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# Revisión del proceso diagnóstico clínico-patológico en linfomas cutáneos B y T: experiencia en el HUA

## Cutaneous B and T lymphomas: review of the clinical and histological diagnostic workup and experience in the HUA


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# ABBREVIATIONS

WHO: World Health Organization

HUA: Hospital Universitario Araba

MF: Mycosis fungoides

FMF: Folliculotropic mycosis fungoides

PCGD-TCL: Primary cutaneous gamma-delta T-cell lymphoma

SPTCL: Subcutaneous panniculitis-like T-cell lymphoma

MRI: Magnetic Resonance Imaging

PCMZL: Primary cutaneous marginal zone (MALT) lymphoma

FNA: Fine-needle aspiration

AITL: Angioimmunoblastic T-cell lymphoma

HRS-cells: Hodgkin and Reed/Sternberg cells

# INDEX

# 1. ABSTRACT

*Background*. The involvement of the skin in neoplastic lymphoproliferative disorders can occur either by an affectation secondary to a systemic neoplasm or by a primary affectation of the skin, which is considered a primary cutaneous lymphoma, a non-Hodgkin lymphoma presenting in the skin with no evidence of extracutaneous disease at the time of diagnosis. Clinical and histological diagnosis of cutaneous lymphoproliferative disorders is a challenge in dermatopathology.

*Objective.* The aim of this paper is to review a selection of cases of cutaneous lymphoma occurred during the past three years in the Hospital Universitario Araba (HUA), focusing on the clinical and pathological diagnostic workup as a whole, to try to meet the difficulties encountered and compare them with those found in the medical literature and highlight the importance of clinical and histological correlation.

*Methods*. Four cases occurred in the HUA during 2015-2018 were selected and compared through a systematic review of cutaneous lymphoma diagnostic workup in articles published in online medical databases and journals.

*Results*. Each case is exposed and analyzed, emphasizing the time elapsed from the first consultation of the patient to final diagnosis, the number of biopsies that each case required, the inquiries made to external centers to confirm the diagnoses and the intervention of different departments. The information found during the bibliographic review of each case is exposed.

*Conclusion*. Clinical and pathological correlation is crucial and should be materialized in the development of sessions and the organization of interdisciplinary committees, in which the diagnosis of cutaneous lymphoma is addressed jointly by the departments involved.

## 2. INTRODUCTION

The involvement of the skin in neoplastic lymphoproliferative disorders can occur either by an affectation secondary to a systemic neoplasm or by a primary affectation of the skin, which is considered a primary cutaneous lymphoma.

The World Health Organization (WHO) defines primary cutaneous lymphomas as non-Hodgkin lymphomas presenting in the skin with no evidence of extracutaneous disease at the time of diagnosis. The revised classification of 2018 includes 18 different subtypes of primary cutaneous lymphoma, representing a quite complex field with many sub-categories (**Table 1**) [2].

| Revised WHO Classification 2018 | (%)* |
|---|---|
| Cutaneous T-cell and NK cell lymphomas | |
| Mycosis fungoides | 39 |
| Mycosis fungoides variants and subtypes | |
| • Folliculotropic mycosis fungoides | 5 |
| • Pagetoid reticulosis | <1 |
| • Granulomatous slack skin | <1 |
| Sézary syndrome | 2 |
| Adult T cell leukemia/lymphoma | <1 |
| Primary cutaneous CD30+ lymphoproliferative disorders | |
| • Primary cutaneous anaplastic large cell lymphoma | 10 |
| • Lymphomatoid papulosis | 15 |
| Subcutaneous panniculitis-like T cell lymphoma | 1 |
| Extranodal NK/T cell lymphoma, nasal type | <1 |
| Primary cutaneous peripheral T cell lymphoma, rare subtypes | |
| • Primary cutaneous gamma-delta T cell lymphoma | <1 |
| • Primary cutaneous aggressive epidermotropic CD8+ T cell lymphoma (provisional) | <1 |

| | |
|---|---|
| • Primary cutaneous CD4+ small/medium-sized pleomorphic T cell lymphoproliferative disorder (provisional) | 3 |
| • Primary cutaneous acral CD8+ T cell lymphoma (provisional) | <1 |
| Primary cutaneous peripheral T cell lymphoma, not otherwise specified | 2 |
| Cutaneous B cell lymphomas | |
| • Primary cutaneous marginal zone B cell lymphoma | 7 |
| • Primary cutaneous follicle center lymphoma | 12 |
| • Primary cutaneous diffuse large B cell lymphoma, leg type | 4 |

**Table 1. Revised WHO Classification 2018: types of primary cutaneous lymphomas and frequencies.**

T-cell cutaneous lymphomas account for 75-80 percent of all primary cutaneous lymphomas. These are a group of neoplasms formed by T lymphocytes with a wide variety in clinical, histological and immunophenotypical presentation, as well as different prognosis [2,3,4]. Mycosis fungoides represents 39 percent of primary cutaneous T-cell lymphomas, being therefore the most frequent cutaneous lymphoma, followed by primary cutaneous CD30+ lymphoproliferative disorders, that encompass lymphomatoid papulosis and primary cutaneous anaplastic large cell lymphoma, which represent 15 and 10 percent of cutaneous T-cell lymphomas, respectively [2,4,6].

B-cell cutaneous lymphomas are a group of neoplasms formed by B lymphocytes that represent 25 percent of all primary cutaneous lymphomas. The three main types of B-cell cutaneous lymphomas are primary cutaneous marginal zone lymphoma, primary cutaneous follicle center lymphoma and primary cutaneous diffuse large B cell lymphoma, leg type [2,3,4].

The skin may also be affected by a systemic process originated elsewhere, but present cutaneous involvement in its development. Both T-cell lymphomas/leukaemias and B-cell lymphomas/leukaemias can present cutaneous involvement, being necessary to differentiate them from a possible primary cutaneous lymphoma.

Clinical and histological diagnosis of cutaneous lymphoproliferative disorders is a challenge in dermatopathology. The arduousness of the process lies not only in differentiating a primary cutaneous process from a systemic disease, and adequately

identifying the subtype to which it belongs, but also in making an accurate differential diagnosis with other benign processes that may present similar features.

The aim of this paper is to review a selection of cases of cutaneous lymphoma occurred during the past three years in the HUA, focusing on the clinical and pathological diagnostic workup as a whole, to try to meet the difficulties encountered and compare them with those found in the medical literature. Of all selected cases, three correspond to primary cutaneous lymphomas of different subtypes, these being mycosis fungoides, primary cutaneous gamma-delta T-cell lymphoma and primary cutaneous marginal zone (MALT) lymphoma; while the other is a systemic lymphoma with cutaneous involvement, from a subtype called T-cell follicular type lymphoma.

## 3. METHODS

This paper is a review of the diagnostic work up followed in four cases of cutaneous lymphoma by the services of Dermatology and Pathology in the HUA.

### 3.1. STUDY DESIGN

A bibliographic study on the clinical and pathological diagnostic work up of cutaneous lymphomas has been carried out, through the systematic review of articles published in online medical databases and journals.

### 3.2. BIBLIOGRAPHIC RESEARCH

The search was based on the literature available in PubMed, UpToDate, Elsevier and the New England Journal of Medicine databases. The articles used in this review were found in the databases cited between July 2018 and March 2019.

The first part of the research was focused on the collection of information on the different classifications and types of cutaneous lymphomas known up to date. Once the clinical cases used in the review were selected, a more specific research was made about each type of cutaneous lymphoma that would be included in the review: mycosis fungoides, primary cutaneous gamma-delta T-cell lymphoma, primary cutaneous marginal zone (MALT) lymphoma and T-cell follicular type lymphoma.

Main criteria used for the election of articles of interest were the publication dates, being the articles of the last 10 years prioritized; the origin of the articles, trying to choose those published in journals of greater impact in Dermatology and Dermatopathology, such as the Journal of the American Academy of Dermatology, Dermatologic Clinics, Diagnostic Histopathology or Seminars in Diagnostic Pathology, among others; and articles signed by authors with extensive knowledge in the field, such as Rein Willemze, Lorenzo Cerroni, Santiago Montes Moreno, and others.

### 3.3. SELECTION OF CASES

Cases of cutaneous lymphoma diagnosed during the past three years (2015-2018) were compiled by the Pathology department, from Vitropath© (Vitro Group, Master Diagnostica S.L), which is an integrated system for the management of this department that covers all of the pathological anatomy workflow (case registration, pending and laboratory work management, carving, reporting and validation of reports). Thirty four cases were located and out of those, four were selected to be reviewed and described more profoundly.

Criteria considered for the inclusion of those cases were, primarily, to reflect the clinical and pathological diagnostic difficulties found in cutaneous lymphoproliferative disorders and the importance in these cases of clinical correlation, being necessary to follow up the biopsies and, in some cases, to review the previous results due to changes in the clinical course of the patient. Some of these cases correspond to commonly found lymphomas in daily practice, such as mycosis fungoides, but others are part of infrequent subtypes.

Of these selected cases, blocks and preparations were removed from the General Archive of Pathology.

### 3.4. REVIEW OF CASES

A critical lecture of articles describing clinical signs, frequent histological findings, immunophenotyping features, molecular characteristics and prognosis of each type of cutaneous lymphoma was made.

Each case was approached individually, approaching first the dermatologic and clinical examination through the reports obtained from the Dermatology department and then the histopathological remarks listed in the reports from the Pathology department.

The review of the cases of Pathological Anatomy consisted in analyzing first the Hematoxylin-Eosin stains, deepening in the morphological study of the samples; the immunohistochemical analysis was then reviewed with the antibodies corresponding to the lymphoid population, T or B-cell according to the case.

Finally, molecular studies carried out by the Molecular Pathology group of the HUA were reviewed. For the molecular diagnosis of T-lymphoma, a fragment of the VJ segment of the hypervariable region of the TCR gene was amplified and for the molecular diagnosis of B-lymphoma, the VDJ segments of the hypervariable region of the immunoglobulin heavy chain (IgH) were amplified, using the PCR technique and subsequent electrophoresis (Kit Master Diagnostica).

Some photographs were taken and included in this research work, from both clinical lesions and histological samples, in order to complete the information and facilitate understanding of the content. Clinical photos were obtained from the photography archives of the Dermatology service and the histological photos were obtained through a Sysmex© slider scanner (Sysmex España S.L).

### 3.5. COMPARISON WITH LITERATURE

Difficulties found during the diagnostic workup of each case in this hospital were compared with those referred in the articles dedicated to cutaneous lymphoma found in medical literature that are included in the bibliography section.

## 4. RESULTS

In this section, the clinical and histopathological development of each selected case is presented, and the information found related to each case in the reviewed medical literature is detailed. Correlation between both is exposed later in the Discussion section.

## 4.1. MYCOSIS FUNGOIDES

### 4.1.1. Case development

In November 2012, a woman on her fifties consulted in the Dermatology department for a tumor lesion in the left inner thigh area, which measured 7x2cm, it was pink colored and infiltrated (**Figure 1A**). A punch biopsy of the lesion was performed and sent to the pathology laboratory, after clinical diagnostic guidance with the suspicion of cutaneous lymphoma, pseudolymphoma, squamous cell carcinoma, dermatofibrosarcoma or leiomyosarcoma. The morphological analysis found a fragment of skin that showed a diffuse lymphoid infiltrate, which occupied the papillary and middle dermis, with focal involvement of the epidermis. It showed lymphoid cells with irregular nuclear contours, with isolated mitosis and occasional eosinophils. Histopathological diagnosis was "*skin with mycosis fungoides in plaque stage*". The sample suggested cutaneous lymphoma and was sent to Valdecilla Hospital (Santander), where they confirmed the diagnosis.

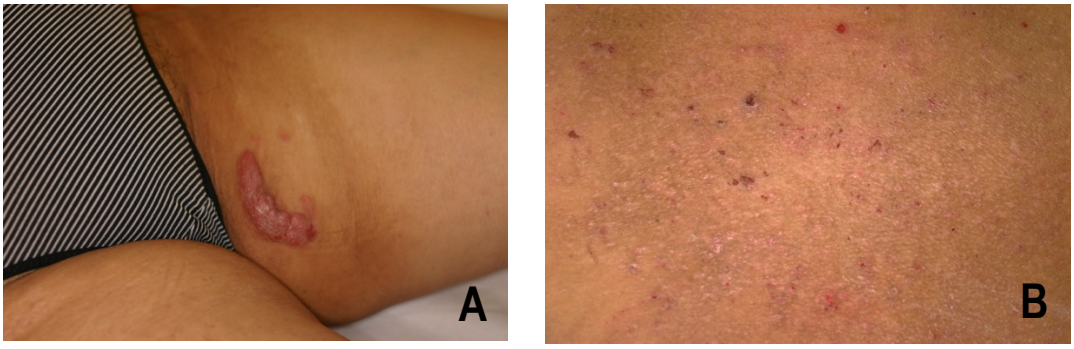



**Figure 1. Mycosis Fungoides. Initial tumor pink-colored lesion, located in the inner thigh area (A) and involvement of the follicles in progression to folliculotropic mycosis fungoides (B).**

In December 2012, a new lesion in the right shoulder developed and, after clinical evaluation, a biopsy was performed. The biopsy showed an irregular and atypical lymphoid infiltrate with an intra-dermal nodular distribution, without evidence of epidermotropism (**Figure 2**). Immunophenotypic analysis showed positivity for CD3-CD4 in 80 percent of the lymphoid component, CD8 in 30-40 percent of the lymphoid component, isolated positivity of CD30 and positivity for CD79α-CD20 in 10 percent of the lymphoid component (**Figure 3, Figure 4, Figure 5**). This was very similar to

the first biopsy but the depth of the infiltrate was more compatible with "*mycosis fungoides in tumor stage*". The patient was assessed by the Hematology department, where they ruled out systemic neoplasm through a bone marrow biopsy that showed no evidence of tumor involvement.

