Journal of Applied Cosmetology

ISSN 2974-6140 (online) ISSN 0392-8543 (print)

Original article

New approach for the treatment of autoimmune diseases

D. Kaliterna[1] and B. Barac[2]

[1]Poliklinika Poliderma, Zagreb, Croatia; [2]Institute of Rheumathology,University of Belgrade, Belgrade, Serbia

Corresponding author:
Dinko Kaliterna, MD,
Ulica grada Vukovara 284d 4. kat
Zagreb (Almeria centar)
e-mail: poliderma@poliderma.hr
Tel: +385 1 4828 961

Keywords: *autoimmune diseases, stem cells, adipose tissue-derived stem cells, ASCs, stem cell treatment, autologous stem cell*

Received: 28 October 2022
Accepted: 11 January 2023

ISSN 2974-6140 (online) ISSN 0392-8543 (print).

ABSTRACT

Autoimmune diseases are due to a dysregulation of the immune system's balance, which attacks healthy cells in the body, leading to tissue damage. They can affect different organs/tissues, and the skin represents one of the most affected. Among the skin-related autoimmune diseases, Cutaneous Lupus Erythematosus (CLE) and Morphea are included. Both conditions present various cutaneous manifestations and may also show systemic involvement. It is noted that they can be challenging to treat, often recalcitrant to currently available therapies, thus causing significant distress to affected patients. The approach of personalised medicine, based on regenerative therapies, has been growing in recent years. The use of autologous adult stem cells has successfully treated autoimmune diseases. In this study, we aimed to evaluate the treatment based on patient-specific adipose-tissue stem cells (ASCs) in our two distinct cohorts of patients affected by CLE (n=20) and Morphea (n=12). Patients presenting only with skin lesions underwent local ASCs therapy by intradermal injection in the affected areas. At the same time, patients who also had extra-cutaneous involvement (complicated cases) received a combined therapy consisting of both intradermal and intravenous injections of ASCs. Overall, after 1 month of treatment, an improvement has been observed in 78% and 73% of subjects treated with intradermal injections and combined therapy, respectively. Notably, a better response to ASCs-based therapy has been reported in CLE patients, reaching an 86% in complicated cases. Our study promotes the application of autologous ASCs as an effective therapeutic strategy in skin-related autoimmune diseases.

INTRODUCTION

Autoimmune diseases affect 5-9% of the world's population. They are due to a dysregulation of the immune system's balance, which attacks healthy cells in the body. It is known that the development of an autoimmune disease requires a combination of genetic susceptibility and environmental factors, which trigger autoimmune pathways, ultimately leading to tissue damage (1, 2).

More than 100 different types of autoimmune diseases have been reported (3), and one tissue commonly affected by autoimmune disorders is the skin. Many skin-related autoimmune diseases include Cutaneous Lupus Erythematosus (CLE) and Morphea or localised scleroderma.

These inflammatory conditions present various cutaneous manifestations and may also show systemic involvement (4, 5). CLE can be divided into three main subtypes: acute (ACLE), subacute (SCLE), and chronic (CCLE) (6). On the other hand, Morphea subtypes include linear, generalised, pan-sclerotic and mixed (7, 8). Current treatment recommendations for the most severe cases of CLE and Morphea are limited, mainly consisting of systemic corticosteroids, traditional immunosuppressants or antimalarials for CLE (9), and a combination of systemic corticosteroids and methotrexate for Morphea (10). However, these are not disease-specific treatments, and both diseases can be challenging to treat, often recalcitrant to currently available therapies, thus causing significant distress to affected patients (11, 12).

Very recently, regenerative therapies based on patient-specific stem cells have been successful in treating autoimmune diseases (13). Stem cells (SCs) are undifferentiated cells responsible for the regeneration and development of organs and tissues, capable of self-renewal and differentiation into multiple cell lineages (14). There are four primary sources of SCs: i) embryonic tissues; ii) fetal tissues; iii) adult tissues; iv) differentiated somatic cells after they have been genetically reprogrammed, i.e. induced pluripotent stem cells (iPSCs) (15). Although adult stem cells have a lower differentiation ability than the other three SC types, these cells can

overcome the ethical and legal issues accompanying the application of embryonic and fetal stem cells and the mutational effects associated with iPSCs (15). Thus, their application has been approved for many conditions, resulting in the most promising, with a solid body of data supporting their safety profiles (16).

