tesia berresten duena. Emaitza horiek bat datoz beste ikerketa berri batzuetan azaldutako datuekin (146, 182-184). Garrantzitsua da aipatzea ikerketa guztiak (gurea barne) GGDren balorazioa egiterakoan etiketen zuntza balioan oinarritzen direla. Aitzitik, 4. eskuizkribuan ikusi den bezala, batzuetan GGPen etiketetan zuntz edukia gutxietsita dago edo ez da agertzen (legediak ez baitu behartzen informazio hori ematera).

Dietaren gantz iturriak eta profila aztertzean, zeliakoek eta kontrolek gantz ase eta monoasegabe kontsumo berdintsua egin arren, gantz poliasegabeen kontsumoa urriagoa da gizon zeliakoen kasuan. Orokorrean, gantz ase eta asegabeen arteko ratioek gomendutakoa betetzen dute baina kolesterolaren kontsumoa gehiegizkoa da bi taldeetan.

Proteinen kasuan, zeliakoak gomendioaren gainetik kokatzen dira eta kontsumoa energia guztiaren %17 da kohorte guztietan. Populazio orokorrean emaitza berdintsuak ikusi daitezke. Horren zioa animalia jatorriko elikagaiak gehiegi hartzea da.

Umeen kohorteari dagokionez, helduaroko antzeko desoreka dietetikoa deskribatu da. Energia gomendioaren %80 besterik ez dute betetzen. Karbohidratoen gomendioa ez da betetzen (energiaren % 45 izanik) eta beharrezkoa baino gantz eta proteina gehiago hartzen dituzte bere dietan, energiaren %40 eta %17 hurrenez hurren. Ildo berean, ikerketa ugarik antzeko ingesta patroia deskribatu dute umeetan (68, 105); alegia, energia kontsumo murriztua (108), karbohidrato konplexu eta zuntza kontsumo txikia (93, 100, 109) eta gantza (93, 100, 108, 110) eta proteina ahorakin altua (108). Gantz kontsumoa ere desorekatua da, gantz ase gehiegi eta asegabe gutxiegi hartuz (100, 111).

Aipagarriak dira Zuccoti eta bere taldeak (101) argitaratutako emaitzek, non ume eta nerabe zeliakoek jarraitzen duten GGD biztanleri orokorraren dietarekin aldaratzen den. Nahiz eta bi taldeek Italian gomendatzen den proteina, gantza eta azukre kontsumoa gainditu, ume zeliakoek energia eta karbohidrato kontsumo are altuagoa zuten, kontrolekin konparatuta, proteinatik eratorritako energia kopurua berdina izanik. Espainian ume zeliakoekin egindako ikerketa batean, diagnostikatu aurretik zeramaten glutendun dieta eta diagnostikoa eta urte batera jarraitzen zuten GGD aldaratu zituzten (185). Lan horrek GGDn gantz monoasegabeen kontsumoa altuagoa zela aurkitu zuen eta gantz aseena, berriz, txikiagoa (185). Kontrara, Babio *et al*.-ek (102) egindako ikerketan adierazi zuten 10-23 urte bitarteko zeliakoek azukre erantsi, gantz total eta eduki proteiko altuko elikagai asko kontsumitzen zituztela, kontrolek baino gehiago. Berriz, almidoietan aberatsak ziren elikagaien kontsumoa baxuagoa zen.

Zuntzari dagokionez, gure ikerketaren ume zeliakoen artean kontsumoa urria da (15g/egun) eta gomendutakoaren azpitik dago, haur-biztanleri orokorrean pareko egoera izanez (93, 100, 102). Egia esan, lan batzuk ez dituzte desberdintasunik antzeman ume zeliakoen eta ez zeliakoen dietak aztertutakoan (100, 102).Ostera, Mariani *et al*.-ek karbohidrato kontsumo txikiagoa eta konkretuki zuntza ingesta askoz txikiagoa topatu zuten zeliako nerabeetan, nerabe kontrolekin erkatzean (93). Hori azaldu daiteke GGD jarraitzeak berekin dakartzalako zuntzan aberatsak diren zerealak ezin kontsumitzea eta zereal finduen presentzia handia (68)

7.taula. Laburpen taula: Espainiako FESNAD Elkartearen energia eta makronutriente gomendioen betetze maila.

		Emakumeak	**Umeak**	**Gizonak**
Energia ingesta	**Gutxiegi**	%30	x/↑	%40
	Egokia	%65		%46
	Gehiegi	%5		%14
Makronutrienteen banaketa:				
Proteinak		x/↓	x/↓	x/↓
Karbohidratoak		x/↑	x/↑	x/↑
Lipidoak		x/↓	x/↓	x/↓
Gantz aseak		x/↓	x/↓	x/↓↓
Zuntzaren kontsumoa	**Egokia**	%4	x/↑↑	%26
	Gutxiegi	%96 (%43 < 15g)		%74 (%28 < 15g)

Notak: x, balioek ez dituzte FESNADeko gomendiok betetzen, ↓ kontsumoa murritzea gomendatzen da; ↑ kontsumoa handitzea gomendatzen da; FESNAD, *Espainiako Nutrizio, Elikadura eta Dietetikako Elkarteen Federazioa.*

3. Mikronutriente kontsumoa

Lehen esan bezala, GGDen ebaluaketa egiteko programa informatikoek GGPen nutrizio-konposizioa espezifikoa izan beharko lukete haien datu basean. Are gehiago, GGPen etiketa gehienek ez dituzte bitamina eta mineral edukiak adierazten. Beraz, ikerketa honetan mikronutriente kontsumoa aztertzeko glutendun homologoetan oinarrituz hurbilketa egin da. Bitaminen eta mineralen ingesta-gomendioa betetzen ote den aztertzeko, erreferentziazko gomendioaren 2/3 (>%67) ezarri da betetze maila egoki bezala (186, 187). Ikerketak agerian jarri du adin talde guztietako parte-hartzaile gehienek D bitamina gutxiegi hartzen dutela eta, gainera, helduengan E bitamina eta iodoa gabezia pairatu ohi dela. Halaber, emakumeen kasuan selenioa eta gizonezkoetan A bitamina eta magnesioa ere urriak dira dietan. Emakumeek eta umeek burdin gabeziak dituzte eta gizonek eta umeek folatoenak. Azkenik, umeek ez dute kaltzio nahikorik kontsumitzen. Ikerketa batzuetan diagnostikoa egiteko momentuan zianokobalaminaren gabezia azpimarratu bada ere (188-190), gure ikerketan inork ez du erakutsi B_{12} bitaminaren ahorakin urria. Egia esan, aipatutako animalia jatorriko elikagaien gehiegizko kontsumoak bitamina honen ekarpena ziurtatzen du, nahiz eta xurgapen portzentajea nolakoa izan den ez ezagutu.

Ikerketa askok azpimarratu dute GGDk mikronutriente-gabeziak aurkez ditzakeela (9, 97, 105, 118). Hori izan daiteke, alde batetik, zerealen hartualdia txikitzen delako eta, bestetik, gari-irinez ekoiztutako produktuak maiz aberastuta daudelako eta GGPk, berriz, ez. Ikusi da GGPek ez dituztela haien homologoek dituzten mikronutriente kopuru berdinak, adibidez, tiamina, erriboflabina, niazina, folato, D bitamina, kaltzio edota burdin gutxiago dutela deskribatu izan da (97, 148). Egoera hori larritzen da kontuan hartzen badugu diagnostikoa eta GGD ezarri baino lehenago nutrienteen xurgapen txarra pairatu egiten dela GZan eta horrek mikronutrienteen gabeziak ekarri ohi duela askotan (79),(191) Gure ikerketako gizonen eta emakumeen GGD erkatzerakoan hainbat ezberdintasun daude mineralei dagokienez, ez ordea bitaminen kontsumoan. Emakumeek hobeto betetzen dute magnesio kontsumoaren gomendioa gizonek baino eta, ostera, burdin, iodo, potasio eta selenio betetze maila okerragoak dituzte.

Datu horiek kontrol taldeekin konparatzen direnean, zeliakoen eta populazio orokorraren gabeziek antzeko patroia jarraitzen dutela antzematen da. Halere, alde esanguratsu batzuk aurki daitezke: E bitamina, niazina, magnesioa eta selenioaren kontsumoa txikiagoa da emakume zeliakoetan eta, aldiz, erriboflabina, B_6 bitamina eta folatoarena nabarmen altuagoak dira. Gizonek ere E bitamina, niazina eta magnesio gutxiago hartzen dute kontrolek baino eta erriboflabina, B_6 bitamina, zink, potasio eta selenio gehiago. Martin *et al*.-ek adierazi zuten B_1, B_2, B_6 bitaminak, azido folikoa, magnesioa eta burdinaren kontsumoa baxuagoa zela zeliako helduen artean biztanleria orokorrean baino (107). Aldiz, Barone *et al*.-ek (165) ikusi zuten kaltzio, D bitamina eta fosforoaren ingesta parekoa zela zeliakoetan eta kontrol taldeetan, biak Italiar ingesta gomendioen barnean kokaturik.

8. taula. Laburpen taula: Espainiako FESNAD Elkartearen mikronutriente-gomendioen bi heren betetzen ez duten pertsonak.

	Emakumeak	**Umeak**	**Gizonak**
Bitaminak			
D	48 %	% 69	% 55
E	% 39	% 57	% 52
A	% 11	% 37	% 19
Folatoa	% 18	% 54	% 24
Mineralak			
Iodoa	% 80	% 93	% 50
Burdina	% 31	% 23	% 0
Selenioa	%31	% 20	% 7
Kaltzioa	% 13	% 51	% 14

9. taula. GGD jarraitzen duten zeliakoen nutrizio egoera eta gehiegizko edo/eta gutxiegizko nutrienteen kontsumoa

Erreferentzia	**Lurraldea**	**Kohortea**	**Nutrizio egoera**	**Ohitura dietetikoak**	
				Gehiegizko kontsumoa	**Gutxiegizko kontsumoa**
Larretxi *et al.* (2018) (192) **1.eskuizkribua**	EAE	Umeak (n=83)	Pisua, GMI eta GM egokiak	Gantza eta proteina	Energia, KH, D bitamina, azido folikoa, Ca eta Fe
Churruca *et al.* (2015) (117) **2.eskuizkribua**	EAE	Helduak (emakumeak) (n=54)	Pisua, GMI eta GM egokiak	Gantza eta proteina	Energia, KH, zuntza (gizon zeliakoek baino gutxiago), E eta D bitamina, folatoa, Ca, Fe,I eta Se
Gonzalez *et al.*(2018) (193) **3.eskuizkribua**	EAE	Helduak (gizonak) (n=42)	GMI eta GM balio altuak	Gantza, proteina eta kolesterola Emakume zeliakoekin erkatuta energia eta gantz ase gehiago hartzen dituzte	KH, zuntza eta gantz poliasegabeak.
Bardella *et al.* (2000) (106)	Italia	Helduak (n=71)	Pisua, GMI, GM, IM eta HM (emakumeetan) kontrolak baino baxuagoak.	Gantza	Energia eta KH
Barone *et al.* (2016) (165)	Italia	Helduak (n=39)	GMI hobetu diagnostikotik. Kontrolekin erkatzerakoan GMI, GM eta HM antzekoak	Gantza	Zuntza

Smecuol *et al.* (1997) (168)	Argentina	Helduak (n=25)	Pisua eta GM handitu eta IM mantendu (ez daude ezberdintasunik GGD zorrotza eta lapsusak egiten dutenen artean)	Ingesta energetiko ↓ GGD zorrotza egiten dutenek lapsusak dituztenek baino	
Ukkola *et al.* (2012) (169)	Finlandia	Helduak (n=698)	GMI hobetu zuten. Pisu baxukoek igo zuten eta gainpisua zutenek jaitsi.	EA	
Wiech *et al.* (2018) (176)	Polonia	Umeak (n=41)	IM eta gorputz-ur balio altuagoak eta GM baxuagoak kontrolak baino GGD zorrotza jarraitzen zuten umeek GM eta IM egokiagoak zituzten lapsusak egiten zutenek baino (ez esanguratsua). Urte bat GGD zorrotzarekin egon ostean neurri antopometrikoak hobetu.	26 ume GGD zorrotza jarraitzen zuten eta 15ek lapsusak zituzten.	
Kautto *et al.* (2014) (111)	Suedia	Nerabeak (n=37)		Gantza, PUFA eta gantza azido α-linolenikoa. Tiamina, erriboflabina, niazina, B6, folato, B12, P, Zn, Se eta Mg.	Proteina eta zuntza
Ohlund *et al.* (2010) (100)	Suedia	Umeak (n=25)	EA	Gantza aseak eta KH sinpleak	Energia (umeen %52an) zuntza, D vitamina, Mg eta Selenio.
Shepherd *et al.* (2013) (96)	Australia	Helduak (n=105 /50 diagostikatu berri eta 55 GGD ≥2urte)	GMI berdintsuak diagnostikatu berriak eta denbora luzez tratatutako zeliakoen artean.		Zuntza, tiamina, foltatoa, A bitamina, Mg, Ca, Fe, eta Zn.
Wild *et al.* (2010) (177)	UK	Helduak (n=139)	EA	KH sinpleak	Mg, Fe, Zn, Mn, Se eta folatoa
Zuccotti *et al.* (2013) (101)	Italia	Umeak (n=18)	Ez dago ezberditasunik	Energia, gantza (baina kontroletan baino baxuagoa) eta KH	
Babio *et al.* (2016) (102)	Espainia	Umeak eta helduak (n=98)	EA	KH sinpleak eta gantzak	Zuntza, azido folikoa, Ca, Fe eta Mg Almidoia duten elikagaiak

Salazar *et al.* (2014) (185)	Espainia	Umeak (n=37)	Neurri antropometriko egokiak	MUFA	Gantz aseak, D vitamina, Zn, Zuntza
Martin *et al.* (2013) (107)	Alemania	Umeak eta helduak (n=80)	EA	Gantza	Zuntza, KH, B1, B2, B6, azido folikoa, Mg eta Fe

Notak: GMI: gorputz masa indizea, GM: gantz masa, IM: ihar masa, HM: hezur masa, KH: karbohidratoak, GGD: gluten gabeko dieta, PUFA: gantza azido poliasegabeak, MUFA: gantza azido monoasegabeak, EA: ez aurkeztua

4. Elikagai taldeak

Elikadura-patroia faktore nagusietakoa da makronutrienteen desorekak edo/eta mikronutrienteen gabeziak azaltzeko. Hori dela eta, zeliakoen ohitura dietetikoak aztertu dira elikagai-kontsumo maiztasunari buruzko galdetegien bitartez. Oro har, zerealen kontsumoa oso eskasa da ikertutako talde guztietan, umeen kohortea izanik hiruretatik kontsumo altuagoa duena. Izan ere, umeek okindegi produktu gehien kontsumitzen dituzte bere eguneroko elikaduran (ogitartekoak, gailetak, gosaltzeko zerealak, e.a). Gure ikerketako emakumeen dietak aztertu ostean agerian jarri zen ia erdiak (%48ak) zereal taldeko ano gutxi bi errazio baino gutxiago- hartu zuten. Hala ere, eta energia osoaren kontsumoa urria zenez gero, energia totalaren %23 zerealetatik datorrela beha daiteke, eta ENIDEko populazio orokorrean, berriz, energiaren %17 (156). Datu horiek iradokitzen dute ikerlanean parte hartu duten emakume zeliakoek ziurrenik nutrizioaren inguruan kezka eta interes handiagoa izango dutela, kontrolek baino eta, ondorioz, ohitura dietetiko arinki osasuntsuagoak. Era berean, zeliakoen gehiengoan barazkien kontsumoa urria bada ere umeak gomendioa okerren betetzen duen taldea da. Fruta ahorakin egokia lortzen dute parte-hartzaileen erdiak, gutxi gora behera. Haragia eta eratorrien kontsumoa, ostera, gehiegizkoa da kasu gehienetan, kezkagarriagoa izanik gizonen artean. Gehiengoak betetzen du esnekien eta arrainen kontsumoa eta, horrekin erlazionaturik, animalia-proteinaren kontsumoa altua da. Adibidez, emakumeen kohortean proteina totalaren %70 inguru animalia jatorrikoa da, %20 esnekietatik eta %50 haragi, arrain eta arrautzatik jasoa hain zuzen ere.

Lekaleei dagokienez, gomendioaren betetze maila oro har egokia da, eta azpimarratzekoa da umeen kohortean ia guztiek asetzen dutela.

Orokorrean, zeliako helduetan emakume eta gizonen elikadura patroia antzekoa da, ezberdintasun bakarra arraina eta arrautzen ahorakinean behatzen da, gizonetan gehiegizko kontsumo portzentaje altuagoak agertuz.

Aztertutako emaitza guzti horiek agerian jarri dute kolektibo zeliakoentzat nutrizio-jarraibide espezifikoak ezartzearen interesa. Esate baterako, karbohidrato konplexuen kontsumoa handitzeko, glutenik gabeko zereal eta pseudozerealei buruz informazioa eman behar zaie, ohiko artoa eta arrozaz aparte (artatxikia, lekaleak, amarantoa, kinoa, artobeltza, teff, edota patatak, besteak beste). Gainera, horiek eguneroko dietan nola sartu azaldu behar zaie. Bestalde, zuntzaren ingestioa areagotzeko, hainbat aholku behar dituzte fruta (ahal bada, azalarekin), barazkiak, ortuariak eta lekaleak gehiago kontsumitzeko. Ez da GGPek dietan izan dezaketen papera ahaztu behar, produktuon etiketak arretaz irakurtzeak duen garrantzia barneratu behar dute, osasungarriagoak diren aukerak erosteko. Animalia jatorriko proteinen ekarpena mugatzeko haragien kontsumoa neurtzeko gomendatuko zaie eta, hartuz gero, aukera koipegabeak lehenetsi ditzaten eskatu. Horrela, murriztu egiten da proteinen eta gantzaren gehiegizko ingestioarekin erlazionatutako gaixotasun kardiobaskularrak (hiperkolesterolemia, hipertentsioa…) izateko arriskua. Aholku hori populazio guztiarentzat interesgarria bada ere, kolektibo zeliakoan are beharrezkoagoa da, gaitz horiek pairatzeko joera handiagoa baita, hainbat ikerketak proposatzen duten moduan (194).

Ume eta nerabe zeliakoen ohitura dietetikoei dagokienez, Mediterranear piramidearen jarraibideekiko atxikidura urria behatu zen.

Helduetan bezala, zereal eta eratorrien taldeko elikagaien kontsumoa eskasa da. Izan ere, %33k bakarrik betetzen dute SENCeko gomendioa (gutxieneko 4 ano/egun zerealen kontsumoa) eta are gehiago, %20k bi ano/egun baino gutxiagoko ahorakina egiten dute. Babiok eta bere taldeak ikusi zuten zeliakoek kontsumitzen zutela almidoi gutxiago (pasta, ogia, opilak) eta proteinadun elikagai gehiago (haragia, arraina, arrautza) biztanleri orokorrak baino (102). Gainera, gure ikerketako ume zeliakoek ez dute lortzen zuntzan aberatsak diren elikagaien kontsumoa betetzea, esate baterako, lekaleak, barazkiak edo/eta fruta. Hortaz, helduetan bezala, proteinen eta gantzaren kontsumoa jaistea eta karbohidratoena eta zuntzarena igotzea gomendatu beharko litzaieke (117).

Ohitura dietetiko osasuntsuekin batera beharrezkoa da ere bizi-ohitura egokiak jarraitzea. Ume eta nerabeen kohortean antzeman da bakarrik gutxi batzuek (%19) betetzen dutela OMEko jarduera fisikoaren gomendioa. Halere, ia inork ez ditu usadio kaltegarriak jarraitzen, hala nola; eskolara gosaldu gabe joatea, astean behin baino gehiagotan opilak kontsumitzea edo/eta sarritan janari azkarreko jatetxeetan jatea.

Ikerketa honen sendotasuna da GGPen nutrizio-osaeran oinarritu izana zeliakoen dieta aztertzeko. Alabaina, produktuon etiketetako datuak erabili izana lanaren muga nagusia da. Izan ere, mikronutriente edukia etiketan adierazten ez denez (195, 196), helduen ataletan glutendun produktu homologoen datuekin estimazio bat egin behar izan da, eta hortaz baliteke mikronutriente kopuruak gainestimatu izana (197). Arazo hori konpontzeko asmoz, umeen kohortean Missbach *et al*.-ek (153) sortutako GGPen mikronutriente edukien taula erabili da azterketa dietetiko zehatzago bat egiteko. Dena den, horiek Alemaniako GGPk dira eta, beraz, desberdintasunak ere aurki daitezke. Horregatik, azkenik EAEn kontsumitzen diren ohiko GGP osasungarriak aukeratu eta horietan bitamina eta mineral ohikoenak analizatu dira, mikronutriente-osaera taula propioa sortzeko.

Bestalde, lan honetan esku hartu duten pertsonak ez dira zoriz aukeratu, parte hartzea bolondresa izan baita. Horrek eragina izan dezake neurtutako ohitura dietetikoetan, halako ikerketetan inplikatzen diren pertsonek elikadurarekiko interes handiagoa izan ohi dute eta. Amaitzeko, diseinu esperimentalean kontrol talde propioa eduki ez izana aipatu behar da, konparatzeko kontrol talde egokiak aurkitu izan baziren ere.

4.1 GGPek GGDren desorekan duten erantzunkizuna

Nahiz eta elikadura patroiak zeliakoen dietan agertzen diren desorekak eta gabeziak hein batean azal ditzakeen, badira ere paper garrantzitsua jokatzen duten beste faktore batzuk. Ikerketa askok jakinarazi dute GGPen nutrizio-osaera ez dela haien glutendun homologoena bezalakoa: batzuetan gantz kopuru handiagoa dute baina karbohidrato, proteina, zuntz eta bitamina eta mineral gutxiago (115, 147, 153, 198) (10. taula). Hortaz, ezin da ukatu produktu horiek GGDn izan dezaketen ondorioa. Hori dela eta, gure ikerketan GGPen nutrizio-osaera aztertzea eta horiek GGDn duten eragina analizatu nahi izan zen (tesi honen bigarren helburu nagusia dena). Horretarako, umeen kohortean GGDtan GGPen nutrizio-osaera ordezkatu zen, glutendun produktu homologoena bere ordez erabiliz. Izan ere, umeen kohortean GGPen kontsumoa garrantzitsua izan zen, hartutako energia osoaren %24 izateraino, beste ikerketa batzuen parekoa (101). Era horretan, glutendun dietaren konposizioa esanguratsuki aldatu zen: karbohidrato eta proteina kopurua igo eta gantz kopurua jaitsi zen. GGPek dietaren profil lipidikoan duten eragina aipatzekoa da, glutendun dietak gantz monoasegabe eta poliasegabe gehiago eta gantz ase gutxiago ditu eta. Mikronutrienteei dagokienez, GGDak E bitamina, zink, potasio eta kaltzio kopuru handiagoak dauzka baina, aldiz, beste hainbat mikronutriente gutxiago (A bitamina, tiamina, erriboflabina, niazina, B6 bitamina, B12 bitamina, folato, biotina, D bitamina, pantotenato, magnesio, sodio, burdin, kupre, iodoa, kloro, manganeso eta selenio).