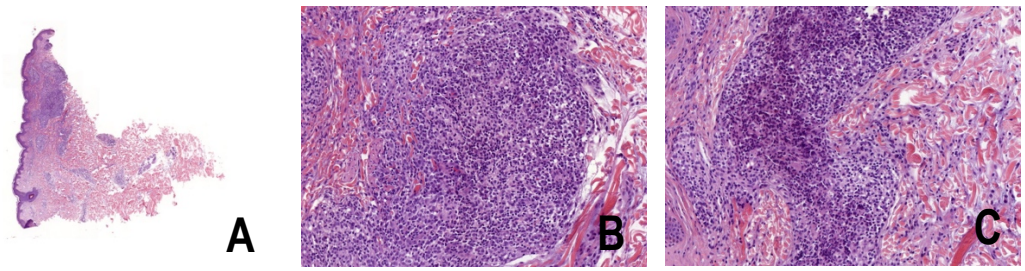



**Figure 2. Images corresponding to the skin biopsy performed in December 2012, stained with hematoxylin-eosin. Panoramic view of shoulder skin (A) that shows an irregular and atypical lymphoid infiltrate with nodular distribution (B, C). Diagnosis was: "*mycosis fungoides in tumor stage*".**

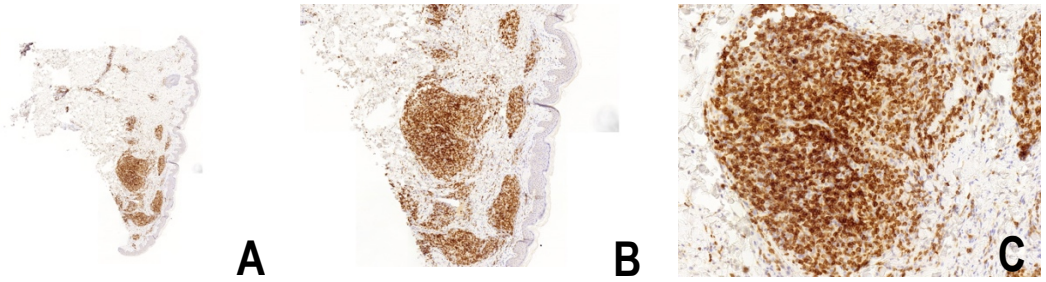



**Figure 3. Images corresponding to the skin biopsy performed in December 2012, in three different magnifications, submitted to CD3 markers, with clear positivity of the nodular lymphoid infiltrates.**

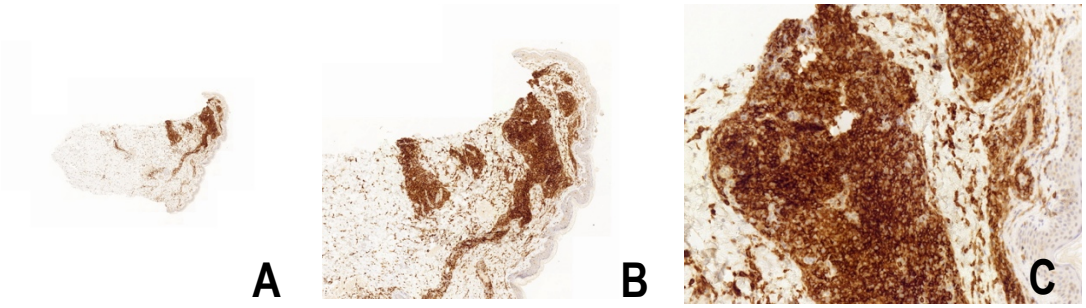



**Figure 4. Images corresponding to the skin biopsy performed in December 2012, in three different magnifications, submitted to CD4 markers, with clear positivity of the nodular lymphoid infiltrates.**

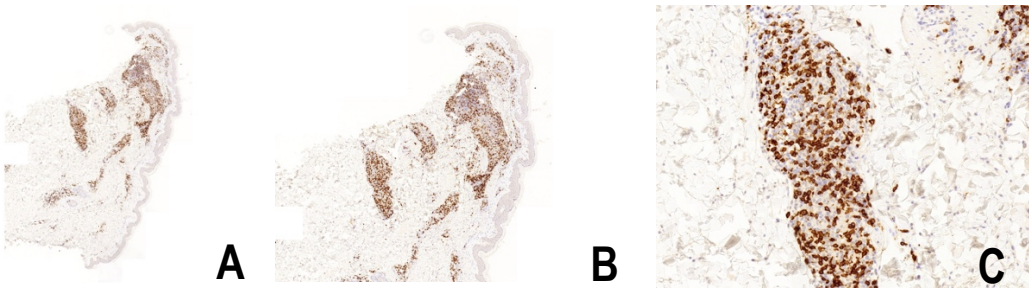



**Figure 5. Images corresponding to the skin biopsy performed in December 2012, in three different magnifications, submitted to CD8 markers, with little positivity of the nodular lymphoid infiltrates.**

In the course of the disease, she presented scorched and lichenified lesions on the back and, to a lesser extent, also in the abdomen, antecubital flexures and thighs. Due to those new clinical findings, in 2013 a new punch biopsy was performed. The new biopsy showed similar findings to the previous biopsies and perpetuated the diagnosis of "*mycosis fungoides in a tumor stage"*. Among the biopsies performed, some lesions were not lymphoma, but rather non-tumoral skin lesions, such as concomitant folliculitis with the neoplastic process.

In 2014, after PUVA treatment with poor results, new tumor lesions that occurred as grouped follicular patches and plaques appeared (**Figure 1B**). The dermatologist performed a biopsy which included skin annexes and abundant subcutaneous cellular tissue. It showed a superficial crust with epidermis of irregular thickness and dermal fibrosis of cicatricial aspect. An irregular lymphoid infiltrate was observed, surrounding the follicles and infiltrating the follicular epithelium, in the form of "Pautrier microabscesses". Immunophenotypic analysis found out positivity for CD3-CD4 in 80% of lymphoid component, highlighting the infiltration of the follicular sheath; isolated CD30, CD79α and CD20 positive accompanying cells. Histopathological diagnosis was informed as "*folliculotropic mycosis fungoides*" (**Figure 6, Figure 7**).

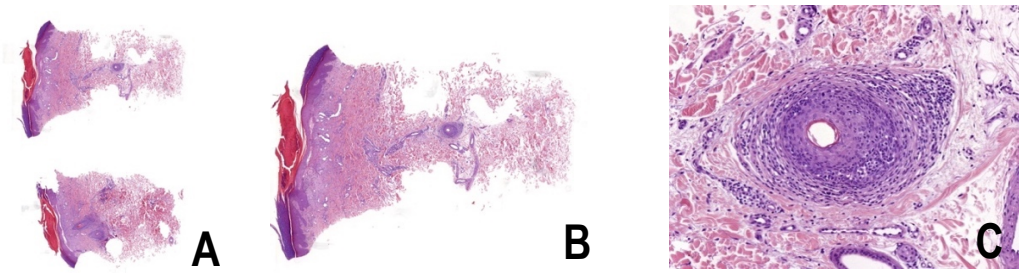



**Figure 6. Images corresponding to the skin biopsy performed in 2014, stained with hematoxylin-eosin. A, B. Panoramic view of fragment of skin with annexes and abundant subcutaneous cellular tissue. C. Irregular and atypical lymphoid infiltrates surrounding a follicle and infiltrating the follicular epithelium. Diagnosis was "*folliculotropic mycosis fungoides*".**

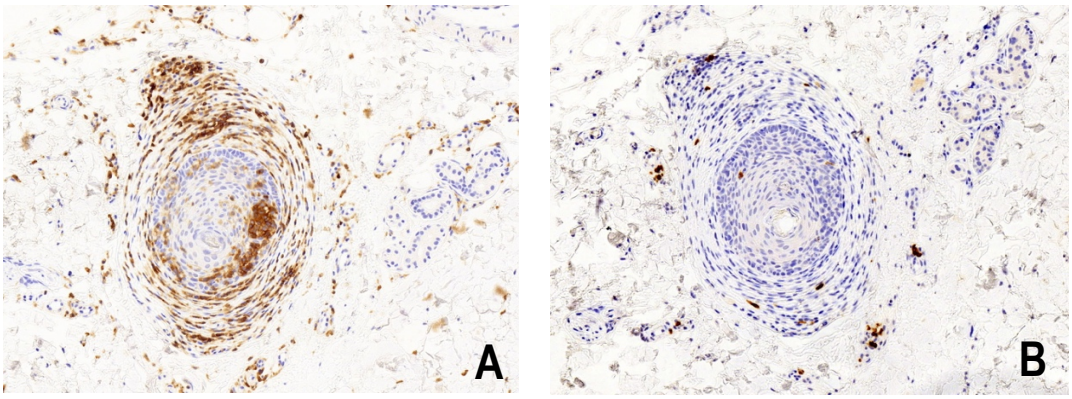



**Figure 7. Images corresponding to the skin biopsy performed in 2014, submitted to CD4 (A) and CD8 (B) markers, showing positivity for the first one among neoplastic lymphoid cells.**

After initiating treatment with interferon-α with partial response and important side effects, biopsies of new lesions appearing in the back and the occipital region were performed. Both biopsies were compatible with infiltration by the already known lymphoma; one of them was "*mycosis fungoides in tumor stage"* and the other corresponded to a "*folliculotropic mycosis fungoides"*.

In June 2015, the patient presented bilateral new mediastinal and hilar pathological lymphadenopathies, together with three new pulmonary micronodules on CT. The patient was referred to the Hematology service, with suspicion of dissemination of the primary cutaneous lymphoma. The lymphadenopathies showed uptake in PET-CT, so in order to categorize these lesions, a transbronchial biopsy (EBUS) was performed.

Samples showed groups of small lymphocytes without atypia and cylindrical contaminating epithelial cells. No cells suspected of malignancy or granulomas were observed, not being possible to rule out sarcoidosis or dissemination due to cutaneous lymphoma. At that time, she also presented new subcutaneous lesions in the left thigh, whose biopsy showed non-necrotizing granulomas, occasionally confluent, delimited by a mild chronic inflammatory infiltrate. The study of SBP, Zielh Neelsen stain and Fite were negative. Molecular studies showed no clonal rearrangement for the TCR gene. The diagnosis was "*non-necrotizing granulomatous inflammation of sarcoid type*" with no infiltration due to lymphoma observed in this sample.

In August 2015, she developed two new lesions in the abdomen and a biopsy was taken from one of them, revealing a fragment of skin where orthokeratosis in epidermis was observed, without evidence of epidermotropism. The underlying dermis was compromised by a dense and diffuse mononuclear infiltrate that affected the entire thickness of the punch, with occasional eosinophils intermixed. Medium-large cells with an irregular nucleus were spotted, some with a visible eosinophilic nucleolus. Additionally, there were some lymphocytes, most of which had an irregular nucleus. Immunophenotypic studies carried out showed that the atypical population of medium-large cells expressed CD4 and CD5 with partial loss of CD3 and CD2 expression and complete loss of CD7 expression. Also, 40% of the cells expressed CD30 weakly. ALK-1 was negative. CD8 and CD79α were positive in the peripheral populations accompanying T and B lymphocytes, respectively. Histopathological diagnosis was "*tumor mycosis fungoides transformed to a large cell, CD30-*" (**Figure 8, Figure 9)**. Given the long evolution of the case, as well as its clinicopathological interest, this biopsy was reviewed jointly with pathologists from the Clinic Hospital (Barcelona).

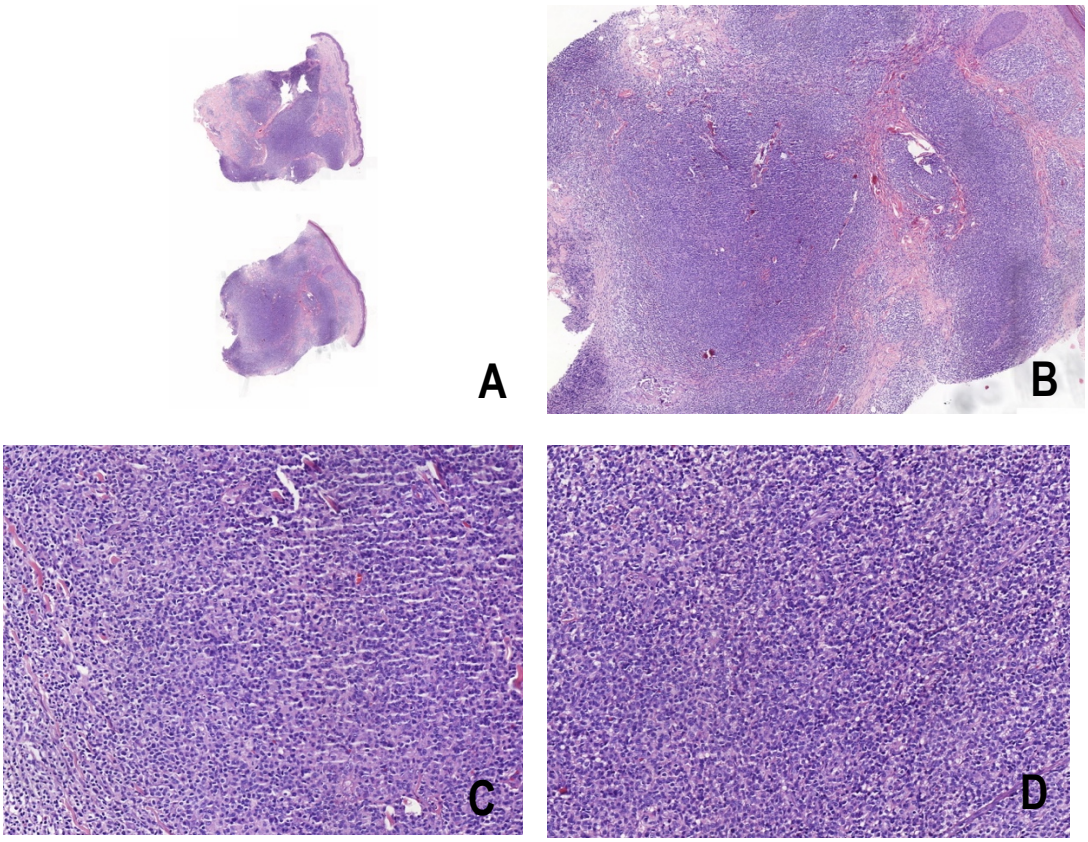



**Figure 8. Images corresponding to the skin biopsy performed in August 2015, stained with hematoxylin-eosin. A. Panoramic view of a skin fragment from abdomen. B, C, D. Increased view of diffuse lymphoid infiltrate with eosinophils intermixed and atypical large cells with irregular nuclei. Diagnosis was "*mycosis fungoides transformed to a large-cell*".**

