2018 will be the 9th year of continuous publication of the Surgical & Cosmetic Dermatology (S&CD)!!

First, I would like to make some brief comments about the changes that the current global scenario of scientific communication presents in its ethical and commercial aspects, and how it has been maintaining and managing this journal so far.

The possibility of obtaining open access articles and charging the authors varying amounts for publication adopted by many journals are the main reasons for these changes that, along with many benefits, also bring some concerns. The main benefits are increasing access to scientific knowledge and author exposure, whereas the main concern refers to the appearance of the perverse, so-called “predatory journals”, which do not evaluate the articles in a legitimate and through fashion, publish fake impact factors and have as their goal to receive the publication fees as fraudulent profit.

Therefore, we highlight S&CD’s attitude, not charging the authors publishing fees since it has institutionally documented sources of funding (Brazilian Society of Dermatology - SBD since the start and, more recently, Brazilian Society of Dermatologic Surgery – SBCD) and abides by the detailed ethical and structural requirements of indexing databases (peer review, publication of articles from different geographical regions and 50% of research articles).

With these principles in mind, we have tried with much effort to manage this journal, which has proved to be encouraging to Brazilian authors, with publications in Dermatologic Surgery, Cutaneous Oncology, Imaging Diagnosis, Technological Advances in Dermatology and Cosmiatry. We have also raised interest from researchers from Latin America, Portugal and the Middle East, without forgetting great names of European Dermatologic Surgery such as Dr. Eckart Haneke, who help us with their experience.

We also count on a strong scientific support from the Ibero-Latin American College of Dermatology and the SBCD, recently strengthened through formal engagements with Dr. Omar Lupi and Dr. Mauro Enokihara, vice-president and president of these respected institutions, respectively.

S&CD is already included among many important regional and international databases such as: REDALYC, LATININDEX, LILACS, SCOPUS and DOAJ (Directory of Open Access Journals), an online directory that promotes access only to good quality and peer-reviewed publications. S&CD is also present in researcher-oriented social media, among them ResearchGate. I would like to personally thank you all, Editorial Board, National and International Review Board, SBD librarians and authors, for the collaboration in this journey towards the realization and publicity of S&CD.

Some barriers are overcome, but there are still many challenges to face.

Profa. Dra. Bogdana Victoria Kadunc
Editor-in-chief of Surgical & Cosmetic Dermatology
Literature review: Auricular disorders Part 1- physical and thermal change related traumas

Revisão da literatura: afecções auriculares parte 1: traumas físicos e por alteração térmica

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ABSTRACT
The ear is a structure with anatomical peculiarities. It is located in an area exposed to trauma and susceptible to a series of dermatoses that are often underdiagnosed by dermatologists and deserve attention. A literature review was carried out including the embryology and anatomy of the auricular pavilion, as well as the diagnosis and treatment of traumatic lesions of physical nature and due to changes in temperature, among others.

Keywords: Anatomy; Ear; Ear diseases

RESUMO
A orelha é estrutura com peculiaridades anatômicas, localizada em área exposta a traumas e suscetível a uma série de dermatoses que merecem atenção, muitas vezes subdiagnosticadas pelos dermatologistas. Realizou-se revisão de literatura incluindo a embriologia e a anatomia do pavilhão auricular, bem como diagnóstico e tratamento das lesões traumáticas de natureza física e por alterações de temperatura, entre outras.

Palavras-chave: Anatomia; Orelha; Otopatias

INTRODUCTION
The ear is divided into three main parts: outer ear, middle ear and inner ear. The outer ear comprehends the pinna and the ear canal.1

The pinna or auricle arises from the second pharyngeal arch and can be seen for the first time in the 6th week of gestation. The lining of the external auditory canal is contiguous with the skin and with the external surface of the tympanic membrane, is ectodermal in origin1 and has modified apocrine glands known as ceruminous glands and sebaceous glands, both associated to hair follicles.2

Due to its anatomical location, the auricle is subject to trauma and exposed to temperature changes, besides presenting many signs associated to different cutaneous and systemic conditions.

THE EAR
The pinna develops from the first pharyngeal arch and is composed primarily of cartilage and skin, which are firmly adhered to the anterior region and, in lesser degree, to the posterior. Due to its anatomical location, the pinna is commonly exposed to the sun the cold. Neoplasms of the cartilage are rare in the pinna; most of them occur in the skin or annexes.

The external auditory canal measures 2-3 cm in length and is made by a cartilaginous portion in a bony portion. Only
the cartilaginous portion has hair follicles, sebaceous and ceruminous glands. In the upper and posterior portion of the canal, the cartilage is incomplete and filled by fibrous tissue. The temporomandibular joint and the parotid gland are found in the anterior region of the auditory canal, and the mastoid bone and the facial nerve are located posteriorly.

The innervation of the pinna and the external auditory canal comes from the great auricular nerve, through the auricular branch of the vagus nerve, auriculotemporal branch of the trigeminal nerve and lesser occipital nerve.\textsuperscript{1}

**EMBRYOLOGY**

The development of the outer ear begins at the end of the 4\textsuperscript{th} week of gestation from six auricular hillocks. Congenital malformations during the fusion of the hillocks are not rare because of their complex development.

The first ones appear on the lower cervical region and elongate towards the skull with the development of the mandible, changing the position of the conchae to the same level as the eye. The lower position of the conchae is frequently associated to malformations, most of them chromosomal.

The external auditory canal is formed from the posterior portion of the first pharyngeal groove inwards as a funneled tube until it reaches the endodermal lining of the tympanic cavity. In the beginning of the 9\textsuperscript{th} week, the epithelial cells of the floor of the ear canal proliferate and form a plaque of the ear canal. This plaque degenerates in in the 7\textsuperscript{th} month, and when it remains, congenital deafness develops.\textsuperscript{1,2}

**EAR ANATOMY**

The outer ear comprises the earlobe inferiorly, a soft and round structure. The outer frame contiguous to the earlobe is a firm structure known as helix. Superior to the helix is the scapha, medially the antihelix (frame opposite to the helix) and in its caudal end the antitragus. Medially to the antihelix, the cartilage forms the concha, and superiorly another concavity known as triangular fossa. The tragus is found medially in the area of attachment of the ear to the cephalic segment around the auditory canal (formed by the tympanic portion of the temporal), which it protects, in the shape of an “S” (Figure 1).

