

# Netherton Syndrome – a therapeutic challenge in childhood

Polina Kostova<sup>1</sup>, Guergana Petrova<sup>1</sup>, Martin Shahid<sup>1</sup>, Vera Papochieva<sup>1</sup>, Dimitrinka Miteva<sup>1</sup>, Ivelina Yordanova<sup>2</sup>, Kossara Drenovska<sup>1</sup>, Irena Bradinova<sup>1</sup>, Camila K. Janniger<sup>3</sup>, Robert A. Schwartz<sup>3</sup>, and Snejina Vassileva<sup>1</sup>

<sup>1</sup>Medicinski universitet-Sofia

<sup>2</sup>Medicinski universitet Pleven Fakultet medicina

<sup>3</sup>Rutgers The State University of New Jersey

March 16, 2024

## Abstract

Netherton syndrome (NS) is a rare autosomal recessive genodermatosis (OMIM #256500) characterized by superficial scaling, atopic manifestations, and multisystemic complications. It is caused by loss-of-function mutations in the SPINK5 gene, which encode a key kallikrein protease inhibitor. There are two subtypes of the syndrome that differ in clinical presentation and immune

## Netherton Syndrome – a therapeutic challenge in childhood

Polina Kostova MD<sup>1</sup>, Guergana Petrova MD PhD<sup>2</sup>, Martin Shahid MD PhD<sup>3</sup>, Vera Papochieva MD<sup>4</sup>, Dimitrinka Miteva MD PhD<sup>5</sup>, Ivelina Yordanova MD PhD<sup>6</sup>, Kossara Drenovska MD PhD<sup>7</sup>, Irena Bradinova MD PhD<sup>8</sup>, Camila K. Janniger MD<sup>9</sup>, Robert A. Schwartz MD, MPH, DSc (Hon)<sup>10\*</sup>, Snejina Vassileva MD PhD<sup>11</sup>

## Affiliations:

1 Pediatric department, Medical University, Sofia; Pediatric clinic, UMHAT Alexandrovska, Sofia, Bulgaria; poli.mu@gmail.com

2 Pediatric department, Medical University, Sofia; Pediatric clinic, UMHAT Alexandrovska, Sofia, Bulgaria; gal\_ps@yahoo.co.uk

3 Department of Dermatology and Venereology, Medical University, Sofia; Dermatology clinic UMHAT Alexandrovska, Sofia, Bulgaria; martin.shahidmd@gmail.com

4 Pediatric clinic, UMHAT Alexandrovska, Sofia, Bulgaria; vera\_pap@abv.bg

5 Pediatric department, Medical University, Sofia; Pediatric clinic, UMHAT Alexandrovska, Sofia, Bulgaria; dimi.sm@abv.bg

6 Department of Dermatology, Venereology and Allergology, Faculty of Medicine, Medical University Pleven, Bulgaria; ivelina\_yordanova@abv.bg

7 Department of Dermatology and Venereology, Medical University, Sofia; Dermatology clinic UMHAT Alexandrovska, 1 Georgi Sofiyski str., 1431 Sofia, Bulgaria; kosara@lycos.com

8 National Genetic Laboratory, Medical University Sofia, University Hospital of Obstetrics and Gynecology “Maichin dom,” Sofia, Bulgaria; bradinova@maichindom.com

9 Dermatology and Pediatrics, Rutgers New Jersey Medical School, Newark, New Jersey, USA; professor@camilajanniger.com

10 Dermatology, Pediatrics, and Pathology, Rutgers New Jersey Medical School, Newark, New Jersey, USA; roschwar@rutgers.edu

11 Department of Dermatology and Venereology, Medical University, Sofia; Dermatology clinic UMHAT Alexandrovska, Sofia, Bulgaria; snejina.vassileva@gmail.com

\* Correspondence: Robert A. Schwartz MD, MPH, DSc (Hon), MAE, FRCP Edin

Professor and Head, Dermatology, Rutgers New Jersey Medical School

185 South Orange Avenue, Newark, New Jersey 07103

roschwar@rutgers.edu

### **Conflict of interest**

The authors declare that the article was written without financial or commercial motives that could represent a potential conflict of interest.