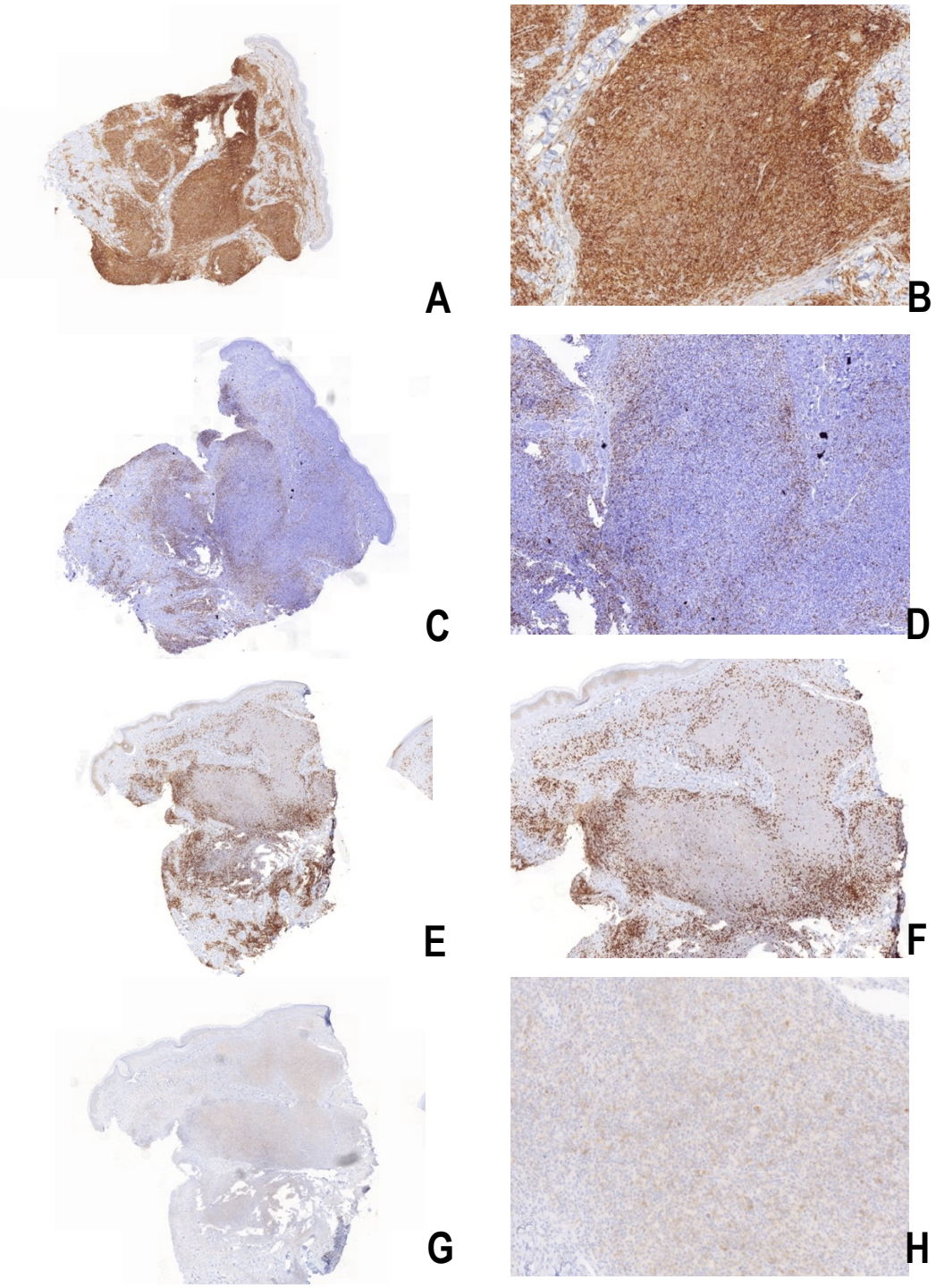



**Figure 9. Images corresponding to the skin biopsy performed in August 2015. A, B. Panoramic and magnified view, respectively, of CD4 markers with evident positivity. C, D. Panoramic and magnified view, respectively, of CD7 markers, which show loss of positivity of neoplastic cells. E, F. Panoramic and magnified view, respectively, of CD8 markers with positivity in accompanying cells. G, H. Panoramic and magnified view, respectively, of CD30 markers, with weak positivity.**

In recent years, biopsies taken after the appearance of new tumor lesions after different treatments (bexarotene, interferon, radiotherapy) perpetuated the diagnosis of "*Mycosis fungoides transformed to large cell lymphoma, CD30-*".

### 4.1.2. Bibliography review

Mycosis fungoides (MF) is the most common type of primary cutaneous T-cell lymphoma, which clinically usually presents in form of patches, plaques or tumors and in more advanced stages may present extracutaneous disease, usually affecting the liver, spleen, lung and peripheral blood [6,7].

*Patches* indicate any size skin lesion without significant elevation or induration, they are usually erythematous lesions, with a predilection for not exposed areas; *plaques* indicate any size skin lesion that is elevated or indurated, they are characterized by infiltrated, irregular and erythematous lesions; *tumors* indicate at least one 1-cm diameter solid or nodular lesion with evidence of depth and/or vertical growth, they are usually solitary and may remain stable for months or grow rapidly. Mucosal involvement has been described in advanced stages, as well as the appearance of erythroderma in approximately 15 percent of mycosis fungoides, which must be differentiated from Sèzary syndrome. Other less frequent signs that can appear in mycosis fungoides are vesicular or purpuric lesions, ichthyosiform skin, keratosis pilaris-like lesions, opportunistic infections or alopecia. One of the most frequently described symptoms in mycosis fungoides is pruritus [4,6,7].

Histopathological characteristics of MF vary according to the stage of the disease. Early stages present a patchy lichenoid or band-like infiltrate in the papillary dermis, where small lymphocytes predominate. In this stage, epidermotropism may be spotted, but Pautrier's microabscesses are rare to find. Some findings that can facilitate diagnosis in this stage are haloed-lymphocytes, exocytosis, lymphocytes on the basal layer of the dermis, epidermal lymphocytes of a larger size than dermal lymphocytes, hyperconvoluted intraepidermal lymphocytes or areas of low spongiosis with numerous lymphocytes. Plaques show a dense lymphocyte infiltrate in the upper dermis, with marked epidermotropism and, in some cases, Pautrier's microabscesses. Although these are considered a classic pathognomonic finding of MF, they are usually

absent in early and tumor lesions. Tumors reveal a nodular or diffuse infiltrate that affects the entire dermis and, occasionally, the subcutaneous fat, with frequent transformation to large cell lymphoma. Large cell transformation is defined as the presence of large lymphocytes exceeding 25 percent of the infiltrate or of large lymphocytes arranged in nodules [6,7].

Mycosis fungoides typically expresses an α/β T-helper phenotype: TCRβ+, TCRγ-, CD2+, CD3+, CD4+, CD5+, CD7-, CD8-, CD45Ro+, TIA1- and most are negative for cytotoxic markers. In tumor stage, lymphocytes may lose expression of CD2, CD3 or CD5 and other less frequent phenotypes have been described, such as CD8+, CD4+/CD8+, CD4-/CD8- or CD56+. Expression of CD30+ has been described in 40-50 percent of large-cell transformed MF, although isolated CD30+ cells are not uncommon in classical MF, increasing in number as the degree of the disease increases [5,6,7].

Most part of MF present clonal rearrangement for the TCR genes, but in early stages it may be difficult to find clonal rearrangements, due to a scarce tumor infiltrate. Likewise, clonal rearrangements can occur in benign reactive processes [5,6,7].

There are three subtypes of mycosis fungoides included in the revised 2018 WHO Classification: folliculotropic mycosis fungoides (FMF), pagetoid reticulosis and granulomatous slack skin. FMF presents with follicular papules, acneiform lesions, keratosis pilaris-like lesions, cysts or indurated plaques and tumors. FMF preferentially involves the head and neck, but cases of lesions on the trunk and limbs as the sole site of involvement have been described. Histologically, it is a variant of MF formed by atypical CD4 T-cells with epidermotropism, which surround the follicles and may or may not be accompanied by mucinosis. Other inflammatory cells, such as plasma-cells or eosinophils, are abundant in the perivascular and perianexal spaces and large-cell transformation is not uncommon. FMF phenotype is the same as that of classical mycosis fungoides [8,9,10].

Patients with limited classic MF have an excellent prognosis. In advanced stages, prognosis is poor, especially for those with skin tumors and/or extracutaneous dissemination. Early stage FMF has a favorable prognosis, similar to that of early

mycosis fungoides, with a five-year survival rate of 94 percent; but advanced-stage FMF presents a five-year survival rate of about a 69 percent. Lack of response to treatment, age over 60 years, elevated levels of lactate dehydrogenase and histological transformation are adverse prognostic data [4,8].

## 4.2. PRIMARY CUTANEOUS GAMMA-DELTA T-CELL LYMPHOMA

### 4.2.1. Case development

In late 2015, a sixty-five-year-old woman was referred to the department of Dermatology for presenting a subcutaneous lesion on the inner side of the left thigh for two months (**Figure 10A**). She was asymptomatic and did not remember any traumatic background. Physical examination showed an erythematous plaque under which an indurated lesion of approximately one cm was palpated. A biopsy was taken and sent for evaluation to the laboratory with the clinical diagnostic suspicion of dermatofibroma.

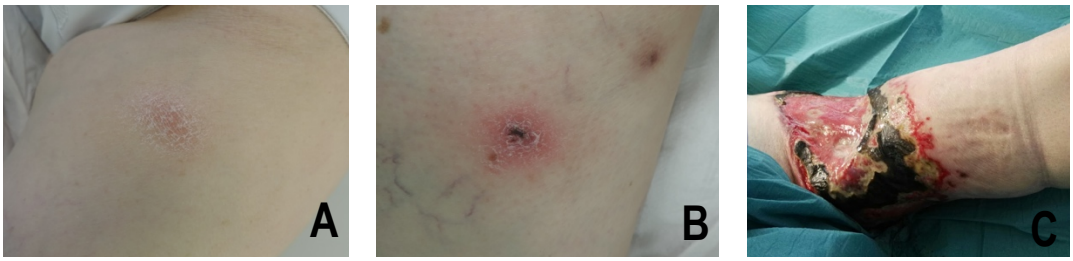



**Figure 10. Primary cutaneous γδ T-cell lymphoma, disease progression. Initial subcutaneous lesion in the inner area of the left thigh (A), nodular and ulcerated lesion in the left thigh with necrotic eschar (B), loss of tissue secondary to cytotoxicity of the lymphoma and debridement (C).**

Pathological study showed a hypodermic increase of fibrous tissue in the septa with presence of an associated inflammatory infiltrate of lymphocytes and macrophages with some giant multinucleated cells, without granulomas. The infiltrate affected the periphery of the lobules, with necrosis of adipocytes giving the diagnosis of "*septal panniculitis*" (**Figure 11**).

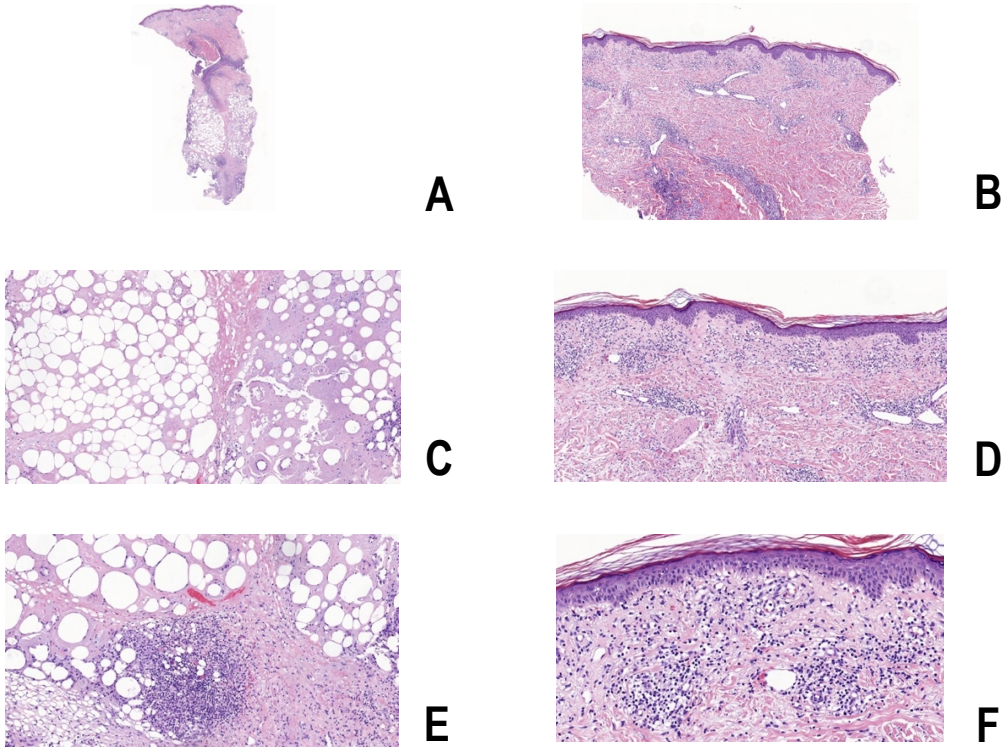



**Figure 11. Images corresponding to the skin biopsy performed in 2015, stained with hematoxylin-eosin, in different magnifications. They show increase of fibrous tissue in the septa, accompanied by inflammatory infiltrates of lymphocytes, macrophages and some giant multinucleated cells and necrosis of adipocytes. Diagnosis was "*septal panniculitis*".**

A month later (January 2016), the biopsied lesion had disappeared and five new subcutaneous and no-painful lesions appeared on the inside of both thighs and one in the right lumbosacral region. None of them was ulcerated, and the patient remained asymptomatic.

In May 2016, new painful and indurated lesions appeared in thighs and abdomen, leaving a scab. A blood analysis was performed finding normal amylase and lipase, negative α1-antitrypsin, normal complement, negative FR, ANA and ENA, and Mantoux was negative as well. Due to changes in the clinical course of the patient, a new biopsy was performed and sent to the laboratory with clinical suspicion of septal panniculitis.

The sample, a fragment of skin of the abdomen, revealed an epidermis with focal vacuolar change. In dermis, perivascular and perifollicular patchy infiltration and

edema were observed, focally interstitial with phenomena of nuclear fragmentation and increase in mucin deposits were also observed. The small dermal vessels showed prominent endothelium without fibrinoid necrosis. The adipose panniculus showed lobular and septal inflammation marked with histiocytes, lymphocytes and occasional polynuclear cells, also some granulomatous structures were identified, without giant cells or changes of necrosis. Ziehl technique was negative, SBP was negative as well and DNA extraction for mycobacterial PCR was also negative. There were no lymphoid accumulations in the adipose panniculus and no atypia was identified. This was summarized as a mixed panniculitis, predominantly lobular with interface dermatitis and deposit of dermal mucin, compatible with "*lupus panniculitis"* (**Figure 12**). This sample and the first biopsy taken in 2015 were sent to the Salamanca Hospital in order to be valued by a reference dermatopathologist, where they confirmed the diagnosis.