Hartara, GGPen osaerak zeliakoen dietan eragin handia daukala ikusita, produktuon zuntza, AI eta mikronutriente edukia neurtu da laborategian. AI, almidoiaren zati txiki bat da, liseritzen ez dena eta osorik heste lodira ailegatzen dena. Bertan mikroflorak substratu gisa erabiltzen du hartziduran. AIren jokaera fisiologikoa zuntza solugarriarenaren antzekoa da, hortaz honek eduki ditzazkeen efektu onuragarriak (kate motzeko gantz azidoen sorrera, gluzemia eta intsulinemia postprandialaren eta lipemiaren jaitsiera) berdinak izango dira (180, 181).

Gure taldeko emaitzek jakinarazi dutenez (**4. eskuizkribua**), GGPek eta glutendun produktuek orokorrean antzeko zuntz eta AI kantitateak dituzte, beste batzuek adierazi duten moduan (146). Are gehiago, ogien kasuan behatu dugu, Segura *et al*.-ek (182) eta Cornicelli *et al*.-ek (199) bezalaxe, gluten gabekoek zuntz-eduki eta AI kantitae handiagoa dutela. Azpimarratzekoa da emaitza horiek orain arteko GGPen zuntza edukiaren datuak kontrajartzen dituztela (115, 116) eta produktuon hobekuntza nutrizionalaren tesia berresten dutela. Halaber, gure ikerketak adierazi duenez, nahiz eta askotan zeliakoek zereal jatorriko elikagai gutxiago kontsumitu, populazio orokorrak baino, ogiaren kontsumoa nahiko antzekoa da bi taldeetan; are gehiago, kasu batzuetan, Zuccoti eta al.-ek beraien ikerketan ikusi zuten moduan, handiagoa ere (101). Hortaz, azpimarratzekoa da elikadura industriak ondo aukeratu duela zein elikagaitan egin nutrizio aurrerapausuak. Are gehiago, egindako hobekuntzek dieta osoan eragin esanguratsua izan dute, GGPak, eta horien artean ogiak, zuntz iturri nagusi bezala agertuz gure emaitzetan. Era berean, Mazzeo *et al*.-ek gluten gabeko ogiek zuntz-ekarpen handia zutela aurkitu zuten, gehiago dena, gehienak zuntz iturri bezala etiketatu zitezkeela (>3 g/100 g) azaldu zuten (197). Dena dela, eta arestian esan bezala, gluten gabeko ogietan oinarria duen zuntzaren kontsumoa ez ezik, nutriente horren iturri naturalak diren elikagaien zuntza sustatzea ere oinarrizkoa da zelikoetan.

Aipagarria da ere gure azterketa analitikoaren bitartez lortutako emaitza eta produktuen etiketan azaldutako datua batzuetan ez dela berdina. Esate baterako, ogien kasuan etiketan agertutako zuntza kopurua analisien bidez lortutako balioaren erdia izan da. Garrantzitsua da, beraz, elikagai horien konposaketaren eta, zehazki, zuntzaren neurketa doia egitea. Horrela, datuok etiketetan eta elikagaien osaera-tauletan eguneratzeko aukera izanik, GGDren ebaluazio egokia egin ahal izateko. Izan ere, gaur egun GGPak zuntz iturri garrantzitsuak dira bai ume, bai heldu zeliakoen GGDtan.

GGPen merkatua zabaldu bada ere, haien bitamina eta mineral edukiari buruzko informazioa mugatua dago (196, 200, 201). Gainera, bibliografian aurki daitezkeen datuak GGPen osagai-zerrendan eta horien osaketan, oinarrituz kalkulatu ohi dira, gainestimazio arazoak ekar dezakeena (197).

Hortaz, lehen esan bezala, datu zehatzagoak jasotzeko eta zeliakoen dietak egokiago baloratzeko bitamina eta mineral ohikoenen azterketa analitikoa burutu zen zenbait GGPtan, ogian, gosarirako zerealetan eta pastan alegia (**5. eskuizkribua)**. Burdina, piridoxina, erriboflabina eta tiamina eduki baxuagoak behatu ziren aztertutako GGP guztietan haien glutendun homologoetan baino. Niazina murrizketa ikusi zen gluten gabeko zereal eta ogietan. Burdinari dagokionez, Rybicka *et al*.-ek (200) antzeko emaitzak lortu zituzten. Aztertutako 408 GGPetatik 273 ez ziren ailegatzen anoa baterako gomendatutako nutriente ekarpenaren %10era eta 23k bakarrik lortzen zuten ekarpen nabaria egitea eguneko ahorakinari (gomendioaren ≥%25 anoako). 368 GGPekin egindako beste ikerketa batean (148) behatu zuten produktuok tiamina, erriboflabina eta niazina eduki txikiagoa zutela ordezkatzen zituzten gari jatorrizko produktuek baino. Emaitza horiek eta ikerketa honetan lortutakoak bat datoz.

Folato kantitatea ogian eta zerealetan txikiagoa zen, manganesoarena pastan eta B5 bitaminarena zerealetan. GGPak ekoizteko eskuarki erabiltzen diren lehengaiak arroz- eta arto- irinak eta jatorri desberdineko almidoiak (patata, artoa…) dira. Dirudienez, proteinetan aberatsak diren frakzioak irinetik erauzteak folatoen murrizketa esanguratsua dakar (201). Gainera, arroz-irina ez da bitamina honetan aberatsa (201). Hala, arroz-irinak %80 folato gutxiago du gari irinak baino.

Azkenik, biotina edukia ezberdina zen elikagai taldeen artean, altuagoa izanik gluten gabeko zerealetan eta baxuagoa pastan glutendun homologoekiko. Kasu batzuetan gluten gabeko zerealak biotinarekin gotortzen zirela behatu zen. Horrek azaltzen du elikagai talde horretan dagoen biotina eduki handia. Halaber, nahiz eta E bitamina kantitatea askoz txikiagoa izan gluten gabeko zerealetan, pasta eta ogien kasuan ez zen ezberdintasunik behatu. Are gehiago, aipagarria da ogi askoren formulazioan behatutako eguzkilore-olioaren erabilera, E bitamina eduki altuagoak sor zituena ale espezifiko horietan.

Azpimarratzekoa da elikagai-industriak GGPen bizi-iraupena eta ezaugarri erreologikoak hobetzeko asmoz egindako esku-hartzeak ondorioak izan dituela produktuon nutrizio-osaeran (202). Hortaz, ekoizpenean GGPen konposizio nutrizionala hobetzeko asmoz neurri ezberdinak proposa daitezke: almidoia osagai nagusi bezala ekiditea, hatzidura erabiltzea, zerealen bertsio integrala aukeratzea, pseudozerealak (artobeltza, kinoa, amarantoa edo teffa) edo/eta lekaleak erabiltzea (203, 204).

Datu analitiko horiek guztiak aplikatuta GGPen mikronutriente edukiak dietetikoki izan dezakeen ohiartzuna aztertzeko aukera paregabea da. Hala, umeen GGD **(2.eskuizkribua)** datu zuzenekin birkalkulatu egin zen. Hots, dieta aztertzeko Missbach-en datu-basean agertzen diren GGPen mikronutrienteen informazio nutrizionalaz (153) baliatu ordez, EAEko produktuen datu analitikoak erabili ziren. Emaitzak antzerakoak izan ziren, datu-baseetan oinarritutakoak muturrekoagoak izan baziren ere. Kaltzioa salbuespena izan zen, mineral honen ahorakina kalkulatzeko datu analitikoa erabiltzen denean umeen dietan gabezia desagertzen baita. Hori datu analitikoa datu-basean (153) agertzen dena baino handiagoa izategatik gertatzen da; ogiaren kasuan, egunean kantitate handian kontsumitzen dena, bikoitza izanik. Horren harira, aipagarri da Europar Batasuneko 2011ko urriaren 25eko 1169 araudiak (64) mineraleei dagokienez onartzen duela -%35 eta +%45 arteko ziurgabetasuna etiketan; horrek azterketa analitikoaren eta Missbach-en datuen arteko aldeak ohikoak direla iradokitzen du. Edonola ere, behatutako kaltzio eduki ezberdintasuna azaltzeko, kontuan hartu behar da ikerlan honetako GGPak EAEn erositakoak direla eta Missbach-enak, berriz, Alemaniako produktuak direla. Azkenik, aipagarria da umeen kohortean kaltzio gabeziarik ez ikustea bat datorrela talde honetan behatu zen den esneki kontsumo egokiarekin.

Aurreko guztia kontuan hartuz, ez dirudi aztertutako GGPek ume zeliakoen dietan behatu diren mikronutriente gabezietan eragina dutenik. Izan ere, zerealak ez dira gutxiegi hartzen diren mikronutrienteen iturri naturala. Hortaz, emaitzok, berriz ere, dieta osasungarria sustaztera garamatzate, fruta eta barazkien kontsumoa areagotzera adibidez.

Azido folikoaren kasuan, GGPetan eduki baxua neurtu zen, glutendun baliokideekiko. Zentzu honetan, azpimarratu behar da Canadan eta EEBBetan gari-irina azido folikoarekin gotortzea derrigorrezkoa dela (205, 206), baina ez, ordea, beste irina alternatibo batzuk aberastea, hala nola GGPetan erabili ohi direnak. Horrela, GGDn ikusitako folato gabezia kontuan hartuz, GGPak edo/eta gluten gabeko osagaiak ere gotortzea interesgarria izan daiteke, ume zeliakoen kasuan gutxienez. Are gehiago, folatoan gotortzeko neurri hori biotinara ere zabaldu daiteke, parte hartzaileengan behatutako gabezi orokortu eta kezkagarrian oinarrituta. Hala, gaur egun hasi dira zenbait gluten gabeko zereal bitamina horrekin gotortzen.

10. taula. GGPen profil nutrizionalen ikerketa batzuen emaitzak, beraien glutendun produktuekin erkatuak (Melini *et al*.-etik moldatuta (207)).

Erreferentzia	Lurraldea	n	Elikagai taldea	GGPen profil nutrizionala
Larretxi *et al*. **(4. eskuizkribua)**	EAE	55 GGP/ 55 GP	Ogia	↑ Zuntza eta AI
			Okindegi produktuak eta tartak	=
			Opilak	=
			Gosari-zerealak eta umeen zereale-formulak	=
			Pasta	=
			Irinak eta oreak	=
			Etiketa eta balio analitikoen ezberdintasunak aurkitu ziren zuntza eta AIren kopuruetan	
Larretxi *et al.* **(5. eskuizkribua)**	EAE	37 GGP/45 GP	Ogia	↑ gantza, E bitamina ↓ proteina, burdina, piridoxina, riboflabina, tiamin eta folatoa
			Zerealak	↑ E bitamina, biotina ↓ burdina, piridoxina, riboflabina, tiamin, B5 eta E bitamina
			Pasta	↑ KH ↓ burdina, piridoxina, riboflabina, tiamin, Mn eta biotina

Erreferentzia	**Lurraldea**	**n**	**Elikagai taldea**	**GGPen profil nutrizionala**
Miranda *et al*. (2014) (115)	EAE	206 GGP/ 289 GP	Ogia	↑ Gantza, gantz asea, gatza ↓ proteina, zuntza
			Pasta	↑ Gantza, gantz asea, gatza ↓ proteina, zuntza
			Okindegi produktuak	↑ Na, kolesterola ↓ proteina, KH
			Marka desberdinen arteko ezberdintasunak	
Missbach *et al*. (2015) (153)	Alemania	63 GGP/ 126 GP	Irinak, ogia eta okindegi produktuak, pasta eta eratorriak, zerealak, opilak eta *snack*ak.	= energia,KH, gantzak, gantz aseak, zuntza eta KH sinpleak ↓ proteina, Na, K
Kulai *et al*. (2014) (147)	Canada	71 GGP/ 60 GP	Ogia	= energia eta gantz aseak ↑ gantzak ↓ proteina
			Pasta	= energia ↑ KH ↓ zuntza, Fe, folatoak
			Gosari-zerealak eta tartak	=
Mazzeo *et al*. (2015) (197)	Italia	60 GGP	Gozokiak	↑ gantza eta KH sinpleak
			Brioche	↑ gatza
			Ogia, pasta, irina,*snack*ak	↑KH eta KH sinpleak Zuntza iturria (>3 g/100 g)

Erreferentzia	**Lurraldea**	**n**	**Elikagai taldea**	**GGPen profil nutrizionala**
Wu *et al*. (2015) (198)			Pasta eta gosari-zerealak	= energia, gantz aseak, Na eta KH sinpleak ↓ proteina
			Ogia	= energia, gantz aseak, Na eta KH sinpleak ↑ zuntza ↓ proteina
			Zereal-barritak, tartak eta opilak	↑ KH sinpleak, gantz asea, gatza
Cornicelli *et al*. (2018) (199)	Italia	235 GGP/ 349 GP	Opilak	= energia ↑ KH, gantz aseak ↓ proteina, zuntza,gatza
			Pasta	= ↑ energia, KH,gantz aseak,gatza ↓proteina,zuntza
			Ogia	= ↑ KH, zuntza ↓energia, proteina (bereziki)
			Ogi txigortua	= energia ↑ KH,gatza ↓proteina (bereziki)

Erreferentzia	**Lurraldea**	**n**	**Elikagai taldea**	**GGPen profil nutrizionala**
Fry *et al*. (2018) (208)	UK	679 GGP/ 1045 GP	Opilak	↑ gatza, KH sinpleak ↓proteina
			Crackers	↑ gatza, KH sinpleak ↓proteina
			Ogi (zuria/integral)	↑ zuntza, gantza eta gantz asea (zuria), KH sinpleak ↓proteina
			Pasta (zuria/integral)	
			Irinak (zuria/integral)	↑ KH sinpleak, gatza (integrala) ↓proteina
			Gosari-zerealak	↑ gatza ↓proteina, zuntza
			Pizza orea	↑ KH sinpleak ↓proteina
Calvo-Lerma *et al*. (2018) (146)	Espainia	621 GGP/ 600 GP	Ogia	↑ gantza eta gantz asea ↓proteina (bereziki)
			Pasta	↓ KH sinpleak eta proteina
			Opilak	↑ gantza ↓proteina

Notak: GGP: gluten gabeko produktuak, GP: glutendun produktuak, KH: karbohidratoak, AI: almidoi iraunkorra

Tesi-lan honek kalitate handiko nutrizio-informazio zabala garatu du GGPen inguruan, oraindik bibliografian murritza dena eta, are gehiago, elikagaien etiketetan edo/eta GGD diseinatzeko eta ebaluatzeko erabili ohi diren datu-base eta tresnetan. Aipatzekoa da beraz, ez dela bakarrik elikagaion osaera aztertzen, horrek duen garrantzi dietetikoa azpimarratzen da. Hala ere, ez da kontuan hartu hain kezkagarria den GGPen bioeskuragarritasuna (209). Lan honen beste ahultasun bat heste-biopsiarik egin ez izana da, ez diagnostikoan ez GGD jarraitzean ere. Hortaz, GGD jarraituz kalte histologikoa arintzen denik ezin da ziurtatu, ezta horrek eragin dezakeen nutrienteen xurgapenaren murrizketa desagertzen denik ere. Nolanahi dela, ikerketa honen helburua GGDk berau pertsona zeliakoan eragin dezakeen gabezia nutrizionak aztertzea izan da, kalte histologikoa egon ala ez.

Funtsean, emaitza horiek guztiek pertsona zeliakoen dieta orekatua ez dela agerian jartzen dute eta, are gehiago, GGPen gehiegizko kontsumoak zenbait nutrizio-desoreka handitzen dituela. Hala ere, ezin da ukatu elikagai-industria, oraindik GGPen kalitate nutrizionala hobetzeko lan handia izan arren, bide onetik doala. Izan ere, gure ikerketan behatu den moduan GGPen zuntz edukia hobetu da azken urteotan glutendun homologoen berdina edo altuagoa izatera ailegatuz. Horrez gain, lan honetan GZan elikagaiak mikronutrienteetan gotortzeko nazioarteko arauak hobetzeko proposamena ere egiten da, ume eta zeliako nerabeei zuzenduta.

Azkenik ezinbestekoa da populazio honen gaineko hezkuntza nutrizionala, GGD osasungarria jarraitzen hezitzeko. Horrela, akatsak detektatu (dietaren urratzeak) eta dieta zuzendu ahal izango dute zeliakoentzat espezifikoak diren jarraibideen bitartez.

SUMMARY

The work carried out in this doctoral thesis can be divided into two main parts. New data on nutritional status and dietary habits of a celiac cohort in the Basque Country are analyzed in the first three manuscripts. The usual defects of the GFD in achieving the nutritional balance and the barriers to follow it are defined by age group and gender (A objective). Then, in the following two manuscripts (manuscripts 4 and 5), the GFP's nutritional composition is analyzed and compared to gluten containing foods. Likewise, the effect that this has on celiac peoples's GFD has been concluded (B objective).

A. Nutritional status and food habits of the celiac population in the Basque Country

1. Antropometric measurements

As may be observed in the anthropometric measurements of the celiacs of this study, most of them have a normal weight. When participants are distributed in three different cohorts, women seem to have a healthier body mass index (BMI) than men and children. In fact, there are a lot of cases of overweight and obesity in the men cohort, with more than a half of them in a high fat percentage, unlike the other two groups. By contrast, 20% of children were underweight. Comparing these results to the data from general population, it can be concluded that the celiac group has lower overweight and obesity prevalence; Furthermore, although the data of men's cohort are less useful, the prevalence of overweight is still lower than in the general population. Bardella *et a*l. published similar results in another study; (106); In short, they described that celiac patients presented lower weight, fat mass (FM), lean mass (LM) and bone mass (BM) than control groups.

As some research has shown, in some cases celiac people do not improve their malnutrition status after carrying out a GFD (mantaining low BMI, FM and bone mineralization) (165-167). Smecuol *et al*'s different results obtained in a study were carried out with 25 newly diagnosed celiac patients (168). This group had low percentages of FM, LM and BM at the beginning but after 37 weeks of GFD treatment, anthropometric measurements were significantly enhanced. In the same vein, Ukkola and colleagues (169) indicated that patients with low weight at the time of diagnosis recovered weight after one year of following a GFD. By contrast, those who were overweight or obese were losing weight because of a better or more balanced GFD. Furthermore, Barone *et al* (165) compared the anthropometric data of 39 celiac people before and after following a GFD. At the time of their diagnosis, most of the celiac patients (82%) had normal or excessive BMI and only 10% of them were malnourished. After following a GFD, there was a significant weight gain in people with normal BMI, without achieving overweight, and half of those with malnutrition (n=4) reached a normal BMI. As shown in other studies, the lack of improvement in the other half (n = 2), could be justified by their inadequate diet or by their concomitant use of certain drugs (170).

As regards sex differences, it is worth mentioning that the result is to be expected, since it is also found in the general population (55, 171-173). The explanation for this may be in part that there is usually a lack of attention and interest in men regarding health, certainly less than that of women, as described in some studies (174, 175).

In general, when celiac people follow a strict GFD they improve their anthropometric measurements. In fact, following a GFD most of the time causes alleviation of the symptoms, leading to recovery of nutritional status. Moreover, this group has a greater sensitivity to food culture, and therefore could have healthier dietary habits. Thus, they have better weight, FM and BMI values than when they started a GFD. In fact, there is sufficient consensus about the following: the earlier the GGD is started, the more rapidly anthropometric measurements will be normalized (91, 176). By contrast, although their nutritional status improves, sometimes it does not reach the level of healthy people (166). This can be explained by the fact that the pathological condition of celiac people sometimes causes a lower bioavailability of nutrients and/or because of unwanted dietary transgressions, as Kautto *et al* suggested (111) in their study.

2. Macronutrient intake

CD treatment can be a dietary challenge for newly diagnosed people. In fact, some of the staple foods in the Mediterranean Diet (MD), such as bread, pasta, cereal and flour and all their derivatives, should be excluded from the GFD. Moreover, all these foods from cereals group are the main source of several macronutrients and micronutrients, which means that the nutritional status of this group can be conditioned by a possible imbalance in the diet and by the lack of some micronutrients (96-98, 113). Thus, agreeing with many other studies (93, 100, 108-110), evaluation of the GFD of the participants in this study revealed that their diet is, in general, unbalanced.

Compared with Dietetic Reference Intake (DRI) (154), it is observed that they had a low energy, complex carbohydrates, and fiber intake and an excessive protein and fat, especially saturated fat intake. In the GFD, macronutrient imbalance is quite predictable, due to the low consumption of cereal and other sources of complex carbohydrates. As in the general population, high consumption of meat and animal origin food was detected.

When men and women who participated in the research were compared, more men consume excessive energy than women, which could justify the higher prevalence of overweight and obesity observed in the men cohort. However, the distribution of macronutrient imbalances is similar in both sexes, as are the dietary fat profiles. Despite the fact that both groups consume excessive fat and cholesterol, men do so more than women.

When comparing celiac and general community nutritional habits similar results, such as macronutrient imbalance (156), are found. Other authors also confirmed similar energy distributions when they compared GFD and control diets (101, 107). Barone *et al*. (165) analyzed the nutritional composition of the GFD by comparing it to a control group. They observed that the amount of energy, carbohydrates and proteins consumed was similar in both groups. However, celiac people consumed more fat and less fiber than the control group. Furthermore, Bardella *et al*. observed that the celiac people had a lower energy and carbohydrate intake than control group, and higher percentage of fat (106). In our study, women consume less energy and macronutrient amounts than controls. In the case of men, there were no notable differences. With regard to carbohydrates, both celiac people and controls were below the recommendation. Although all of them achieve the Minimum DRI (130g/day), celiac people consume fewer grams than the controls. In the case of celiac people, only 42% of total energy is obtained from carbohydrates, and similar results are seen in the general population.