Interestingly, adult stem cells are located in almost all organs and tissues (17). However, those derived from adipose tissue, i.e. adipose stem cells (ASCs), seem to be the most advantageous for clinical application, mainly because they can be easily harvested in higher quantities and minimally invasive ways (18, 19). In addition, they can be applied in autologous form.

Given all the abovementioned considerations, we aimed to evaluate the treatment based on patient-specific ASCs in our two distinct cohorts of patients affected by CLE and Morphea, respectively.

MATERIALS AND METHODS

Study population

We enrolled 20 patients diagnosed with CLE and 12 with Morphea who underwent skin biopsy in our medical clinic in Zagreb.

Both patients with CLE and Morphea had typical skin lesions. Therefore, patients presenting only with skin lesions were defined as "not-complicated" (n=20). At the same time, those who also had extra-cutaneous involvement were defined as "complicated" (total n=12). All patients have written informed consent before starting the treatment.

Preparation for the stem cell treatment

Prior to starting therapy, patients underwent t all or some of the following procedures according to their health conditions:

- Detox infusions: anti-inflammatory properties and power to remove heavy metals, reduce the virus load and repair the cell membranes, thus restoring balance in the body.
- Ozone therapy: anti-inflammatory effect and ability to inactivate viruses.
- Hemotherapy and/or laser blood irradiation: anti-inflammatory effect.
- Ultraviolet blood irradiation and/or photodynamic therapy: the ability to inactivate viruses.

Processing of autologous ASCs

Each adipose tissue was micronised, resized and filtered using the instrument Adinizer®, composed of different sizes of sharp blade-edge stainless steel mesh discs, which harvested fat into a fine, homogenised fat tissue without emulsification.

After this process, the ASCs were processed by the Automatic Cell Station (ACS) from the Bsl company, an integrated system of specially designed centrifuge and incubator, in which ASCs are processed with the collagenase in just 20 min. The average number of processed ASCs was 30 million, with 100 % preserved vitality. After the processing, ASCs were immediately ready for intravenous application or intradermal injection.

Autologous ASCs treatment

All patients were offered to undergo stem cell treatment. In particular, not-complicated patients were offered to undergo ASCs local therapy by intradermal injection in the affected areas. On the other side, complicated patients were offered to undergo a combined ASCs-based therapy, consisting of both intradermal and

intravenous IV injections of stem cells. Patients were evaluated before and after 1 month of the stem cell treatment.

RESULTS

CLE patients

Patients with CLE (n=20) had a mean age of 32±6 years. Out of 20 patients, 16 were females, and 4 were males. All patients showed the typical CLE-related skin lesions, whilst 8 subjects also showed extra-cutaneous involvement (complicated patients), including arthralgias (n=4), mild kidney failure (n=2), pericarditis (n=1) and pleurisy (n=1). As reported in the Methods section, not-complicated patients were offered stem cell local injections in the affected skin areas. In contrast, complicated patients were offered to undergo a combination of intradermal/IV stem cell therapy.

The outcome of stem cells therapy in CLE patients

All patients underwent stem cell local injections in the affected skin areas, while 7/8 complicated cases also underwent IV stem cell therapy. One patient with mild kidney failure refused IV treatment. Patients with pericarditis and pleurisy were also treated with acetylsalicylic acid.

After 1 month of stem cell therapy, 80% (n=16/20) of total patients showed an improvement in CLE-related skin lesions, while 4 patients showed skin disease stability (Fig. 1A). No patients reported exacerbation of skin lesions or the onset of new skin lesions in a previously healthy skin area. Concerning 7 complicated patients treated with the combined therapy, we observed that 3 patients reported an improvement, 1 case with no variation of the pain related to arthralgia, 1 patient with mild kidney failure showed a slight decrease in creatinine values and a slight increase in glomerular filtration rate value. In comparison, 2 patients with pericarditis and pleurisy had amelioration of their conditions (Fig. 1B); overall, 86% (n=6/7) of complicated cases showed improved disease-related conditions (Fig. 1B).