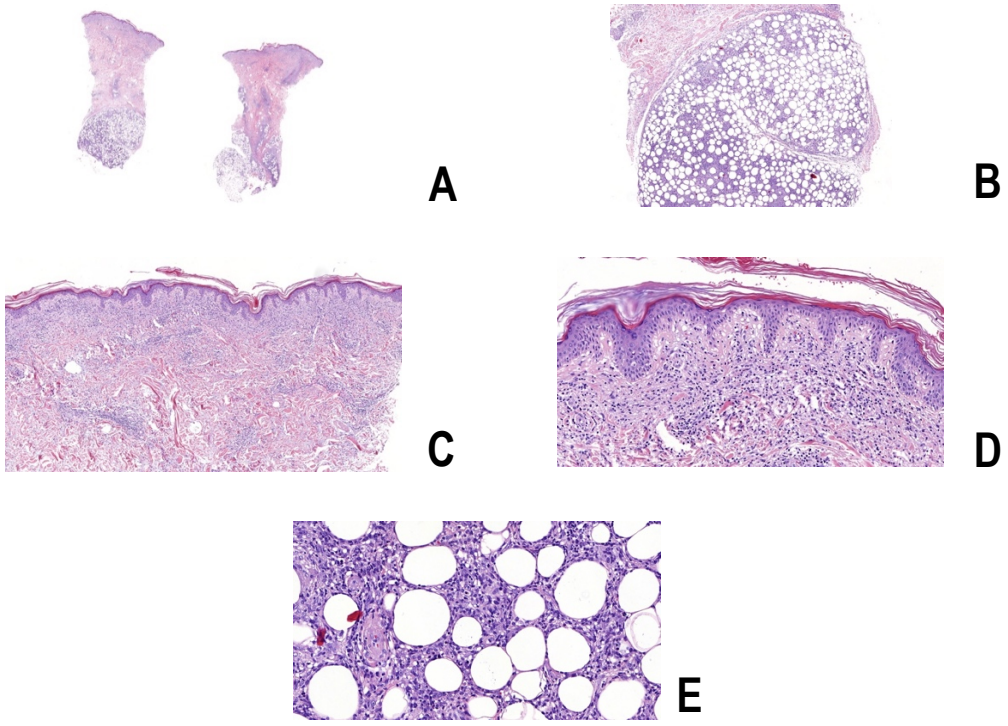



**Figure 12. Images corresponding to the skin biopsy performed in May 2016, stained with hematoxylin-eosin, in different magnifications. A. Panoramic view of fragment of skin from the abdomen. The adipose panniculus showed lobular and septal inflammation marked with histiocytes, lymphocytes and polynuclear cells, with some granulomatous structures (B, E), with interface dermatitis and deposit of dermal mucin (C, D). Diagnosis at that moment was "*lupus panniculitis*".**

The patient returned to consultation in September 2016, presenting a nodular and ulcerated lesion on the left thigh, that had appeared in January, over which a necrotic ulcer showed up (**Figure 10B**). The patient complained of asthenia, sore throat and fever of 38ºC. Due to clinical and lesional changes, a new biopsy was performed and sent to the laboratory with clinical suspicion of lupus panniculitis.

This biopsy, a fragment of skin of the left thigh showed a necrotic epidermal surface; the dermis was widened and widely occupied, with fibrosis and abundant inflammatory infiltrate of acute interstitial distribution, with sitriking phenomena of nuclear fragmentation. Some nuclei were discretely enlarged, with occasional hyperchromasia. Dermal vessels were affected, with transmural inflammation, without findings of frank fibrinoid necrosis. Isolated adipocytes surrounded by the changes described above were observed. No granulomatous structures were observed, PAS staining, colloidal iron and alcian blue did not provide data of interest. Histological diagnostic impression at this point was "*dermal necrosis with accompanying vascular involvement"*. Because of the lack of pathological or clinical suspicion of lymphoma, no lymphoid markers were performed at that moment (**Figure 13**).

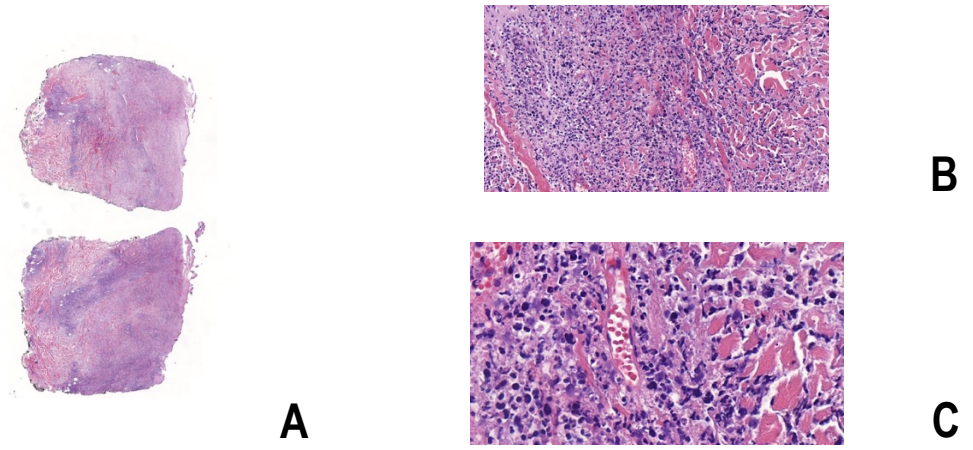



**Figure 13. Images corresponding to the skin biopsy performed in September 2016, stained with hematoxylin-eosin. Necrotic epidermal surface (A); dermis occupied by inflammatory infiltrate, with some hyperchromatic nuclei and affected vessels, with transmural inflammation (A, B, C). Diagnosis was "*dermal necrosis with accompanying vascular involvement*".**

The 26th September of 2016, the patient was admitted at the Emergency department, in charge of the Internal Medicine service, due to general deterioration and fever. She was treated with piperacilin-tazobactam and vancomycin was added in the absence of response. An MRI of the left thigh was requested and an involvement of subcutaneous cellular tissue and signs of myositis without abscess or osteomyelitis were observed. A CT scan was performed, but no suspicious images were seen, except for splenomegaly. The Hematology service ruled out hematological neoplasia, since the bone marrow biopsy requested was negative. The Traumatology service decided to debride the injury and refer the patient to the Plastic Surgery Department of Donostia Hospital (Donostia - San Sebastián). The samples taken at Donostia Hospital during the months of October and November 2016 showed chronic and acute inflammation or panniculitis, without evidencing neoplasia.

During her stay in Donostia Hospital, she had to be transferred to the ICU, due to acute renal failure secondary to septic shock and DVT in the right internal iliac vein and left gonadal vein. She was placed on hemodialysis and another debridement of the lesion was performed (**Figure 10C**).

After the development of a hemophagocytic syndrome, clinical services informed the Pathology department, that carried out an exhaustive review of all the biopsies and pieces obtained so far. Signs of atypia were observed in the lymphoid cellularity and new markers were performed with the result of positivity for CD2, CD3, CD56 and granzyme B, and loss of markers for CD4, CD5, CD7 and CD8. PD1, CD57, CD30 and TIA-1 were negative. Ki67 proliferative index was 30 percent. HIS EBER was also negative (**Figure 14)**.

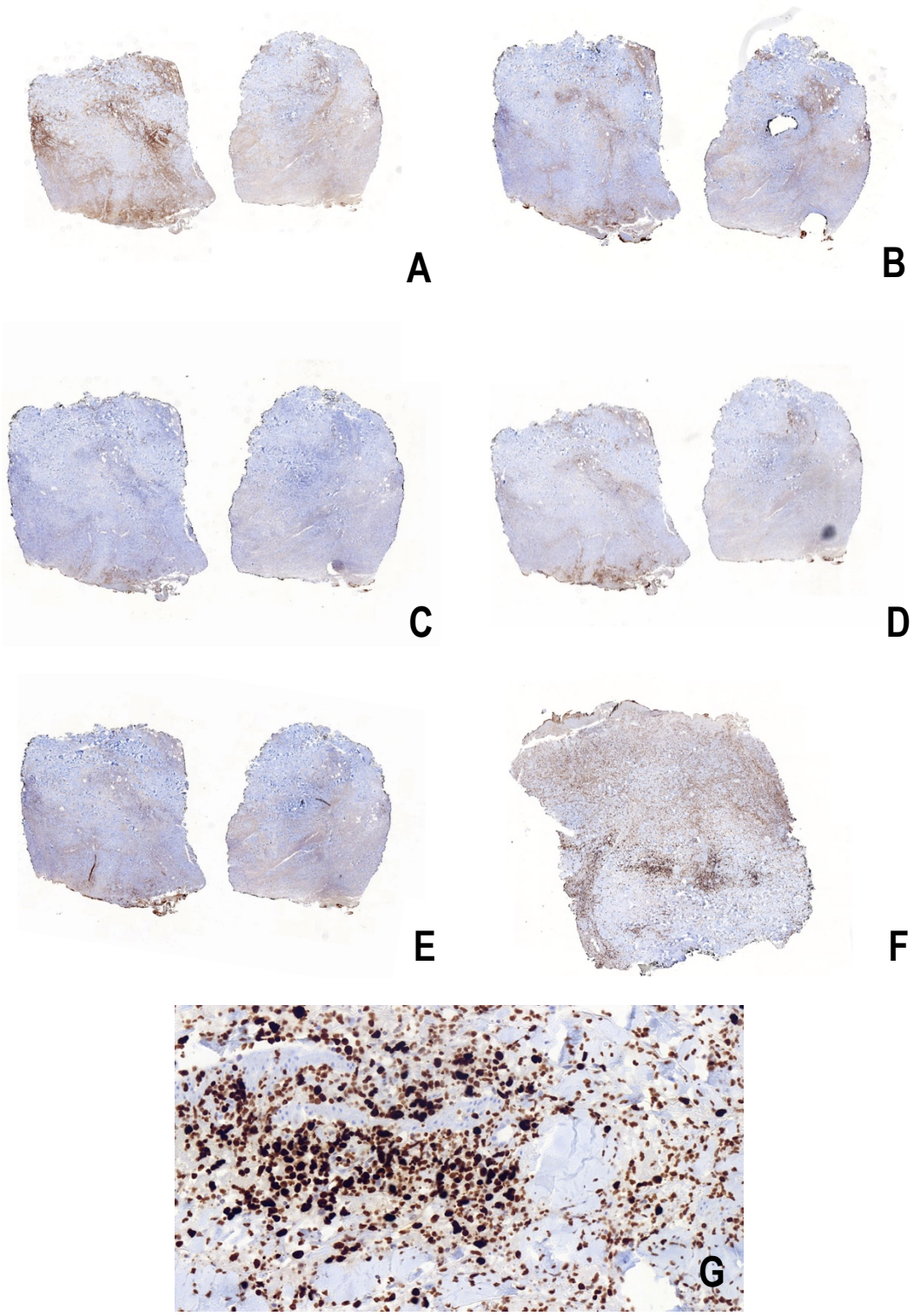
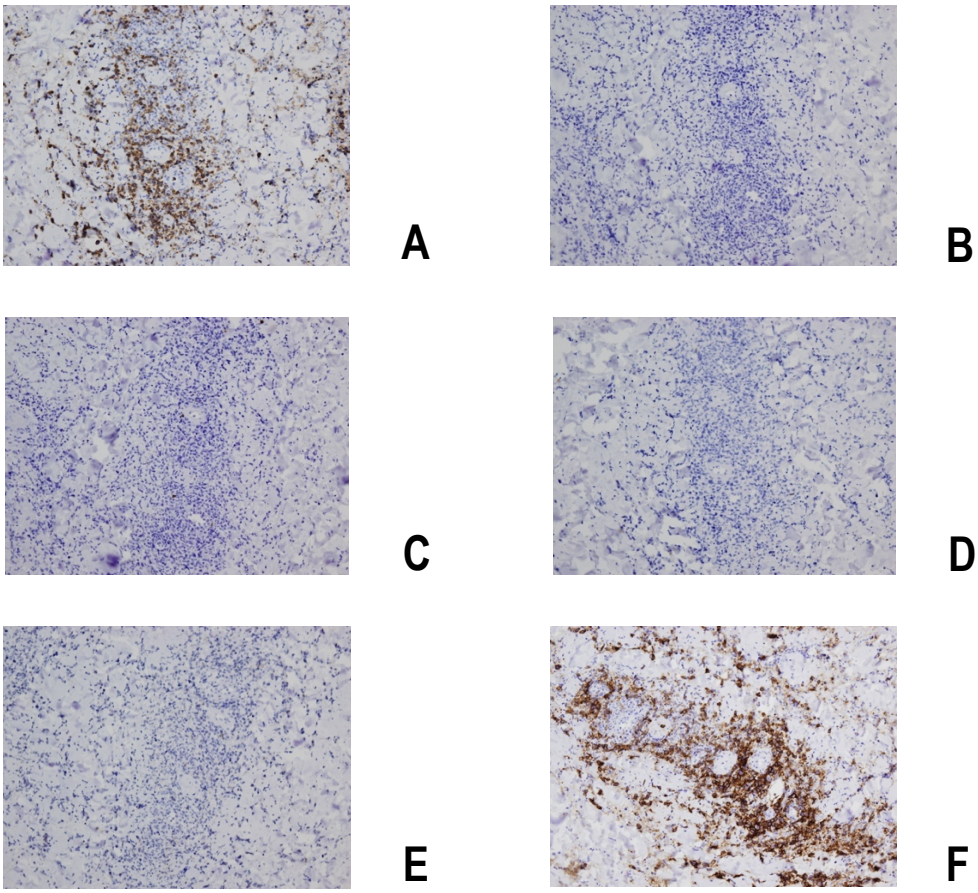



**Figure 14. Images corresponding to the reviewed biopsy performed in September 2016, submitted to different specific markers, which show positivity for CD3 (A), and loss of markers for CD4 (B), CD5 (C), CD7 (D) and CD8 (E). Ki67 proliferative index was 30 percent (F, G).**

This review led to a change in the diagnostic orientation of the case, proposing the possibility of a lymphoproliferative process and, given the initial panniculitis and the clinicopathological course of the disease, the proposed entities were "*primary cutaneous gamma-delta T-cell lymphoma"* and "*subcutaneous panniculitis-like T-cell lymphoma"*. Because more specific markers were needed, the samples were sent the 30th November 2016 to the Valdecilla Hospital (Santander) for diagnostic confirmation. They confirmed the previous findings and the specific immunohistochemical study showed positivity of the atypical cells for CD3, CD56, Granzyme, Perforin, TCR gamma, Ki66 of 70%, with negativity for CD20, CD4, CD8, CD30, TCRβF1, PD1, EBV-LMP1 and EBER. Histopathological diagnosis was "*subcutaneous thigh tissue with primary cutaneous gamma-delta T-cell lymphoma*", confirming the diagnostic suspicion of our hospital.