### **Ethics statement**

Written informed consent has been obtained from the parents for the publication of all data and images from the family's archives contained in this article.

### **Data availability statement**

The medical records for this article are not publicly available for reasons of patient anonymity.

### **Funding**

The authors did not receive any financial support for this case report. The authors have no conflicts of interest to declare.

### **Acknowledgments**

The authors thank the patient and his relatives for permission to publish this case report. Adherence to treatment would not have been possible without the support of the child's family. It is to the family's credit that the child can enjoy lasting well-being. Respect for their endless love and care every day.

### **Abstract**

Netherton syndrome (NS) is a rare autosomal recessive genodermatosis (OMIM #256500) characterized by superficial scaling, atopic manifestations, and multisystemic complications. It is caused by loss-of-function mutations in the SPINK5 gene, which encode a key kallikrein protease inhibitor. There are two subtypes of the syndrome that differ in clinical presentation and immune profile, ichthyosiform erythroderma and ichthyosis linearis circumflexa. NS is a multisystemic disease with numerous extracutaneous manifestations. Current therapy for patients with NS is mainly supportive, as there is no curative or specific treatment, especially for children with NS, but targeted therapies are being developed. We describe an eight-year-old boy with genetically proven NS treated with intravenous immunoglobulin for recurrent skin and systemic infections from infancy, growth retardation, and associated erythroderma. Under this therapy, his skin status, infectious exacerbations, and quality of life all improved. Knowledge of the cytokine-mediated pathogenesis of NS and the development of new biologic drugs open new possibilities for NS patients. However, the different therapeutic options have been applied in a limited number of cases, and variable responses have been shown. Randomized controlled trials with a sufficient number of patients stratified and treated according to their specific immune profile and clinical phenotype are needed to evaluate the safety and efficacy of treatment options for patients with NS.



**Keywords :** Netherton syndrome, children, infections, treatment, ichthyosis linearis circumflexa, ichthyosiform erythroderma

**Word Count:** 2407

## Introduction

Netherton syndrome (NS) is a rare autosomal recessive genodermatosis (OMIM #256500) characterized by superficial scaling, atopic manifestations, and multisystemic complications, a triad of congenital ichthyosiform erythroderma or ichthyosis linearis circumflexa, hair shaft abnormalities, and immune dysregulation. Its incidence is one in 200 000 newborns. It may represent up to 18% of infants with congenital erythroderma.<sup>1</sup> NS is caused by loss-of-function mutations in Serine Protease Inhibitor Kazal type 5 (SPINK5) gene, which is located on chromosome 5q31-32 and encodes Lympho-Epithelial Kazal-type-related protease inhibitor (LEKTI).<sup>2</sup> To date, a total of 108 variants have been found, encompassing all types of mutations.<sup>3</sup> LEKTI, expressed in the thymus and epithelia, is a direct inhibitor of kallikrein-related peptidases and cathepsin G and affects epidermal desquamation and altered skin barrier function. Skin manifestations in NS include ichthyosiform erythroderma (IE) or ichthyosis linearis circumflexa (ILC), a specific hair shaft defect called trichorrhexis invaginata (bamboo hair), and an atopic diathesis. Indeed, NS is a multisystemic disease with characteristic cutaneous and numerous extracutaneous manifestations.<sup>4</sup> Diagnosis of NS can be enhanced with histologic examination;<sup>5</sup> immunostaining shows the absence or reduction of LEKTI expression in the granular layer of the epidermis and in the hair follicles.<sup>6</sup> Abnormalities of the hair shaft could be detected in only 20-50% of affected hairs.<sup>7</sup> Laboratory findings show peripheral eosinophilia and elevated IgE levels. It is very difficult to distinguish atypical NS from other types of ichthyosis; detection of gene mutations in SPINK5 is crucial for diagnosis. Current therapy for patients with NS is mainly supportive, as there is no curative or specific treatment, especially for children with NS, but targeted therapies are being developed.

We describe a child with genetically proven Netherton syndrome, for whom intravenous immunoglobulin for 5 months proven beneficial.