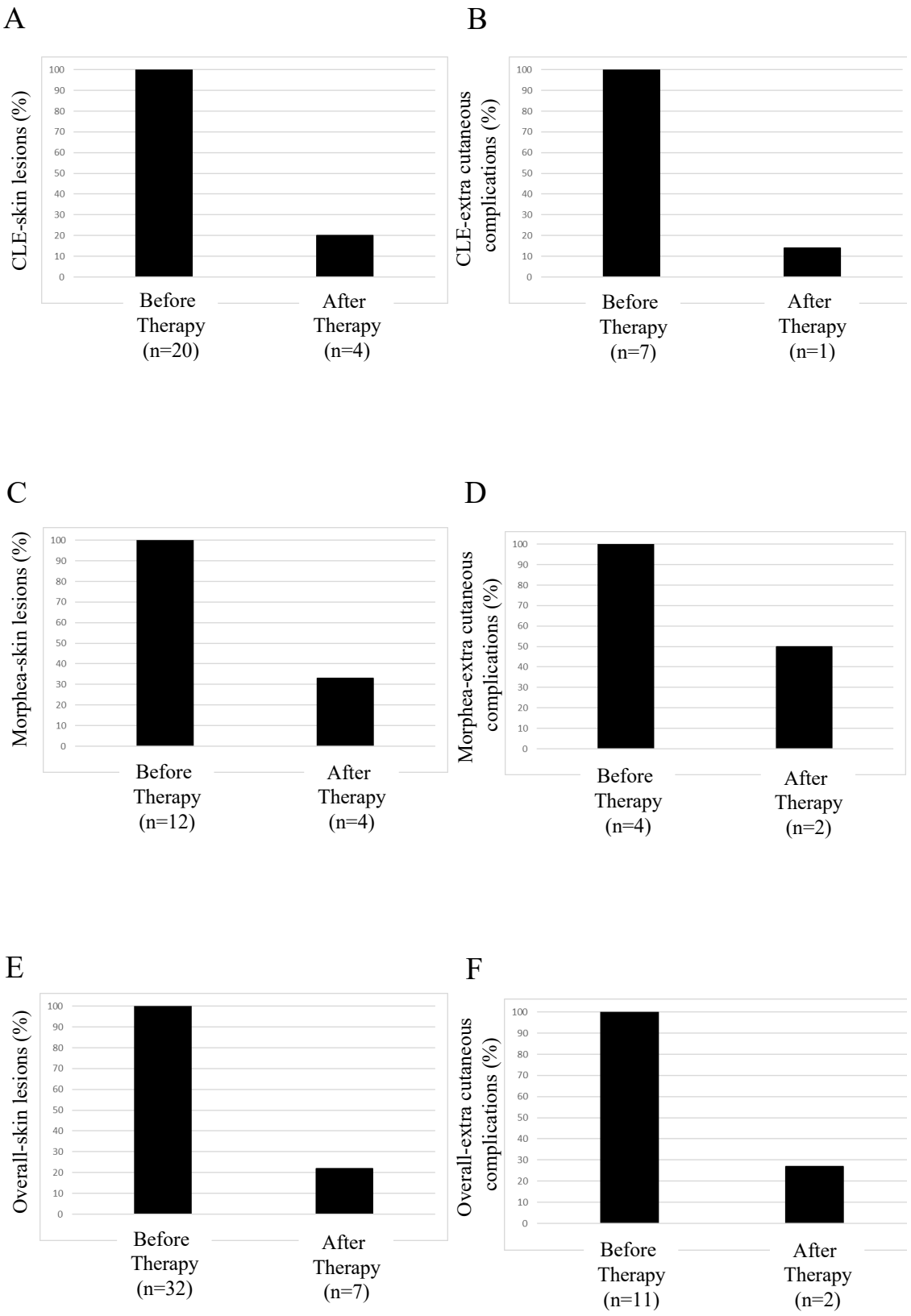

Fig. 1. *Percentage of skin lesions and extra-cutaneous complications before and after stem cell therapy. Panels A and B refer to patients affected by Cutaneous Lupus Erythematosus (CLE), panels C and D to Morphea patients and panels E and F to the overall population (CLE + Morphea patients).*

Morphea patients

Patients with morphea (n=12) had a mean age of 39±7 years. Out of 12 patients, 10 were females, and 2 were males. All patients showed morphea-related skin lesions (morphea plaques), while 4 subjects referred to joint contractures as extra-cutaneous involvement (complicated patients).

The outcome of stem cells therapy in Morphea patients

All patients agreed to undergo stem cell local injections in the affected skin areas, and complicated patients accepted to undergo the combined therapy. After 1 month of stem cell therapy, 67% (n=8/12) of patients showed an improvement in morphea-related skin lesions (Fig. 1C). Concerning 4 complicated patients, 2 reported an improvement and 2 no variation in symptoms related to joint contractures (Fig. 1D). Overall, 50% (n=2/4) of complicated cases showed an improvement in disease-related conditions.