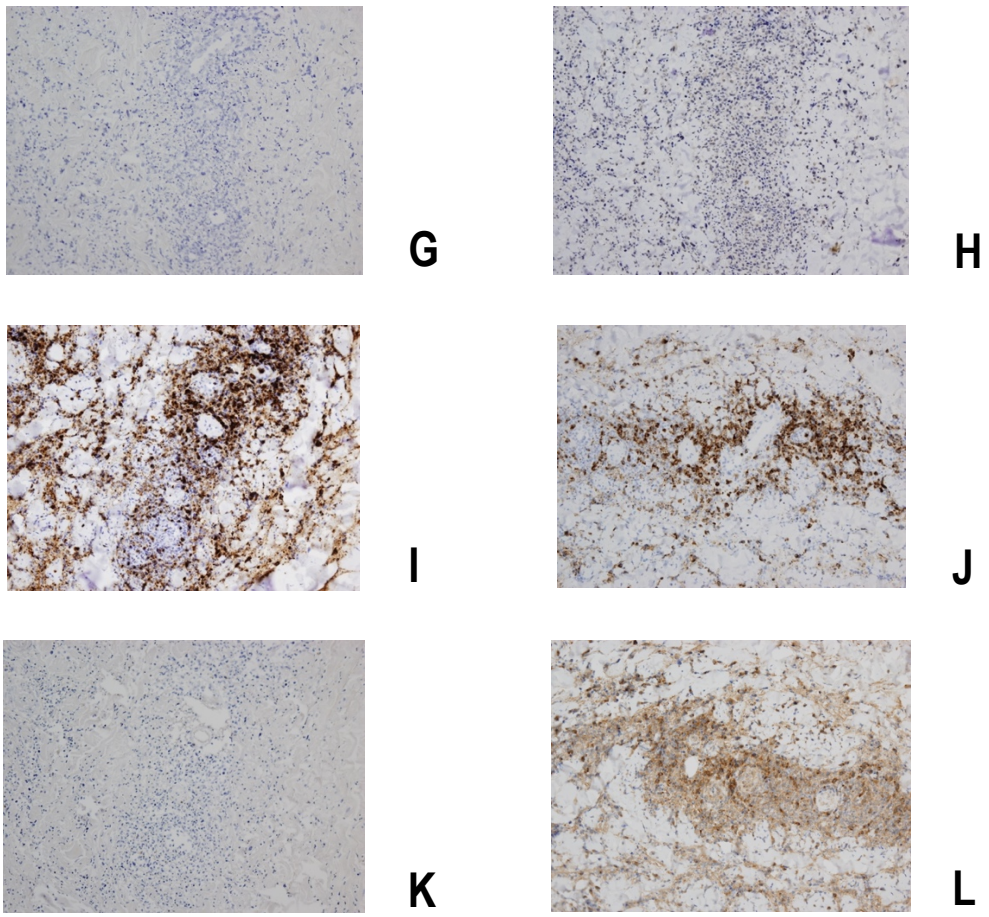



**Figure 16. Images corresponding to new specific markers applied to the previous biopsy, resulting in positivity of the atypical cells for CD3 (A), CD56 (F), Granzyme (I), Perforin (J), TCR gamma (L) and negativity for CD20 (D), CD4 (B), CD8 (C), CD30 (E), TCRβF1 (K), PD1, EBV-LMP1 (H) and EBER (G).**

In December 2016, the patient was transferred to the Hematology service of the HUA. When the Dermatologist assessed the patient, she was disoriented in time and space. The patient died days later.

### 4.2.2. Bibliography review

Primary cutaneous γδ T-cell lymphoma (PCGD-TCL) is a neoplasm composed of a clonal proliferation of mature and activated γδ T-cells, with a cytotoxic phenotype. PCGD-TCL is a rare lymphoma, accounting for <1 percent of all cutaneous lymphomas [3,4,10,11,19].

PCGD-TCL predominantly affects adults, with an average age of diagnosis of 60 years, although pediatric cases (12-15 years) have been described [3,4,10,11]. It presents

with ulcero-necrotic plaques, nodules or tumors that affect predominantly the extremities, although other sites may be involved [3,4,10,11,18,19,20]. Cases with MF-like or psoriasis-like erythematous and scaly patches, that later evolve into aggressive lesions with ulceration have been described [12]. Dissemination to mucosal and other extranodal sites is common, but lymph nodes, spleen and bone marrow are not involved usually [3,4,11,19]. Patients may present a hemophagocytic syndrome (25-30%), more particularly in those with panniculitis involvement, and accompanying B symptoms such as fever, night sweats and/or weight loss [3,4,5,11,18,19,20].

Histologically, PCGD-TCL is characterized by presenting epidermotropic, dermal and subcutaneous histological patterns, solely or, more usually, in combination [3,4,5,10,11,18,19,20]. Epidermal involvement can go from a mild epidermotropism to marked pagetoid reticulosis-like infiltrates, which may associate with intraepidermal vesiculation and necrosis [11,19]. Dermal involvement can occur from mild perivascular to deep nodular to diffuse lymphoid infiltrates [18]. Subcutaneous involvement is characterized by infiltration of the fat lobes with focal involvement of the septa and rimming of individual adipocytes, in a way that may be confused with the subcutaneous panniculitis-like T-cell lymphoma (SPTCL), but it shows more accentuated infiltration of the dermis and/or epidermis [4,5,18,19,20]. Neoplastic cells are medium-large sized, with pleomorphic nuclei and coarsely clumped chromatin. Angiocentricity, angiodestruction and necrosis are common, due to the cytotoxic phenotype of this lymphoma [3,4,5,10,11,18,19,20]. Some reported features that could complicate the diagnosis of primary cutaneous γδ T-cell lymphoma are mucin deposition, simulating lupus erythematosus, or subcutaneous infiltrates with minimal atypia and a histiocytic mixture, simulating a non-specific panniculitis, which eventually ends up showing more specific biopsy findings [10].

PCGD-TCL cells express a TCR γδ+, TCR αβ-, CD2+, CD3+, CD5-, CD7+/- and CD56+ phenotype, with marked expression of cytotoxic proteins, including Granzyme B, perforin and TIA1. Most cases are double negative for CD4 and CD8, although cases that express CD8 positivity have been described, as well as cases of TCR γδ and TCR αβ co-expression. Molecular studies typically show a monoclonal rearrangement of the TCR genes [3,4,5,10,11,18,19,20].

Primary cutaneous γδ T-cell lymphoma is an aggressive lymphoma usually associated with therapy resistance and a median survival of 12-15 months. Some studies have shown that subcutaneous fat involvement have a poorer prognosis, although cases with indolent behavior have been reported [3,4,10,11,13,18].

## 4.3. PRIMARY CUTANEOUS MARGINAL ZONE (MALT) LYMPHOMA

### 4.3.1. Case development

In July 2013, an eighty-eight-year-old man was referred to the Dermatology department for presenting pruritic lesions on the trunk, upper limbs and scalp that lasted two months (**Figure 17**). On examination, he presented indurated, erythematous-violaceous lesions, with a deep component, without epidermal involvement. The patient was afebrile and had no accompanying symptoms. In the blood analysis, he had high IgE levels, the rest being normal. Two biopsies were taken from the back and sent to the laboratory with the clinical differential diagnosis of prurigo, cutaneous lymphoma, pseudolyphoma, Hailey-Hailey disease and bullous pemphigoid disease.

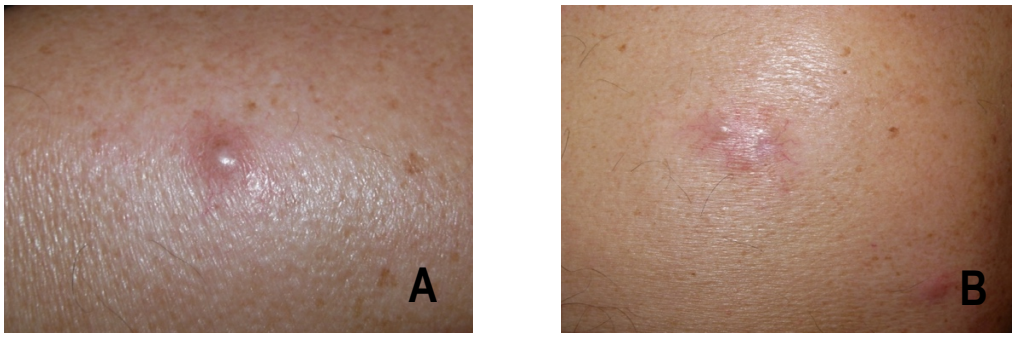



**Figure 17. Cutaneous B-cell marginal zone lymphoma. Indurated, erythematous-violaceous lesions, with a deep component, without epidermal involvement (A, B).**

Histologic study showed two skin fragments that included skin adnexa and abundant subcutaneous cellular tissue. Both showed a lymphoid infiltrate of nodular, perivascular and periadnexal disposition, with exocytosis and isolated spongiosis without evident epidermotropism. No vasculitis or granulomas were present. Immunophenotypic analysis was positive for CD3 and CD4 in 80% of the cells, CD79α and CD20 were positive in more than 50% of the cells, CD30 was positive in

isolated cells, CD43 and BCL-2 were also positive, favoring the diagnosis of "*primary cutaneous marginal zone (MALT) lymphoma"* (**Figure 18, Figure 19, Figure 20**).

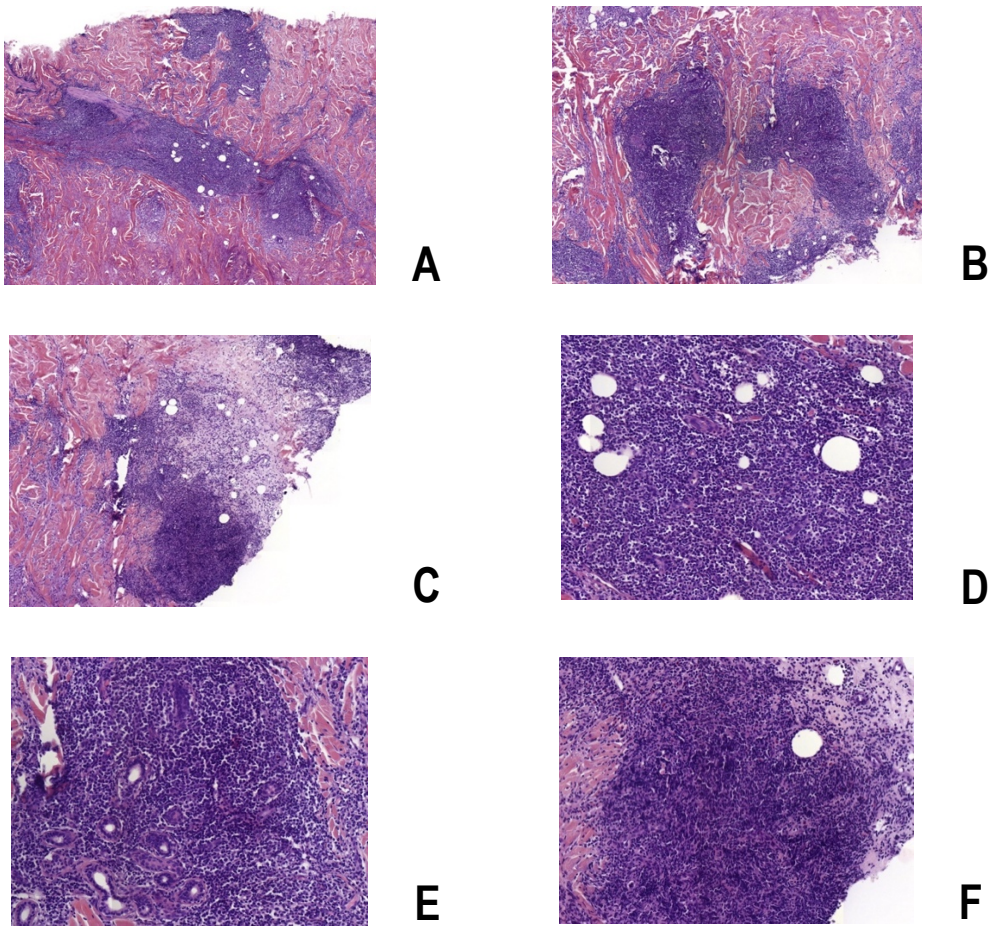



**Figure 18. Images corresponding to the skin biopsy performed in July 2013, stained with hematoxylin-eosin, that included skin adnexa and subcutaneous cellular tissue, showing a lymphoid infiltrate of nodular, perivascular and periadnexal disposition, with exocytosis and isolated spongiosis.**

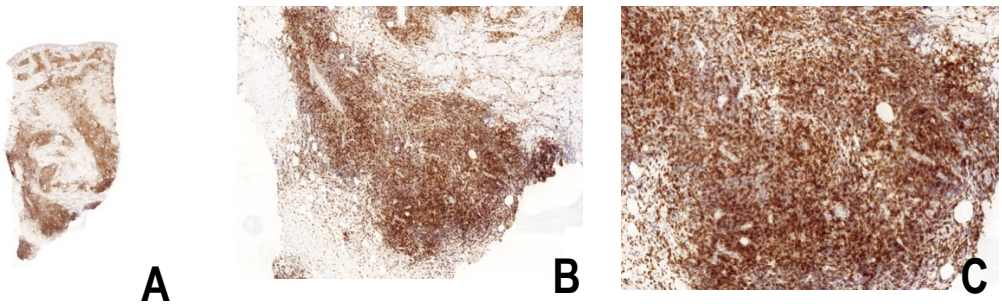



**Figure 19. Images corresponding to the skin biopsy performed in July 2013, submitted to CD3 markers, that show clear positivity of neoplastic cells.**

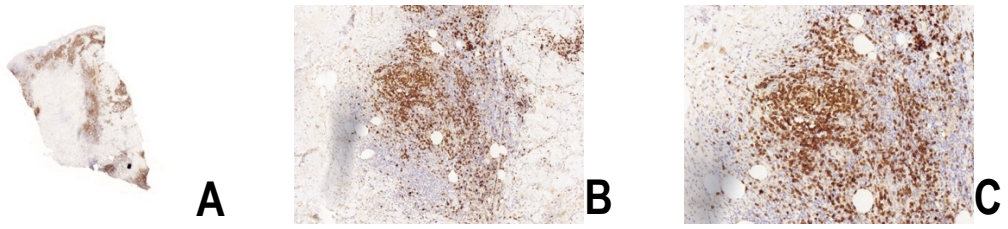



**Figure 20. Images corresponding to the skin biopsy performed in July 2013, submitted to CD79α markers, that reflect positivity in 50 percent of neoplastic cells.**

The patient was referred to the Hematology department to determine whether it was a primary cutaneous lymphoma or a cutaneous affectation secondary to a systemic lymphoma. The myelogram did not present significant morphological alterations. The bone marrow biopsy showed interstitial and nodular infiltration by a B CD79α and Pax5 positive lymphoma, which indicated infiltration of bone marrow by lymphoma, elevating diagnosis to a "*primary cutaneous marginal zone (MALT) lymphoma, stage IV, due to skin and bone marrow involvement*" (**Figure 21**).