In this study, celiac people ate less fiber than controls did. What is more, celiac women, especially the youngest ones, had lower intake than celiac men did. Which is to say, even though the fiber intake was very small in both sexes, the men's group achieved a better fiber recommendation (26% of men vs. 4% of women who fulfil the recommendation).

Other previous studies have shown that GFD is lower in fiber than a gluten containing diet. Wild *et al* (177) showed that fiber intake of celiac women was significantly lower compared to data from dietary surveys in the United Kingdom's population. Martin *et al* found the same results by comparing their men cohort data with the German community (107). In fact, a low fiber intake can be associated with the consumption of GFPs, because the formulation of these products is often low in fiber to avoid the risk of cross contamination in the production process (178).

Even so, in recent years, interest in GFP has increased, as has a concern to improve their quality. In this way, the food industry has begun to use new components in the production of GGP (179) to improve their organoleptic and nutritional value. The new components will try to correct the faulty composition of the GFPs seen in the past (115).

Among these new raw materials can be cited resistant starch (RS), a source of fiber. The physiological behavior of RS is similar to soluble fiber, due to its beneficial effects (180, 181). Data obtained from our analytical analyzes (**manuscript 4**) indicates that gluten-free bread has a greater amount of fiber and RS than gluten-free bread, confirming the thesis of nutritional improvement in GFPs. These results coincide with the data described elsewhere (146, 182-184). It is important to mention that all research (including ours) is based on the value of fiber that appears on the label of the GFP. However, as described in the 4th manuscript, in some cases the fiber content of GFP labels is either underestimated or does not appear (because the law does not require this information).

By analyzing the dietary fat sources and the lipid profile of the GFD versus the control's diet, it may be observed that while the intake of saturated and monounsaturated fatty acids (MUFA) is similar in both groups, the celiac men consume fewer polyunsaturated fatty acids (PUFA). On the whole, the ratio of saturated/unsaturated fats met the recommendations but the intake of cholesterol was excessive in both groups.

In the case of protein intakes, celiac people are above the recommendation and their consumption is 17% of total energy in the three cohorts. In general, there are similar results in the general population. One of the reasons to justify this may be excessive intake of food of animal origin.

Regarding the children's cohort, a similar dietary imbalance to that in adults has been observed. They fulfilled only 80% of the energy intake recommendation. The carbohydrate intake recommendation is not fulfilled (with 45% of energy) and they consumed more fat and protein than they should, 40% and 17% of total energy respectively. In the same vein, many studies have described similar intake patterns in children (68, 105), which is to say reduced energy intake (108), low carbohydrate and fiber (93, 100, 109) and high fat (93, 100, 108, 110) and protein consumption (108). Fat consumption is also unbalanced, with excessive saturated fat and low unsaturated fats (100, 111).

It is worth mentioning the results published by Zuccoti *et al*. (101) , where the GFD followed by adolescent and celiac children was compared with the diet of the general population in Italy. Although both groups exceeded the Italian recommended protein, fat and sugar intake, celiac children had a much higher energy and carbohydrate intake than the control group, with the same amount of energy derived from proteins. In a study carried out in Spain, they compared the diet of a group of celiac children before diagnosis and after one year of following the GFD (185). This work found that uptake of monounsaturated fats was higher in the GFD, while saturated fat consumption was lower (185). By contrast, Babio *et al*. (102) indicated that celiacs between 10 and 23 years old consumed more added sugars, total fat and high-protein foods than controls. Nevertheless, the consumption of food rich in starches was lower.

As far as fiber is concerned, in our study celiac children consumed a small amount of it (15g/day) which was lower than the recommended; nevertheless, it is a similar amount to the uptake of the general child population (93, 100, 102). In fact, some studies did not detect any differences between the diets of celiac and non-celiac children (100, 102). However, Mariani *et al*. found a much lower carbohydrate consumption and, in particular, a lower intake of fiber in celiac adolescents, when compared to controls (93). This can be explained by the fact that following a GFD excludes high-fiber cereals and includes the presence of refined cereals (68) .

Table 7. Summary table. Spanish Society Recommendations (FESNAD) compliance concerning energy and macronutrient intake.

		Women	**Children**	**Men**
Energy intake	**Insufficient**	30%	x/↑	40%
	Adequate	65%		46%
	Excessive	5%		14%
Distribution of macronutrients:				
Proteins		x/↓	x/↓	x/↓
Carbohydrates		x/↑	x/↑	x/↑
Lipids		x/↓	x/↓	x/↓
Saturated fats		x/↓	x/↓	x/↓↓
Fiber intake	**Appropriate**	4%	x/↑↑	26%
	Insufficient	96% (43 %< 15g)		74% (28% < 15g)

Notes: x, values do not achieve Spanish Society (FESNAD) recommendations, ↓ suggests an intake reduction; ↑ suggests an intake increase; FESNAD, Federation of Spanish Societies of Nutrition and Dietetics.

3. Micronutrient intake

As mentioned earlier, the computer software to evaluate celiac diets should have specific nutritional composition of the GFPs in their database. Moreover, most GFPs labels do not indicate the content of vitamins and minerals. Therefore, in this study the analysis of the micronutrient intake of the participants was carried out based on estimation with homologous gluten-containing products. In order to analyze whether the intake of vitamins and minerals is appropriate or not, the compliance of 2/3 (> 67%) of the recommendation was established, as elsewhere (186, 187) . he research reveals that almost all participants took too little vitamin D and adults cohort suffered from vitamin E and iodine deficiency. In the case of women, selenium uptake was low and in men vitamin A and magnesium are also scarce in the diet. Women and children presented iron deficiencies and men and children also folate deficiencies. Finally, children did not consume enough calcium. Although some studies have underlined the lack of cyanocobalamin in celiac people at the time of diagnosis (188-190), none of our participants showed low vitamin B_{12} intake. In fact, the excessive consumption of food with an animal origin assures the contribution of this vitamin, even if the absorption level is unknown.

Many studies have revealed that GFD can present micronutrient deficiencies (9, 97, 105, 118). This may happen because, on the one hand, GGD reduces both cereal type and intake, and on the other hand, wheat flour products are often enriched with micronutrients and GFP, often made by maize or rice, does not. It has also been observed that GFPs do not have the same amounts of micronutrients as their counterparts, such as thiamine, riboflavin, niacin, folate, vitamin D, calcium, or iron (18, 48). This situation is even more worrying if we take into account that at the time of diagnosis a lot of celiac people have a poor absorption of nutrients, which often results in a lack of micronutrients.

When comparing the GFD of men and women in our research, there were differences in minerals, but not in the consumption of vitamins. Women reached better intake recommendations for magnesium than men, but worse iron, iodine, potassium, and selenium ones.

When these data were compared with control groups, the deficiencies of celiac and general population seem to follow a similar pattern. However, some significant differences could be found: vitamin E, niacin, magnesium and selenium consumption are lower in celiac women, while riboflavin, vitamin B6 and folate were significantly higher. Men also took less vitamin E, niacin and magnesium than controls and riboflavin, vitamin B6, zinc, potassium and more selenium. Martin *et al*. indicated that B1, B2, B6 vitamins, folic acid, magnesium and iron consumption were lower among celiac adults than in the general population (107). At the same time, Barone and colleagues (165) found that calcium, vitamin D and phosphorus intake were comparable in celiac and control groups, both following Italy's intake recommendations.

Table 8. Summary table. Recommendation for the micronutrient intake of the Spanish FESNAD Association (people who do not achieve 2/3 of the recommendation).

	Women	**Children**	**Men**
Vitamins			
D	48%	69%	55%
E	39%	57%	52%
A	11%	37%	19%
Folate	18%	54%	24%
Minerals			
Iodine	80%	93%	50%
Iron	31%	23%	0%
Selenium	31%	20%	7%
Calcium	13%	51%	14%

Table 9. Nutritional status of people following a GFD and excessive or low nutrient intake

Reference	Country	Cohort	Nutritional status	Dietary habits	
				Excessive consumption	Low consumption
Larretxi *et al.* (2018) (192) **1st manuscript**	Basque Country	Children (n=83)	Appropriate weight, BMI and FM	Lipids and protein	Energy, CH, D vitamin, Folic acid, Ca and Fe
Churruca *et al.* (2015) (117) **2nd manuscript**	Basque Country	Adult (women) (n=54)	Appropriate weight, BMI and FM	Lipids and protein	Energy, CH, fiber (less than celiac men), vitamin E and vitamin D , folate, Ca, Fe, I and Se
Gonzalez *et al.*(2018) (193) **3rd manuscript**	Basque Country	Adults (men) (n=42)	BMI and FM high values	Lipids, protein and cholesterol. Comparing them with celiac women energy and saturated fat intake is higher	CH, fiber and PUFA
Bardella *et al.* (2000)(106)	Italy	Adults (n=71)	Weight, BMI, FM, IM eta HM (women) lower than controls.	Lipids	Energy and CH

Barone *et al.* (2016) (165)	Italy	Adults (n=39)	Better BMI than at diagnosis. Similar BMI, FM eta HM than controls	Lipids	fiber
Smecuol *et al.* (1997) (168)	Argentina	Helduak (n=25)	Weight and FM increased and LM mainteined (no differences between strict GFD and dietary lapses)	Lower energy intake celiac people who follow strict GFD than those with dietary lapses.	
Ukkola *et al.* (2012) (169)	Finland	Adults (n=698)	Better BMI after following GFD.	Not shown	
Wiech *et al.* (2018) (9)	Poland	Children (n=41)	LM and water weight higher values and lower FM than controls Children with strict GFD better FM and LM than children with dietary lapses. After one year on a GFD anthropometric measurements improve.	26 children follow a strict GFD and 15 have dietary lapses.	
Kautto *et al.* (2014) (111)	Sweden	Adolescents (n=37)		Lipids, PUFA and α-linolenic acid, tiamin and riboflabin, niacin, vitamin B6, folate, B12, P, Zn, Se and Mg.	
Öhlund *et al.* (2010) (100)	Sweden	Children (n=25)	Not shown	Saturated fat and sugar	Energy (52% of children) fiber, vitamin D, Mg and Se.
Shepherd *et al.* (2013) (96)	Australia	Adult (n=105 /50 diagostikatu berri eta 55 GGD ≥2urte)	Similar BMI between newly diagnosed and long-term treated celiacs.		Fiber , tiamin, folate, vitaminA, Mg, Ca, Fe, and Zn.
Wild *et al.* (2010) (177)	UK	Adult (n=139)	Not shown	Sugar	Mg, Fe, Zn, Mn, Se and folate
Zuccotti *et al.* (2013) (101)	Italy	Children (n=18)	No differences	Energy, lipids (but lowe than controls) and CH	
Babio *et al.* (2016) (102)	Spain	Children and adult (n=98)	Not shown	Sugar and lipids	Fiber, folic acid, Ca, Fe and Mg Starchy food

Salazar *et al.* (2014) (185)	Spain	Children (n=37)	Appropriate anthropometric measures	MUFA	SFA, Vitamin D, Zn, fiber
Martin *et al.* (2013) (107)	Germany	Children and adult (n=80)	Not shown	Lipids	fiber, CH, vitamins B1, B2, B6, folic acid, Mg and Fe

Notes: BMI: body mass index, FM: fat mass, LM: lean mass, BM: bone mass, CH: carbohydrates, GFD: gluten free diet, PUFA: polyunsaturated fatty acids, MUFA: monounsaturated fatty acids

4. Food category

The dietary pattern is one of the main factors for explaining macronutrient imbalances and micronutrient deficiencies in the GFD. For this reason, the dietary habits of the celiac people were analyzed through food frequency questionnaires in this study. In general, consumption of cereals was very low in all the cohorts studied, children being the cohort with the highest consumption. In fact, children consumed the most bakery products in their daily food (sandwiches, biscuits, breakfast cereals, etc.). After analyzing the diets of our women, it was observed that almost half of them (48%) consume a small amount of cereals, less than two servings per day. However, since total energy consumption is scarce, 23% of total energy can be obtained from cereals, while in the general community of the ENIDE, 17% of energy is used (22). These data suggest that the celiac women involved in the study probably had greater concern and interest in nutrition than controls, and, consequently, slightly healthier dietary habits. Similarly, in the majority of celiac people consumption of vegetables was scarce too, children being the cohort with worst achievement of the recommendation. Around half of the participants fulfilled recommendations for fruit intake. By contrast, the consumption of meat and derivatives was excessive in most cases, especially among men. Most people also fulfilled the consumption of dairy products and fish, and consequently, animal protein consumption was high. For example, in women's cohort about 70% of dietary protein was from an animal origin, 20% from dairy products and 50% from meat, fish and eggs. With regard to legumes, the compliance grade was appropriate, and it should be emphasized that almost all children fulfilled this recommendation.

In general, the dietary pattern of celiac women and men were similar, the only difference was observed in fish and eggs, which showed higher percentages of excessive consumption in men.

All these results show the importance of implementing specific nutritional guidelines for celiac people. For example, in order to increase the consumption of complex carbohydrates, information about gluten-free cereals and pseudo-cereals should be provided, in addition to traditional gluten-free cereals such as corn and rice (cornstarch, legumes, amaranth, quinoa, cornstarch, teff, or potatoes), as well as how these should be included in the daily diet. Furthermore, in order to increase the intake of fiber, they need some tips on how to consume more fruit (if possible, with skin), vegetables, vegetables and legumes. The role of GFP in the diet should not be forgotten, showing the importance of reading product labels to buy healthier options. In order to decrease the intake of animal protein they will be recommended to control their meat consumption and to choose lower fat content products. This might reduce the risk of cardiovascular disease associated with excessive protein and fat intake (hypercholesterolemia, high blood pressure ...). Although this advice is interesting for all the population, it is even more necessary in the celiac collective, which is more likely to suffer from these diseases, as proposed by various authors (194).

As regards the dietary habits of the celiac children and adolescents, poor adhesion to the Mediterranean pyramid was observed. As in the case of adults, the consumption of food from cereals and derivatives group was inadequate. In fact, only 33% of them fulfilled the SENC recommendation (minimum 4 portions/day cereal consumption) and 20% of them even consumed less than 2 portions/day. Babio *et al*. saw that celiac people ate fewer starches (pasta, bread, cakes) and more protein food (meat, fish, eggs) than the general population (102). Additionally, the celiac children of our study did not achieve the consumption of food rich in fiber, such as legumes, vegetables and/or fruit. Therefore, as in adults, it would be recommendable to decrease protein and fat consumption and increase carbohydrate and fiber intake (117).

Apart from having healthy dietary habits it is also necessary to lead a healthy lifestyle. It has been observed that only a few people (19%) in the cohort of children and adolescents achieved the WHO recommendation for physical activity. However, almost no one followed unhealthy habits, such as going to school without having breakfast, consuming cakes for more than one a week and/or eating often in fast food restaurants.

The strength of this research is based on the use of GFP's nutritional composition to analyze the celiac's diet. However, simultaneously, the main limitation of this research has been the usage of label's data which has not micronutrient content of GFPs (195, 196). In fact, an estimation using the label data of their gluten-containing counterparts was made in order to analyze the participants' diet, which may over- or underestimate the amount of micronutrient intake (197). In order to solve this problem, a table of micronutrient content of the GFPs created by Missbach *et al*.(153) was used to make a more detailed dietary analysis in children cohort. However, this database was made with GFPs produced in Germany, so micronutrient content can be slightly different from those found and produced in Spanish. For this reason, in the end we selected some of the normally consumed GFPs to analyze the most common vitamins and minerals in the laboratory and to create our own database. Another limitation of this work is that the selection of people who participated in this research was not randomized, as they were volunteers. This fact could affect the registered dietary habits of the participants, because people who attended are usually more interested in nutrition than the rest of the population is. Finally, it is worth mentioning that there is no own control group in the experimental design, although adequated controls had been found.

4.1 Role of GFPs in GFD imbalance

Even though the dietary pattern may explain to some extent the imbalances and deficiencies that could appear in the GFD, other factors also play a significant role. Many studies have reported that GFP's nutritional composition is not like their gluten-containing counterparts, as sometimes they have a higher amount of fat but less carbohydrate, protein, fiber, and vitamins and minerals (115, 147, 153, 198) (table 10). Therefore, the impact that these products may have on GFD cannot be denied. For this reason, we wanted to analyze the GFP's nutritional composition in our study and observe the effect of these on the GFD (the second main objective of this thesis). To this end, the GFD of the children cohort was taken and all the GFPs were replaced by similar gluten-containing counterparts. In fact, GFP consumption in the children's cohort was important, up to 24% of the total energy consumed, results that were in good accordance with other studies (101).

Thus, the composition of the GFD changed significantly: carbohydrate and protein percentages decreased and the fat percentage increased. It is important to mention the effect of GFP on the lipid profile of the GFD, because the gluten containing diet had more MUFA and PUFA and less saturated fat. With reference to micronutrients, GFD contained more vitamin E, zinc, potassium and calcium, but it also had fewer micronutrients (Vitamin A, Thiamin, Riboflavin, Niacin, Vitamin B6, Vitamin B12, Folate, Biotin, Vitamin D, Pantothenate, Magnesium, sodium, iron, cupper, iodine, chlorine, manganese and selenium).

In view of the fact that GFD is highly influenced by the composition of these products, fiber, RS and micronutrient content were measured in the laboratory. RS is a small part of the starch that cannot be digested and reaches the large intestine intact, where colon microflora use it as a substrate in fermentation. The physiological behavior of RS is similar to that of soluble fiber, so that the beneficial effect of these (short fatty acid synthesis, control of glycaemia and postprandial insulinemia and lipid lowering effect) may be the same (180, 181)

As reported by our team (**manuscript 4**), nowadays GFPs and gluten containing products have generally similar fiber and RS content, in agreement with other authors (146). In addition, we have also observed, in line with Segura *et al*.(182), and Cornicelli *et al*. (199), that in the case of breads, gluten-free have a higher contain of fiber and RS than gluten-containing. It is worth pointing out that these results contrast the results of the GFP fiber content that were up to date (115, 116) and that they confirm the view about the nutritional improvement of the GFPs. Our research also has indicated that although many celiac people consumed less cereal-derived food than the general population, bread consumption was quite similar in both groups, even higher in some cases, as Zuccoti and others observed in their research (101). This, it is remarkable that food industry selected an appropriate target to perform nutritional improvements. Moreover, such amelioration had a noticeable impact on total diet; in fact, GFPs, breads particularly, appeared as an important fiber source in our data. Likewise, Mazzeo *et al*. discovered that gluten-free breads were a good source of fiber and, moreover, that almost all of them could be labelled as a “fiber source” (> 3 g / 100 g) (197). Nevertheless, and as mentioned, fiber coming from not only GF breads, but also from other natural important fiber sources should be enhanced.

It is also worth mentioning that the results obtained from our analytical analysis and the information described in the labeling of products are not the same. For example, the content of fiber in the label for the breads was half the value obtained through analysis. It is important, therefore, to know the composition of these foods and, specifically, the correct measurement of the fiber. So that, after revising label information and updating food composition databases, the GFD of celiac people can be properly evaluated. In fact, today GFPs are important sources of fiber both in children’s and in adult’s GFD.

Despite the growing market of the GFP, their data on vitamin and mineral content is still limited (196, 200, 201). Moreover, the data found in the literature are usually calculated from ingredients and their composition databases, which has been proposed to lead to overestimation (197).

Thus, as we have already said, in order to obtain better and more accurate data and to make a more adequate evaluation of celiac’s diets, our team decided to carry out an analytical study of the most common vitamins and minerals (**manuscript 5**). Lower iron, piridoxin, riboflavin and thiamin content was found in the three analyzed GFP groups. Niacin reduction was observed in GF flakes and breads. With regard to iron, similar results were found by Rybicka *et al.* (200). She described that from 408 analyzed GFP, 273 fulfilled less than 10% of recommended nutrients intake per portion and only 23 products were high contributor to the daily intake (over 25% of recommendation intake per portion). In a research performed with 368 GFP, including flours, breads, pasta and cold cereals, overall it was observed that this kind of products contained lesser amounts of thiamin, riboflavin and niacin, than the wheat product they were intended to replace (148). These results are in line with the obtained in the present study.

Folate content was lower in GF flakes and breads but only reached statistically significance in the last one, manganese amount was lower only in GF pasta, and that of vitamin B5 in GF flakes Commonly used ingredients for GFP are maize and rice flours as well as a variety of starches (potato, corn), among others. It seems that removal of protein-rich fractions from flours may result in dramatic depletion of folates (201). Additionally, rice flours are not very rich in this vitamin (201). In fact, we calculated an almost 80% reduction of folate content in rice flour with respect to wheat flour (p=0.05).

Finally, biotin content differed widely among groups, being higher in cereal flakes and lower in pasta GFP than in their counterparts. Besides, we found that some GF cereals were fortified with biotin, thus explaining its bigger content in this GF food group. Similarly, although vitamin E contribution from GFP was lower in flakes, no differences were observed in pasta and bread. Moreover, it is remarkable that some breads showed a formulation with sunflower oil, which led to higher vitamin E content in those specific stuffs.

It is important to point out that food technology interventions to improve the shelf life and rheological properties of GFP have influenced their nutritional profile (202). In order to avoid the absence of the mentioned micronutrients without fortifying foodstuffs, different strategies can be proposed: excluding starch as a major ingredient, sourdough fermentation, and using less popular grain GF flour such as that from pseudocereals (buckwheat, quinoa, amaranth and teff), including wholemeal form of gluten-free cereals (203, 204).

Taking into account all these analytical data, we achieved an unique opportunity to analyze the potential effect of GFP's micronutrient content in celiacs diet. In this way, GFD composition of children (**manuscript 2**) was recalculated with analitycal data. So that, instead of using the nutritional information of Missbach's micronutrients database to analyze the diet, the analytical data of GFPs from Basque Country were used. The results were similar, although those deficiences calculated with data from Missbach database (153) were more accentuated. Calcium entailed the exception because when its analytical value is used to evaluate the intake of calcium in the celiac children's diet, the observed deficiency dissapeared. That happens because calcium analytical data is higher than that of Missbach *et al*. (153); concretely, in the case of bread, which is very consumed, the content is twice the amount from the data-base.