The muscles of the ear are rarely appreciated because of their rudimentary features but need to be known and described. They can be considered muscles of facial expression belonging to a system of sphincters, which in many animals is quite distinct with functions of movement of the ears or closure of the auditory canal during hibernation.\textsuperscript{1,2} The ear is intensely vascularized, particularly in the lobe, and the vessels can be found at its base.

**TRAUMA/HEMATOMA**

Trauma of the pinna frequently occurs due to contusion, laceration or loss of the whole pinna. In some cases, the pinna can be resutured successfully. The reconstructive technique must follow the basic principles of plastic surgery. The cutaneous suture should be performed cautiously with meticulous control of bleeding, because the skin is juxtaposed to the underlying cartilage. Areas that are extremely macerated should be removed with the aim of minimizing deformities and scars.

Hematoma (Figure 2) of the pinna occurs not only in wrestlers, boxers, rugby, soccer players and other sports, but it is also common in children. Hematomas are common in contact sports such as wrestling, due to the exposure of the ears. When the ear is struck, a detachment between the perichondrium and the cartilage can occur, creating a space filled with blood, forming a hematoma. Since the cartilage does not have its own blood supply, this space can lead to necrosis or infection.

Trauma is the most common cause of hematomas although blood dyscrasias can lead to small hemorrhages. The external region of the pinna is the most affected area due to its exposed location. Blood builds up quickly after a trauma, dissecting the tissue between the perichondrium and the cartilage, originating a purple collection involving the whole pinna. If this collection is not treated early, the blood rearranges into a fibrous mass, leading to cartilage necrosis by regional ischemia. The cicatricial mass, particularly after repetitive trauma, creates a deformity known as “cauliflower ear”.\textsuperscript{3}

Treatment is based on prompt drainage of the accumulated blood. Due to the possibility of developing perichondritis, it must be done under sterile conditions. Large-spectrum antibiotics are used, including those with action against Pseudomonas aeruginosa pre- and post-operatively. The incision should be parallel to the helix, in the scapha, and must be large enough to allow complete aspiration of the hematoma, as early as possible to avoid its rearrangement. In this case, curettage would be needed. Small drains can be inserted to prevent a new collection of blood or serum forming, however, they must not be left for longer than 48 hours because of the risk of infection.

A pressure dressing must be adapted to the shape of the ear over the area of the hematoma and another in the posterior region of the pinna, and a suture that runs through the whole pinna can be used. It should be kept in place for at least 48 hours. Small residual collections should be drained according to sterile conditions.

**Figure 1:** Schematic representation of the surface anatomy of the ear.
techniques. Antibiotic is used for 5 to 7 days, and perichondritis should be treated immediately.

LACERATIONS AND ABRASIONS

Pinna lacerations require meticulous and thorough closure, realigning the fragments and maintaining its outline. A comparison with the opposite ear should be made so as to maintain symmetry as much as possible. Lacerations and abrasions should be treated under local anesthesia, however, in children and in severe or extensive cases general anesthesia is the best option. Cases of laceration and rupture of the earlobe are common due to trauma of earring piercings.4

The wound should be inspected and thoroughly and insistently washed to remove any foreign or necrotic material. A debridement must be performed (Figure 3) in the most conservative way possible. Simple skin lacerations are generally closed with 6-0 nylon sutures and the cartilage with absorbable sutures. It might be necessary to remove a small strip of cartilage along the laceration to facilitate closure of the perichondrium. The original shape of the border of the helix must be maintained.

Pressure dressings are required for 24 hours, and oral antibiotics for 1 week. When the laceration extends to the auditory canal, a dressing is applied inside the canal to help mold it after suturing, keeping it in place for several weeks in order to avoid stenosis.

Abrasions should be cleaned and covered with gauze, antibiotic and occlusive dressing for 24 hours. After removal, the wound is treated only with plain dressings until reepithelization occurs. If there is intense pain under the dressing, it should be removed immediately and the wound should be thoroughly inspected in search of signs of ischemia, hematoma or infection. When suturing is needed, it should start from the most difficult area of surgical access, such as the auditory canal and concha, finishing the surgery in the periphery. In some cases of extensive lacerations, wedge resection or minimal debridement is required, and the edges should be brought close in a way as not to alter the normal curvature of the ear.

In extensive abrasions or when small pieces of skin are lost, the lesion must be exhaustively cleaned and occlusive dressing with vaselined rayon gauze must be applied. Epithelization will occur satisfactorily in 7 to 10 days, since the ear has a great ability of secondary healing.

In the cases with significant loss of skin and exposure of the perichondrium, skin grafts are used for the repair. The preferred donor site is the contralateral postauricular region because it has similar color and texture.

When there is loss of perichondrium with exposure of the cartilage, the following can be used: wedge resection and primary closure, local flap, excision of the cartilage and skin graft, or maintenance of the open wound. Wedge resection with primary closure is used when the lesion is small or has a peripheral location.

When local flaps are used, the preferred area is the postauricular region. In some cases, the flap can be taken to the exposed area and, in a second stage, return it to its origin using a skin graft in the stump area. In other cases, the cartilage can be resected with a graft placed in the stump area. This procedure has the advantage of involving only one stage.6

Another alternative would be to maintain the wound open, frequently changing the dressing with gauze soaked in saline, waiting for the formation of granulation tissue and performing secondary repair subsequently.

PARTIAL LOSS OF THE PINNA

When there is a substantial detachment of the ear but the blood flow is preserved by a small pedicle, the tissues should be sutured to their original locations, with as little trauma as possible. For the fixation of the cartilaginous framework, 6-0 monofilament nylon sutures should be used with the goal of keeping it in position. It is necessary to reevaluate tissue viability 48 hours after the initial procedure.