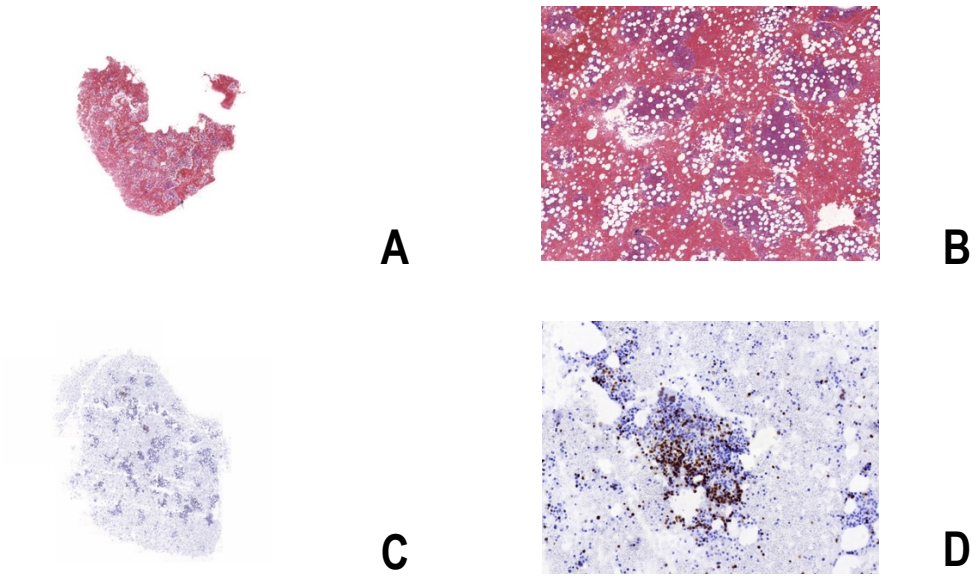



**Figure 21. Images corresponding to the bone marrow biopsy, stained with hematoxylin-eosin, showing an interstitial and nodular lymphoid infiltrate (A, B) that presents Pax5 positivity (C, D).**

In December 2013, a new biopsy of the left frontal area was performed due to the development of a new papule, to rule out another type of lesion (basal cell carcinoma) and confirm it was a lymphoma lesion. The sample revealed a diffuse dermal lymphoid

infiltrate, with a morphologic and immunohistochemical profile similar to the previous biopsy, compatible with the diagnosis of "*primary cutaneous marginal zone (MALT) lymphoma*".

In September 2018, a biopsy of a new erythematous-violaceous nodular lesion on the right cheek was performed. Histological study showed dermal infiltration by a medium-sized lymphoid population, with irregular and atypical nuclei; focally monocytoid appearance, which was arranged in a nodular and diffuse pattern between collagen bands. It cleared the cutaneous annexes for the most part, and respected the papillary dermis and epidermis, reaching the reticular dermis in all its thickness. It was accompanied by some scattered eosinophils, and no accompanying plasma cells were identified. Immunohistochemical study showed positivity for CD20 and CD79α in tumor cells, with aberrant expression of CD43 and positivity for BCL-2; CD3 was positive in the accompanying T component. Diagnosis was "*skin with morphological and immunohistochemical findings compatible with infiltration due to his primary cutaneous marginal zone (MALT) lymphoma, previously diagnosed*" (**Figure 22**).

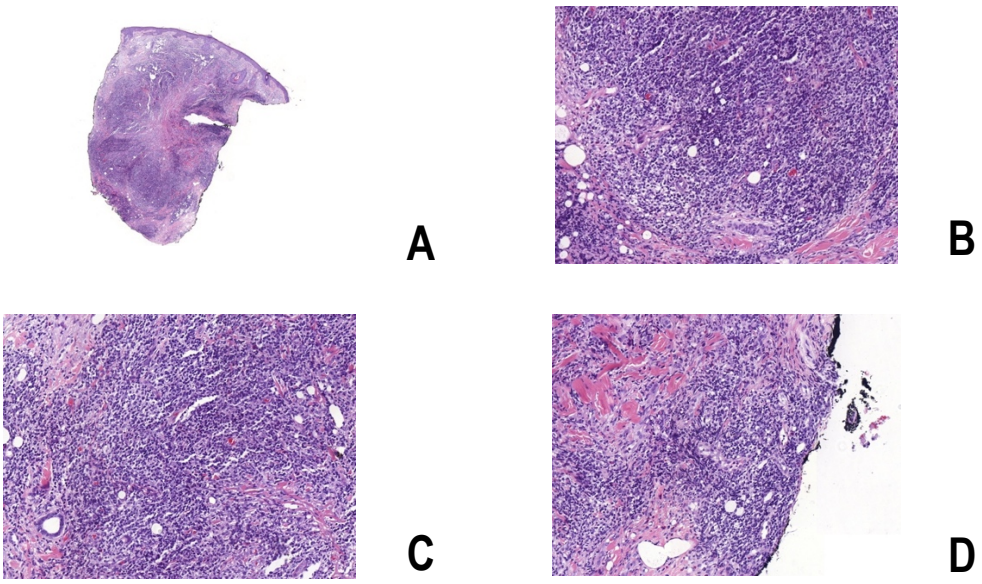



**Figure 22. Images corresponding to the skin biopsy performed in September 2018, stained with hematoxylin-eosin, which showed dermal infiltration by a medium-sized lymphoid population, with irregular and atypical nuclei, arranged in a nodular and diffuse pattern between collagen brands, compatible with infiltration by primary cutaneous marginal zone (MALT) lymphoma.**

Until now, the patient has received treatment with Rituximab and Leukeran, and he is stable, with no dermatological lesions or clinical symptoms related to lymphoma.

### 4.3.2. Bibliography review

Primary cutaneous marginal zone (MALT) lymphoma (PCMZL) is an indolent B-cell lymphoma composed of neoplastic small B cells, plasma cells and reactive T cells. It accounts for 7 percent of all primary cutaneous lymphomas [3] and 30-40 percent of all primary cutaneous B-cell lymphomas [4]. It most commonly affects adults, with an average age at diagnosis of 55 years, with male preponderance [4, 26].

PCMZL usually presents violaceous red papules, plaques or nodules, located on the trunk or upper extremities, that can be pruritic and seldom painful [3,4,24,25,26]. These lesions are more often multifocal, but may also be isolated. Lesions may wax and wane, and can undergo spontaneous regression, which has been associated with anetoderma [3,4]. Recurrences are common, present in 40 percent of cases, more frequently in patients with multiple lesions, but extracutaneous dissemination is exceptional [26]. In recent studies, this lymphoma has been associated with gastrointestinal disorders and autoimmune diseases, as well as with *Borrelia burgdorferi* infection in Europe [4,24].

Histologically, PCMZL presents with nodular or diffuse lymphocytic infiltrates in the dermis, separated from the epidermis by a prominent narrow layer (Grenz zone) and can sometimes extend to the subcutaneous fat. The infiltrate is composed by small lymphocytes, plasma cells and follicles with reactive germinal centers, admixed with numerous reactive T cells. Plasma cells are frequently located in the periphery of the infiltrate and in the subepidermal compartment [3,4,5,24,25,26]. Dutcher bodies (PAS+, immunoglobulin containing, intranuclear inclusions) and Russel bodies (intracytoplasmic inclusions) may be observed [24,25]. It rarely shows colonization of follicular structures, lymphoepitelial lesions or transformation into a diffuse large B cell lymphoma [25].

PCMZL neoplastic cells express positivity for markers CD19, CD20, CD22, CD79α and BCL-2; and express negativity for CD3, CD5, CD10, Cyclin D1 and BCL-6 markers. Reactive germinal centers cells present a CD10, BCL-6 positive and BCL-2

negative phenotype. Plasma cells express CD138 and CD79α, but are negative for CD20 and present restriction of light chains in the immunohistochemical study in 70-75 percent of cases [3,4,5,24,25]. Two subsets of primary cutaneous marginal zone lymphoma have been described: a heavy chain class-switched form (more frequent), and a non class-switched form. The first one expresses IgG, reveals abundant reactive T lymphocytes and few neoplastic B cells. The latter expresses IgM and is usually accompanied by a diffuse proliferation of B cells, expresses CXCR3 and is associated with *Borrelia burgdorferi* infection [3,4].

The clonal rearrangement of heavy and light chain genes of immunoglobulins by PCR helps confirm the diagnosis [26]. In some cases, translocations t(14;18), t(3;14) and t(11;18) have been reported, but it is rare [25].

Primary cutaneous marginal zone (MALT) lymphoma has a favorable prognosis, with a 5-year survival rate of >98 percent, although recurrences are common. Extracutaneous dissemination is rare, but more frequently appears in multifocal disease, non-class-switched form and in cases with transformation [3,4,25].

## 4.4. T-CELL FOLLICULAR TYPE LYMPHOMA

### 4.4.1. Case development

In March 2015, a sixty-year-old woman consulted in the Dermatology department for a generalized pruritus process in trunk, upper limbs and scalp, that lasted two years, despite treatment with antihistamines (**Figure 23**). She presented lymphopenia and a previously performed CT scan showed splenomegaly; therefore, she was being followed-up by the Hematology department. She started phototherapy treatment, but did not produce any improvement.

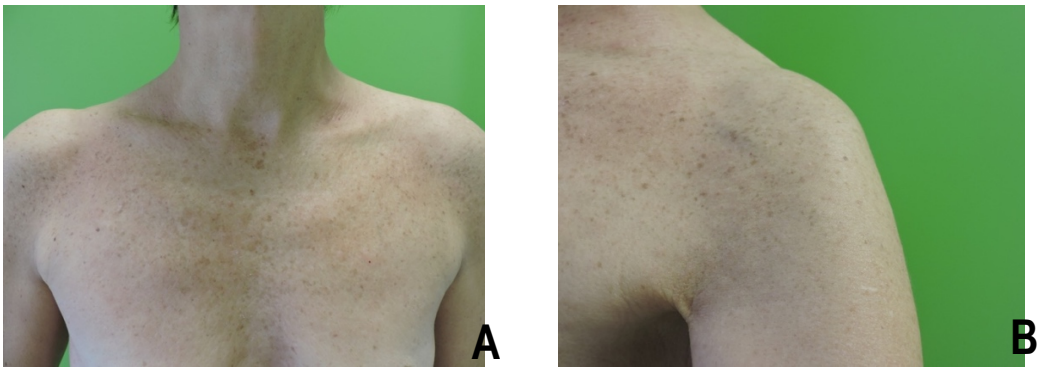



**Figure 23. T-cell follicular type lymphoma. Skin with signs of dryness and slight scratching marks (A, B).**

During the course of the disease, the patient presented a submental adenopathy, a PAAF was performed. Histological analysis showed lymphocytes in different evolutionary stages, histiocytes and non-necrotizing microgranulomatous formations. Immunophenotypical study was positive for CD68 and negative for CD30. At that point, the diagnosis was found negative for malignant cells, compatible with "*non-necrotizing microgranulomatous lymphadenitis*".

After several topical treatments, the patient returned to consultation in July 2017 due to worsening of the pruritus process. The submental adenopathy had increased in size, she continued with lymphopenia and, in the CT requested by Hematology, she presented with enlarged splenomegaly and multiple lymphadenopathies. Along with the Hematology service, the decision was to remove the adenopathy and analyze it to rule out a lymphoma. Also, three skin biopsies from the scapular area, neck and shoulder were performed and sent to the laboratory. Later in August, two submental nodes were removed by the Maxillofacial Surgery department.

Histological study of the three cutaneous biopsies revealed in dermis, and varyingly affecting its entire thickness, an inflammatory infiltrate that formed non-necrotizing granulomas constituted by occasionally multinucleated histiocytes, which were surrounded by a lymphocyte corona. No atypia suggesting malignancy was observed in any of the three biopsies submitted. Immunohistochemical techniques PAS, Giemsa, Grocott, Silver methamine, Zielh and Fite were negative for identification of causal agent. Despite negativity, the process suggested an infectious process, DNA was extracted and sent to the Microbiology department for PCR for Leishmania and

atypical mycobacteria, being the result negative. Histological sections of submental lymph nodes showed abundant small granulomas, not necrotizing, sarcoid type and few eosinophils, that suggested the diagnosis of "*non-necrotizing chronic granulomatous lymphadenitis"* (**Figure 24**).

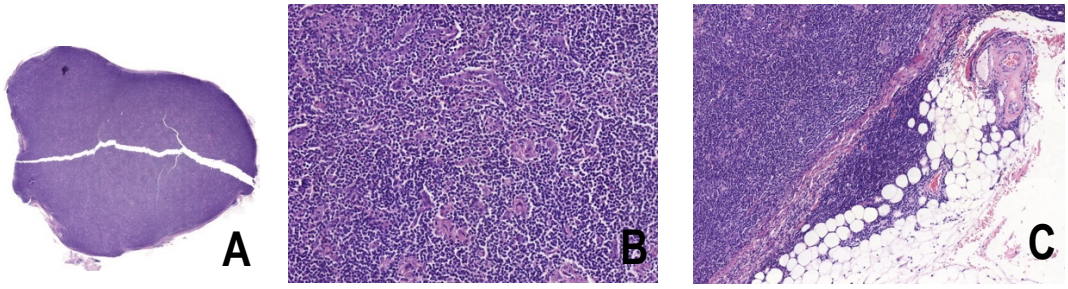



**Figure 24. Images corresponding to the submental-lymph node biopsy performed in 2015, stained with hematoxylin-eosin. Panoramic view of the ganglion (A), showing abundant microgranulomas not necrotizing, sarcoid type and a few eosinophils (B). In the magnified image, the lymphocytes surpass the ganglion capsule (C).**

In September 2017, other infectious processes were discarded and epicutaneous tests were performed, being these negative. In the month of October, due to the patient's torpid evolution, all the biopsies performed on the skin and lymph nodes were carefully reviewed and the case was presented in dermatopathology session, where a new diagnostic orientation of the case was proposed. The possibility of a lymphoproliferative process accompanied by granulomas was suggested. Given the histological characteristics observed during the review of the case, "*angioimmunoblastic T-cell lymphoma"* was thought to be a possible diagnosis and new analysis were performed. Immunohistochemical study showed expression of CD3, partial CD4, PD1, BCL6 and partial CD7 expression by neoplastic cells. No expression of CD8 or CD10 were observed. EBV-EBER was negative and follicular dendritic cell expansion was not observed.