In this sense, it is worth mentioning that the EU 1169 regulations on the 25th of October 2011 (64) accept for minerals an uncertainty from minus 35% to plus 45% in the label; This suggests that differences in analytic measures and Missbach data can be acceptable. In any case, in order to explain the observed inequality of calcium content, it should be taken into account that the GFPs in this study are purchased in the Basque Country and that Missbach database is considering German products. Lastly, it can be highlighted that this new data on the lack of calcium is in accordance with the appropriate intake of dairy products that are seen in the **manuscript 2**.

Taking this results into account, it does not seem that analyzed GFP groups contribute to the detected micronutrient deficits in celiac children's diet. In fact, cereals are not considered as natural sources for these micronutrients. Thus, this scenario reinforces the idea of promoting healthy GFD trough general recommendations such as greater consumption of vegetables and fruits.

In the case of folic acid, we observed a lower content of this vitamin in GFP than in their gluten containing equivalents. In this regard, in Canada and EEUU (205, 206) the fortification of wheat flour with folic acid is mandatory, but not for other alternative flours, as the ones used in GFP. Taking into account the observed folate deficiency in GFD, this fortification in GFP or ingredients could be of interest for celiac children. Folate fortification measure could be extended also to biotin, which wide diet-deficiency in celiac population was alarming. In fact, some of the analyzed GF cereals were supplemented with this vitamin.

Table 10: Review of the nutritional composition of GFP vs GCP (Adapted from Melini *et al*.)

<table>
<tr><th>Reference</th><th>Country</th><th>n</th><th>Food category</th><th>Nutritional profile of GFP</th></tr>
<tr><td rowspan="7">Larretxi et al. (4th manuscript)</td><td rowspan="7">Basque Country</td><td rowspan="7">55 GFP/ 55 GCP</td><td>Bread</td><td>↑ Fiber and RS</td></tr>
<tr><td>Bakery products and cakes</td><td>=</td></tr>
<tr><td>Pastries</td><td>=</td></tr>
<tr><td>Breakfast cereals and infant cereal formula</td><td>=</td></tr>
<tr><td>Pasta</td><td>=</td></tr>
<tr><td>Flours and doughs</td><td>=</td></tr>
<tr><td colspan="2">Regarding fiber and RS contain of the GFP, differences have been found between label and analytical data</td></tr>
<tr><td rowspan="3">Larretxi et al. (5th manuscript)</td><td rowspan="3">Basque Country</td><td rowspan="3">37 GFP/45 GCP</td><td>Bread</td><td>↑ lipids, vitamin E
↓ protein, iron, piridoxin, riboflavin and thiamin and folate</td></tr>
<tr><td>Cereals</td><td>↑ vitamin E, biotin
↓ iron, piridoxin, riboflavin, thiamin, B5, and bitamin E</td></tr>
<tr><td>Pasta</td><td>↑ CH
↓ iron, piridoxin, riboflavin, thiamin, manganese and biotin</td></tr>
</table>

Reference	Country	n	Food category	Nutritional profile of GFP
Miranda *et al.* (2014)(115)	Basque Country	206 GFP/ 289 GCP	Bread	↑ Total fat, saturated fat and salt ↓ protein and fiber
			Pasta	↑ Total fat, saturated fat and salt ↓ protein, fiber
			Bakery products	↑ Na and cholesterol ↓ protein and CH
			Differences between brands	
Missbach *et al.* (2015) (153)	Germany	63 GFP/ 126 GCP	Flours, bread and bakery products, pasta and derivatives, cereals, cookies and snacks	= energy, CH, total fat, saturated fat, fiber and sugar ↓ protein, Na and K
Kulai *et al.* (2014) (147)	Canada	71 GFP/ 60 GCP	Bread	= energy and saturated fat ↑ Total fat ↓ protein
			Pasta	= energy ↑ CH ↓ fiber, Fe, folate
			Breakfast cereals and cakes	=
Mazzeo *et al.* (2015) (197)	Italy	60 GFP	Sweets	↑ Total fat and sugar
			Brioche	↑ salt
			Bread, pasta, flour, snacks	↑ CH and sugar Fiber source (>3 g/100 g)

Reference	**Country**	**n**	**Food category**	**Nutritional profile of GFP**
Wu *et al*. (2015) (198)			Pasta and breakfast cereals	= energy, saturated fat, Na and sugar ↓ protein
			Bread	= energy, saturated fat, Na and sugar ↑ fiber ↓ protein
			Cereal bars, cakes and cookies	↑ sugar, saturated fat and salt
Cornicelli *et al*. (2018) (199)	Italy	235 GFP/ 349 GCP	Cookies	= energy ↑ CH, saturated fat ↓ protein, fiber and salt
			Pasta	= ↑ energy, CH, saturated fat and salt ↓protein and fiber
			Bread	= ↑ CH and fiber ↓energy, protein (specially)
			Toast bread	= energy ↑ CH and salt ↓protein (specially)

Reference	**Country**	**n**	**Food category**	**Nutritional profile of GFP**
Fry *et al*. (2018) (208)	UK	679 GFP/ 1045 GCP	Cookies	↑ salt and sugar ↓protein
			Crackers	↑ salt and sugar ↓protein
			Bread (white/brown)	↑ fiber, total fat, saturated fat (white) and sugar ↓protein
			Pasta (white/brown)	
			Flours (white/brown)	↑ sugar and salt (brown) ↓protein
			Breakfast cereals	↑ salt ↓protein and fiber
			Pizza dough	↑ sugar ↓protein
Calvo-Lerma *et al*. (2019) (146)	Spain	621 GFP/ 600 GCP	Bread	↑ Total fat and saturated fat ↓protein (specially)
			Pasta	↓ sugar and protein
			Cookies	↑ Total fat ↓protein

Notes: GFP: gluten-free products, GCP: gluten containing products.

This thesis has developed a high quality nutritional information on GFPs, which is still scarce in the bibliography, and even in food labels, databases and tools used for the design and evaluation of GFD. It is noteworthy, therefore, that it is not only the evaluation of the nutritional composition of the GFPs, but also its dietary importance. Even so, the bioavailability of GFP, which is a mtter of concern (209), is not considered in this work. Another weakness of this work was not to have the intestinal biopsy not at the time of the diagnosis neither during the follow-up of the GFD. Therefore, there is no evidence to confirm an alleviation of the histological damage that can assure an optimal nutrient absorption. Nevertheless, the objective of this research was to analyze the nutrient deficiencies that GFD could cause to celiac people themselves, whether or not their histologic damage persists.

In conclusion, all these results revealed both that the diet of celiac people is not balanced as well as that an excessive consumption of GFPs increases some nutritional imbalances. However, it cannot be denied that, though the food industry still has a great deal of work to improve, the nutritional quality of GFPs it is progressing well. Indeed, as we havedescribed, the content of fiber in some GFPs has enhanced in recent years leading to the same or higher amount that their gluten counterparts. Besides, this research work also proposes to improve the international standards for the food fortification with some micronutrients for celiac children and adolescences

Finally, nutritional education among this population is essential to continue learning how to reach a healthy GFD. In this way, voluntary or unvoluntary mistakes can be detected (dietary transgressions) and corrected through specific dietary guidelines for celiac people.

NUTRIZIO HEZKUNTZA

NUTRIZIO HEZKUNTZA

Tesi honen zeharkako helburua lortutako emaitza guztiak gizarteari helaraztea izan da. Zehazki, pertsona zeliakoei eta zeliakiaren inguruko osasun profesional guztiei, kolektibo honen bizi-kalitatea hobetzeko asmoz.

Tesi honetan erakutsi den moduan, GGD jarraitzeak zailtasun erantsia dakar dieta orekatu baten gomendioak betetzerakoan, zeliakoek glutendun zerealak dietatik baztertu behar baitituzte. Zerealen elikagai taldeak gure patroi dietetikoaren oinarria izanik, hauek deseztatzeak desoreka nutrizionalak eta mikronutrieen gabeziak jasateko arriskua areagotzen du (210). Balizko gabeziak garaiz identifikatzea garrantzizkoa da, zeliakia estuki lotuta baitago dietarekin erlazionatutako beste zenbait patologiekin, hala nola, gaixotasun kardiobaskular, anemia edo/eta osteoporosiarekin (70-72). Hori dela eta, ezinbestekoa da zeliakoek GGD zorrotza jarraitzea sintomak ezabatzeko eta dieta honen barnean onar daitezkeen elikagaien nutrizio-osaera sakon ezagutzea hauen aukera egokia egiteko eta GGD zorrotza ez jarraitzeak dakartzan arriskuak sahiestea.

Zentzu horretan, gure taldeak 2014 urtean argitaratutako artikulu batean (115) agerian jarri zuen GGPen eta bere glutendun homologoen artean dauden desberdintasun nutrizionalak. Halaber, tesi honetan behatu den moduan, zeliakoen makronutriente ingesta eta elikagai taldeen kontsumo-banaketa ezberdina da populazio orokorrarekin aldaratzen denean. Zeliakoek karbohidrato konplexu gutxiago eta gantz gehiago kontsumitzen dituzte eta zailtasunak dituzte aukera dietetiko egokiena hautatzeko desoreka ekiditeko, glutendun osagaiak oso hedatuta baitaude elikaduran.Gutxi balitz, GGPen etiketek ez dute mikronutriente edukia jasotzen eta horrek, hortaz, iturri dietetiko horietatik kontsumitutako mikronutriente kopurua estimazio zuzena eragozten du. Bestalde, elikagai industrian glutenik gabeko zerealak ez dira mikronutrienteekin gotortzen, glutenarekin kutsatzeko arriskutik babesteko.Kontuan hartuta populazio orokorrean urriak ohi diren mikronutrienteetan gotortzen direla, erraz nabaritu daiteke zeliakoek gabezi-gaixotasunak pairatzeko arriskua handiagotuta dutela. Aurreko guztia dela eta, kolektibo zeliakoak behar dituen jarraibide dietetikoak bereziatuak izan behar dira.

Horiek horrela, tesi honetan aurkitutako zailtasunetako bat zeliakoen dietak egoki ebaluatzea izan zen. Izan ere, hauxe zen hain zuzen ere momenturaino dietista-nutrizionistek GGD diseinatu edo/eta ebaluatu nahi zuten bakoitzean aurkitzen zuten traba. Berez, orduraino existitzen ziren programa informatikoak ez ziren egokiak GGD ebaluatzeko, beren datu-baseetan ez zegoelako GGPen informazio nutrizionalik. Horrek zeliakoen dietaren nutrizio-edukia oker estimatzea zekarren, glutendun homologoetan oinarrituta egin ohi baita. Muga hori gainditzeko asmoz, hainbat GGPren nutrizio-osaera biltzen zuen datu-basea sortu genuen, hasiera batean “Alimentación y Salud” software komertizalean erantsi genuena (AyS, Software, Tandem Innova, Inc., Huesca, Espainia). Horretarako, lehenik eta behin industriak gehien ekoizten dituen GGPen laginketa egin zen, 20 merkataritza-marka desberdinen 700 GGP baino gehiago bilduz. Horien artean okindegi produktuak, pasta, haurrentzako hauts-zerealak, animalia jatorrizko produktu ekoiztuak, *snack*ak, e.a. aukeratu ziren. Geroago, etiketan agertutako energia eta makronutrienteen inguruko nutrizio-informazioa bildu zen eta, etiketa nahikoa izan ez zenean, ekoizleei zuzenean eskatu zitzaien epostaren bitartez. Mikronutrienteen edukiari dagokionez, produktuon etiketetan halakorik adierazten ez denez, Missbach *et al*.-ek (153) sortutako GGPen mikronutriente-osaera biltzen duen tauletan oinarritu ginen gure ikerketan parte hartzen zuten zeliakoen dieta era zuzenago eta zehatzago batean aztertu ahal izateko. Ostean, datu-baseko produktuen langiketa berria egin zen, gehien kontsumitzen diren GGPak aukeratuz: ogia, pasta eta zerealak, alegia. Laborategi-analisiak burutu ziren mikronutriente-edukiaren taula propioa sortzeko.

Lan hori guztia erraldoia da eta GGDrekin ondo lan egiteko beharrezkoa bada ere, orduraino ez zegoen merkatuan horretarako tresnarik. Hortaz, bildutako datu guztiekin *"Gluten -free diet"* softwarea garatzea erabaki genuen; GGD espezifikoak diseinatzeko eta ebaluatzeko erreminta berezia, irekia eta kolektibo zeliakoaren behar batzuk asetzeko helburua duena. Tresna berriak dieta espezifikoak eta orekatuak aztertu eta diseinatzeko balio du, pauta dietetiko zehatzak ematea erraztuz. Aplikazioak funtzio bikoitza dauka: alde batetik, informatzailea da, dietaren nutrizio-osaera zehazten baitu, eta, bestetik, hezitzailea, erabiltzaileak dieta orekatuari buruz ikasteko aukera baitu (adibidez, ebaluatutakoa kolore desberdinez adierazten dira ondo eta gaziki doitutako nutrienteak). Softwarearen jasotzaileak hainbat izan daitezke: kolektibo zeliakoa eta inguruko banakoak, esan bezala, osasun arloko profesionalak, elikagaien industriako kideak edo/eta ikerlariak, besteak beste.

***"Gluten-free diet"* softwarea garatzeko prozesua:**

1. Software berriaren datu basearen garapena: GGPen eta elikagai orokorren nutrizio-osaera. Guk sortutako GGPen nutrizio-osaera biltzen zuen datu basea gainontzeko elikagaien informazioarekin osatu zen. Hori lortzeko,

eskaera egin ostean, ACOESANek (Agencia Española de Consumo, Seguridad Alimentaria y Nutrición) Espainiako Elikagaien Konposizio Datu-Basea (BEDCA) erabiltzeko baimena eman zigun gure ikerketa taldeari. . Ondoren, datu-basea egokitu, GGPenarekin batu software berrian txertatzeko prest utzi zen. Datu-base anitz hau da software honen funtsa.

2. Software honek dietak disenatu edo/eta ebaluatzea helburu du. Horretarako, energi kopurua, makronutrienteen banaketa aztertu eta mikronutrienteen ahorakin maila neurtzen du. Horretaz gain, kontsumitutako elikagai taldeak zehazten ditu.

3. Softwarearen erabiltzaileak:

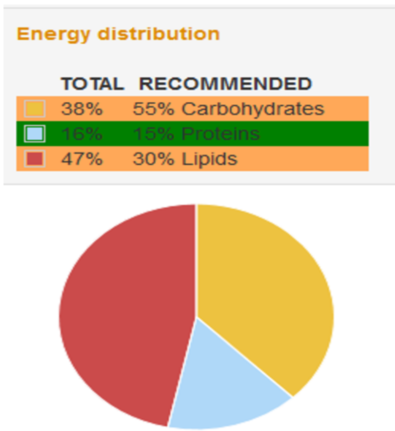
Softwareak bi erabitzaile profil bereizten ditu; norbanakoa (kolektibo zeliakoa) eta osasun arloko profesionala (8. Irudia)

8. irudia. Eratutako software berriaren erabiltzaileen aukerak

3.1 Osasun-profesionala: pazienteguztien historia klinikoak gorde ditzake. Pertsona zeliako bakoitzari bisitan hartutako datu antropometrikoak, datu analitikoak, sintomen galdetegia eta galdetegi dietetikoak (24 orduko galdetegia, elikagaien kontsumo-maiztasunari buruzko galdetegia eta GGDrekiko atxikimendu galdetegia) edukiko ditu bere atalean.

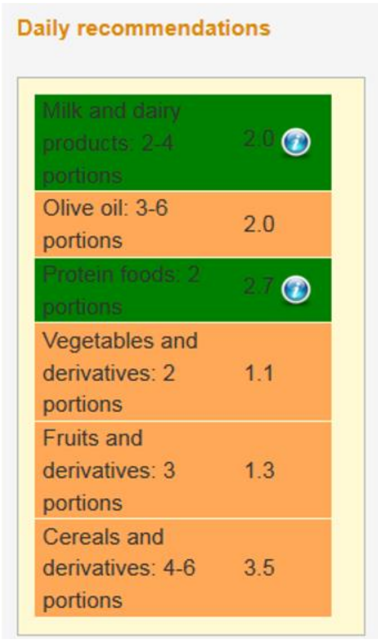
3.2 Pertsona zeliakoa: aurreko profilaren berdintsua da baina kasu honetan erabiltzaileak bere datuak sartzeko eta ebaluatzeko aukera bakarrik izango du. Erabiltzaile honen ezaugarria da hezi beharra duela, hortaz, beregan pentsatzen erramintak argia eta hezigarria izan behar du.

Hortaz,“Gluten-free diet” softwareak, dieta diseinatu eta ebaluatzeaz gain, nutrizio-hezkuntza eskaintzen du. Horrela, erabiltzaileak 24 orduko galdetegia betetzen duen bitartean pantailaren eskuinaldean informazio nutrizionala agertuko zaio. Energiaren banaketa “gazta” itxurako grafika batean adierazten da, batera dieta orekatua jarraitzeko FESNADeko gomedioak islatuz (154). Halaber, nutriente edo nutriente-talde baten gomendioa betetzen denean berde kolorez behatuko da, betetzen ez denean, berriz, gorriz edo laranjaz, segun eta betekinzun maila. Horrela erabiltzaileak bere dieta orekatua den ala ez erraz jakingodu.



9. irudia. GGDren makronutrienteen banaketaren adibide bat

Horren azpian bi taula azaltzen dira, batak dietaren energia, makronutriente eta mikronutriente edukia aurkeztuz eta besteak elikagai taldeen kontsumoa. Azken horretan zeliakoak kontsumitzen dituen elikagai-anoa kopurua SENCek emandako gomendioekin alderatuko da (154). Berriz ere, gomendioa betetzekotan berdez ikusiko da eta, egingo ez balu, ordea, laranjaz edo gorriz. Halaber, esnekietan eta proteinadun elikagai-taldeetan agertzen den ikurra klikatuz gero, likagai-talde bereko aukera baliokide desberdinak dituen taulak azaltzendira.



10. irudia. Elikagai taldeen gomendioen betetze maila

4. Softwarea balioztatu

Programaren fidagarritasuna, errepikakortasuna eta zehaztasuna konprobatzeko hainbat dieta ebaluatu ziren, bai horretarako bereziki diseinatutako dieta berriak zein gure ikerketetako 24 orduko galdetegietatik jasotako GGDk. Hasteko, ezaugarrik ezberdinak zituzten dietak (orekatuak, hiperkalorikoak, hiperproteikoak, hiperlipidikoak, etab.) sortu ziren eta eskuz baloratu ziren. Horrela, alde batetik software berriak kalkuluak ondo egiten dituen ala ez aztertu daiteke eta, bestetik, akatsak egotekotan, konkretuki zein nutrienteen kalkuluetan agertzen den akatsa jakingo dugu. Hortaz, programak egoki funtzionatzen duela iradoki daiteke. Halaber, emaitzak Alimentación y Salud programaren bidez lortutakoekin aldaratu ziren eta ez zen ezberdintasun esanguratsurik aurkitu. Ondoren, zeliakoen 70 galdetegi dietetiko bildu (n=35 heldu eta n=35 ume) eta elikagaien kontsumoa programa berriarekin aztertu zen . Horren ostean, dieta horiek bikoiztu ziren glutendun dietak sortzeko. Horretarako, hauetan agertzen ziren GGPak glutendun produktu homologoengatik ordezkatu ziren eta dieta horiek guztiek (glutendunak eta glutengabekoak) esandako programa informatiko komertzialaren ere analisatu ziren hurrengo aldaraketak eginez:

11. taula. Balidazioan egindako konparaketa motak

KONPARAKETA	**HELBURUA**
GGD (“Gluten Free Diet”) vs GD (“Gluten Free Diet”)	Bi dieten arteko desberdintasunak behatu
GGD (“Gluten Free Diet”) vs GGD (beste programa bat)	Software berria kalkuluak ondo egiten dituela berrestu
GGD (“Gluten Free Diet”) vs GD (beste programa bat)	Merkatuan dauden programak baliagarriak ez direla zeliakoen dietak ebaluatzeko.

Notak: GGD: gluten gabeko dieta, GD: glutendun dieta

Emaitzek erakutsi dute dieta berdina programa desberdinekin aztertutakoan nabari aldatzen direla energia eta nutriente ekarpenak, agerian usten duena GGD berariazko programa batekin ebaluatzearen garrantzia. Horren isla da, egun, zenbait ospitaletan eta zeliakoen elkarteetan “Gluten-free diet” softwarea erabiltzen ari direla zeliakoen dietak aztertzeko.

Softwarea doako zerbitzua da eta UPV/EHUko Glutena Aztertzeko Laborategiaren web orrialdean kokatuta dago, baita hurrengo estekan ere: *http://www.ehu.eus/dieta-singluten/*

Sortutako GGPen nutrizio-osaera biltzen duen datu-baseak eta “Gluten-free diet” softwareak ahalbidetu dute GGDen ebaluazio zuzena egitea. Tesi honen emaitzak populazioari transmititzeko nahiaz, lana erabilgarri izan zedin eta, ondorioz, baliteke, pertsona zelikoen osasuna eta bizi-kalitatea hobetzeko, “Zeliakoarentzako elikadura orekatuaren gida” sortu eta argitaratu genuen, EZErekin lankidetzan (211). Gida horretan zeliakoek egiten dituzten ohiko akatsen adibideak adierazi eta horiek ekidin edo zuzentzeko dieta eredu ezberdinak eskaintzen dira.

12.taula. Dieten ebaluazioak egin ondoren aurkitutako zeliakoen elikadura patroiaren ezaugarriak eta probetxugarri baliabideak egoera hobetzeko.