## Case History/Examination

An 8-year-old boy, born from a second normal pregnancy, with no history of collodion membrane at birth, was admitted to the neonatology ward at 5 days of age for generalized skin erythema with fine scaling. His cutaneous eruption was complicated by a secondary infection. A diagnosis of erythrodermia ichthyosiformis congenita was rendered (Figure 1). Subsequently, he was repeatedly hospitalized for cutaneous flares, which manifested with severe erythroderma with areas with fissures and erosions with serous exudate (Figure 2). Because of the severe pruritus, intravenous treatments with antibiotics and human albumin, topical corticosteroids, emollients, and oral antihistamines were administered. Despite this treatment, the pruritus and the severe erythrodermic state involving the trunk, limbs, the head, and ears were unaltered. The child's hair was sparse, lusterless, brittle, and fragile. Trichoscopy revealed trichorrhexis invaginata, plus golf tee and matchstick hairs (Figure 3). The nails were without eponychium. Over the years, he suffered from acute tubulo-interstitial nephritis, chickenpox, bronchiolitis, and pneumonia. Genetic testing subsequently confirmed two mutations in the SPINK5 gene: the pathogenic variant NM\_006846.3:c.1530C > A p.(Cys510Ter) and the probable pathogenic variant c.420del p(Ser141ProfaTer5). Systemic treatment with acitretin, oral cholecalciferol, antihistamines and emollients was initiated. After three months of therapy with acitretin, only worsening of the dermatologic condition was observed, and this therapy was terminated.

At 8 years of age, the child was readmitted to the pediatric clinic with fever up to 39 degrees Celsius for two weeks, with a flare of his cutaneous eruption, evidence of leukocytosis and increased inflammatory activity, with inconclusive evidence of pneumonia from the radiographs. The boy weighed 21 kg (5 P, -1.68 z) with a height of 122 cm (11 P, -1.23 z), and a BMI of 11 P, -1. Axillary and inguinal lymph nodes were enlarged in the form of protruding bundles, with a soft-elastic consistency, slightly painful on palpation. Respiration was attenuated vesicular, with fine crackles in the left base. The heart rate and rhythm were normal, with a holosystolic murmur 3/6 degrees at the left sternal border. There was a hepatomegaly with a sharp liver border at 3 cm below the costal margin, firm consistency, not painful on palpation. The spleen was not

enlarged.

## Methods (Differential diagnosis, investigation and treatment)

Laboratory tests revealed a five-fold increase in liver transaminases, and a decrease in serum albumin, creatinine, and uric acid levels. Furthermore, discrete increase in D-dimers and fibrinogen and very low levels of 25-OH-vitamin D (15 nmol/l) were observed. Elevation of procalcitonin (three times above normal), C-reactive protein and serum IgE level ( $> 2500$  IU/ml) were noted. The ALEX combined allergy test (295 allergens) revealed evidence of sensitization to birch, alder, hazelnut, beech, strawberry, dog, and cat. The child was not found to have allergic symptoms during the pollen season or allergic reactions to foods or medications. The child has no atopic predisposition, in that there is no family history of asthma, eczema, and hay fever. *Citrobacter spp* was isolated from nasal and wound swabs examined as an outpatient. *Staphylococcus aureus* was isolated from throat swabs and hemoculture during hospitalization. Consultation with a pediatric gastroenterologist revealed that the increase in serum transaminases was most likely during the course of infection. Echocardiography showed mild subaortic stenosis, not advanced in its dynamics, with mild hypertrophy of the left ventricle and upper diastolic size of the same. Mild aortic regurgitation, mitral valve prolapse with mild mitral regurgitation; preserved ventricular function. The child has no cognitive impairment or developmental delay.

Treatment with an intravenous antibiotic for 7 days, an oral antihistamine, and emollients was given. It was continued at home with an oral antibiotic and antifungal, a probiotic, and oral vitamin D administration. Because of the subjective discomfort the child experienced with topical application of methylprednisolone aceponate milk, therapy with a cooling cream containing clobetasol propionate cream once daily and an emollient once daily was recommended. Administration of IVIG was suggested as part of the systemic therapy.