Strauch3 observed that the ear survived when the pedicle was made of only one portion of the lobule or of a small segment close to the insertion of the pinna on the cephalic seg-
Auricular disorders: physical and thermal traumas

The ideal treatment in partial avulsions with cartilage exposure should aim for its immediate closure. The cartilage can be preserved with a graft or a partially adhered segment, covered by adjacent flap or buried under the flap (pocket technique). Traumas can range from partial to total loss of the pinna. After anesthesia, cleaning and debridement, as already described, the extension of tissue loss and the state of the remaining structure determines if the wound can be sutured primarily or if it will need reconstruction.

Loss of skin with intact perichondrium can be repaired with a partial or total graft. Postauricular skin provides tissue that matches the skin of the pinna; supraclavicular areas can also be used. If there is loss of skin and perichondrium, a vascularized tissue is needed to cover the cartilage. We can then use a flap from the pre or postauricular area but preauricular flaps can seem thick for the lateral aspect of the pinna and have less than satisfactory cosmetic results. If the area of cartilage exposed is small, it can easily be excised and the edges of the wound sutured together. If none of these options is possible, we can wait for tissue granulation, maintaining with local dressings until a graft can be done.

Reconstruction of total tissue loss, such as the helix, earlobe and segmental defects affecting the upper, middle or lower aspect of the pinna can be quite complex or require multiple steps. The method of choice for reconstruction will depend on the size of the defect. Defects with less than 2 cm can be transformed into small ellipses and closed primarily. Benchamam et al. described a technique (Antia-Buch) with the use of upper and lower chondrocutaneous flaps of the helix adjacent to the defect. After incision of the skin and cartilage of the lateral aspect of the pinna, the skin of the medial aspect is widely undermined allowing advancement of the flaps over the defect. These advancement techniques can correct large defects, however, sometimes the reduction in size of the ear is not acceptable. In these cases, a reconstruction in stages must be performed, bringing another tissue to the defect to reconstruct the helix. There is a variety of techniques for rotation of tubular flaps for this purpose. Another option involves advancement of the skin of the medial aspect of the pinna and of the postauricular area, that can be combined to cartilage grafts. This technique can be used in larger ear lesions. When multiple surgical steps are needed, the cartilage must not be left exposed. It can be covered embedding it in the postauricular skin where the affected area is washed, cleaned, dermabrased and anatomically sutured to the remaining ear stump. Therefore, a postauricular pocket or tunnel is created, which protects this stump. Two weeks later, the embedded portion is re-exposed in order to complete epithelization. This provides an adequate protection, and the overlying skin can be used for the first phase of the reconstruction. Cartilage from the contralateral concha can be used for a composite graft. This technique can also be used to reconstruct the ear with a larger segment loss.

TOTAL AVULSION OF THE PINNA

In the literature, the success of reimplantation of completely avulsed was possible with aggressive treatment with intravenous antibiotics, anticoagulants and vasodilators. Venous congestion should be relieved through multiple punctures or incisions to allow drainage.

Microsurgery is performed whenever possible nowadays, with new vascular anastomosis conducted with microsurgical techniques. Anticoagulation, antibiotic therapy, frequent dressings and venous drainage techniques are also needed.

The part amputated should be transported in sterile conditions, cooled and resutured as quickly as possible. When it is not possible to rescue it, we can opt for fixed prosthesis, osseointegrated implants or total reconstruction of the pinna.

Ear amputations have an extremely complex surgical solution. Until recently, the only possible option was saving the cartilaginous framework.

Careful attempts of saving the extremely damaged cartilaginous framework can make reconstruction difficult. This occurs because the traumatized or damaged cartilage generates inadequate details or protrusions.

Current reconstruction techniques using autogenous cartilage grafts covered by flaps of the fascia of the temporal muscle offer better results than those achieved with attempts of conservation of cartilage in distant sites.

In amputation cases that are treated with placement of the denuded cartilage underneath the postauricular flap and release of the flap in 2 to 4 weeks, late retraction of the inserted segment is seen, as well as loss of the outline and contour of the ear.

Reconstructions with autogenous cartilage using a segment of the contralateral concha or rib cartilage associated to proper cutaneous coverage frequently yield more acceptable ears from the cosmetic point of view.

Reimplantation of large segments or of the whole ear is sometimes destined to failure, except if the recipient area is first prepared to increase vascularization.

ANIMAL BITES

Basic concepts of repair of ear lesions are described previously with some specific considerations. First, identify the animal (including human) and extension of the lesion. As with other wounds, photographic documentation is necessary, as well as tetanus prophylaxis and investigation regarding rabies prophylaxis.

All patients should be treated with antibiotic. Such lesions are contaminated by aerobic and anaerobic microorganisms, the most frequent being detected in human bites: S. aureus, Pasteurella multocida, Streptococci, Bacteroides and Fusobacterium sp. Penicillin or ampicillin covers most pathogens, including P. multocida. Amoxicillin/clavulanate and ticarcillin have a broader spectrum of action, including S. aureus and beta-lactamase positive. Treatment begins with thorough cleaning and irrigation of the wound. If it is very large and requires surgical reconstruction, the timing must be considered (early/late). It is controversial, but late reconstruction in cases of severe human bites or in those that occurred many hours before is possible if it is earlier than 5 hours, with the wound clean and able to be primarily closed. In
case the repair cannot be performed immediately, local dressings should be applied until there is granulation tissue in the area, and it is clean and free of infections.