Ikerketaren bidez ikusitakoa	**Zeliakoari helarazitako informazioa**	**Baliabidea**
1. Dieta desorekatua makronutrienteei eta zuntzari dagokionez: proteina ↑, KH ↓, zuntz ↓, gantza ↑↑, gantz-asea ↑↑. 2. Mikronutriente batzuen gabeziak: D, E eta A bitaminak, Azido folikoa, Fe, Ca, I eta Se 3. Elikagai-taldeen kontsumo gomendioen haustea.	Zer da dieta orekatua? Zer dira nutrienteak? Non daude? Zer elikagai talde daude? Nola lortu dieta orekatu bat? Zer elikagai sartu dieta bakoitzean? Egunerokorako aholku erabilgarriak	Gida Dietak diseinatzeko eta ebaluatzeko softwarea Aurrez-aurreko tailerrak

"Gluten-free diet" erremintaren garapena eta zeliakoen "Zeliakoarentzako elikadura orekatuaren gida" tesi honen transferentzia-ekarpen garrantzitsuenak izan dira. Izan ere, tesi honen eskuizkribuetan agertzen diren emaitzei esker garatu baitira. Lehen esan bezala, baliabideok oso onuragarriak dira, bai zeliakoentzat, bere osasunean eta bizi kalitatean eragina daukatelako, bai gizarte orokorrarentzat ere, GZ eta GGD kontzeptuak zabaltzen baititu. Erantzunkizun sozialen arloan, horiek ezagutzeak besteekiko enpatia eta zeliakoen inklusioa errazten baitu

GLUTEN GABEKO DIETAREKIKO ATXIKIMENDUA

ADHERENCE TO THE GLUTEN-FREE DIET

GGDrekiko atxikimendua

Askotan aipatu den moduan, GZa tratatzeko eraginkorra den tratamendu bakarra bizitza osorako GGD da. Izan ere, dietarekiko atxikimendu zorrotzak zeliakiaren antigorputzen normalizazioa, heste meharreko mukosaren osatzea eta sintomen arintzea dakar (12, 98, 212, 213). Ildo horretan, tratamenduaren konplimendu egokiak zeliakiarekin erlazionatzen diren zenbait gaixotasunen garapena ekiditen du (124, 213). Halere, maiz zeliakoentzat tratamendua ondo jarraitzea zaila suertatzen da, dieta hau betetzeko zailtasun sozialengatik, GGPen ezaugarri organoleptiko eskasagoengatik, kostu ekonomiko altuagatik, eskuragarritasun mugatuagatik edo/eta desegokia den etiketatuagatik (214). Horretaz gain, GGDrekiko atxikimendu eskasa beste faktore batzuekin erlazionatzen da: adina diagnostikoaren momentuan (esate baterako, nerabezaroan oso ohikoa da GGD urratzea), gaixotasunaren inguruko ezagutza eskasa eta faktore psikologikoak besteak beste (90, 124, 215).

Tesi honen helburu nagusia EAEko zeliakoen dieta nutrizionalki aztertzea izan da, bai kualitatiboki eta baita kuantitatiboki ere. Aitzitik, ez da neurtu parte-hartzaileen dietarekiko atxikimendu zehatza, ezta GZarekin erlazionatutako antigorputzen odol-mailak neurtu ere. Horixe lanaren muga bat izan da. Izan ere, egungo Europar jarraibideek tTG antigorputzen (Ac) neurketa gomendatzen dute GGDren arrakasta monitorizatzeko eta tratamendua jarraitzen den bitartean normalizazio serologikoa lortzen dela bermatzeko (11, 42). Rubio-Tapia *et al.*-ek argitaratu zuten moduan, urtebetez GGD jarraitu ondoren serologia proba positiboa izateak transgresio dietetikoak jazo direla iradokitzen du (42). Heste mukosaren egoerari dagokionez, zeliako helduen %65ak osatze histologikoa lortzen du bi urtez GGD zorrotza jarraituta (75-77) Umeen kasuan azkarrago gertatzen da (77), bigarren urterako %88-96 osatzen da (77, 78) eta 5 urteetan %100 (77). Gure lanaren bigarren muga da pazienteen sintomei buruzko informazioa bildu ez izana, ez diagnostikoan ezta ikerlana aurrera zihoanean ere.

Horiek horrela, tesi honetan GZaren ikuspegi anitzagoa lantzeko interesgarria iruditu zitzaigun Tampereko *The Celiac Disease Research Center*en (*CeliRes*en) egonaldia egitea, Katri Kaukinen Doktorearen zuzendaritzapean. CeliRes gunean disziplina anitzeko ikerketak burutzen dira, betiere glutenak eragindako gaixotasunen inguruan, azterketa epidemiologiko eta klinikotik ikerketa genetiko, immunobiologiko edo/eta mikrobiologikora hedatuz.

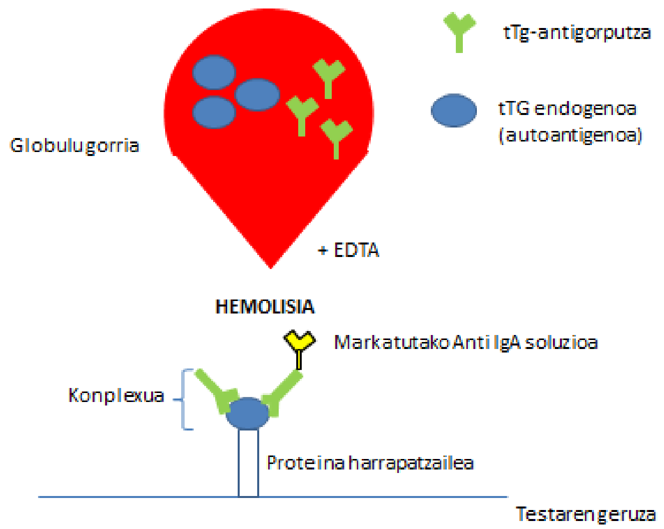
CeliResen egindako lanaren helburua honakoa izan zen: Finlandiako zeliako helduen kohorte baten ohitura dietetikoak eta horiekin erlazionatzen diren ezaugarri klinikoak aztertzea. Azken hori tesia osatzeko asmoz garatu zenez, ohitura dietetikoei dagokienez GGDrekiko atxikimendua eta dietaren segurtasuna arriskuan jarri dezaketen zerealen kontsumoa izan ziren aztergai, oloa eta gari-almidoia. Ezaugarri klinikoak antigorputz neurrien eta sintomen arabera aztertu ziren.

Parte-hartzaileak eta prozedura:

Tampereko Unibertsitate Ospitaleak eta Unibertsitateak elkarlanean burututako ikerketa bateko kohortea aztertu zen. Kohorteak 864 partehartzaile zituen, %75 emakumeak eta %25 gizonezkoak, eta adina, batez beste, 53 urtekoa da. Bolondresak egunkarian jarritako albisteen bitartez edo Finlandiako Zeliakoen Elkartearen laguntzaz errekrutatu ziren.

Ikerketa honetan biopsia probaz diagnostikatutako eta adin nagusiak ziren zeliakoak hautatu ziren, Tampere ospitaleko ikerlanean engaiatu ziren pazienteen serie prospektiboetatik aukeratuta.

Lehenengo, parte-hartzaileei analisi serologikoak egin zitzaizkien. Alde batetik, odol-proba tradizionalen bitartez eta ELISA metodoa erabiliz, EMA eta tTG antigorputz mailak neurtu zitzaizkien. Positibotzat hartu ziren EMA antigorputza 1 ≥ 5 izatea eta tTG-2- antigorputza ≥ 30 izatea. Bestetik, POCT testaren *(Point-of-Care test)* bidez GZaren kasu positiboak diagnostikatu ziren. POCT testa atzamarretik erauzitako odol tanta batean tTG2 antigorputzen neurketan datza, erreakzio immunologikoek gidatuta. Test azkarra eta erraza da, 5 minututan egin baitaiteke, medikuaren edo nutrizionistaren kontsultan nahiz etxean egingarria. Orain arte probatu diren POCT testak diagnostiko momentuan antigorputzak detektatzeko egokiak izan dira (216-219).



11. irudia. POCT testaren mekanismoa

Halaber, parte-hartzaileen hurrengo datu demografiko eta klinikoak bildu ziren: sexua, adina (elkarrizketa egin zitzaien egunean eta diagnostikoan), diagnostikoa non egin zitzaien, aurkezpen klinikoa diagnostikoan, sintoma motak, sintomen iraupena edo/eta larritasuna (bai diagnostikoa egin aurretik baita GGD jarraitu ostean ere). Azkenik, zeliakoen txosten kliniko osoak birpasatu ziren diagnostiko momentuko laborategiko proba serologikoen emaitzak edukitzeko.

Dietarekiko atxikimendua aztertzeko, lehenik parte-hartzaileei diagnostikoan jasotako aholku dietetikoaren inguruan galdetu zitzaien. Ondoren, transgresio dietetikoei buruz zuzenean itaundu zitzaien eta hurrengo sailkapena egin zen zeliakoen erantzunen arabera: dieta zorrotza (urtean lapsus gutxi batzuk), noizean behingo transgresioak (hilabetean behin) eta glutena hartzen dutenak (hilabetean maiz). Amaitzeko, GGD maneiatzeko gaitasuna eta olo eta almidoi kontsumoari buruzko informazioa bildu zen.

Aipagarri da errekrutamendua eta datu bilketa Pirkanmaa barrutiko Ospitaleko Etika Batzordearen jarraibideak errespetatuz egin zirela eta parte-hartzaile guztiek idatzizko baimen informatua eman zutela.

<u>Analisi estatistikoa:</u>

Analisi estatistikoa IBMren SPSS programa estatistikoaren 23. bertsioa erabiliz burutu zen (IBM Inc., Armonk, NY, USA). P< 0,05 balioak onartu ziren esanguratsu bezala. Emaitzak adierazteko aldagai binomiko eta kategorikoak parte-hartzaile zenbakiaren (n) eta ehunekoen bidez aurkezten dira tauletan. Chi-karratu testa erabiliz aztertu izan dira.

<u>Emaitzak eta eztabaida:</u>

Ikerketaren datuak bildu ostean parte-hartzaileak hiru kohortetan sailkatu ziren dietarekiko atxikimenduaren arabera: 1) dieta zorrotza jarraitzen zutenak, 2) dieta-lapsusak noizean behin zituztenak eta 3) glutendun dieta normala hartzen zutenak (lapsus dietetikoak maiz egiten zituztenak).

13. taula. GGD jarraitzen duten heldu zeliakoen historia dietetikoaren eta datu biokimikoen hainbat emaitza.

	Dieta zorrotza (n=823)		**Lapsusak noizean behin (n=22)**		**Glutendun dieta (n=4)**		**P**
	n	**%**	**n**	**%**	**n**	**%**	
Aholku dietetikoa							*0,068*
Ez	164	21	1	5	2	50	
Bai	620	79	20	95	2	50	
GGD jarraitzeko gaitasuna	772	96	16	84	2	67	***0,002***
GGD							*0,228*
< 2 urte	97	12	5	23	0	0	
≥ 2 urte	724	88	7	77	4	100	
Oloa kontsumitzen du	670	82	18	82	4	100	*0,648*
Almidoia kontsumitzen du	670	82	20	91	2	50	*0,136*
EMA-ag positiboa	82	10	9	41	1	25	***<0.001***
tTG2-ag positiboa	221	27	11	50	1	25	***0,006***
POCT positiboa	90	11	4	18	2	50	***0,028***
Egungo sintomak							***0,004***
Batere	620	77	10	45	2	67	
Arinak	177	22	10	45	1	33	
Larriak	12	2	2	9	0	0	

Notak: GGD: gluten gabeko dieta; POCT: *point-of-care test*¸ EMA-ag: Antigorputz Endomisiala; tTG2-ag transglutaminasa 2 antigorputza

Azpimarratzekoa da Finlandiako zeliako gehienek (%97k) GGD zorrotza daramatela eta gutxi batzuk (%3k) bakarrik izaten dituztela lapsusak. Datu hori bat dator 2015an See *et al*-ek (214) aipatutakoarekin lurralde desberdinen GGDrekiko atxikimenduaren *review*an. Horretan ikusi zuten Finlandia, Suedia eta EEBB atxikimendu zorrotza altuena zuten lurraldeak zirela (%90tik gora). Beste lurralde batzuetan, berriz, Frantzia edo Italian esaterako, dietarekiko atxikimendua esanguratsuki jeisten da, %42-62 bitartean egonik. Aztertutako kohortearen atxikimendu ezin hobea diagnostikoaren momentuan aholku dietetikoa jaso zutelako azal daiteke. Lapsus dietetikoak dituztenen artean diagnostikoaren unean aholku dietetikoa eman zen, aitzitik, portzentaje handiago batek bi urte baino gutxiago darama diagnostikatua; beraz, baliteke dieta oraindik guztiz kontrolatuta ez edukitzea. Glutena sarritan kontsumitzen zutenen artean erdiek bakarrik jaso zuten aholku dietetikoa osasun langileen eskutik diagnostikoaren momentuan. Halere, kohorte honek pairatzen zituen sintomen datuak aztertzerakoan gehiengoak sintomarik ez edo sintoma arinak zituela behatu zen

Izan ere, bibliografian aipatzen da dietistarekin elkarrizketak egitea pazienteak GGDri ondo atxikitzeko prozedura-gakoa dela, *gold standarra* esan zitekeen (74). Gainera, dietarekiko atxikimendua neurtzeko galdetegi desberdinak egon arren, hauek ez dute nahi gabeko gluten ahorakina erakusten, hortaz eskarmentua duen dietistaren aholkua ezinbestekoa da (220, 221). Umeekin egindako beste ikerketa batean (222) honakoa behatu zuten: aldizka jarraipena jasotzen zuten umeek atxikimendu egokia lortzeko aukera bikoizten zutela, jarraipenik ez zutenekin (edo ia ez zutenekin) aldaratuta.

Halaber, kohorteak jasotako aholku dietetikoa xehetasunez aztertzean (14. taula) frogatu zen kasu gehienetan dietisten eskutik etorri zela.

14. taula. Jasotako aholku dietetikoaren jatorria

	Dieta zorrotza (n=823)		**Lapsusak noizean behin (n=22)**		**Glutendun dieta (n=4)**	
Aholkua nork	***n***	***%***	***n***	***%***	***n***	***%***
Ez	164	21	1	5	2	50
Dietistak	527	67	18	86	2	50
Medikuak	46	6	0	0	0	0
Erizainak	17	2	1	5	0	0
CDA-k	21	3	0	0	0	0

Notak: CDA: *Celiac Disease Association*

Halere, Suedian Johansson eta al.-ek (223) egindako ikerketan GGDrekiko atxikimenduan aholku dietetikoa guztiz beharrezkoa zela behatu bazen ere, dietistak edo medikuak emateak ez zuen alderik.

Posiblea litzateke zeliakoek aitortutako atxikimendua erreala ez izatea; izan ere, lapsus dietetiko gehienak nahi gabekoak ohi dira (214). Aitzitik, Finlandiako zeliakoetan jasotako atxikimenduaren datuak antigorputzen datuekin berresten ziren, See *et al*.-en reviewak islatzen duen moduan. Horrela, atxikimenduaren arabera sailkatutako taldeak erkatzean EMA eta tTG antigorputzetako ezberdintasun esanguratsuak zeuden, positibotasuna askoz altuagoa izanda lapsusak dituztenen artean. POCT testean berdina gertatu zen , desberdintasuna hain nabaria ez bazen ere.

Ulergarria da GGD era egokian jarraitzeko gai sentitzen ote diren galderari erantzutean desberdintasun esanguratsuak aurkitzea, balio askoz handiagoak behatuz dieta zorrotza jarraitzen zutenen artean, lapsusak egiten zutenekin konparatuz.

Bestalde, azpimarratu behar da parte-hartzaile gehienek oloa eta gari-almidoia kontsumitzen zutela GGDren barne. Finlandian GGDn oloa hartzea baimenduta dago (224), segurutzat eta nutrizionalki mesedegarritzat hartzen delako. Alde batetik, oloa zuntza disolbagarriaren iturri garrantzitsua da eta, hortaz, janondoko gluzemia mantenduz intsulinaren beharra jaitsi edota kolesterolaren xurgapena murriztu egiten ditu (225). Bestetik, mineralen eta bitaminen iturri aberatsa ere bada (114). Are gehiago, GGDan oloa sartzea dietarekiko atxikimendua hobetu omen duela deskribatu da (226) Almidoiari dagokionez, eztabaida handia sortu izan da betidanik osagai honek GGDn izan dezakeen segurtasunaren inguruan. Lurralde batzuk, Finlandia barne, gari-almidoian oinarritutako GGPak erabili izan dituzte hainbat hamarkadatan zehar (227). Izan ere, gariaren frakzio proteinaduna (toxikoa dena alegia) almidoitik guztiz banatzen dela uste baita. Hala ere, ikerlari batzuen aburuz karbohidratoak eta proteinak banatzeko teknikak ez daude nahikoa garatuta eta proteinen kutsadura xumea egon liteke almidoian (228).

Esandakoaz aparte, 2009 urtetik aurrera Europar jarraibideak (58, 61) betez gluten gabeko etiketa edukitzeko produktuek 20 mg/kg baino gutxiago izan behar dute. Logikoki, betekizun hori gari-almidoia osagaia bezala erabiltzen duten GGPek ere kunplitu behar dute. Aipagarri da, lege hau indarrean sartu baino lehen Eskandinabian eta Erresuma Batuan 200 mg/kg-rainoko gari-almidoizko produktuak kontsumitzen zituztela zeliakoek, heste meharreko kalte histologikorik somatu gabe edo/eta erikortasuna handitu gabe (227, 229-231). Horretan oinarrituta eta dieta ahal den eta gutxien mugatzeko, Eskandinaviako lurraldeetan gari-almidoian oinarritutako produktuen kontsumoa ohikoa da. Gure lurraldean, egun, almidoia kontsumitzea segurutzat jotzen da, are gehiago, hainbat GGPren osagaia dugu.

Azkenik, ikerketa egin bitartean zeliakoek pairatzen zituzten sintomei dagokienez, GGD zorrotza jarraitzen duten hiru laurdenek baino gehiagok ez zituzten inongo sintomarik eta, berriz, lapsusak zituztenen artean erdiek baino gehiagok sintoma arinak edo larriak sufritzen zituzten. Dena den, aipagarria da glutena bere dietan kontsumitzen zutenen %67k ez zuela sintomarik aitortzen. Beraz, lehen esan bezala, pentsa daiteke hori litekeela GGD ez jarraitzearen kausa, hots, ez zutela ondoeza sentitzen eta horregatik ez ziotela garrantzi nahikorik ematen dietari (232).

Ondorioak:

GZaren diagnostikoarekin batera GGDri buruzko hezkuntza nutrizionala jasotzeak berebiziko garrantzia du; izan ere, dietarekiko atxikimendua bermatzeko bidea baita. Urriegia bada gutxiegizko atxikimenduak GZaren sintomak luzatzea eta balizko beste gaixotasunak agertzeko arriskua areagotzea ekar ditzake. Ondorioz, GZan eskarmentua duten dietistak beharrezkoak dira gaixotasunaren tratamendu eta jarraipenean. Sintomak ere, jakina, atxikimendua modulatzeko oinarrizkoak izango dira.

Ezaguna da POCT testa erabilgarria da GZaren kasu positiboak diagnostikatzeko (216, 219). Halaber, ikerketa honetan transgresio dietetikoak berresteko tresna bezala erabili da. POCTen emaitzak proba serologiko tradizionalen emaitzekin erkatu direnean dietarekiko atxikimendua zaintzeko tresna erabilgarria (erreza, azkarra eta ez inbasiboa) izan daitekeela behatu da; dena den, emaitza gehiago behar dira testaren erabilera hau sustatu aurretik.

Adherence to the Gluten-Free Diet

It is well known that the only effective treatment for celiac disease (CD) is a strict lifelong gluten-free diet (GFD). In fact, strict obedience of a GFD will result in remission in symptomatic, serological and histological aspects (98, 212, 213). In that sense, many disease complications can be avoided by appropriate adherence to this diet (124, 213). However, sometimes celiac patients find it hard to strictly follow the diet due to social limitations such as organoleptic characteristics, high price, limited availability and inappropriate labelling (214). In addition, low adherence to the GFD can be related to other factors such as: age, diagnosis time (teenagers usually do more diet transgressions), scarce knowledge about the disease and psychological factors, among others (90, 124, 215).

The aim of this thesis is to evaluate, both quantitatively and qualitatively, the diet of celiac people from the Basque Country. However, neither adherence of the participants to the GFD nor celiac disease related serum antibodies have been measured, which is one of the limitations of this study. In fact, nowadays European Guidelines recommend the measurement of tTG antibodies to monitor the success of GFD and to guarantee serologic normalization during treatment (11, 42). Rubio-Tapia *et al.* published that the maintenance of positive serologic test despite following a GFD for one year suggests dietary lapses (42). Concerning small-bowel mucosal, 65% of celiac adults achieve a histological recovery after following a strict GFD for two years (75-77). In the case of children this happens more rapidly (77). By the second year, 88-96% (77, 78) of them have the mucosa healed and in five years time all of them (77). Another limitation of this thesis would be not to include information about symptoms of the patients, neither at the time of the diagnosis nor during treatment.

Thus, it was thought that in order to include a more global perspective of CD in this doctoral thesis, it would be interesting to conduct a research stay at The Celiac Disease Research Center in Tampere (CeliRes) under the direction of Dr. Katri Kaukinen. The CeliRes is a multidisciplinary research center that carries out different studies in the field of gluten-related diseases, including epidemiological and clinical studies and genetic, immunobiological and/or microbiological research.

The purpose of the work developed during the visit to The CeliRes was to analyze the dietary habits of a Finnish adult celiac cohort and the clinical characteristics that are related to them. As the study of the dietary habits of a Finnish cohort was developed with the intention of complementing athis doctoral thesis, the consumption of cereals (oats and starch) that could endanger the safety and/or the adherence to the GFD were also analyzed. Clinical characteristics were evaluated according to the antibody measures and symptoms.

Patients and study design:

The nationwide cross-sectional study was conducted in Tampere University Hospital and the University of Tampere. The patient cohort comprising of 864 celiac disease patients, 75% women and 25% men, with median age of 53 years old, were collected from a prospective patient series. The participants were recruited via newspaper advertisements and with the help of local and national celiac disease societies. Inclusion criteria were age ≥ 18 years and biopsy-proven celiac disease diagnosis.

First, IgA class antiendomysialantibodies (EMA) and serum IgA class tissue antitransglutaminase antibodies (tTG2) were measured by enzyme-linked inmmunosorbent assay (ELISA); a dilution 1: ≥5 and a unit value ≥ 30 were considered positive, respectively. In the case of selective IgA deficiency, the corresponding IgG class antibodies were determined. Furthermore, positive CD cases were determined by POCT test *(Point-of-Care test).* The POCT test is based on the measurement of tTG2 antibodies extracted from a whole blood sample. This is a quick and easy test, which can be performed in 5 minutes at the doctor's or nutrition consultant's office or even at home. So far, POCT tests have been tested for detecting antibodies at the time of diagnosis (216-219).