In a period without underlying infection, immunologic status was examined, which showed a total T lymphocyte count in the reference range with an increase in the cytotoxic suppressor subpopulation. While B lymphocytes and NK cells were within reference ranges, there was increased percentage of CD57+CD8+ cells. Serum immunoglobulins were normal with the exception of greatly increased IgE levels. The accumulation of CD8+CD57+ T cells was attributed to repeated antigen stimulation during chronic infection, these cells being the most effective ones to fight chronic viral infections due to their high expression of interferon- $\gamma$ , granzyme B, and perforin, as well as their marked cytolytic activity.<sup>8</sup>

## Conclusion and Results (Outcome and follow-up)

Due to recurrent skin and systemic infections in infancy and associated erythroderma, the child was started on intravenous immunoglobulin, as subcutaneous administration was judged not possible in this child, to reduce infectious exacerbations, improve local skin status, and improve quality of life. Our patient, like most NS patients, has normal serum IgG levels, suggesting that the improvement with replacement therapy in these patients is more likely due to the immunomodulatory effects of IVIG. Recent data show that the number of pediatric patients with NS treated with immunoglobulins was 14, of whom 12 showed a positive clinical effect, with the dose administered ranging from 400 to 500 mg/kg per month and the duration of treatment from 2 months to 5 years.<sup>9</sup> A recent meta-analysis comparing different IVIG infusion regimens in patients with Kawasaki disease found that high-dose IVIG regimens are likely to be more effective and may even reduce the need for additional treatment compared to medium- or low-dose IVIG regimens, with little or no difference in the number of adverse events between groups.<sup>10</sup> We decided to test whether the higher IVIG dose would not be more effective in patients with NS. We started with 250 mg/kg/day for 3 days or 750 mg/kg/month administration and have not observed any adverse effects so far.

During the follow-up period, normalization of liver transaminases was noted, and another interesting phenomenon was the sustained decrease in total IgE ( $> 2500 - > 484$ .  $5 - > 256 - > 183$ .  $3 - > 160$ .  $2 - > 129.2$  IU/ml). It may represent a marker of favorable therapeutic response. No such positive trend was observed with vitamin D supplementation, and the child maintained reduced levels (51 nmol/l) despite oral supplementation. In the last five months since receiving replacement therapy, the child gained two kilograms

and grew three centimeters (BMI 18 P, -0.91 z). Furthermore, decrease in the incidence of infectious exacerbations was noted in addition to significant reduction of the Netherton Area and Severity Assessment (NASA) score (66 to 36 points) (Figure 4).

## Discussion

Netherton syndrome is a rare and severe autosomal recessive genodermatosis in the group of congenital ichthyosis. Clinically, it is first evident as the triad of ichthyosiform erythroderma (IE), trichorrhexis invaginata (bamboo hair), and atopic diathesis.<sup>4</sup> IE is present at birth or shortly thereafter as generalized erythroderma with desquamation. In some individuals, the erythrodermic endotype may persist throughout life, but it more often is periodically or completely replaced by another endotype called ichthyosis linearis circumflexa (ICL), manifested by polycyclic and serpiginous migratory erythematous plaques with double-edged scales at the periphery.

Due to the disrupted epithelial barrier, the neonatal period is crucial and life-threatening complications such as electrolyte imbalances, including dehydration, hypothermia, gastrointestinal symptoms, skin/respiratory/systemic infections, and sepsis may occur due to increased transepidermal water loss and superficial scaling of the skin.<sup>4</sup> Patients are highly susceptible to cutaneous and systemic infections. Immunophenotyping of patients from NS revealed no evidence of immunodeficiency. Variable T- and B-cell imbalances are found in a limited number of patients, but these changes are likely secondary to chronic skin infections and inflammation.<sup>11</sup> However, the most recent International Union of Immunological Societies (IUIS) classification of inborn errors of immunity places NS in the group of hyper-IgE syndromes (HIES), which are combined immunodeficiencies with associated or syndromic features.<sup>12</sup> Due to increased catabolic rate, malabsorption, chronic inflammation and recurrent infections, these children suffer from failure to thrive, growth retardation and short stature. Extracutaneous manifestations such as acute pancreatitis, hypothyroidism, thymic atrophy, and acute bilateral renal vein thrombosis have been rarely reported.<sup>4</sup> Cognitive impairment and developmental delays have been identified in some patients. Adult patients may develop cutaneous squamous cell carcinoma.<sup>13</sup>

The diagnosis of NS can be enhanced with histologic examination of involved skin showing detachment of the stratum corneum, hyperkeratosis, acanthosis, focal hypergranulosis, and superficial perivascular lymphocytic infiltration.<sup>5</sup> Laboratory findings show peripheral eosinophilia and elevated IgE levels. It is very difficult to distinguish atypical NS from other types of ichthyosis; detection of gene mutations in SPINK5 is essential for diagnosis.