**LESIONS BY THERMAL CHANGES**

**Cold or freezing**

Localized trauma resulting from exposure to the cold, being tissue loss due to direct cellular and vascular damage. Animal studies show that after freezing, chondrocytes show immediate evidences of injury followed by minimal changes in the epidermal cells. However, the microvasculature shows dramatic changes, with detachment of endothelial cells from the internal elastic lamina (ice crystal formation), stasis and extravasation of red blood cells, interstitial edema, low flow and tissue necrosis. Release of arachidonic acid metabolites, thromboxane A2 and prostaglandins worsen the necrosis and act in the regulation of capillary permeability even further. Crystallization of the intracellular fluid can be primarily responsible for this. The ear becomes edematous, erythematous, soft and can present bullae with fluid discharge through the skin. In the final stages of a cold burn, regional capillaries become filled with red blood cells due to the low flow through them and loss of serum due to abnormal permeability. Recovery is slow due to obstruction, thrombosis and ischemia, that lead to necrosis of the affected tissue.

Treatment is based on the severity of the lesion. The ear should be treated gently to minimize injury in a tissue already damaged and devitalized. Massage and other manipulations should be avoided. Allow ear temperature to gradually return to body temperature. When vasodilation occurs, pruritus and intense pain will occur, which should be controlled with analgesics. In children, restraining the hands is necessary. In severe cases or in those with prolonged exposure, the use of anticoagulants to minimize thrombosis and intravascular coagulation is recommended, and this treatment should be continued until the exact damaged area is identified. In all cases, combined estrogens and bioflavonoids can be used intravenously to improve abnormal capillary permeability. Antibiotics might be necessary to prevent infection of the devitalized tissue. If necrosis of some areas of the pinna becomes obvious, we must wait until it is completely defined before surgical excision is performed. Spontaneous detachment of the devitalized tissue allows for minimal surgery and a better cosmetic result. If there is suggestion of infection and gangrene, surgical excision must be performed immediately.

Cold can also be an important factor in juvenile spring eruption, chondrodermatitis nodularis helicis, chilled blains, lupus erythematosus and cryoglobulinemia. Lesions similar to those caused by the cold can be triggered by the excessive use of ethyl chloride spray for ear piercings. 

**BURNS**

The pinna is particularly susceptible to thermal trauma because of its exposed location and to the minimal amount of subcutaneous tissue between the skin and perichondrium. Tissue can be lost by direct thermal trauma and progressive dermal ischemia seems to be the result of a complex interaction of factors, including increased vascular permeability with significant edema formation and release of chemical mediators with potent effects on vascular function. The damaged tissue functions as a substrate for infections and, finally, cartilage loss and subsequent auricular deformity.

First degree burns affect only the epidermis. The ear becomes erythematous, warm and painful to touch, and a watchful waiting approach must be used. Second degree burns involve the epidermis and part of the dermis and can be subdivided into deep and superficial. Third degree burns or full-thickness are those where there is complete destruction of the epidermis and dermis. Many times, the differentiation between a partial deep burn and a full-thickness burn can be challenging. Mild edema can occur with a deep burn and a full-thickness burn might not present with pain or tissue dissection, leading to partial or total amputation of the ear. The lack of pain sensation does not help to determine the depth of the burn, because it varies in second and third degree burns. The main caution in cases of second and third degree burns is the prevention of chondritis, a serious infection that also leads to loss of cartilage. A meticulous care of the wound should be undertaken, and local pressure avoided. Purdue described regimens that involve gentle washing of the ear and use of antibacterial soaps once or twice a day, followed by the application of topical antibacterial creams, reducing the incidence of chondritis to 0% to 3%. Exposed cartilage due to severe burns should be covered with vascularized tissue in the attempt of preserving it.

**ARTIFACTUAL OR IATROGENIC LESIONS**

The majority of other forms of trauma tend to be artifactual or iatrogenic. Piercing can lead to infection, and the complications from this procedure can reach up to 34%, with keloid formation, nickel contact dermatitis and earlobe pseudo-lymphoma. Rupture of the tympanic membrane and acquired atresia of external auditory canal usually result from surgical trauma or inadequate attempts to remove foreign bodies or impacted cerumen. Contracture of the external auditory canal can follow burns.

**ACANTHOMA FISSURATUM**

Fissured lesion, sometimes presenting as an erythematous, scaly papule resembling an actinic keratosis. It occurs behind the ear or on the upper portion of the ear, at the junction with the
Auricular disorders: physical and thermal traumas

Scalp and is caused by ill-adjusted eyeglass temples. The differential diagnosis includes basal cell carcinoma, actinic keratosis and early stage squamous cell carcinoma.5,10

FOREIGN BODY

Foreign bodies in the ear canal are a common scenario at the emergency departments, more commonly in those younger than 5 years of age. They can be asymptomatic or painful, cause inflammation or infection of the external auditory canal,6 otitis externa and tinnitus.11,12

In childhood, most correspond to small objects and grains, and in adults, the cotton in cotton buds. Treatment is washing the ear or surgery. The list of objects that can be found in the ear canals is varied, being impacted cerumen a common cause of irritation. On the other hand, little cerumen, particularly in women, can lead to pruritus. One must be careful and attentive with the excessive use of cotton, excessive cleaning or scratching of the ear. The use of metallic objects can lead to contact dermatitis. Abrupt attempts to remove the object lead to lesions of the isthmus or other areas. If the foreign body is smaller than the opening of the canal, it can be removed with Hartmann forceps. Larger objects should be “fished” with a handle or, in some cases, with injection of water through the wall similarly to cerumen removal, or even aspirated with appropriate devices. In the case of insects, they should first be killed with a cotton plug embedded in ether, chloroform or alcohol for 5 minutes and then washed out. If the child is agitated, general anesthesia is needed. Many cases of trauma of the tympanic membrane and ossicles take place with children’s movements during this type of procedure. When objects are found beyond the isthmus, its removal must be done with anesthesia. Flies can deposit their eggs in the ear and the resulting myiasis can lead to pain, inflammation and, occasionally, more severe complications.

Loose hairs in the ear canal can also be the cause for noises in the ear.

CONCLUSIONS

This review evidences a great variety of conditions that can affect the ear. Even though we sometimes have the impression that diseases affect the ear because it is part of the skin, what is true in many cases, others refer exclusively to this region and its own features and function. We addressed dermatitis, trauma, infections, congenital malformations and benign and malignant tumors, making this subject extremely interesting for the dermatologist and also surgeons; a broader knowledge allows opening up a range of possible differential diagnoses and treatment options.