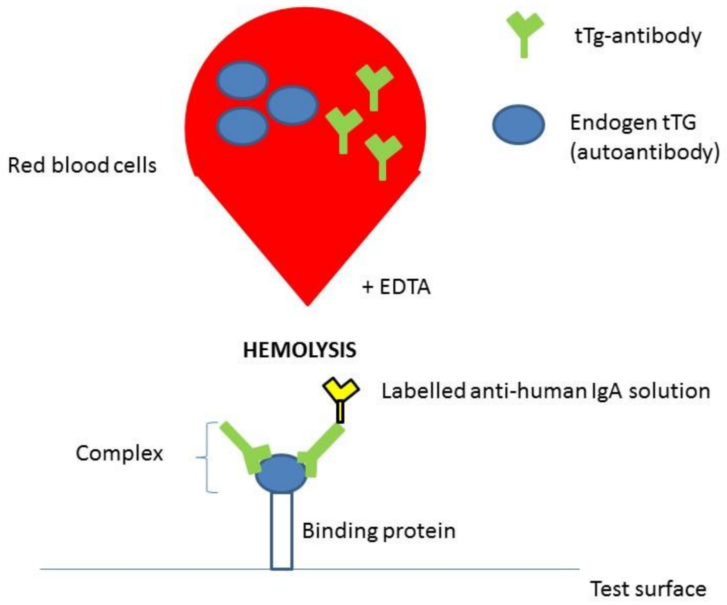


Figure 11. The mechanism of the rapid whole blood test (POCT)

Likewise, the following demographic and clinical data were gathered for all the participants: gender, age (at the interview and diagnosis time), diagnosis place, clinical presentation in the diagnosis, symptoms type, duration and/or severity of symptoms (both before diagnosis and while on a GFD). Finally, the complete hospital reports were reviewed in order to obtain the results of laboratory serological tests at the time of the diagnosis

In order to analyze the adherence to the GFD, first participants were asked about the dietary advice received at the time of diagnosis. After that, a history of occasional or regular consumption of gluten-containing products and oats was assessed by a dietary interview and depending on their answer the participants were classified into three different cohorts: strict GFD (few dietary lapses in a year), occasional lapses (dietary lapses once a month) and normal gluten intake (more than one intake of gluten a month). Finally, the ability to handle the GGD and information about oats and starch consumption were collected.

It is worth mentioning that recruitment and data gathering were carried out in compliance with the protocol approved by the Ethical Committee of the Pirkanmaa Hospital District and that all study subjects gave written informed consent.

Statistical Analysis:

Statistical analysis was made using the Statistical Package for Social Sciences version 23 (IBM Inc., Armonk, NY, USA). A < 0,05 was considered significant in all analyses. Binomial and categorical variables are presented as number of subjects (n) and percentages in tables. Variables were analyzed using Chi-square.

Results and discussion:

After collecting all the data, participants were classified into three cohorts according to their dietary adherence: 1) those who followed strict GFD, 2) those who had occasional dietary lapsus and 3) those who had a regular gluten intake (frequent dietary lapses).

Table 13. Dietetic habits, serological measurements and clinical symptoms of adults celiac patients on a GFD.

	Strict GFD (n=823)		**Ocasional lapses (n=22)**		**Normal gluten intake (n=4)**		**P-value**
	n	**%**	**n**	**%**	**n**	**%**	
Received dietary advice							*0,068*
No	164	21	1	5	2	50	
Yes	620	79	20	95	2	50	
Capable of managing GFD	772	96	16	84	2	67	***0,002***
GFD							*0,228*
< 2 years	97	12	5	23	0	0	
≥ 2 years	724	88	7	77	4	100	
Use of oats	670	82	18	82	4	100	*0,648*
Use of starch	670	82	20	91	2	50	*0,136*
EMA-ab titers positivity	82	10	9	41	1	25	***<0.001***
tTG2-ab positivity	221	27	11	50	1	25	***0,006***
POCT positivity	90	11	4	18	2	50	***0,028***
Current symptoms							***0,004***
None	620	77	10	45	2	67	
Slight	177	22	10	45	1	33	
Serious	12	2	2	9	0	0	

Notes: GFD: gluten-free diet; POCT: *point-of-care test*¸ EMA-ab: anti-endomysial antibodies; tTG2-ab: anti-Transglutaminase-2 antibody

Notably, most Finnish celiac patients (97%) follow a GFD and only a few (3%) have dietary lapses. These data are in good accordance with the review in 2015 by See *et al*. about adherence to GFD across different European countries. In this review, they observed that Finland together with Sweden and USA had the highest adherence level to GFD (over 90%). By contrast, in other countries, such as France or Italy dietary compliance is significantly lower, between 42-62%. The high adherence of the cohort studied can be explained by dietary advice given at the time of diagnosis. Dietary advice was also given in the cohort with dietary lapses. However, a large percentage of people in this group were diagnosed less than two years ago; so that GFD may not be fully controlled yet. Only half of those who consumed gluten normally received dietary advice from healthcare personnel at the time of diagnosis. Nevertheless, when analyzing the data of the symptoms in this cohort we can observed that almost all of them have no or slight symptoms.

Similarly, other authors say that the role of a trained dietitian in celiac disease at the moment of the diagnosis is a key procedure for better adherence to the GFD, which could be called *Gold standard* (74). Additionally, although there are different questionnaires for measuring adherence to the GFD, these do not take into account inadvertent gluten intake so the dietitian's advice is essential (220, 221). Another study carried out in children (222) observed that, when comparing celiac children who had been regularly monitored with those who did not, the children in follow-up had twice the chance of achieving good adhesion to the GFD.

Likewise, in the detailed analysis of the dietary advice received by the cohort (table 14), it was shown that in most cases such advi ce came from the dietician.

Table 14. Dietary advice received

	Strict GFD (n=823)		**Occasional lapses (n=22)**		**Normal gluten intake (n=4)**	
Received dietary advise	***n***	***%***	***n***	***%***	***n***	***%***
No	164	21	1	5	2	50
Dietitian	527	67	18	86	2	50
Doctor	46	6	0	0	0	0
Nurse	17	2	1	5	0	0
CDA	21	3	0	0	0	0

CDA: *Celiac Disease Association*

However, despite the fact that dietary counseling was absolutely necessary for the good adherence to GFD, Johansson *et al*. (223) found that there were no differences between the adherence when the dietitian gave counseling or when the physician did.

Furthermore, it could be possible that the strict adherence to the GFD declared by the participants was not real since most dietary lapses are involuntary (214). Nevertheless, these data were confirmed by measurement of the antibodies. Thus, in the comparison of the groups classified by adherence, there were significant differences in EMA and tTG antibodies, with much higher positivity among the celiac people who had dietary lapses. In POCT test the result was similar, although the difference between the cohorts was less obvious.

It is understandable to find significant differences between the three cohorts when we analyzed the response to the question about the capability of carrying out a GFD. A higher percentage of celiac who are on a strict

GFD feel more confident following the GFD that those ones that have dietary lapses.

Furthermore, it should be emphasized that most of the participants consumed oats and wheat starch in the GFD. In Finland, including oat in the GFD is allowed (224), as it is considered safe and nutritionally beneficial. On the one hand, oats are an important source of soluble fiber and therefore help in maintaining low postprandial glucose which reduces the need for insulin and also cholesterol absorption (225). On the other hand, it is also a rich source of minerals and vitamins (114). Moreover, the inclusion of oats in the GFD has been described as improving adherence to the diet (226). Regarding wheat starch, there has been a great debate about the safety of this component in the GFD. Some countries, including Finland, have used wheat starch in the GFPs for decades (227). In fact, it is believed that the protein fraction of wheat (which is toxic for celiacs) is removed entirely from starch. By contrast, some researchers pointed out that carbohydrates and protein separation techniques were not sufficiently developed and there may be a low level of protein contamination in starch (228).

Notwithstanding, since 2009 the European guidelines (58, 61) have said that a product can be labelled as “gluten-free” when it has less than 20 mg/kg of gluten. Logically, this requirement is also mandatory with GFPs that use wheat starch as an ingredient. It is noteworthy that before this law became effective celiac people from Scandinavia and the United Kingdom consumed wheat products containing up to 200 mg/kg of gluten, and no histological damage in the small-bowel mucosa and/or increased morbidity were detected (227, 229-231). Based on this, and in order to restrict the food included in a GFD as little as possible, Scandinavian countries use wheat starch as a part of their diet. In our country, nowadays, the consumption of starch is considered safe and, what is more, it is a component of some GFPs.

Finally, concerning the symptoms suffered by celiac people during the study, more than three quarters of celiac people following a strict GFD did not have any symptoms, while more than half of those who have dietary lapses suffered from slight or severe symptoms. However, it is important to highlight that 67% of those usually consuming gluten in their diet did not recognize any symptoms. Therefore, as we have already said, it might be thought that this could be the cause of their not adhering to the GFD in that as they did not feel discomfort, they did not attach enough importance to the GFD (232).

Conclusions:

Together with diagnosis, receiving nutritional education about the GFD is essential in order to guarantee its good adherence. If this education is not enough, the symptoms can be prolonged and the risk of other potential illnesses increased. As a result, a trained dietitian is required in the treatment and monitoring of celiac disease. Furthermore, symptoms are necessary for monitoring adherence.

It is well known that the POCT test is useful for the diagnosis of positive CD case (216, 219). Likewise, this study has been used as a tool to confirm dietary transgression. When POCT's results are compared to the results of traditional serological tests, it has been observed that it is a useful (easy, fast and non-invasive) tool for measuring adherence to diet; However, more results are needed before the implementation of this test.

ONDORIOAK

CONCLUSIONS

ONDORIOAK

1. Euskal Autonomi Erkidegoko (EAEko) ume eta nerabe zeliakoen gluten gabeko dieta (GGD) ez da orekatua, MDrekiko atxikimendu txikia erakutsiz. Horren arrazoi nagusiak bi dira: ohitura dietetiko ezegokiak, populazio orokorrean gertatzen denaren antzera, eta GGPen kontsumo altua. Izan ere, azken horiek GGDren zenbait ezaugarri are gehiago desorekatzen baitituzte.

2. Oro har, EAEko emakume zeliakoen antropometria populazio orokorrarena baino hobea bada ere, jarraitzen duten GGD desorekatua da, bai kuantitatiboki eta baita kualitatiboki ere. Hala ere, badirudi egiten dituzten akatsak ez direla gluten gabeko produkturen (GGDren) berezitasun espezifikoen ondorio, populazio orokorraren ohitura ezegokiak errepikatzen baitituzte.

3. EAEko gizon zeliakoen kopuru esanguratsu batek nutrizio-egoera ezegokia aurkezten du. Horren arrazoia da jarraitzen duten GGD ez orekatua, bai kuantitatiboki eta baita kualitatiboki ere. Dietaren akats gehienak populazio orokorrean ere gertatzen dira, gizarte osoaren ohitura ezegokiak arazoen abiapuntu izanik. Gutxi batzuk dagozkio GGDri bakarrik, mikronutrienteei dagozkienak bereziki.

4. Heldu zeliakoen nutrizio-egoerari dagokionez, emakumeena gizonena baino hobea da. Emakume zeliakoek beren dieta zaintzeko ardura handiagoa dute. Horren isla da GGDren nutrizio-osaera bien kasuan

desorekatua izanik, gizonen dieta gomendioetatik gehiago urruntzen dela, lipidoen profilari dagokionez batez ere.

Orokorrean, pertsona zeliakoen dieta ohitura desegokiak eta nutrizio-gabeziak zuzentzeko aholku dietetiko espezifiko eraginkorra ematea gomendatzen da. Horrela, merkatuan berriak diren gluten gabeko zerealak eta pseudozerealak ezagutarazi eta horien kontsumoa sustatu egin behar da. Jakina, aurrekoaz gain barazki, fruta eta lekaleen kontsumoa bermatuko da. Arrakasta lortzeko pertsona hauei jarraipen dietetikoa egitea ezinbestekoa izango da.

5. Itxarondakoaren aurka, GGPen eta glutendun homologoen zuntz eta almidoi iraunkor (AI) edukia antzekoa da, ogietan izan ezik, non are kantitate handiagoak neurtu diren. Hori azken urteetan produktuon nutrizio-ezaugarriak hobetu izanaren ondorioa da. Are gehiago, behatutako konposatu hauei dagozkien dieta desorekak, ez zaizkio produktu hauei sor, bibliografian iradoki izan denaren kontra.

6. Espero zenarekin ados, GGPek mikronutriente gutxiago dituzte haien homologoek baino. Hala ere, orokorrean ez dirudi hori denik GGDren gabezien erantzulea. Alabaina, produktuon osaera hobetze aldera, bai foliko zein biotinaren gotortzea haztatu behar da.

7. EAEko ume eta nerabeek jarraitzen duten GGDren zenbait desoreka hein batean GGPei esleitu ahal zaizkie, gehiegizko lipido-kontsumoa eta horien profil-ezegokia batez ere.

Laburbilduz, elikadura industriak pertsona zeliakoen osasunean duen erantzukizunari ihardetsiz, azken urteotan GGPen kalitate nutrizionala hobetu du hainbat osagaietan, hala nola zuntza, AI eta zenbait mikronutrienteetan. Aitzitik, oraindik GGPen mikronutriente batzuen edukia eta profil lipidiko osasungarriagoa lortzeko lan egin behar da.

Tradizionalki gluten eza ziurtatzea izan da elikadura industriaren kezka. Egun, GGPen osagaiak seguruagoak izanik, industria produktuon nutrizio-kalitateari ere erreparatzen hasi zaio. Zenbait elikagai eta nutrienteetan hobekuntza esanguratsuak lortu diren arren, erronka honi eustea ezinbestekoa da detektatutako nutrizio-beharrak asetzeko. Are gehiago, lan hori guztia gizarteratzen ahalegina egin behar da, elikagaien etiketetako informazioa osatuz adibidez.

8. Finlandiako zeliakoek GGDri atxikimendu ezin hobea aurkeztu diote. Horrela, gaixotasun zeliakoaren sintomak murriztu eta balizko gaixotasun batzuk agertzeko arriskua saihesten dute. Atxikimendua nutrizio-hezkuntza egokiari sor zaio, besteak beste.

Badirudi POCT testa atxikimenduaren jarraipena egiteko tresna egokia izan daitekeela, oraindik bere erabiliera berretsi behar bada ere.

CONCLUSIONS

1. The gluten free diet (GFD) of celiac children and adolescents from the Basque Country is not balanced, adhering only slightly to the Mediterranean Diet. There are two main reasons for this: inadequate dietary habits, similar to those of the general population, and high consumption of gluten free products (GFP). In fact, the nutritional composition of the GFP can make the diet even more unbalanced.

2. In general, although the anthropometry of celiac women in the Basque Country is better than that of the general population, their diet continues to be unbalanced, both quantitatively and qualitatively. However, it seems that the mistakes that they make cannot be largely attributable to the specific GFD, since they repeated the unsatisfactory dietary habits of the general population.

3. A significant number of celiac men in the Basque Country find themselves in an inadequate nutritional status. The reason for this is that GFD is not balanced, both quantitatively and qualitatively. Most of the dietary mistakes detected also happen in the general population, as a result of inadequate dietary habits. Only a few results from following a GFD, especially those related to micronutrients.

4. As regards the nutritional status of the celiac adults, women are healthier than men. Celiac women are more responsible about their diet. This is reflected in the fact that GFD's nutritional composition is inadequate in both

cases, but men's diet is further from the recommendations, especially in the lipid profile.

In general, it is recommendable to provide a specific and effective dietary recommendation for celiac people with inadequate habits and nutritional deficiencies. Along these lines, it is necessary to promote the consumption of new gluten-free cereals and pseudo-cereals on the market. Of course, in addition, the consumption of vegetables, fruit and vegetables should be guaranteed. Dietary monitoring of celiac people is essential in order for success.

5. Contrary to expectations, fiber and resistant starch (RS) content of GFPs and their gluten containing counterparts are similar, except in bread, where even larger quantities have been measured. This has been the consequence of improving the nutritional characteristics of these products in recent years. Furthermore, the dietary imbalances of these compounds are not caused by GFPs, as has been suggested in the bibliography.

6. As expected, GFPs have lower amount of micronutrients than their homologues. However, overall, this fact does not seem to be responsible of GFD’s deficiencies. Nevertheless, in order to improve their nutritional composition, it should be considered the enrichment with folic and biotin.

7. Dietary imbalances observed in the GFD of children and adolescents from the Basque Country can be attributed partly to GFPs, particularly excessive lipid intake and its inadequate profile.

In summary, food industry, in response to the responsibility it has in celiac people's health, in recent years has improved GFP's nutritional quality in various components, such as fiber, RS and some micronutrients. However, it is still necessary to continue working on the content of other GFP's micronutrients and to get a healthier lipid profile in them.

Traditionally, the food industry has worried about guaranteeing the absence of gluten contamination in GFPs. Nowadays, when the ingredients are safer, they also have started looking after the nutritional quality of these products. Although significant improvements in some products and nutrients have been achieved, maintaining this challenge is essential to achieve the nutritional needs detected. Furthermore, it is important to transfer all the extracted knowledge to the society, through measures such as completing GFP's label information for example.

8. Adherence of celiac people from Finland to the GFD is excellent. In this way, they reduce the symptoms of celiac disease and prevent the risk of other possible illnesses. Among other reasons, their good adherence to the diet is due to sufficient nutritional education.

It seems that POCT test can be a good tool for monitoring the adherence to the diet, even if its use has to be still confirmed.

ERREFERENTZIAK

REFERENCES

1. Dowd B, Walker-Smith J. Samuel Gee, Aretaeus, and the coeliac affection. Br Med J. 1974;2(5909):45-7.
2. Gee S. On the coeliac disease. St Barthol Hosp Rep. 1888; 24:17-20
3. Herter CA. On Infantilism from Chronic Intestinal Infection . New York : Mac. Millan Comp. .
4. Blanco A. Enfermedad celíaca Evolución histórica de los conocimientos sobre la enfermedad celíaca. En: Arranz E, Bernardo D, Blanco A, Calvo C, Chirdo FG, Fernández F et al. Enfermedad celíaca. Introducción al conocimiento actual de la enfermedad celíaca. Ed. Ergon;2009. p. 1-28
5. Dicke WK. Coeliac disease. Investigation of the harmful effects of certain typesof cereal on patients with coeliac disease (Thesis). University of Utrecht, The Netherlands, 1950 (in Dutch).
6. van Berge-Henegouwen GP, Mulder CJ. Pioneer in the gluten free diet: Willem-Karel Dicke 1905-1962, over 50 years of gluten free diet. Gut. 1993;34(11):1473-5.
7. VAN DE KAMER JH, WEIJERS HA, DICKE WK. Coeliac disease. IV. An investigation into the injurious constituents of wheat in connection with their action on patients with coeliac disease. Acta Paediatr. 1953;42(3):223-31.
8. DICKE WK, WEIJERS HA, VAN DE KAMER JH. Coeliac disease. II. The presence in wheat of a factor having a deleterious effect in cases of coeliac disease. Acta Paediatr. 1953;42(1):34-42.
9. Anderson CM, Frazer AC, French JM, Gerrard JW, Simmons HG, Smellie JE.Coeliac disease: gastro-intestinal studies and the effects of dietary wheat flour. Lancet 1952; i: 836-42.
10. PAULLEY JW. Observation on the aetiology of idiopathic steatorrhoea; jejunal and lymph-node biopsies. Br Med J. 1954;2(4900):1318-21.
11. Husby S, Koletzko S, Korponay-Szabó IR, Mearin ML, Phillips A, Shamir R, et al. European Society for Pediatric Gastroenterology, Hepatology, and Nutrition guidelines for the diagnosis of coeliac disease. J Pediatr Gastroenterol Nutr. 2012;54(1):136-60.
12. Green PH, Cellier C. Celiac disease. N Engl J Med. 2007;357(17):1731-43.
13. Lionetti E, Catassi C. New clues in celiac disease epidemiology, pathogenesis, clinical manifestations, and treatment. Int Rev Immunol. 2011;30(4):219-31.
14. Nevoral J. Celiac Disease in Children: What Has Changed? : International Journal of Celiac Disease; 2014. p. 18-23.

15. A P, L R. Epidemiology of celiac disease and non-celiac gluten-related disorders. In: Arranz E, Fernández-Bañares F, Rosell C, Rodrigo L, Peña A, editors. Advances in the understanding of gluten related pathology and the evolution of gluten free foods. Barcelona: OmniaScience; 2015. p. 27-73.
16. Catassi C, Rätsch IM, Gandolfi L, Pratesi R, Fabiani E, El Asmar R, et al. Why is coeliac disease endemic in the people of the Sahara? Lancet. 1999;354(9179):647-8.
17. Lionetti E, Gatti S, Pulvirenti A, Catassi C. Celiac disease from a global perspective. Best Pract Res Clin Gastroenterol. 2015;29(3):365-79.
18. Simón E, Larretxi I, Churruca I, Lasa A, Bustamante MÁ, Navarro V, et al. Nutritional and Analytical Approaches of Gluten-Free Diet in Celiac Disease. Springer; 2017.
19. Silano M, Agostoni C, Sanz Y, Guandalini S. Infant feeding and risk of developing celiac disease: a systematic review. BMJ Open. 2016;6(1):e009163.
20. Szajewska H, Shamir R, Mearin L, Ribes-Koninckx C, Catassi C, Domellöf M, et al. Gluten Introduction and the Risk of Coeliac Disease: A Position Paper by the European Society for Pediatric Gastroenterology, Hepatology, and Nutrition. J Pediatr Gastroenterol Nutr. 2016;62(3):507-13.
21. Kemppainen KM, Lynch KF, Liu E, Lönnrot M, Simell V, Briese T, et al. Factors that Increase Risk of Celiac Disease Autoimmunity Following a Gastrointestinal Infection in Early Life. Clin Gastroenterol Hepatol. 2016.
22. Narang M, Puri AS, Sachdeva S, Singh J, Kumar A, Saran RK. Celiac Disease and H. Pylori Infection in Children: Is there any Association? J Gastroenterol Hepatol. 2016.
23. Vivas S, Ruiz de Morales JM, Fernandez M, Hernando M, Herrero B, Casqueiro J, et al. Age-related clinical, serological, and histopathological features of celiac disease. Am J Gastroenterol. 2008;103(9):2360-5; quiz 6.
24. celíaca GdtdPpedpdle. Protocolo para el diagnóstico precoz de la enfermedad celíaca. 2018.
25. Vriezinga SL, Schweizer JJ, Koning F, Mearin ML. Coeliac disease and gluten-related disorders in childhood. Nat Rev Gastroenterol Hepatol. 2015;12(9):527-36.
26. Rostom A, Dubé C, Cranney A, Saloojee N, Sy R, Garritty C, et al. The diagnostic accuracy of serologic tests for celiac disease: a systematic review. Gastroenterology. 2005;128(4 Suppl 1):S38-46.
27. Weile B, Hansen BF, Hägerstrand I, Hansen JP, Krasilnikoff PA. Interobserver variation in diagnosing coeliac disease. A joint study by Danish and Swedish pathologists. APMIS. 2000;108(5):380-4.