The outcome of stem cells therapy in the overall population

Considering both patients with CLE and morphea, who agreed to undergo local and, when appropriate, local/IV stem cell therapy, we observed the following results. Of the 32 patients treated with local injection of stem cells, 78% (n=25/32) showed an improvement in affected skin areas (Fig. 1E). By stratifying the study population in not-complicated and complicated cases, we observed that: i) 80% of total patients only affected by skin lesions (n=16/20), who were treated with intradermal injection of stem cells, showed improvement of the skin lesions; ii) 73% of total patients, who also exhibited an extracutaneous involvement (n=9/11) and received the combined intradermal/IV injection of stem cells, reported an improvement of the disease (Fig. 1F).

DISCUSSION

Autoimmune diseases occur when the body's immune system attacks its tissue due to an inappropriate immune response, considering the body cells as self-antigens. The loss of immune homeostasis and an inflammatory dysregulation underlying these autoimmune conditions lead to tissue damage by activating autoantibodies and autoreactive T lymphocytes. Autoimmune diseases can affect different organs/tissues, and the skin represents one of the most affected. Among the skin-related autoimmune diseases, CLE and Morphea are included. Both conditions are characterised by simultaneous activation of the innate and adaptive immune system (8), leading to an autoimmune response against the own epidermis, depending on the combination of individual genetic susceptibilities and different environmental triggers, such as UV rays, trauma and radiation (20, 21). In addition, extra-cutaneous manifestations may occur, resulting in a more severe phenotype (4, 5).

Early diagnosis and treatment are necessary to minimise damage in severe patients. However, it is worth noting that currently, there are no specifically approved treatments for these diseases.

In this context, the application of stem-cell therapy could be useful. SCs have the stronger power of self-healing, thus keeping the organism in balance. Consequently, SCs are the key to recovering from a specific disease. Very recently, adult SCs have shown promise as a type of cell-based therapy option for autoimmune diseases, thanks to their immunomodulatory properties (22). Furthermore, according to the literature (13), autologous SCs, i.e. derived from the same patient, hold a safe option for patients with a severe autoimmune disease without the risk of any immune rejection and allogenic sensitisation. In addition, it has been reported that SCs from adipose tissue seem to be the most promising stem cell types, as they are available in high quantities and can be accessed easily and non-invasively (18, 19).

Here, we report the application of autologous ASCs in two cohorts, including 20 patients with CLE and 12 Morphea cases. Considering the overall population treated with stem cell therapy (patients with CLE and morphea, complicated or not), we found that after 1 month of stem cell treatment, 78% of the subjects treated with local injections showed an improvement in the skin lesions. In contrast, 73% of the subjects treated with the combined therapy (intradermal/IV stem cell injection) reported improved symptoms related to extra-cutaneous complications (Fig. 1E-F). The rest of the sample showed stability of the disease, and, notably, no patients reported worsening skin lesions. In particular, regarding patients with CLE, 80% showed an improvement in CLE-related skin lesions, while 4 patients showed skin disease stability (Fig. 1A). Concerning the complicated CLE patients, who accepted the combined therapy, 2/3 of them with pain related to arthralgia reported an improvement, and the remaining one had no variation; one patient with mild kidney failure showed a slight improvement in kidney function.

In contrast, the patients with pericarditis and pleurisy had amelioration of their conditions. Considering the latter patients, it is worth noting that we cannot exclude that the improvement of pericarditis and pleurisy could be related to the concomitant treatment with acetylsalicylic acid. On the other hand, considering patients with Morphea, 67% of subjects showed an improvement in morphea-related skin lesions, while in the remaining cases, the disease was stable with no worsening (Fig. 1C). Concerning the complicated Morphea cases, half reported an improvement and half no variation in symptoms related to joint contractures (Fig. 1D).

Overall, ASCs therapy improves skin lesions in 80% and 67% of cases with CLE and Morphea, respectively. The better improvement of skin lesions in patients with CLE than in those with morphea could be due to the higher grade of sclerosis in the skin lesions related to morphea. In the case of extra-cutaneous manifestations, we observed an overall improvement in 86% and 50% of cases with CLE and Morphea, respectively. This difference may be due to the low number of Morphea complicated cases (n=4).

In conclusion, autologous stem cell-based therapy represents the highest level of modern medicine. Our study promotes the application of autologous ASCs as an effective therapeutic strategy in autoimmune diseases. Further studies in larger cohorts will validate this regenerative medicine's efficacy and safety profiles, extending it to other skin-related autoimmune diseases.