REFERENCES


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Treatment of viral warts with intralesional bleomycin

ABSTRACT

Viral warts are one of the most prevalent dermatoses. The clinical picture varies from a single lesion with spontaneous cure to multiple recalcitrant lesions. Bleomycin sulfate is a cytotoxic action drug approved for the chemotherapeutic treatment of some malignancies. There are a number of studies that have been carried out during the last 45 years demonstrating its usefulness in dermatology, especially in intralesional therapy for viral warts, meaning it is an excellent option for lesions in difficult-to-handle topographies and for cases that do not respond to other approaches.

Keywords: Bleomycin; Drug therapy; Viral wart

INTRODUCTION

Bleomycin was originally isolated from the fungus Streptomyces verticillus, in 1962, by Umezawa et al. This glycopeptide has antibacterial and antiviral effects, however, it is very useful because of its cytotoxic effect. It was approved by the Food and Drug Administration (FDA) for systemic treatment of testicular carcinoma, malignant pleural effusion, Hodgkin and non-Hodgkin lymphoma. It was approved as adjuvant chemotherapy for the treatment of some advanced form of cutaneous squamous cell carcinoma.2-4

Despite not being approved for intralesional treatment, there are many case reports and studies in dermatology demonstrating efficacy and safety for the treatment of many conditions, such as viral warts, keloids, hemangiomas, vascular malformations and some malignant neoplasms (basal cell carcinoma, keratoacanthoma, squamous cell carcinoma and Kaposi sarcoma), for example.3,5,6

Pharmacodynamics

Knowing its low absorption when administered orally, intravenous, intramuscular or subcutaneous administration should be used in cases of systemic treatment, according to the approach protocol.3

Due to its low transepidermal absorption, a very high dose is required to achieve minimum tissue concentration when applied topicaly.3 Therefore, intralesional injection becomes an option for
the treatment of localized lesions. It is considered that systemic absorption is minimal after one intralesional injection.7,8

James et al.9 dosed the serum level of bleomycin in seven patients with palmar viral warts after injection of one unit (U) of the drug. Serum concentration ranged from 7.5 to 113.5ng/ml and from 4.9 to 34.8ng/ml after 45 and 120 minutes, respectively.

It is mainly excreted through the kidneys, but liver participation is suggested. There is also enzymatic destruction with bleomycin hydrolase, enzyme found in many tissues, in varying concentrations.3

Mechanism of action

It acts mainly via cleavage of the DNA strand. In the presence of oxygen, Fe2+ and a reducing agent, bleomycin transfer electrons from Fe3+ to oxygen, generating reactive oxygen species. The free radical causes oxidative damage to the nucleotides, cleaving the DNA strand.7 There is also degradation of cellular RNA.11 Another mechanism of action related to a good therapeutic response in viral warts is endothelial damage.11 Direct effect of bleomycin on human papillomavirus (HPV) was not described.

Because it is a hydrophilic substance, its permeability through the cell membrane is low. Mizuno e Ishida12 demonstrated that the association of bleomycin with lidocaine, procaine, tetracaine, dibucaine, butacaine or ethanol generates an increased cytotoxic effect. These substances would cause a disorganization of the cell membrane and, therefore, increased transmembrane permeability.3,12

Pharmaceutical form

The preparation commercially available is bleomycin sulfate, lyophilized powder, in vial/ampoule with 15U.3,4

Viral warts

Cutaneous viral warts are a common, benign skin infection, caused by HPV, that most frequently affects hands and feet (Figure 1).13,14 The presentation and severity can vary from a single lesion with spontaneous resolution to multiple, chronic lesions. They affect any age group, with a prevalence ranging from 5% to 30% in children and young adults.13-15

Immunosuppressed individuals are highly susceptible, with long standing lesions and reduced response to treatment.13,15 Besides aesthetic and functional limitations, we should bear in mind the association between HPV and cutaneous squamous cell carcinoma.15 Therefore, it is important to biopsy chronic verrucous lesions in adults, since they can represent malignant epithelial neoplasms.

Cryotherapy, salicylic acid, lactic acid, glutaraldehyde, imiquimod, electrocautery, surgical excision, curettage, podophyllotoxin, cantharidin, 5-fluorouracil (topical or intralesional), immunotherapy, photodynamic therapy and pulsed dye laser are described as therapeutic options.3,13,14

Intralesional bleomycin for the treatment of viral warts

The first description of intralesional bleomycin for the treatment of viral warts was in the 1970s, and since then many studies have confirmed its efficacy and safety. For many authors, this is an excellent therapeutic option for difficult-to-approach sites, particularly the periungual region (Figures 2 and 3), and for recalcitrant lesions, even in immunosuppressed individuals.3,8,9,11,13,14,16,17

Dhar et al.18 assessed 73 patients between 5 and 50 years of age, with a total of 155 warts, comparing cryotherapy with intralesional bleomycin. The cure rate in the bleomycin group was of 94.9%; in the group treated with cryotherapy, 76.5%.

In a similar study, Adalatkhah et al.19 demonstrated cure in 86% and 68% of the cases treated with bleomycin and cryotherapy, respectively. Rossi et al.20 demonstrated a cure rate of 82%, 2.5-fold higher than in the placebo group.