There is currently no ideal treatment, particularly in childhood.<sup>14</sup> The first successful therapy of a genetic disorder, specifically ichthyosis linearis circumflexa (Netherton syndrome), was achieved by the legendary Edmund Klein and associates, including a coauthor of this current paper (RAS),<sup>15</sup> in the late 1970s utilizing oral low dose cyclophosphamide, possibly related to selective effects of this agent on lymphocyte subpopulations and portending many other options. However, this syndrome tends to improve as patients age, but the clinical course may be disturbed by intermittent exacerbations. The predominant therapy for patients with NS is mostly supportive, as there is neither curative nor specific therapy specifically for children with NS, but targeted therapies are currently in development. The challenge is to find therapies that both repair the disrupted skin barrier and stop chronic inflammation. Current treatment options include skin cleansing, bleach baths, emollients/moisturizers, calcineurin inhibitors and oral antihistamines. Antibiotics, topical corticosteroids, and intravenous immunoglobulins (IVIG) can be used during exacerbations. The likely mechanism of action of intravenous immunoglobulin is the ability of IgG to opsonize bacteria, stimulate phagocytosis, and reduce the incidence of infection and inflammation. Immunoglobulin replacement therapy is preferred in severe cases of NS with frequent infections and failure to thrive and can be used at any age. Many reports describe a remarkable clinical response with no significant adverse effects.<sup>14</sup> Primarily, a reduction in recurrent infections and improvement in skin condition have been reported, as the therapy is well tolerated by patients (mainly neonates and children with NS). The disadvantage of the therapy is the cost and the fact that it has to be performed in a hospital.<sup>4</sup> Systemic therapy in NS patients with chemically synthesized vitamin A derivatives has shown a variable degree of efficacy and tolerability. Partial improve-

ment or exacerbation has been reported for acitretin or isotretinoin. The side effects with bone toxicity and teratogenicity and questionable therapeutic response outweigh the benefits of taking these drugs.<sup>4</sup> In our patient, treatment with acitretin was also discontinued due to worsening of the condition. Another modern approach to consider is the use of topical tacrolimus.<sup>16</sup> There is a wide range of potential target therapies such as kallikrein inhibitors, monoclonal antibodies blocking IL-36, IL-17, IL-12/IL-23, IL-4/IL-13 (targeting allergic manifestations), IL-9, JAK/STAT inhibitors, complement inactivation. None of these treatment options represents a complete cure from NS. Of the therapies used to date, anti-IL-17A therapy may be the most effective. Currently, some gene therapy approaches are being developed.

The best therapeutic strategy for the moment may be a combination of anti-inflammatory therapy and therapy that restores the disrupted skin barrier in NS patients. Unfortunately, all therapeutic options have been applied only in a limited number of cases, and variable responses have been shown.<sup>15</sup> Limited data published to date suggest that biologics therapy is safe and often effective in adults and children with NS, but long-term efficacy and tolerability remain unknown.<sup>14</sup> Randomized controlled trials with sufficient numbers of patients stratified and treated according to their specific immune profile and clinical phenotype are needed to assess the safety and efficacy of treatment options for patients with NS.<sup>16</sup> Studies based on stratification of patients using omics-based techniques such as genomics, transcriptomics, proteomics, metabolomics, and bioinformatics would thus be a step toward precision medicine in Netherton Syndrome.

Thus, Netherton syndrome is multisystemic disease with considerable clinical variability. Based on the reported cases and the authors' recommendations, we recommend consideration of IVIG therapy, based on this child's favorable response noting reduced recurrent severe infections and enhanced growth. If his condition worsens, we would consider therapy blocking IL-17A, which may be the best therapeutic option for improving persistent inflammatory skin diseases and/or preventing acute relapses. Insights into the cytokine-mediated pathogenesis of NS and the increasing use of biologic therapies are opening new possibilities for NS patients. However, the various therapeutic options have been used in a limited number of cases and have shown variable responses. There is a great need for randomized controlled trials with enough patients stratified and treated according to their specific immune profile and clinical phenotype to evaluate the safety and efficacy of treatment alternatives for patients with NS.