REFERENCES

1. Hocking AM, Buckner JH. Genetic basis of defects in immune tolerance underlying the development of autoimmunity. Front Immunol. 2022; 13:972121.
2. Vojdani A, Vojdani E. The Role of Exposomes in the Pathophysiology of Autoimmune Diseases I: Toxic Chemicals and Food. Pathophysiology. 2021; 28(4):513-543.
3. Autoimmune Disease List. [(accessed on 2 November 2021)]. Available online: https://www.autoimmuneinstitute.org/resources/autoimmune-disease-list/ [Ref list].
4. Condie D, Grabell D, Jacobe H. Comparison of outcomes in adults with pediatric-onset morphea and those with adult-onset morphea: a cross-sectional study from the morphea in adults and children cohort. Arthritis Rheumatol. 2014; 66(12):3496-504.
5. Petty AJ, Floyd L, Henderson C, Nicholas MW. Cutaneous Lupus Erythematosus: Progress and Challenges. Curr Allergy Asthma Rep. 2020; 20(5):12.
6. Okon LG, Werth VP. Cutaneous lupus erythematosus: diagnosis and treatment. Best Pract Res Clin Rheumatol. 2013; 27(3):391–404.
7. Zulian F, Athreya BH, Laxer R, et al. Juvenile localised scleroderma: clinical and epidemiological features in 750 children. An international study. Rheumatology (Oxford). 2006; 45(5):614-20.

8. Li SC, Torok KS, Pope E, et al. development of consensus treatment plans for juvenile localised scleroderma: a roadmap toward comparative effectiveness studies in juvenile localised scleroderma Arthritis Care Res (Hoboken). 2012; 64(8):1175-85.
9. Quain RD, Werth VP. Management of cutaneous dermatomyositis: current therapeutic options. Am J Clin Dermatol. 2006; 7(6):341–351.
10. Laxer RM, Zulian F. Localized scleroderma. Curr Opin Rheumatol. 2006; 18(6):606-613.
11. Elhage KG, Zhao R, Nakamura M. Advancements in the Treatment of Cutaneous Lupus Erythematosus and Dermatomyositis: A Review of the Literature. Clin Cosmet Investig Dermatol. 2022; 15:1815-1831.
12. Wenzel D, Haddadi NS, Afshari K, Richmond JM, Rashighi M. Upcoming treatments for morphea. Immun Inflamm Dis. 2021; 9(4):1101-1145.
13. Mohammedsaleh ZM. The use of patient-specific stem cells in different autoimmune diseases. Saudi J Biol Sci. 2022; 29(5):3338-3346.
14. Zakrzewski W, Dobrzyński M, Szymonowicz M, Rybak Z. Stem cells: past, present, and future. Stem Cell Res Ther. 2019; 10(1):68.
15. Bacakova L, Zarubova J, Travnickova M, et al. Stem cells: their source, potency and use in regenerative therapies with focus on adipose-derived stem cells - a review. Biotechnol Adv. 2018; 36(4):1111-1126.
16. Hoang DM, Pham PT, Bach TQ, et al. Stem cell-based therapy for human diseases. Signal Transduct Target Ther. 2022; 7(1):272.
17. Ding DC, Shyu WC, Lin SZ. Mesenchymal stem cells. Cell Transplant. 2011; 20(1):5-14.
18. Frölich K, Hagen R, Kleinsasser N. Mesenchymale Stammzellen aus Fettgewebe (ASC) - Grundlagen und Anwendung in der HNO-Heilkunde [Adipose-derived stromal cells (ASC) - basics and therapeutic approaches in otorhinolaryngology]. Laryngorhinootologie. 2014; 93(6):369-380.
19. Varghese J, Griffin M, Mosahebi A, Butler P. Systematic review of patient factors affecting adipose stem cell viability and function: implications for regenerative therapy. Stem Cell Res Ther. 2017; 8(1):45.
20. Dharamsi JW, Victor S, Aguwa N, Ahn C, Arnett F, Mayes MD, Jacobe H. Morphea in adults and children cohort III: nested case-control study-the clinical significance of autoantibodies in morphea. JAMA Dermatol. 2013; 149(10):1159-65.
21. Kuhn A, Wenzel J, Bijl M. Lupus erythematosus revisited. Semin Immunopathol 2016; 38(1):97–112.
22. Nawab K, Bhere D, Bommarito A, Mufti M, Naeem A. Stem Cell Therapies: A Way to Promising Cures. Cureus. 2019; 11(9):e5712.