Besides common and palmoplantar warts, there are studies showing efficacy for the treatment of anogenital warts, with a cure rate of up to 70%.21

Since intralesional bleomycin is a modality considered to be off-label, there is no standardized dilution. Most authors describe the use of this drug in varying concentrations, from 1 to 1.5U/ml:

15ml saline 0.9% for 15U bleomycin = 1U/ml.2
5ml distilled water for 15U bleomycin. Mix 1/3 of this solution with 2/3 2% lidocaine = 1U/ml.18,22
4 ml saline 0.9% + 6ml 2% lidocaine for 15U bleomycin = 1.5U/ml.21

The needle should be inserted at the base of the lesion and the injection continued until the local blanching is achieved,18,21 This is an important sign that the medication was correctly injected.3

Most articles mention that the ideal dose should range between 0.1 and 0.3U/lesion, and 3U is the maximum dose recommended per treatment. Usually, 2 or 3 treatments are required, every 3 or 4 weeks.3,18,21,23
For lesions on the hands and/or feet, in the first week after the procedure, the formation of hematic and darkened crusts is expected. This is due to the absolute reduction in blood flow, which results in necrosis and subsequent disappearance of the lesion (Figure 4). In general, there is resolution in 2 to 4 weeks. Facial warts regress gradually and the lesions disappear without crust formation. Blood flow is probably reduced after injection; however, it is not completely blocked due to the rich vasculature of this area. Induction of endothelial apoptosis and direct keratinocyte injury can result in wart regression without necrosis or eschar.

Application of a drop of bleomycin solution on the surface of the lesion, followed by multiple punctures is an alternative to intralesional therapy that was described as translesional multipuncture. Khalid et al. used this technique in 15 patients, using 0.1U/ml bleomycin solution and a 27G needle. The dose used ranged between 0.3 and 0.6ml per treatment. After 4 weeks, another treatment was performed in cases that persisted or recurred. They observed that 46.6%, 73.3% and 86.6% had a good response after 1, 3 and 6 months after the procedure, respectively.

Side effects
Pain is one of the main limiting factors of this treatment, which can last up to 72 hours, with a peak at the time of injection. This would be one of the reasons why some authors use lidocaine as diluent, reducing pain during and immediately after the procedure.
Intralesional Bleomycin: warts

Other common side effects of the intralesional therapy with bleomycin are: erythema, edema, ulceration, hematic crust formation and eschars.\textsuperscript{2,3,16}

Transient hypopigmentation or hyperpigmentation can occur (particularly in phototype IV, V or VI patients), atrophic or hypertrophic scars and hematomas. Less frequently, Raynaud phenomenon, digit necrosis, ungual dystrophy and loss of the nail were described after injection of periungual warts.\textsuperscript{2,4,9} Although rare, flagellate dermatitis can also occur with intralesional therapy.\textsuperscript{3,24}

There are no reports of systemic complications with the use of intralesional bleomycin, except for flu-like symptoms, only described in the treatment of hemangiomas or vascular malformations.\textsuperscript{7,21,25}

Options to reduce pain during intralesional injection\textsuperscript{3,26}

Application of topical anesthetic or local injection
Using small diameter needles (27G or 30G)
Substitution of the needle when multiple punctures are necessary
Stretching or pinching adjacent skin
Applying ice or specific vibration devices
Inject slowly
Avoid a large volume by application area

Contraindication
Hypersensitivity or idiosyncratic reaction to the medication, Raynaud phenomenon, peripheral vascular disease, pregnancy (category D) and breastfeeding.\textsuperscript{3,27}

There are no safety studies regarding intralesional use in the pediatric age group.\textsuperscript{3}

CONCLUSION
Despite being described for many years as an effective and safe therapeutic approach for viral warts, intralesional injection of bleomycin remains an off-label option. Studies revealed a high cure rate even in cases refractory to other therapies, in lesions on difficult-to-handle sites and in immunosuppressed patients. Pain is the main limiting factor of this procedure.

We highlight the importance of histopathology in the evaluation of long standing verrucous lesions in adults, because they can represent a squamous cell carcinoma and not only a plain viral wart.

REFERENCES
19. Adalatkhah H, Khalilollahi H, Amini N, Sadeghi-Bazargani H. Compared therapeutic efficacy between intralesional bleomycin and cryothera-
21. Lee JY, Kim CW, Kim SS. Preliminary study of intralesional bleomy-

23. Nunley JR, Wolverton SE. Medicamentos Sistêmicos. In: Bologna JL, Jo-
25. Horbach SE, Rigter IM, Smitt JH, Reekers JA, Spuls PI, van der Horst CM. In-
26. Park KK. Minimize that "pinch and burn": tips and tricks to reduce injec-
27. BRASIL, Ministério da Saúde. Secretaria da Atenção à Saúde. Departa-

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Investigation on the use of 3% and 5% retinoic acid in peeling solution as a drug delivery agent after percutaneous induction of collagen with needles (IPCA®): safety profile and application protocol

Investigação sobre o uso do ácido retinoico a 3% e a 5% em soluções para peeling como agente para drug delivery após indução percutânea de colágeno com agulhas (IPCA®): perfil de segurança e protocolo de uso

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ABSTRACT

Introduction: Retinoic acid in peeling solution is widely used in the treatment of photaging. To date, the degree of sterility of these solutions or the safety of their use in skins whose integrity has been lost through microneedling interventions is unknown.

Objectives: To evaluate the bactericidal potential of 3% and 5% retinoic acid in peeling solution, with and without a colored vehicle, as well as the safety and tolerance to its administration immediately after application with microneedles.

Methods: Samples of 3% and 5% retinoic acid solution, with and without a colored vehicle, prepared by two dispensing pharmacies (A and B) were exposed to Pseudomonas aeruginosa and Staphylococcus aureus colonies. These solutions were used as drug delivery agents after percutaneous induction of collagen with needles.

Results: The samples evaluated in D0, D30, D60 and D90 indicated the presence of bactericidal capacity of the tested agents. The use of the solutions following intervention with microneedles was well tolerated and yielded satisfactory results.

Conclusion: The retinoic acid peeling solution can be safely used following procedures that lead to a loss of integrity of the skin barrier. The absence of adverse effects and good results yielded by the procedure suggest that the association of microneedling and retinoic acid peeling is an innovative, reproducible and safe proposal.

Keywords: Chemexfoliation; Therapeutics; Tretinoin

RESUMO

Introdução: O ácido retinoico em solução para peelings é amplamente usado no tratamento do fotonevelhamento. Até o presente momento não conhecemos o grau de esterilidade dessas soluções ou a segurança de seu uso em peles cuja integridade tenha sido perdida por intervenções com microagulhas.