Samples were sent to Valdecilla Hospital (Santander), reference center in the diagnosis of lymphomas, for extension of special techniques battery that could confirm or rule out the diagnostic suspicion. In this center, more specific immunohistochemical markers were added and lymphoma was confirmed: "*lymph node with peripheral*

*lymphoma of follicular T-cell phenotype"*; "*skin with infiltration by T-cell follicular lymphoma"*. Both presented clonal rearrangement for the TCR-gamma gene.

In November 2017, an iliac crest biopsy was performed, revealing nodular infiltration by a T-cell lymphoma, accompanied by microgranulomas (**Figure 25**).

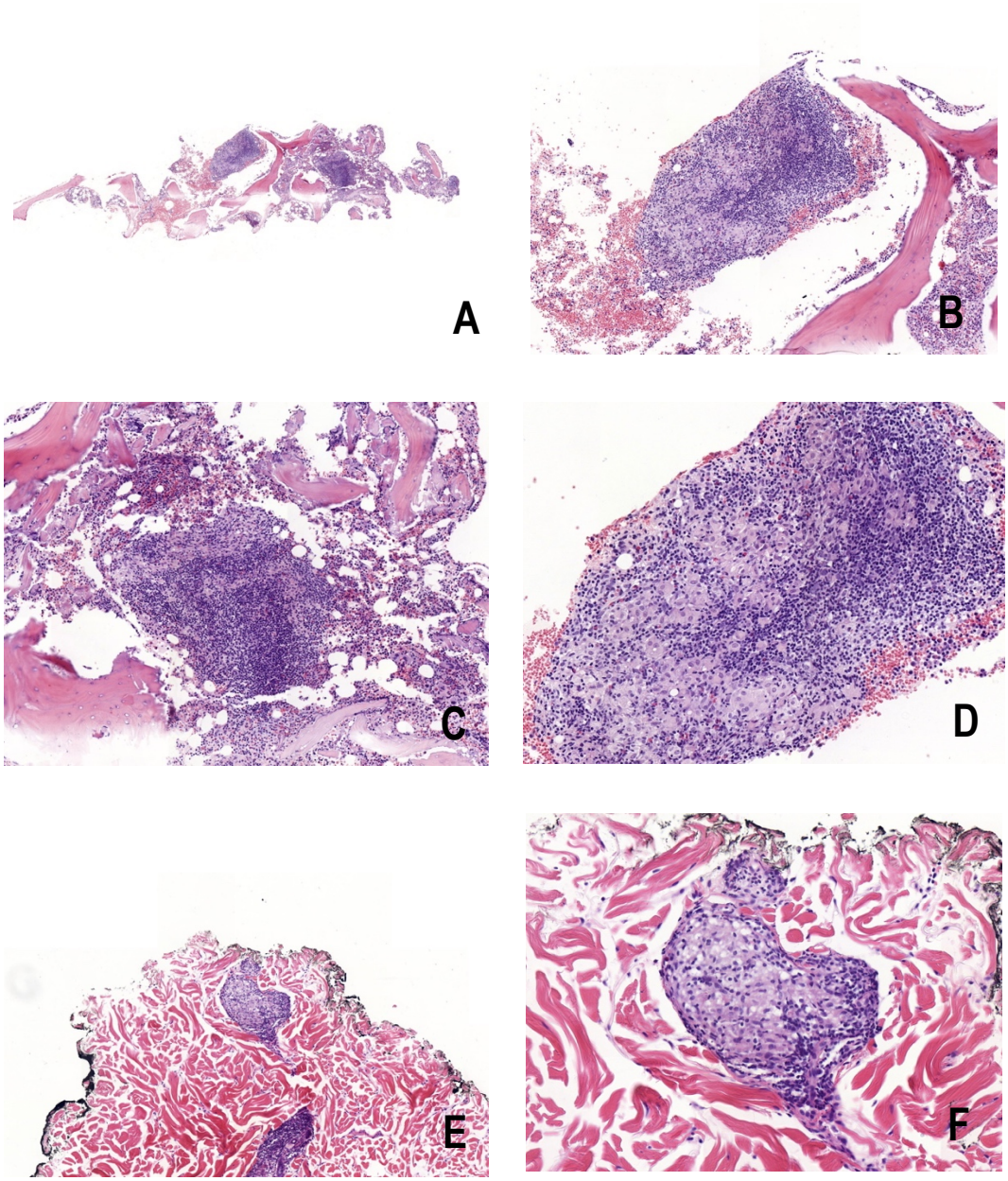



**Figure 25. Images corresponding to the bone marrow biopsy performed in November 2017, stained with hematoxylin-eosin. Overview of bone barrow (A), showing an infiltration similar to that found in the skin biopsy, with nodular infiltration by a T-cell lymphoma (B, C, D), accompanied by microgranulomas (E, F).**

The patient remains currently stable, although she continues with itching associated with the neoplastic process.

### 4.4.2. Bibliography review

Nodal T-cell lymphomas are a heterogeneous group of neoplasms, which include the subtypes of angioimmunoblastic T-cell lymphoma (the most frequent), T-cell follicular type lymphoma and nodal peripheral T-cell lymphoma with T-follicular helper phenotype, no otherwise specified. These last two subtypes were included in the 2016 WHO Classification as provisional entities [3].

T-cell follicular type lymphoma is a node-based lymphoid neoplasm of T-follicular helper cells, with a predominantly follicular growth pattern lacking characteristic features of angioimmunoblastic T-cell lymphoma [29].

This rare lymphoma can present with lymphadenopathies, hepatosplenomegaly and cutaneous rash, as well as laboratory abnormalities similar to those produced by angioimmunoblastic T-cell lymphoma. However, unlike the latter, cases with limited disease and absence of B symptoms have been reported [29].

T-cell follicular type lymphoma is characterized by follicular or perifollicular growth of neoplastic cells with T-helper phenotype, and lacks diffuse polymorphic infiltration and vascularization, characteristic features of angioimmunoblastic T-cell lymphoma [4]. Also, large B-cells may be seen, including HRS-like cells [29].

Immunophenotypic studies reveal the expression of at least two of the following markers: PD1, CD10, BCL6, CXCL13, ICOS, SAP and CCR5. Of these, PD1 and ICOS present >90 percent sensitivity, but are not specific since may also be present in activated T-cells. On the other side, CXCL13 and CD10 are more specific markers for T-cell follicular type lymphoma [4, 29]. Large B-cells in interfollicular areas usually express pan-B markers and are EBV+. HRS-like cells express CD30+ and are usually EBV+, although they may be EBV- and rosetted by neoplastic T-follicular helper cells. Molecular analysis shows clonal rearrangement of TCR genes, and approximately 20 percent of cases harbor a t(5;9) translocation [29].

Recent studies about T-cell follicular type lymphoma have shown that the behavior was aggressive, with 50 percent of patients dying after a median follow up of 24 months [29].

## 4.5. GLOBAL STUDY OF ALL CASES

### 4.5.1. Time elapsed to final diagnosis

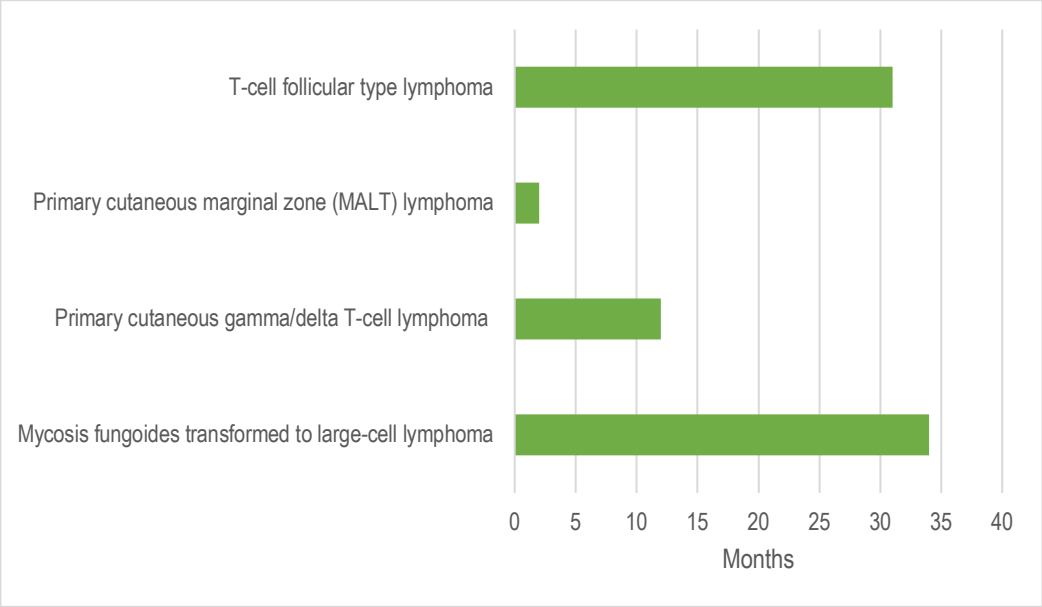



**Figure 26. Time elapsed from first consultation until histological final diagnosis. Mycosis fungoides transformed to large-cell lymphoma: 34 months; Primary cutaneous gamma/delta T-cell lymphoma: 12 months; Primary cutaneous marginal zone (MALT) lymphoma: 2 months; T-cell follicular type lymphoma: 31 months.**

The diagnosis of classic MF was obtained from the first biopsy, but the clinical picture was evolving, ending in a folliculotropic mycosis fungoides transformed into a large-cell lymphoma (CD30-), which was diagnosed 34 months after the patient's first consultation. Primary cutaneous gamma-delta T-cell lymphoma, which is a rare lymphoma, happened with a rapidly progressive clinical picture and the diagnosis was delayed for 12 months. In the case of primary cutaneous marginal zone (MALT) lymphoma, the diagnosis required 2 months, less than in the rest of cases. T-cell follicular type lymphoma, an extremely rare lymphoma, recently included in the WHO Classification, delayed the diagnosis for 31 months.

### 4.5.2. Required procedures to achieve a final diagnosis

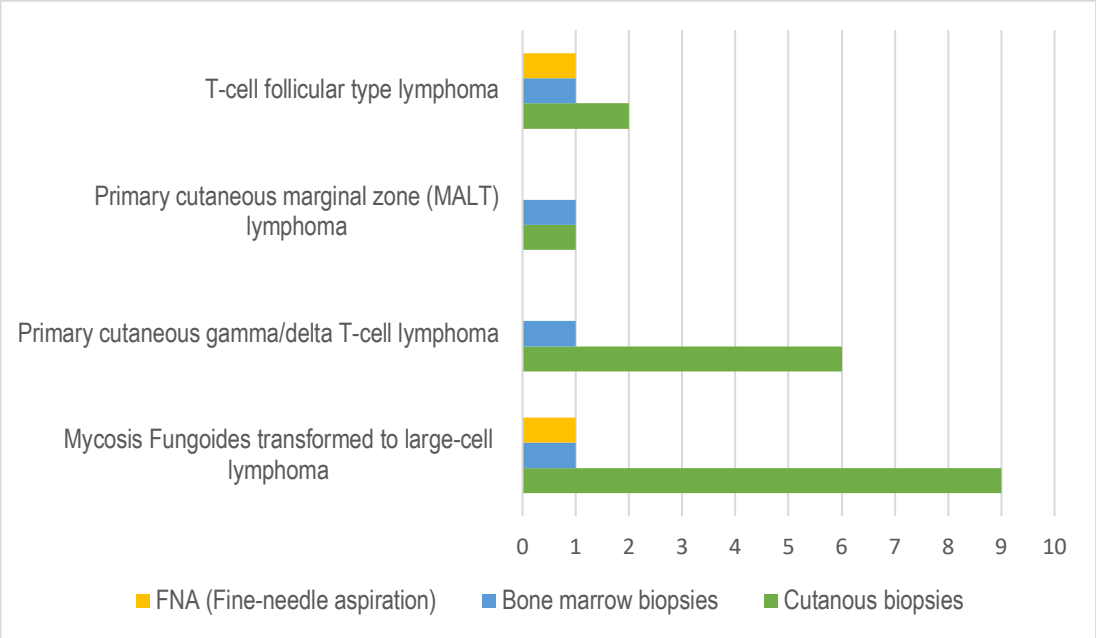



**Figure 27. Required procedures to achieve a final diagnosis. Mycosis fungoides: 9 cutaneous biopsies, 1 bone marrow biopsy and 1 FNA (Fine-needle aspiration); Primary cutaneous gamma/delta T-cell lymphoma: 6 cutaneous biopsies and 1 bone marrow biopsy; Primary cutaneous marginal zone (MALT) lymphoma: 1 cutaneous biopsy and 1 bone marrow biopsy ; T-cell follicular type lymphoma: 2 cutaneous biopsies, 1 bone marrow biopsy and 1 FNA (Fine-needle aspiration).**

As explained in the sections corresponding to each case, some of the lymphomas required several biopsies to reach a definitive diagnosis. It may happen, as in the case of mycosis fungoides, that it is necessary to perform several biopsies to verify the evolution of the disease or, as in the primary cutaneous gamma-delta T-cell lymphoma, to reach a diagnosis.

In addition, in these cases other complementary tests, such as bone marrow biopsies or FNAs are usually necessary to rule out other diagnoses.