## References

- 1 . Pruszkowski A, Bodemer C, Fraitag S et al. Neonatal and infantile erythrodermas: a retrospective study of 51 patients. *Arch Dermatol* 2000; 136: 875–880.
- 2 . Chavanas S, Bodemer C, Rochat A et al. Mutations in SPINK5, encoding a serine protease inhibitor, cause Netherton syndrome. *Nat Genet* 2000; 25: 141–142.
- 3 . The Human Gene Mutation Database (HGMD). <https://www.hgmd.cf.ac.uk/ac/gene.php?gene=SPINK5> (20 March 2023, date last accessed).
- 4 . Petrova E, Hovnanian A. Advances in understanding of Netherton syndrome and therapeutic implications. *Expert Opin Orphan Drugs* 2020; 8: 455–487.
- 5 . Hovnanian A. Netherton syndrome: skin inflammation and allergy by loss of protease inhibition. *Cell Tissue Res* 2013; 351: 289–300.
- 6 . Bitoun E, Micheloni A, Lamant L et al. LEKTI proteolytic processing in human primary keratinocytes, tissue distribution and defective expression in Netherton syndrome. *Hum Mol Genet* 2003; 12: 2417–2430.
- 7 . Srinivas SM, Hiremagalore R, Suryanarayan S et al. Netherton syndrome with pili torti. *Int J Trichology* 2013; 5: 225–226.
- 8 . Huang B, Liu R, Wang P et al. CD8+CD57+ T cells exhibit distinct features in human non-small cell lung cancer. *J Immunother Cancer* 2020; 8: e000639.

- 9 .** Nouwen AEM, Schappin R, Nguyen NT et al. Outcomes of Systemic Treatment in Children and Adults With Netherton Syndrome: A Systematic Review. *Front Immunol* 2022; 13: 864449.
- 10 .** Broderick C, Kobayashi S, Suto M et al. Intravenous immunoglobulin for the treatment of Kawasaki disease. *Cochrane Database Syst Rev* 2023; 2023: CD014884.
- 11 .** Eränkö E, Ilander M, Tuomiranta M et al. Immune cell phenotype and functional defects in Netherton syndrome. *Orphanet J Rare Dis* 2018; 13: 213.
- 12 .** Tangye SG, Al-Herz W, Bousfiha A et al. Human Inborn Errors of Immunity: 2022 Update on the Classification from the International Union of Immunological Societies Expert Committee. *J Clin Immunol* 2022; 42: 1473–1507.
- 13 .** van der Voort EAM, Prens EP. Netherton syndrome with multiple non-melanoma skin cancers. *Acta Derm Venereol* 2013; 93: 727–728.
- 14 .** Pontone M, Giovannini M, Filippeschi C et al. Biological treatments for pediatric Netherton syndrome. *Front Pediatr* 2022; 10: 1074243.
- 15.** Klein E, Hahn GM, Solomon JA, et al. Explorations of antimitotic agents in the treatment of a congenital disease, ichthyosis linearis circumflexa. *J Surg Oncol.* 1979;11:85-8
- 16.** Suga Y, Tsuboi R, Hashimoto Y, et al. A case of ichthyosis linearis circumflexa successfully treated with topical tacrolimus. *J Am Acad Dermatol.* 2000;42:520-2
- 17 .** Nouwen AEM, Schappin R, Nguyen NT et al. Outcomes of Systemic Treatment in Children and Adults With Netherton Syndrome: A Systematic Review. *Front Immunol* 2022; 13: 864449.
- 18 .** Clinical Trials register - Search for Netherton Syndrome. <https://www.clinicaltrialsregister.eu/ctr-search/search?query=Netherton+Syndrome> (20 March 2023, date last accessed).

**Images (provided as a separate files)**







FIGURE 1 Photos taken during the stay in the neonatology ward showing diffuse redness and scaling all over the body.











FIGURE 2 Photos taken over the years during exacerbations.