Objetivos: Avaliar o potencial bactericida do ácido retinoico 3% e 5% em soluções para peelings com e sem tonalizante, bem como a segurança e tolerância de sua administração imediatamente após o tratamento com microagulhas.

Métodos: Amostras de solução de ácido retinoico 3% e 5%, com e sem tonalizante, oriundas de duas farmácias de manipulação (A e B) foram expostas a colônias de Pseudomonas aeruginosa e Staphylococcus aureus. Essas soluções foram usadas como drug delivery após indução percutânea de colágeno com agulhas.

Resultados: As amostras avaliadas em D0, D30, D60 e D90 mostraram capacidade bactericida sobre os agentes testados. O uso das soluções após a intervenção com microagulhas foi bem tolerado e apresentou resultados satisfatórios.

Conclusão: A solução de ácido retinoico para peelings pode ser utilizada com segurança após procedimentos que levem à perda da integridade da barreira cutânea. A ausência de efeitos adversos e os bons resultados do procedimento permite sugerir a associação de microagulhamento e peeling de ácido retinoico como uma proposta inovadora, reproduzível e segura.

Palavras-chave: Abrasão química; Terapêutica; Tretinoína
INTRODUCTION

The proposal of ablative treatments aiming at stimulating and remodeling the collagen is advocated by dermatologists for a long time. It is known that mechanical or chemical removal of the epidermis favors cytokine release and migration of inflammatory cells that result in the replacement of the damaged tissue by a remodeled tissue. The use of retinoic acid as a chemical peel agent has been proposed for lightening, rejuvenation and improvement of the texture of the skin.

By stimulating cell turn over, transcutanous elimination of pigments and a moderate collagen remodeling, retinoic acid enables softening of fine wrinkles, treatment for melasma and makes shallow scars more superficial, as well as improves the aspect of stretch marks. This agent is conventionally used in concentration ranging from 3% to 5%, in isolation or associated to other chemical peel substances such as Jessner’s solution, applied immediately before. Classified as a superficial peel, it aims at removing the stratum corneum, with injury to the epidermis, reaching the basement membrane and leading to repercussions in the dermis. The big advantage of retinoic acid chemical peel is its relative safety in all phototypes, limiting the risk of complications when all necessary recommendations for an ablative procedure are taken into consideration. The epidermis, stripped off its basement membrane, is replaced by a tissue seen as cicatricial, with flattening of the dermal papillae. An inflammatory response in the dermis is triggered by the destruction of the epidermis, leading to the production of parallel thick collagen bundles, different to the interlacing collagen network found in normal skin. Studies have revealed that transforming growth factor-β (TGF-β) plays an important role in the first 48 hours of scar formation. Whereas TGF-β1 and TGF-β2 promote formation of cicatricial collagen, TGF-β3 seems to promote regeneration and healing of the wound due to more physiologic collagen, almost with no feature of the pre-existing one. In an attempt to achieve a shorter time for resolution after the procedure and reduce the risk of complications, we currently observe a trend to indicating procedures that avoid deep epithelization. Percutaneous collagen induction with needles proposes the stimulation of collagen production preventing deep epithelization. Percutaneous Collagen Induction, first assessed by the African plastic surgeon Des Fernandes, whose studies with 480 patients with scars, wrinkles and laxity yielded good results, has been used all over the world. In Brazil, Emerson Lima registered the name percutaneous collagen induction with needles (IPCA®). This intervention produces hundreds of microlesions through a roller with microneedles and results in partial integrity of the cutaneous barrier, with the dissociation of keratinocytes as a target, and stimulates the release of cytokines such as mainly interleukin-1α, but also interleukin-8, interleukin-6, TNF-α and GM-CSF. This process results in dermal vasodilation and keratinocyte migration to repair the epidermal damage. In an experimental study, the relationship between the length of the needle and the depth of the injury caused using pig skin, which is considered similar to human skin. From the results, the authors proposed a classification into mild, moderate and deep injury, related to the length of the needle and its ability to cause the planned damage (Box 1). Considering the diffuse erythema with few petechiae seen in moderate injuries, a favorable condition to receive an active substance, this article proposes the use of the IPCA® technique, associated to retinoic acid chemical peel as drug delivery, with the objective to optimize results of procedures to improve the quality of the skin. We assumed that an active substance applied as drug delivery over the skin that lost its integrity due to a previous procedure needs to be sterile. Until now, studies published do not offer support about the safety of using retinoic acid solution, commonly used for chemical peels, after IPCA®. Therefore, we began our investigation with the evaluation of this active substance.

OBJECTIVES

This study aimed at evaluating the bactericidal action of 3% and 5% retinoic acid solutions used for superficial chemical peels, compounded by two different independent pharmacies, as well as if the sterility of these solutions would be affected by adding coloring, by time after compounding or by storage conditions.

Another objective was to evaluate the safety of the above-mentioned solutions, used in procedures where skin integrity was lost, allowing their use as drug delivery.

METHODS

Evaluation of the safety of 3% and 5% retinoic acid as drug delivery agents.

Tinted and non-tinted retinoic acid solutions at 3% and 5% in alcohol where evaluated, compounded in two different pharmacies: A (Roval) and B (Pharmapele), both in Recife (PE), Brazil, with a shelf life of 90 days, keep in room temperature (RT) and at 4°C. The objective was to investigate sterility and bactericidal action.

Sterility of the four solutions from each pharmacy was evaluated at the same day of compounding, with seeding of an amount of 100µL from each sample in BHI agar (Brain Heart Infusion) and incubated at 37°C for 24 hours, observing bacterial growth.

On the same occasion, bactericidal action was initially tested with an amount of 100µL of retinoic acid from each packaging, diluted in 480µL of BHI medium plus 20µL of a 24-hour culture of Pseudomonas aeruginosa (Figure 1). Retinoic acid was diluted five times in this procedure. Bactericidal action of retinoic acid was also evaluated in the second and third months.