28. Leffler DA, Schuppan D. Update on Serologic Testing in Celiac Disease. Am J Gastroenterol. 2010;105(12):2520-4.
29. Moscoso J, R Q. Enfermedad Celíaca: revisión. Rev Méd Clín Condes. 2015;26:613-27.
30. Spatola BN, Kaukinen K, Collin P, Mäki M, Kagnoff MF, Daugherty PS. Persistence of elevated deamidated gliadin peptide antibodies on a gluten-free diet indicates nonresponsive coeliac disease. Aliment Pharmacol Ther. 2014;39(4):407-17.
31. Leffler D, Schuppan D, Pallav K, Najarian R, Goldsmith JD, Hansen J, et al. Kinetics of the histological, serological and symptomatic responses to gluten challenge in adults with coeliac disease. Gut. 2013;62(7):996-1004.
32. Raju SA, Mooney PD, Aziz I, Kurien M, Sanders DS. Letter: gluten challenge in the era of noncoeliac gluten sensitivity--a change in clinical practice? Aliment Pharmacol Ther. 2016;43(5):656.
33. Arranz E, Garrote J. HLA en la enfermedad celíaca. 2014.
34. Villanacci V, Ceppa P, Tavani E, Vindigni C, Volta U, (GIPAD) GIPAD, et al. Coeliac disease: the histology report. Dig Liver Dis. 2011;43 Suppl 4:S385-95.
35. Blanco A. Enfermedad celíaca Evolución histórica de los conocimientos sobre la enfermedad celíaca. En: Arranz E, Bernardo D, Blanco A, Calvo C, Chirdo FG, Fernández F et al. Enfermedad celíaca. Introducción al conocimiento actual de la enfermedad celíaca. Ed. Ergon;2009. p. 1-28.
36. World Gastroenterology Organisation. WGO Practice Guideline - Celiac Disease. Available from: http://www.worldgastroenterology.org/guidelines/global-guidelines/celiac-disease
37. Cheng J, Brar PS, Lee AR, Green PH. Body mass index in celiac disease: beneficial effect of a gluten-free diet. J Clin Gastroenterol. 2010;44(4):267-71.
38. Goel G, King T, Daveson AJ, Andrews JM, Krishnarajah J, Krause R, et al. Epitope-specific immunotherapy targeting CD4-positive T cells in coeliac disease: two randomised, double-blind, placebo-controlled phase 1 studies. Lancet Gastroenterol Hepatol. 2017;2(7):479-93.
39. Kaukinen K, Lindfors K, Mäki M. Advances in the treatment of coeliac disease: an immunopathogenic perspective. Nat Rev Gastroenterol Hepatol. 2014;11(1):36-44.
40. Kurppa K, Collin P, Mäki M, Kaukinen K. Celiac disease and health-related quality of life. Expert Rev Gastroenterol Hepatol. 2011;5(1):83-90.
41. Nachman F, del Campo MP, González A, Corzo L, Vázquez H, Sfoggia C, et al. Long-term deterioration of quality of life in adult patients

with celiac disease is associated with treatment noncompliance. Dig Liver Dis. 2010;42(10):685-91.
42. Rubio-Tapia A, Hill ID, Kelly CP, Calderwood AH, Murray JA, Gastroenterology ACo. ACG clinical guidelines: diagnosis and management of celiac disease. Am J Gastroenterol. 2013;108(5):656-76; quiz 77.
43. Guandalini S, Polanco I. Nonceliac gluten sensitivity or wheat intolerance syndrome? J Pediatr. 2015;166(4):805-11.
44. Sapone A, Bai JC, Ciacci C, Dolinsek J, Green PH, Hadjivassiliou M, et al. Spectrum of gluten-related disorders: consensus on new nomenclature and classification. BMC Med. 2012;10:13.
45. Molina-Infante J, Santolaria S, Sanders DS, Fernández-Bañares F. Systematic review: noncoeliac gluten sensitivity. Aliment Pharmacol Ther. 2015;41(9):807-20.
46. B C. A clinical update: non-celiac gluten sensivity. 2014.
47. Shewry PR, Halford NG. Cereal seed storage proteins: structures, properties and role in grain utilization. J Exp Bot. 2002;53(370):947-58.
48. Hausch F, Shan L, Santiago NA, Gray GM, Khosla C. Intestinal digestive resistance of immunodominant gliadin peptides. Am J Physiol Gastrointest Liver Physiol. 2002;283(4):G996-G1003.
49. Shan L, Molberg Ø, Parrot I, Hausch F, Filiz F, Gray GM, et al. Structural basis for gluten intolerance in celiac sprue. Science. 2002;297(5590):2275-9.
50. Sciarini, L.S., Ribotta, P.D., León, A.E. et al. Food Bioprocess Technol (2010) 3: 577. https://doi.org/10.1007/s11947-008-0098-2 Influence of Gluten-free Flours and their Mixtures on Batter Properties and Bread Quality
51. Matos ME, Rosell CM. Understanding gluten-free dough for reaching breads with physical quality and nutritional balance. J Sci Food Agric. 2015;95(4):653-61.
52. Real A, Comino I, de Lorenzo L, Merchán F, Gil-Humanes J, Giménez MJ, et al. Molecular and immunological characterization of gluten proteins isolated from oat cultivars that differ in toxicity for celiac disease. PLoS One. 2012;7(12):e48365.
53. Comino I, Real A, de Lorenzo L, Cornell H, López-Casado M, Barro F, et al. Diversity in oat potential immunogenicity: basis for the selection of oat varieties with no toxicity in coeliac disease. Gut. 2011;60(7):915-22.
54. Thompson T, Lee AR, Grace T. Gluten contamination of grains, seeds, and flours in the United States: a pilot study. J Am Diet Assoc. 2010;110(6):937-40.

55. Comino I, Moreno MeL, Real A, Rodríguez-Herrera A, Barro F, Sousa C. The gluten-free diet: testing alternative cereals tolerated by celiac patients. Nutrients. 2013;5(10):4250-68.

56. Arentz-Hansen H, Fleckenstein B, Molberg Ø, Scott H, Koning F, Jung G, et al. The molecular basis for oat intolerance in patients with celiac disease. PLoS Med. 2004;1(1):e1.

57. Steinert RE, Raederstorff D, Wolever TM. Effect of Consuming Oat Bran Mixed in Water before a Meal on Glycemic Responses in Healthy Humans-A Pilot Study. Nutrients. 2016;8(9).

58. Codex Alimentarious. CODEX STAN 118-1979. Standard for Foods for Special Dietary Use for Persons Intolerant to Gluten. 2008 Revision.

59. Akobeng AK, Thomas AG. Systematic review: tolerable amount of gluten for people with coeliac disease. Aliment Pharmacol Ther. 2008;27(11):1044-52.

60. Catassi C, Fabiani E, Iacono G, D'Agate C, Francavilla R, Biagi F, et al. A prospective, double-blind, placebo-controlled trial to establish a safe gluten threshold for patients with celiac disease. Am J Clin Nutr. 2007;85(1):160-6.

61. Commission Regulation (EC) No. 41/2009 of January 2009 concerning the composition and labelling of foodstuffs suitable for people intolerant to gluten. Off J Eur Union, L16; (January 21,2009).

62. Commission Implementing Regulation (EC) No. 828/2014 of 30 July 2014 on the requirements for the provision of information to consumers on the absence or reduced presence of gluten in food. OFF J Eur Union, L228; (July 31, 2014)

63. Bustamante M, Fernández-Gil MP, Churruca I, Miranda J, Lasa A, Navarro V, et al. Evolution of Gluten Content in Cereal-Based Gluten-Free Products: An Overview from 1998 to 2016. Nutrients. 2017;9(1).

64. Commission Regulation (EU). No. 1169/2011 of European Parliament and of the Council on the provision of foodinformation to consumers.Off. J. Eur. Union L2011,304, 18–63.

65. Scherf KA, Poms RE. Recent developments in analytical methods for tracing gluten. J Cereal Sci. 2016; 67: 112-22

66. Dewar DH, Amato M, Ellis HJ, Pollock EL, Gonzalez-Cinca N, Wieser H, et al. The toxicity of high molecular weight glutenin subunits of wheat to patients with coeliac disease. Eur J Gastroenterol Hepatol. 2006;18(5):483-91.

67. Shewry. the structural and evolutonary relationships of the prolamin storage proteins of barley, rye and wheat.

68. Vici G, Belli L, Biondi M, Polzonetti V. Gluten free diet and nutrient deficiencies: A review. Clin Nutr. 2016;35(6):1236-41.

69. Midhagen G, Hallert C. High rate of gastrointestinal symptoms in celiac patients living on a gluten-free diet: controlled study. Am J Gastroenterol. 2003;98(9):2023-6.
70. Holmes GK, Prior P, Lane MR, Pope D, Allan RN. Malignancy in coeliac disease--effect of a gluten free diet. Gut. 1989;30(3):333-8.
71. Haines ML, Anderson RP, Gibson PR. Systematic review: The evidence base for long-term management of coeliac disease. Aliment Pharmacol Ther. 2008;28(9):1042-66.
72. Olmos M, Antelo M, Vazquez H, Smecuol E, Mauriño E, Bai JC. Systematic review and meta-analysis of observational studies on the prevalence of fractures in coeliac disease. Dig Liver Dis. 2008;40(1):46-53.
73. Pink IJ, Creamer B. Response to a gluten-free diet of patients with the coeliac syndrome. Lancet. 1967;1(7485):300-4.
74. See J, Murray JA. Gluten-free diet: the medical and nutrition management of celiac disease. Nutr Clin Pract. 2006;21(1):1-15.
75. Galli G, Esposito G, Lahner E, Pilozzi E, Corleto VD, Di Giulio E, et al. Histological recovery and gluten-free diet adherence: a prospective 1-year follow-up study of adult patients with coeliac disease. Aliment Pharmacol Ther. 2014;40(6):639-47.
76. Lanzini A, Lanzarotto F, Villanacci V, Mora A, Bertolazzi S, Turini D, et al. Complete recovery of intestinal mucosa occurs very rarely in adult coeliac patients despite adherence to gluten-free diet. Aliment Pharmacol Ther. 2009;29(12):1299-308.
77. Wahab PJ, Meijer JW, Mulder CJ. Histologic follow-up of people with celiac disease on a gluten-free diet: slow and incomplete recovery. Am J Clin Pathol. 2002;118(3):459-63.
78. Vécsei E, Steinwendner S, Kogler H, Innerhofer A, Hammer K, Haas OA, et al. Follow-up of pediatric celiac disease: value of antibodies in predicting mucosal healing, a prospective cohort study. BMC Gastroenterol. 2014;14:28.
79. Wierdsma NJ, van Bokhorst-de van der Schueren MA, Berkenpas M, Mulder CJ, van Bodegraven AA. Vitamin and mineral deficiencies are highly prevalent in newly diagnosed celiac disease patients. Nutrients. 2013;5(10):3975-92.
80. Malandrino N, Capristo E, Farnetti S, Leggio L, Abenavoli L, Addolorato G, et al. Metabolic and nutritional features in adult celiac patients. Dig Dis. 2008;26(2):128-33.
81. Abenavoli L, Delibasic M, Peta V, Turkulov V, De Lorenzo A, Medić-Stojanoska M. Nutritional profile of adult patients with celiac disease. Eur Rev Med Pharmacol Sci. 2015;19(22):4285-92.

82. Corazza GR, Valentini RA, Andreani ML, D'Anchino M, Leva MT, Ginaldi L, et al. Subclinical coeliac disease is a frequent cause of iron-deficiency anaemia. Scand J Gastroenterol. 1995;30(2):153-6.
83. Capristo E, Mingrone G, Addolorato G, Greco AV, Corazza GR, Gasbarrini G. Differences in metabolic variables between adult coeliac patients at diagnosis and patients on a gluten-free diet. Scand J Gastroenterol. 1997;32(12):1222-9.
84. Bodé S, Hassager C, Gudmand-Høyer E, Christiansen C. Body composition and calcium metabolism in adult treated coeliac disease. Gut. 1991;32(11):1342-5.
85. Mazure RM, Vazquez H, Gonzalez D, Mautalen C, Soifer G, Cataldi M, et al. Early changes of body composition in asymptomatic celiac disease patients. Am J Gastroenterol. 1996;91(4):726-30.
86. Capristo E, Addolorato G, Greco AV, Gasbarrini G. Body composition and energy metabolism in celiac disease. Am J Gastroenterol. 1998;93(9):1596-7.
87. Capristo E, Addolorato G, Mingrone G, De Gaetano A, Greco AV, Tataranni PA, et al. Changes in body composition, substrate oxidation, and resting metabolic rate in adult celiac disease patients after a 1-y gluten-free diet treatment. Am J Clin Nutr. 2000;72(1):76-81.
88. Capristo E, Malandrino N, Farnetti S, Mingrone G, Leggio L, Addolorato G, et al. Increased serum high-density lipoprotein-cholesterol concentration in celiac disease after gluten-free diet treatment correlates with body fat stores. J Clin Gastroenterol. 2009;43(10):946-9.
89. Hallert C, Grant C, Grehn S, Grännö C, Hultén S, Midhagen G, et al. Evidence of poor vitamin status in coeliac patients on a gluten-free diet for 10 years. Aliment Pharmacol Ther. 2002;16(7):1333-9.
90. Kurppa K, Lauronen O, Collin P, Ukkola A, Laurila K, Huhtala H, et al. Factors associated with dietary adherence in celiac disease: a nationwide study. Digestion. 2012;86(4):309-14.
91. Barera G, Mora S, Brambilla P, Ricotti A, Menni L, Beccio S, et al. Body composition in children with celiac disease and the effects of a gluten-free diet: a prospective case-control study. Am J Clin Nutr. 2000;72(1):71-5.
92. Radlović N, Mladenović M, Leković Z, Zivanović D, Brdar R, Radlović V, et al. Effect of gluten-free diet on the growth and nutritional status of children with coeliac disease. Srp Arh Celok Lek. 2009;137(11-12):632-7.
93. Mariani P, Viti MG, Montuori M, La Vecchia A, Cipolletta E, Calvani L, et al. The gluten-free diet: a nutritional risk factor for adolescents with celiac disease? J Pediatr Gastroenterol Nutr. 1998;27(5):519-23.

94. Dickey W, Kearney N. Overweight in celiac disease: prevalence, clinical characteristics, and effect of a gluten-free diet. Am J Gastroenterol. 2006;101(10):2356-9.
95. Valletta E, Fornaro M, Cipolli M, Conte S, Bissolo F, Danchielli C. Celiac disease and obesity: need for nutritional follow-up after diagnosis. Eur J Clin Nutr. 2010;64(11):1371-2.
96. Shepherd SJ, Gibson PR. Nutritional inadequacies of the gluten-free diet in both recently-diagnosed and long-term patients with coeliac disease. J Hum Nutr Diet. 2013;26(4):349-58.
97. Saturni L, Ferretti G, Bacchetti T. The gluten-free diet: safety and nutritional quality. Nutrients. 2010;2(1):16-34.
98. Caruso R, Pallone F, Stasi E, Romeo S, Monteleone G. Appropriate nutrient supplementation in celiac disease. Ann Med. 2013;45(8):522-31.
99. Myléus A, Ivarsson A, Webb C, Danielsson L, Hernell O, Högberg L, et al. Celiac disease revealed in 3% of Swedish 12-year-olds born during an epidemic. J Pediatr Gastroenterol Nutr. 2009;49(2):170-6.
100. Ohlund K, Olsson C, Hernell O, Ohlund I. Dietary shortcomings in children on a gluten-free diet. J Hum Nutr Diet. 2010;23(3):294-300.
101. Zuccotti G, Fabiano V, Dilillo D, Picca M, Cravidi C, Brambilla P. Intakes of nutrients in Italian children with celiac disease and the role of commercially available gluten-free products. J Hum Nutr Diet. 2013;26(5):436-44.
102. Babio N, Alcázar M, Castillejo G, Recasens M, Martínez-Cerezo F, Gutiérrez-Pensado V, et al. Patients With Coeliac Disease Reported Highter Consumption of Added Sugar and Total Fat Than Healthy Individuals. J Pediatr Gastroenterol Nutr. 2016.
103. Erdem T, Ferat Ç, Nurdan YA, Halime E, Muhammed Selçuk S, Hamza K, et al. Vitamin and mineral deficiency in children newly diagnosed with celiac disease. Turk J Med Sci. 2015;45(4):833-6.
104. García-Manzanares A, Lucendo AJ. Nutritional and dietary aspects of celiac disease. Nutr Clin Pract. 2011;26(2):163-73.
105. Penagini F, Dilillo D, Meneghin F, Mameli C, Fabiano V, Zuccotti GV. Gluten-free diet in children: an approach to a nutritionally adequate and balanced diet. Nutrients. 2013;5(11):4553-65.
106. Bardella MT, Fredella C, Prampolini L, Molteni N, Giunta AM, Bianchi PA. Body composition and dietary intakes in adult celiac disease patients consuming a strict gluten-free diet. Am J Clin Nutr. 2000;72(4):937-9.
107. Martin J, Geisel T, Maresch C, Krieger K, Stein J. Inadequate nutrient intake in patients with celiac disease: results from a German dietary survey. Digestion. 2013;87(4):240-6.

108. Dell'Olio D, Palma L, Malorgio E, Ansaldi Balocco N. [What do celiac children eat? Dietary analysis of a group of children with celiac disease on a diet]. Minerva Gastroenterol Dietol. 1995;41(4):269-73.
109. Hopman EG, le Cessie S, von Blomberg BM, Mearin ML. Nutritional management of the gluten-free diet in young people with celiac disease in The Netherlands. J Pediatr Gastroenterol Nutr. 2006;43(1):102-8.
110. Ferrara P, Cicala M, Tiberi E, Spadaccio C, Marcella L, Gatto A, et al. High fat consumption in children with celiac disease. Acta Gastroenterol Belg. 2009;72(3):296-300.
111. Kautto E, Ivarsson A, Norström F, Högberg L, Carlsson A, Hörnell A. Nutrient intake in adolescent girls and boys diagnosed with coeliac disease at an early age is mostly comparable to their non-coeliac contemporaries. J Hum Nutr Diet. 2014;27(1):41-53.
112. Enghardt BH, Pearson M, Becker W. Dietary habits and nutrient intake in Swedish children 4 years old and school children in grades 2 and 5 (Riksmaten – barn 2003). Uppsala: Uppsala, National Food Administration (Livsmedelsverket); 2006
113. Whitton C, Nicholson SK, Roberts C, Prynne CJ, Pot GK, Olson A, et al. National Diet and Nutrition Survey: UK food consumption and nutrient intakes from the first year of the rolling programme and comparisons with previous surveys. Br J Nutr. 2011;106(12):1899-914.
114. Størsrud S, Hulthén LR, Lenner RA. Beneficial effects of oats in the gluten-free diet of adults with special reference to nutrient status, symptoms and subjective experiences. Br J Nutr. 2003;90(1):101-7.
115. Miranda J, Lasa A, Bustamante MA, Churruca I, Simon E. Nutritional differences between a gluten-free diet and a diet containing equivalent products with gluten. Plant Foods Hum Nutr. 2014;69(2):182-7.
116. Pellegrini N, Agostoni C. Nutritional aspects of gluten-free products. J Sci Food Agric. 2015;95(12):2380-5.
117. Churruca I, Miranda J, Lasa A, Bustamante M, Larretxi I, Simon E. Analysis of Body Composition and Food Habits of Spanish Celiac Women. Nutrients. 2015;7(7):5515-31.
118. Samasca G, Sur G, Lupan I, Deleanu D. Gluten-free diet and quality of life in celiac disease. Gastroenterol Hepatol Bed Bench. 2014;7(3):139-43.
119. Janatuinen EK, Pikkarainen PH, Kemppainen TA, Kosma VM, Järvinen RM, Uusitupa MI, et al. A comparison of diets with and without oats in adults with celiac disease. N Engl J Med. 1995;333(16):1033-7.
120. Janatuinen EK, Kemppainen TA, Julkunen RJ, Kosma VM, Mäki M, Heikkinen M, et al. No harm from five year ingestion of oats in coeliac disease. Gut. 2002;50(3):332-5.