### 4.5.3. Inquiries made to external centers

| | Reference hospitals | Reason for consultation |
|---|---|---|
| Mycosis fungoides | Valdecilla Hospital (Santander) | Complementary morphological study<br><br>Confirmation of diagnosis of mycosis fungoides |
| | Clinic Hospital (Barcelona) | Confirmation of diagnosis of mycosis fungoides transformed to large-cell lymphoma, CD30- |
| Primary cutaneous gamma-delta T-cell lymphoma | Salamanca Hospital | Diagnostic compatibility with lupus panniculitis |
| | Valdecilla Hospital (Santander) | Study of specific markers (CD3, CD56, Granzyme, Perforin, TCR gamma, Ki66, CD20, CD4, CD8, CD30, TCRBF1, PD1, EBV-LMP1 and EBER)<br><br>Confirmation of diagnosis of primary cutaneous gamma-delta T-cell lymphoma |
| Primary cutaneous marginal zone (MALT) lymphoma | None. | |
| T-cell follicular type lymphoma | Valdecilla Hospital (Santander) | Study of special techniques<br><br>Confirmation of diagnosis of nodal T-cell lymphoma with a T-follicular helper cell phenotype |

**Table 2. Inquiries made to external centers. Mycosis fungoides: Valdecilla Hospital (Santander), Clinic Hospital (Barcelona); Primary cutaneous gamma/delta T-cell lymphoma: Salamanca Hospital, Valdecilla Hospital (Santander); Primary cutaneous marginal zone (MALT) lymphoma: none; T-cell follicular type lymphoma: Valdecilla Hospital (Santander)**

The need for confirmation or the application of specific markers by reference centers reflects the specificity and difficulty of these diagnoses.

# 5. DISCUSSION

## 5.1. MYCOSIS FUNGOIDES

With the clinical suspicion of cutaneous lymphoma due to the tumor lesion presented by the patient, among other differential diagnoses such as pseudolymphoma, squamous cell carcinoma, dermatofibrosarcoma or leiomyosarcoma, the initial performed biopsy showed a histologic morphology compatible with MF, the most frequent type of primary cutaneous lymphoma. Diffuse lymphoid infiltrate in the papillary dermis, epidermotropism and Pautrier's microabscesses were clear findings of the suspicion diagnosis and the latter, a pathognomonic sign, as exposed in the medical literature [3,4,5,6,7].

In the successive biopsies of 2013 and 2014, morphological and immunohistochemical changes were observed, suggesting progression of the disease. Thus, the presence of isolated CD30+ cells has been observed in cases of indolent mycosis fungoides, but the increase in number of CD30+ cells has been related to advanced disease and is more frequently observed in late phases [5,6,7]; affectation of the subcutaneous cellular tissue in mycosis fungoides in tumor stage has been related to large cell transformed mycosis fungoides [6,7].

As described in several articles and described in biopsies analyzed after 2014, folliculotropic mycosis fungoides presented with an irregular lymphoid infiltrate that surrounded the follicles with a similar phenotype to that found in previous biopsies, which later presented mucinous degeneration. FMF in this case, which was derived from mycosis fungoides in tumor stage, has been associated with worse prognoses at 5 years [4,5,7,8,9,35].

In June 2015, due to the development of hilar and mediastinal lymphadenopathies, together with three pulmonary micronodules, forcing a distinction between sarcoidosis or dissemination of cutaneous lymphoma. In addition, new lesions that emerged during the same year revealed a granulomatous presentation. Kogut et al. described a case of granulomatous mycosis fungoides, a rare subtype of MF that is estimated to represent 6,3 percent of cases [37]. Also, in older studies, Brincker described granulomatous

reactions in 7.3 percent of cases of non-Hodgkin lymphoma, most of them with a sarcoid presentation [38].

In August 2015, after the development of new lesions in the abdomen, a change in the components of the lymphoid infiltrate was described in the biopsy performed, showing medium to large-sized cells and CD30 + expression in 40 percent of those, considering the diagnosis of *mycosis fungoides transformed into a large cell lymphoma*, *CD30-*. As exposed in the results section, transformation into a large cell lymphoma is defined as the presence of lymphocytes 4 times the size of a normal lymphocyte, present as micronodules or occupying more than 25 percent of the infiltrate. This transformation is indicative of an accelerated and more aggressive course, with decreased survival. Salhaney et al. described large oval nuclei, prominent and/or multiple nucleoli and moderate to abundant amphophilic cytoplasm. A certain degree of epidermotropism can be observed, as well as cerebriform cells scattered throughout the periphery of the transformed cells. In immunohistochemical analysis, a CD68+ phenotype was described, identifying a histiocytic component and excluding granulomatous mycosis fungoides; CD4+ is frequently found, with usual loss of panT-cell antigens, but CD8+ or CD4-/CD8- have also been observed. CD30+ cells may be seen, usually representing less than 75 percent of the infiltrate; if they exceed 75 percent of the infiltrate, it may be impossible to differentiate the transformed mycosis fungoides from a CD30 positive lymphoproliferative disorder arising in a patient with mycosis fungoides [3,7,8,9,10].

The results of this review reflect the difficulty that exists in the diagnostic workup of mycosis fungoides, despite being the most frequent type of cutaneous. For years (2012-2015), histological diagnosis was changing, which led to the performance of numerous skin biopsies and inquiries to an external reference center in cutaneous lymphoma (Valdecilla Hospital, Santander). Therefore, communication between both Pathology and Dermatology departments was essential.

## 5.2. PRIMARY CUTANEOUS GAMMA-DELTA T-CELL LYMPHOMA

A sixty-five-year-old patient complained of a very nonspecific lesion, which appeared as an indurated erythematous plaque on the inner area of her left thigh, that was finally diagnosed as *primary cutaneous gamma-delta T-cell lymphoma*. Several studies coincide in the average age of diagnosis (60 years), as well as with the usual location of presentation of this lymphoma, the extremities [3,4,10,11,18,19,20]. Likewise, cases with psoriasis-like presentation have been described, with erythematous plaques that evolve later to ulcerated lesions [12], which is quite consistent with the clinical evolution of this patient. In September 2016, the patient started with symptoms of fever, sore throat and asthenia, which could be interpreted as accompanying B symptoms, also described in the articles reviewed in this paper [3,4,5,11,18,19,20].

Histologic diagnosis of this lymphoma was complex, due to the successive initial biopsies that simulated other possible clinical conditions, such as septal panniculitis or lupus panniculitis. In the reviewed literature related to PCGD-TCL, the findings of mucin deposits or subcutaneous infiltrates with minimal atypia and histiocytosis are described as complications during the diagnostic workup [10], which can cause doubts precisely with the diagnoses identified in this case. Aguilera et. al describe in a Journal of the American Academy of Dermatology article published in 2007, a case in which a *primary cutaneous gamma-delta T-cell lymphoma* presented as a lupus panniculitis [17]. Another case was published in this same journal in 2018, in which a *primary cutaneous gamma-delta T-cell lymphoma* presented as a cytophagic histiocytic panniculitis, a rare and usually ulcerated type of panniculitis [22].

Once a lymphoproliferative process was suspected, the studied biopsies could be confused with a *subcutaneous panniculitis-like T-cell lymphoma* [4,5,18,19,20], a lymphoma with a TCR αβ- phenotype, and forced to include it as differential diagnosis of PCGD-TCL until subjecting the samples to specific markers that provide a definitive diagnosis. Willemze exposes in an article of 2017 the different features of lymphomas with panniculitic presentation, in which he includes the *subcutaneous panniculitis-like T-cell lymphoma* and the *primary cutaneous gamma-delta T-cell lymphoma* [19].

Specific immunohistochemical analysis performed at the reference center, clarified the diagnosis, as positive markers typically found in PCGD-TCL, such as TCR γδ, CD3, CD56, Granzyme B, perforin or TIA 1 were observed [3,4,5,10,11,18,19,20]. Molecular studies of the biopsies reviewed between November and December 2016 showed monoclonal rearrangement of the TCR genes, an expected finding in the study of this type of lymphoma [3,4,5,10,11,18,19,20].

The exposed results reflect the complexity involved in arriving at a prompt and accurate diagnosis of a *primary cutaneous gamma-delta T-cell lymphoma*. The lack of availability of specific markers, the presence of TCR γδ phenotype in some indolent lymphomas, as well as the inexperience in the interpretation of this type of infiltrates, have made this a challenging diagnosis. In these cases, correlation between morphological, immunophenotypic and molecular data is essential, but so is the clinical and pathological correlation to reach a definitive diagnosis in the shortest possible time.

### 5.3. PRIMARY CUTANEOUS MARGINAL ZONE (MALT) LYMPHOMA

An eighty-eight-year-old man consulted for presenting several pruritic, indurated, erythematous-violaceous lesions on the trunk, upper extremities and scalp that had lasted two months. Lesions presented in cases of *primary cutaneous marginal zone (MALT) lymphoma* are described as pruritic violaceous papules, plaques or nodules, that usually appear in the upper extremities [3,4,24,25,26], which correspond to those why the patient consulted.

Histologic study of the biopsy corresponding to the first lesions showed a nodular, perivascular and perianexial lymphoid infiltrate, with exocytosis and isolated spongiosis without epidermotropism. The reviewed literature morphologically describes the PCMZL as a nodular or diffuse lymphocytic infiltrate in dermis, separated from the epidermis, that rarely shows colonization of follicular structures [3,4,5,24,25,26], which can be observed in other types of extranodal marginal zone lymphoma [25].

Immunophenotypic studies performed in the reference center to which the biopsy was sent, showed positivity for B-cell markers (CD20 and CD79α), as well as positivity for BCL2, orienting the diagnosis towards cutaneous B cell lymphoma, as described in reviews dedicated to this type of lymphoma [3,4,5,24,25]. In addition, clonal studies also indicated clonal rearrangement for the IgH gene, corresponding to B lymphoma [26].

The bone marrow study indicated infiltration by PCMZL, a necessary differential diagnosis from systemic lymphoma. Patients with cutaneous involvement due to a systemic marginal zone lymphoma tend to be older, and lesions preferentially occur in the head and neck. Morphological analysis is similar to other types of extranodal marginal zone lymphoma, but as said before, *primary cutaneous marginal zone (MALT) lymphoma* do not usually show infiltration of the follicles. Most marginal zone lymphomas express IgM, whereas *primary cutaneous marginal zone (MALT) lymphoma* usually expresses IgG, IgA or IgE [25].

These results reflect how communication and joint approach of patients with cutaneous lymphoma by the services of Dermatology, Pathology and, in many cases, Hematology, is fundamental to reach an accurate diagnosis.

### 5.4. T-CELL FOLLICULAR TYPE LYMPHOMA

In this case, a sixty-year-old woman consulted the Dermatology department for generalized pruritus on the trunk, upper extremities and scalp, which had lasted for the past two years. Chronic pruritus in Dermatology consultations is possibly one of the challenges the clinician faces in daily life, since the range of pathologies in which it presents is very extensive, from banal to potentially aggressive diseases. In an article published in the New England Journal of Medicine in April 2013, chronic pruritus is associated with a marked reduction in quality of life, insomnia and mood alterations, including anxiety and depression. In this same study, possible causes of chronic pruritus are described, including dermatological and myeloproliferative disorders, and it is considered a priority to determine the underlying cause of chronic pruritus (**Figure 28**) [39]. In addition to pruritus, the patient presented with splenomegaly and several

lymphadenopathies, all of it described in reviews about *angioimmunoblastic T-cell lymphoma* and/or *T-cell follicular lymphoma* [29,32].

As stated in the results section, the first cytology performed of a submental lymph node in May 2017 was negative for malignant cells, which hindered the diagnosis workup. Later in July of that same year, the ganglion was removed and abundant microgranulomas of sarcoid type were observed, without necrosis and some eosinophils. In a reviewed article, it was described that neoplastic cells could go unnoticed, due to pronounced inflammatory background [29]. In this case, the presence of microgranulomas made the diagnosis hard, suggesting chronic non-necrotizing granulomatous lymphadenitis. It was with the clinical worsening of the patient, and after discussed in Dermatopathology session, when the possibility of a T-cell lymphoproliferative process was raised. Pujol et al. described four cases of patients diagnosed with *nodal and/or extranodal non-Hodgkin lymphoma* and *angioimmunoblastic T-cell lymphoma* who developed specific cutaneous involvement and prominent epithelioid cell and/or granulomatous inflammation [30]. In another of the reviewed articles, non-necrotizing granulomas are described as a possible presentation in chronic lymphocytic leukaemias, and specifies regarding the cutaneous presentation that granulomas may precede the histologic findings of lymphoma and can mimic non-necrotizing granulomatous entities [31].

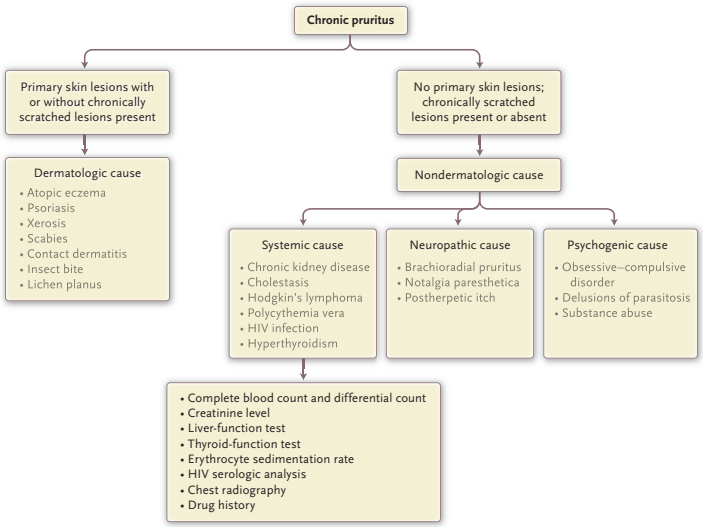



**Figure 28. Diagnostic workup for chronic pruritus. Figure obtained from Yosipovitch G, Bernhard J. Chronic pruritus. N Engl J Med 2013; 368: 1629.**

The diagnostic suspicion was *angioimmunoblastic T-cell lymphoma* (AITL), the most frequent type of nodal T-cell lymphomas with a T-follicular helper-cell phenotype. *T-cell follicular type lymphoma* is a very rare subtype, included in the WHO Classification in 2016, as a provisional entity, therefore, it is a lymphoma of which there is scarce clinical and pathological experience.

## 6. CONCLUSIONS

The aim of this review is to emphasize the difficulty of diagnosing this type of neoplasms, both on the clinical and on the histopathological side. All this has been reflected in the time elapsed from the first consultation of the patient to final diagnosis, the number of biopsies that each case required, the consultations made to external centers to confirm the diagnoses and the intervention of Dermatology, Pathology, Hematology and Radiology services, principally.

For all these reasons, clinical and pathological correlation is crucial and should be materialized in the development of sessions, mainly between the Dermatology and Pathology departments, and the organization of interdisciplinary committees, in which the diagnosis of cutaneous lymphoma is addressed jointly.

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