FIGURE 3 Trichoscopy and clinical photos taken at admission to the pediatric clinic.





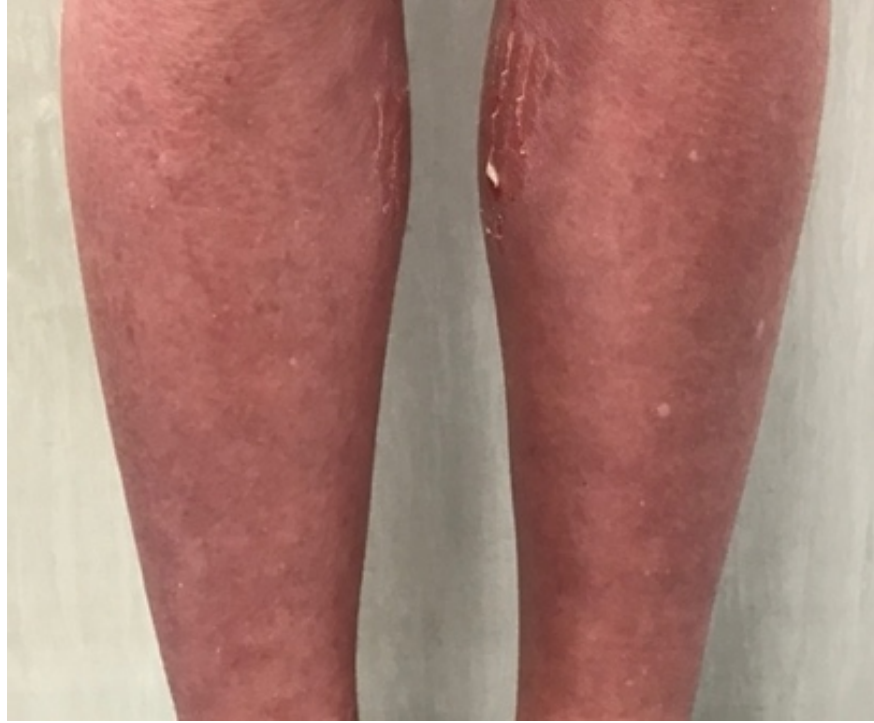




FIGURE 4 Photos from the hospital stay for the fourth infusion

**ORCID numbers:**

Polina Kostova 0000-0002-3012-1861

Guergana Petrova 0000-0002-3012-1861

Martin Shahid 0000-0002-3012-1861

Vera Papochieva

Dimitrinka Miteva 0000-0003-2885-6479

Ivelina Yordanova 0000-0002-3012-1861

Kossara Drenovska 0000-0002-8254-1630

Irena Bradinova 0000-0002-3012-1861

Camila K. Janniger 0000-0002-6873-2465

Robert A. Schwartz 0000-0003-3036-3825

Snejina Vassileva 0000-0002-3012-1861

1. Please provide the below following statement in the manuscript: Consent statement

Written informed consent has been obtained from the parents for the publication of all data and images from the family's archives contained in this article.

- Author contribution statement

CASRAI CRediT Taxonomy: authors' contribution(s) to the submitted manuscript are attributed as below. Submitting Authors may provide Author Contributions at original submission but MUST provide the information at revised submission. At revision submission, all authors should check the contributions carefully as, if your manuscript is accepted, this information will be included in the published article: CRediT Taxonomy

Polina Kostova Conceptualization, Data curation, Formal analysis, Methodology, Supervision, Visualization, Writing – original draft, Writing – review & editing Guergana Petrova Writing – original draft, Writing – review & editing Martin Shahid Writing – original draft, Writing – review & editing Vera Papochieva Writing – original draft, Writing – review & editing Dimitrinka Miteva Writing – original draft, Writing – review & editing Ivelina Yordanova Writing – original draft, Writing – review & editing Kossara Drenovska Data curation, Investigation, Project administration, Supervision, Validation, Writing – original draft, Writing – review & editing Irena Bradinova Writing – original draft, Writing – review & editing Camila K. Janniger Writing – original draft, Writing – review & editing Robert A. Schwartz Supervision, Writing – original draft, Writing – review & editing Snejina Vassileva Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Project administration, Resources, Supervision, Writing – original draft, Writing – review & editing

