121. Högberg L, Laurin P, Fälth-Magnusson K, Grant C, Grodzinsky E, Jansson G, et al. Oats to children with newly diagnosed coeliac disease: a randomised double blind study. Gut. 2004;53(5):649-54.
122. Hoffenberg EJ, Haas J, Drescher A, Barnhurst R, Osberg I, Bao F, et al. A trial of oats in children with newly diagnosed celiac disease. J Pediatr. 2000;137(3):361-6.
123. Lee AR, Ng DL, Zivin J, Green PH. Economic burden of a gluten-free diet. J Hum Nutr Diet. 2007;20(5):423-30.
124. Hall NJ, Rubin G, Charnock A. Systematic review: adherence to a gluten-free diet in adult patients with coeliac disease. Aliment Pharmacol Ther. 2009;30(4):315-30.
125. Fabiani E, Taccari LM, Rätsch IM, Di Giuseppe S, Coppa GV, Catassi C. Compliance with gluten-free diet in adolescents with screening-detected celiac disease: a 5-year follow-up study. J Pediatr. 2000;136(6):841-3.
126. Viljamaa M, Collin P, Huhtala H, Sievänen H, Mäki M, Kaukinen K. Is coeliac disease screening in risk groups justified? A fourteen-year follow-up with special focus on compliance and quality of life. Aliment Pharmacol Ther. 2005;22(4):317-24.
127. Ciacci C, Iavarone A, Mazzacca G, De Rosa A. Depressive symptoms in adult coeliac disease. Scand J Gastroenterol. 1998;33(3):247-50.
128. Ciacci C, D'Agate C, De Rosa A, Franzese C, Errichiello S, Gasperi V, et al. Self-rated quality of life in celiac disease. Dig Dis Sci. 2003;48(11):2216-20.
129. Leffler DA, Edwards-George J, Dennis M, Schuppan D, Cook F, Franko DL, et al. Factors that influence adherence to a gluten-free diet in adults with celiac disease. Dig Dis Sci. 2008;53(6):1573-81.
130. Fera T, Cascio B, Angelini G, Martini S, Guidetti CS. Affective disorders and quality of life in adult coeliac disease patients on a gluten-free diet. Eur J Gastroenterol Hepatol. 2003;15(12):1287-92.
131. Edwards George JB, Leffler DA, Dennis MD, Franko DL, Blom-Hoffman J, Kelly CP. Psychological correlates of gluten-free diet adherence in adults with celiac disease. J Clin Gastroenterol. 2009;43(4):301-6.
132. Ciacci C, Cirillo M, Cavallaro R, Mazzacca G. Long-term follow-up of celiac adults on gluten-free diet: prevalence and correlates of intestinal damage. Digestion. 2002;66(3):178-85.
133. Tanpowpong P, Broder-Fingert S, Katz AJ, Camargo CA. Age-related patterns in clinical presentations and gluten-related issues among children and adolescents with celiac disease. Clin Transl Gastroenterol. 2012;3:e9.

134. Nelson M, Mendoza N, McGough N. A survey of provision of dietetic services for coeliac disease in the UK. J Hum Nutr Diet. 2007;20(5):403-11.
135. Pelegrí Calvo C. Evaluación del estado de salud y nutricional de pacientes celiacos de la Comunidad Valenciana.Tesis doctoral. Universidad de Valencia; 2011
136. Cranney A, Zarkadas M, Graham ID, Butzner JD, Rashid M, Warren R, et al. The Canadian Celiac Health Survey. Dig Dis Sci. 2007;52(4):1087-95.
137. Rodrigo-Sáez L, Fuentes-Álvarez D, Pérez-Martínez I, Alvarez-Mieres N, Niño-García P, de-Francisco-García R, et al. Differences between pediatric and adult celiac disease. Rev Esp Enferm Dig. 2011;103(5):238-44.
138. Zarkadas M, Dubois S, MacIsaac K, Cantin I, Rashid M, Roberts KC, et al. Living with coeliac disease and a gluten-free diet: a Canadian perspective. J Hum Nutr Diet. 2013;26(1):10-23.
139. Ciacci C, Iavarone A, Siniscalchi M, Romano R, De Rosa A. Psychological dimensions of celiac disease: toward an integrated approach. Dig Dis Sci. 2002;47(9):2082-7.
140. Ciacci C, Ciclitira P, Hadjivassiliou M, Kaukinen K, Ludvigsson JF, McGough N, et al. The gluten-free diet and its current application in coeliac disease and dermatitis herpetiformis. United European Gastroenterol J. 2015;3(2):121-35.
141. Zarkadas M, Cranney A, Case S, Molloy M, Switzer C, Graham ID, et al. The impact of a gluten-free diet on adults with coeliac disease: results of a national survey. J Hum Nutr Diet. 2006;19(1):41-9.
142. Pelegrí C, Mañes J, Soriano JM. [Translation and adaptation to Spanish language of the quality of life questionnaire for celiac people called Canadian Celiac Health Survey]. Nutr Hosp. 2014;30(4):941-4.
143. Casellas F, López Vivancos J, Malagelada JR. Current epidemiology and accessibility to diet compliance in adult celiac disease. Rev Esp Enferm Dig. 2006;98(6):408-19.
144. Mustalahti K, Lohiniemi S, Collin P, Vuolteenaho N, Laippala P, Mäki M. Gluten-free diet and quality of life in patients with screen-detected celiac disease. Eff Clin Pract. 2002;5(3):105-13.
145. Shah S, Akbari M, Vanga R, Kelly CP, Hansen J, Theethira T, et al. Patient perception of treatment burden is high in celiac disease compared with other common conditions. Am J Gastroenterol. 2014;109(9):1304-11.
146. Calvo-Lerma J, Crespo-Escobar P, Martínez-Barona S, Fornés-Ferrer V, Donat E, Ribes-Koninckx C. Differences in the macronutrient and dietary fibre profile of gluten-free products as compared to their gluten-containing counterparts. Eur J Clin Nutr. 2019.

147. Kulai T, Rashid M. Assessment of Nutritional Adequacy of Packaged Gluten-free Food Products. Can J Diet Pract Res. 2014;75(4):186-90.
148. Thompson T. Thiamin, riboflavin, and niacin contents of the gluten-free diet: is there cause for concern? J Am Diet Assoc. 1999;99(7):858-62.
149. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a standard definition for child overweight and obesity worldwide: international survey. BMJ. 2000;320(7244):1240-3.
150. Cole TJ, Flegal KM, Nicholls D, Jackson AA. Body mass index cut offs to define thinness in children and adolescents: international survey. BMJ. 2007;335(7612):194.
151. Gallagher D, Heymsfield SB, Heo M, Jebb SA, Murgatroyd PR, Sakamoto Y. Healthy percentage body fat ranges: an approach for developing guidelines based on body mass index. Am J Clin Nutr. 2000;72(3):694-701.
152. Russolillo, G.; Marques, I. Food Portion Sizes Album; Imagen Comunicación Multimedia: Madrid, Spain, 2008; p. 186
153. Missbach B, Schwingshackl L, Billmann A, Mystek A, Hickelsberger M, Bauer G, et al. Gluten-free food database: the nutritional quality and cost of packaged gluten-free foods. PeerJ. 2015;3:e1337.
154. FESNAD. Dietary reference intakes (DRI) for spanish population, 2010. Actividad Dietética. 2010;14(4):196-7.
155. Aranceta J, Serra-Majem L, Population WPftDoF-BDGftS. Dietary guidelines for the Spanish population. Public Health Nutr. 2001;4(6A):1403-8.
156. Nutritional Assessment Studies: Spanish Population Dietary Intakes. National Health Survey. ENIDE. Available online: http://aesan.msssi.gob.es/AESAN/web/evaluacion_riesgos/seccion/estudios_evaluacion_nutricional.shtml (accessed on 25 May 2015)
157. Gobierno Vasco (2005) Encuesta de nutrición 2005. Departamento de Sanidad del Gobierno Vasco, Vitoria-Gasteiz
158. International A. Methods 968.06 & 992.15. Official Methods of Analysis of AOAC International, 18th Edition, 2005 (Revision 3, 2010).2005. p. p129.
159. International A. AOAC Official Method, 991.43. Total, soluble, and snsoluble dietary fiber in foods. Official Methods of Analysis of AOAC International, 18th Edition, 2005 (Revision 3, 2010). 1994.
160. McCleary B, Monaghan D. Measurement of resistant starch. Journal of Aoac International. 2002;85(3):665-75.
161. AESAN/BEDCA. Base de datos española de composición de alimentos v 1.0 [Available from: http://www.bedca.net.

162. Roe M, Pinchen H, Church S, Finglas P. McCance and Widdowson's The Composition of Foods Seventh Summary Edition and updated Composition of Foods Integrated Dataset. Nutrition Bulletin. 2015;40(1):36-9.
163. Australian Food Composition Database (Januari 2019). Available from: http://www.foodstandards.gov.au/science/monitoringnutrients/afcd/Pages/default.aspx
164. United States Department of Agriculture. Agricultural Research Service. USDA Food Composition Databases. Available from: https://ndb.nal.usda.gov/ndb/search/list.
165. Barone M, Della Valle N, Rosania R, Facciorusso A, Trotta A, Cantatore FP, et al. A comparison of the nutritional status between adult celiac patients on a long-term, strictly gluten-free diet and healthy subjects. Eur J Clin Nutr. 2016;70(1):23-7.
166. Laurikka P, Salmi T, Collin P, Huhtala H, Mäki M, Kaukinen K, et al. Gastrointestinal Symptoms in Celiac Disease Patients on a Long-Term Gluten-Free Diet. Nutrients. 2016;8(7).
167. Mahadev S, Murray JA, Wu TT, Chandan VS, Torbenson MS, Kelly CP, et al. Factors associated with villus atrophy in symptomatic coeliac disease patients on a gluten-free diet. Aliment Pharmacol Ther. 2017;45(8):1084-93.
168. Smecuol E, Gonzalez D, Mautalen C, Siccardi A, Cataldi M, Niveloni S, et al. Longitudinal study on the effect of treatment on body composition and anthropometry of celiac disease patients. Am J Gastroenterol. 1997;92(4):639-43.
169. Ukkola A, Mäki M, Kurppa K, Collin P, Huhtala H, Kekkonen L, et al. Changes in body mass index on a gluten-free diet in coeliac disease: a nationwide study. Eur J Intern Med. 2012;23(4):384-8.
170. Kabbani TA, Goldberg A, Kelly CP, Pallav K, Tariq S, Peer A, et al. Body mass index and the risk of obesity in coeliac disease treated with the gluten-free diet. Aliment Pharmacol Ther. 2012;35(6):723-9.
171. Peralta M, Ramos M, Lipert A, Martins J, Marques A. Prevalence and trends of overweight and obesity in older adults from 10 European countries from 2005 to 2013. Scand J Public Health. 2018;46(5):522-9.
172. Rodríguez-Rodríguez E, López-Plaza B, López-Sobaler AM, Ortega RM. [Overweight and obesity among Spanish adults]. Nutr Hosp. 2011;26(2):355-63.
173. Gutiérrez-Fisac JL, Banegas Banegas JR, Artalejo FR, Regidor E. Increasing prevalence of overweight and obesity among Spanish adults, 1987-1997. Int J Obes Relat Metab Disord. 2000;24(12):1677-82.

174. Sharp P, Bottorff JL, Hunt K, Oliffe JL, Johnson ST, Dudley L, et al. Men's Perspectives of a Gender-Sensitized Health Promotion Program Targeting Healthy Eating, Active Living, and Social Connectedness. Am J Mens Health. 2018;12(6):2157-66.
175. Galdas PM, Cheater F, Marshall P. Men and health help-seeking behaviour: literature review. J Adv Nurs. 2005;49(6):616-23.
176. Więch P, Chmiel Z, Bazaliński D, Sałacińska I, Bartosiewicz A, Mazur A, et al. The Relationship between Body Composition and a Gluten Free Diet in Children with Celiac Disease. Nutrients. 2018;10(11).
177. Wild D, Robins GG, Burley VJ, Howdle PD. Evidence of high sugar intake, and low fibre and mineral intake, in the gluten-free diet. Aliment Pharmacol Ther. 2010;32(4):573-81.
178. E. Gallagher, T.R. Gormley, E.K. Arendt. Recent advances in the formulation of gluten-free cereal-based products. Trends in Food Science and Technology, 15 (2004), pp. 143-152
179. Stantiall SE, Serventi L. Nutritional and sensory challenges of gluten-free bakery products: a review. Int J Food Sci Nutr. 2018;69(4):427-36.
180. Moraes C, Borges NA, Mafra D. Resistant starch for modulation of gut microbiota: Promising adjuvant therapy for chronic kidney disease patients? Eur J Nutr. 2016;55(5):1813-21.
181. Fuentes-Zaragoza, E.; Riquelme-Navarrete, M.J.; Sánchez-Zapata, E.; Pérez-Álvarez, J.A. 470 Resistant starch as functional ingredient: A review. Food Research International 2010, 43, 931-471 942.
182. Segura ME, Rosell CM. Chemical composition and starch digestibility of different gluten-free breads. Plant Foods Hum Nutr. 2011;66(3):224-30.
183. Murphy MM, Douglass JS, Birkett A. Resistant starch intakes in the United States. J Am Diet Assoc. 2008;108(1):67-78.
184. A.A. Alsaffar Effect of food processing on the resistant starch content of cereals and cereal products – a review. Int. J. Food Sci. Technol., 46 (2011), pp. 455-462
185. Salazar J, Espín B, Rodríguez A, Argüelles F, García R, Rubio M, et al. Valoración nutricional de la dieta sin gluten. ¿Es la dieta sin gluten deficitaria en algún nutriente? An Pediatr. 2015;83:33–9.
186. Roman-Vinas, B.; Ribas Barba, L.; Ngo, J.; Martinez-Gonzalez, M.A.; Wijnhoven, T.M.; Serra-Majem, L.Validity of dietary patterns to assess nutrient intake adequacy. Br. J. Nutr. 2009, 101 (Suppl. 2), S12–S20
187. Marshall TA, Stumbo PJ, Warren JJ, Xie XJ. Inadequate nutrient intakes are common and are associated with low diet variety in rural, community-dwelling elderly. J Nutr. 2001;131(8):2192-6.

188. Dahele A, Ghosh S. Vitamin B12 deficiency in untreated celiac disease. Am J Gastroenterol. 2001;96(3):745-50.
189. Harper JW, Holleran SF, Ramakrishnan R, Bhagat G, Green PH. Anemia in celiac disease is multifactorial in etiology. Am J Hematol. 2007;82(11):996-1000.
190. Dickey W. Low serum vitamin B12 is common in coeliac disease and is not due to autoimmune gastritis. Eur J Gastroenterol Hepatol. 2002;14(4):425-7.
191. Schøsler L, Christensen LA, Hvas CL. Symptoms and findings in adult-onset celiac disease in a historical Danish patient cohort. Scand J Gastroenterol. 2016;51(3):288-94.
192. Larretxi I, Simon E, Benjumea L, Miranda J, Bustamante MA, Lasa A, et al. Gluten-free-rendered products contribute to imbalanced diets in children and adolescents with celiac disease. Eur J Nutr. 2018.
193. González T, Larretxi I, Vitoria JC, Castaño L, Simón E, Churruca I, et al. Celiac Male's Gluten-Free Diet Profile: Comparison to that of the Control Population and Celiac Women. Nutrients. 2018;10(11).
194. Lebwohl B, Cao Y, Zong G, Hu FB, Green PHR, Neugut AI, et al. Long term gluten consumption in adults without celiac disease and risk of coronary heart disease: prospective cohort study. BMJ. 2017;357:j1892.
195. Staudacher HM, Gibson PR. How healthy is a gluten-free diet? Br J Nutr. 2015;114(10):1539-41.
196. Gliszczyńska-Świgło A, Klimczak I, Rybicka I. Chemometric analysis of minerals in gluten-free products. J Sci Food Agric. 2018;98(8):3041-8.
197. Mazzeo T, Cauzzi S, Brighenti F, Pellegrini N. The development of a composition database of gluten-free products. Public Health Nutr. 2015;18(8):1353-7.
198. Wu JH, Neal B, Trevena H, Crino M, Stuart-Smith W, Faulkner-Hogg K, et al. Are gluten-free foods healthier than non-gluten-free foods? An evaluation of supermarket products in Australia. Br J Nutr. 2015;114(3):448-54.
199. Cornicelli M, Saba M, Machello N, Silano M, Neuhold S. Nutritional composition of gluten-free food versus regular food sold in the Italian market. Dig Liver Dis. 2018;50(12):1305-8.
200. Rybicka I. The Handbook of Minerals on a Gluten-Free Diet. Nutrients. 2018;10(11).
201. Yazynina E, Johansson M, Jägerstad M, Jastrebova J. Low folate content in gluten-free cereal products and their main ingredients. Food Chemistry. 2008;111(1):236-42.

202. Gobbetti M, Pontonio E, Filannino P, Rizzello CG, De Angelis M, Di Cagno R. How to improve the gluten-free diet: The state of the art from a food science perspective. Food Res Int. 2018;110:22-32.
203. Rybicka I, Gliszczyńska-Świgło A. Minerals in grain gluten-free products. The content of calcium, potassium, magnesium, sodium, copper, iron, manganese, and zinc. Journal of Food Composition and Analysis. 2017;59:61-7.
204. Rybicka I, Gliszczynska-Swiglo A. Gluten-Free Flours from Different Raw Materials as the Source of Vitamin B. J Nutr Sci Vitaminol (Tokyo). 2017;63(2):125-32.
205. Government of Canada 2018 eta Us Department of Health and Human Services 2015:Government of Canada. 2018. In: Laws J, ed. Food and drug regulations. Ottawa, Ontario, Canada 2001, 4, 1403–1408.

206. US Department of Health and Human Services. 2015. Questions and answers on FDA's fortification policy – guidance for ndustry. In: Food and Drug Aministration. Available at https://www.fda.gov/media/94563/download (accesed on 2019 May 23)
207. Melini V, Melini F. Gluten-Free Diet: Gaps and Needs for a Healthier Diet. Nutrients. 2019;11(1).
208. Fry L, Madden AM, Fallaize R. An investigation into the nutritional composition and cost of gluten-free versus regular food products in the UK. J Hum Nutr Diet. 2018;31(1):108-20.
209. Suliburska J, Krejpcio Z, Regula J, Grochowicz A. Evaluation of the content and the potential bioavailability of minerals from gluten-free products2013. 75-80 p.
210. Lindfors K, Ciacci C, Kurppa K, Lundin KEA, Makharia GK, Mearin ML, et al. Coeliac disease. Nat Rev Dis Primers. 2019;5(1):3.
211. A.Lasa, I.Churruca, J. Miranda, I.Larretxi, M.A. Bustamante, E.Simón; Zeliakoarentzako elikadura orekatuaren gida. ISBN/ISSN: 978-84-695-9829-0 Publicada con la colaboración de EZE (Asociación de Celíacos de Euskadi) y con la financiación del Departamento de Salud del Gobierno Vasco. 2014.
212. Pietzak MM. Follow-up of patients with celiac disease: achieving compliance with treatment. Gastroenterology. 2005;128(4 Suppl 1):S135-41.
213. Niewinski MM. Advances in celiac disease and gluten-free diet. J Am Diet Assoc. 2008;108(4):661-72.
214. See JA, Kaukinen K, Makharia GK, Gibson PR, Murray JA. Practical insights into gluten-free diets. Nat Rev Gastroenterol Hepatol. 2015;12(10):580-91.

215. Jackson PT, Glasgow JF, Thom R. Parents' understanding of coeliac disease and diet. Arch Dis Child. 1985;60(7):672-4.
216. Raivio T, Kaukinen K, Nemes E, Laurila K, Collin P, Kovács JB, et al. Self transglutaminase-based rapid coeliac disease antibody detection by a lateral flow method. Aliment Pharmacol Ther. 2006;24(1):147-54.
217. Raivio T, Korponay-Szabó I, Collin P, Laurila K, Huhtala H, Kaartinen T, et al. Performance of a new rapid whole blood coeliac test in adult patients with low prevalence of endomysial antibodies. Dig Liver Dis. 2007;39(12):1057-63.
218. Raivio T, Korponay-Szabó IR, Paajanen T, Ashorn M, Iltanen S, Collin P, et al. Comparison of a novel whole blood transglutaminase-based ELISA with a whole blood rapid antibody test and established conventional serological celiac disease assays. J Pediatr Gastroenterol Nutr. 2008;47(5):562-7.
219. Korponay-Szabó IR, Raivio T, Laurila K, Opre J, Király R, Kovács JB, et al. Coeliac disease case finding and diet monitoring by point-of-care testing. Aliment Pharmacol Ther. 2005;22(8):729-37.
220. Leffler DA, Dennis M, Edwards George JB, Jamma S, Magge S, Cook EF, et al. A simple validated gluten-free diet adherence survey for adults with celiac disease. Clin Gastroenterol Hepatol. 2009;7(5):530-6, 6.e1-2.
221. Biagi F, Andrealli A, Bianchi PI, Marchese A, Klersy C, Corazza GR. A gluten-free diet score to evaluate dietary compliance in patients with coeliac disease. Br J Nutr. 2009;102(6):882-7.
222. Bardella MT, Molteni N, Prampolini L, Giunta AM, Baldassarri AR, Morganti D, et al. Need for follow up in coeliac disease. Arch Dis Child. 1994;70(3):211-3.
223. Johansson K, Malmberg Hård Af Segerstad E, Mårtensson H, Agardh D. Dietitian visits were a safe and cost-effective form of follow-up care for children with celiac disease. Acta Paediatr. 2019;108(4):676-80.
224. Aaltonen K, Laurikka P, Huhtala H, Mäki M, Kaukinen K, Kurppa K. The Long-Term Consumption of Oats in Celiac Disease Patients Is Safe: A Large Cross-Sectional Study. Nutrients. 2017;9(6).
225. Clemens R, van Klinken BJ. The future of oats in the food and health continuum. Br J Nutr. 2014;112 Suppl 2:S75-9.
226. Peräaho M, Kaukinen K, Paasikivi K, Sievänen H, Lohiniemi S, Mäki M, et al. Wheat-starch-based gluten-free products in the treatment of newly detected coeliac disease: prospective and randomized study. Aliment Pharmacol Ther. 2003;17(4):587-94.
227. Kaukinen K, Collin P, Holm K, Rantala I, Vuolteenaho N, Reunala T, et al. Wheat starch-containing gluten-free flour products in the treatment

of coeliac disease and dermatitis herpetiformis. A long-term follow-up study. Scand J Gastroenterol. 1999;34(2):163-9.

228. Thompson T. Wheat starch, gliadin, and the gluten-free diet. J Am Diet Assoc. 2001;101(12):1456-9.

229. Ciclitira PJ, Cerio R, Ellis HJ, Maxton D, Nelufer JM, Macartney JM. Evaluation of a gliadin-containing gluten-free product in coeliac patients. Hum Nutr Clin Nutr. 1985;39(4):303-8.

230. Collin P, Reunala T, Pukkala E, Laippala P, Keyriläinen O, Pasternack A. Coeliac disease--associated disorders and survival. Gut. 1994;35(9):1215-8.

231. Collin P, Mäki M, Kaukinen K. Complete small intestine mucosal recovery is obtainable in the treatment of celiac disease. Gastrointest Endosc. 2004;59(1):158-9; author reply 9-60.

232. Ukkola A, Mäki M, Kurppa K, Collin P, Huhtala H, Kekkonen L, et al. Patients' experiences and perceptions of living with coeliac disease - implications for optimizing care. J Gastrointestin Liver Dis. 2012;21(1):17-22.