<table>
<thead>
<tr>
<th>Features of the stimulus</th>
<th>Needle length</th>
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<tbody>
<tr>
<td>Mild injury</td>
<td>0,25 ± 0,5mm</td>
</tr>
<tr>
<td>Moderate injury</td>
<td>1 ± 1,5mm</td>
</tr>
<tr>
<td>Deep injury</td>
<td>2 ± 2,5mm</td>
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</table>

Box 1: Classification of the intensity of injury caused by microneedling
of shelf life. This time, the product was not diluted, as it is used for chemical peels and recommended by the manufacturers, at 3% and 5%, tinted and non-tinted, stored at RT and at 4°C (fridge temperature recommended by the pharmacies for conservation of the product). A quantity of 500µL from each jar was placed into sterile microtubes, inoculated with P. aeruginosa or S. aureus colony, and part of the homogenized product was seeded into BHI agar plates in three situations: soon after homogenization, and one and two hours after. As control, bacteria were also inoculated into sterile saline. Plates were incubated at 37°C for 24 hours, when bacterial growth was observed (Figure 2).

In the first seeding for evaluation of the product sterility, no bacterial growth was observed in any of the plates, what confirmed the sterility of the product in all packages tested. In the tests performed in the first month, when the product was diluted, bacterial growth was seen in many situations. In view of this, in the second and third months of shelf life tests were performed as recommended by the manufacturer and as it is conventionally used for chemical peels. From the seeding soon after homogenization of the inoculum there was growth only in saline and some jars of retinoic acid. Seeding in the first and second hours only showed growth in saline (Figure 2), confirming the bactericidal action of tinted and non-tinted 3% and 5% retinoic acid recommended for use. It was also seen that the product is not changed whether stored at room temperature or at 4°C (Table 1). Of note is the bactericidal effect of agents tested with S. aureus, even considering that this bacterium has thicker cell membrane and is supposedly more resistant.

**Protocol proposed for the association of IPCA® with retinoic acid chemical peel**

In view of the proven bactericidal action of tinted and non-tinted 3% and 5% retinoic acid solutions, we proposed its use as drug delivery agent in association with IPCA®. For this purpose, we proposed moderate injury (Figure 3) for the application of tinted 5% retinoic acid (Figure 4), considering that with this end point the skin would be subject to receiving the active substance with no interference of a more intense bleeding as the one seen in deep injuries. Twelve volunteers participated in this assessment, with complaints of acne scars and photoaging, between 21 and 38 years of age, seen at the Outpatient Clinic of Cosmiatry of the Santa Casa de Misericórdia do Recife. The study was conducted according to the recommendations of the 1996 declaration of Helsinki, modified in 2013, and authorized by the Ethics Committee of the institution. The procedure was conducted under topical anesthesia with 4% lidocaine cream (Dermomax® Aché, São Paulo, Brazil) applied 40 minutes before the intervention. A device with needle length of 1.5mm (Dr.Roller® Mooham Enterprise Co. Gyeonggi-do South Korea, Anvisa n.80669000001). We performed back and forth movements, forming horizontal lines that were subsequently crossed by vertical and diagonal lines until a diffuse erythema developed all over the face, with mild pin-point bleeding, characterizing moderate injury (Figure 3).

Immediately after, a tinted 5% retinoic acid solution was applied using a sterile brush all over the area treated (Figure 4) and left for 2 hours, when it was removed with water and liquid soap, at home. It was recommended the patients did not apply any products on the skin for the following 8 hours, using SPF > 50 sunscreen. All subjects evaluated were photographed before and after 30 days of the procedure by the same researcher, with the same camera and using the same light. Eight days after the
procedure, the patients returned to the clinic to evaluate tolerability of the intervention.

RESULTS

In the eighth day post-procedure there were no complaints of discomfort, burning, erythema or peeling in the 12 patients treated. Two reported mild burning between 48 and 72 hours after the intervention and one reported intense peeling after 72 hours accompanied by erythema. None of the subjects had to take time off work in the period after the procedure, and all continue to use only sunscreen until eight days after. In the 30-days follow-up appointment, all reported lightening, increased glow and vigor, with softening of superficial scars and fine wrinkles. Two independent evaluating dermatologists who analyzed the standardized photographs, considered that the intervention resulted in an overall improvement of the quality of

<table>
<thead>
<tr>
<th>Samples</th>
<th>Identification</th>
<th>Storage</th>
<th>TEST 2nd month S. aureus</th>
<th>TEST 2nd month P. aeruginosa</th>
<th>TEST 2nd month S. aureus</th>
<th>TEST 2nd month P. aeruginosa</th>
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<td>4A</td>
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<tr>
<td>8A</td>
<td>5% non-tinted</td>
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the skin in the whole group treated. On physical examination, lightening, improved texture, increased glow and closure of pores. All in the group chose the option “very satisfied” when answering the evaluation questionnaire, with the options of little satisfied, moderately satisfied, very satisfied. No side effects such as persistent erythema, hypersensitivity, irritation, pruritus or post-inflammatory hyperpigmentation were seen. Six of the 12 patients had moderate peeling even after seven days of the procedure. None of them had to stop their daily activities because of the intervention.

**DISCUSSION**

The benefits of IPCA® on the correction of scars, wrinkles and laxity are well established, as well as the ability of retinoic acid peel in providing cosmetic improvement of the quality of the skin. Until present, retinoic acid peel has been proposed to intact skin, when there is no need to question its sterility, even less its bactericidal action, because of the protection offered by the cutaneous barrier. To our knowledge, this is the first investigation study that evaluated the destructive ability of retinoic acid over two common bacteria in the environment, *S. aureus and P. aeruginosa*. Despite studies on the viability of microorganisms, no reports were found on the sterility of retinoic acid solutions used for chemical peels.8,9 According to our results, it was possible to use this active substance safely, even after disruption of skin integrity with a moderate injury caused by microneedles. This proposal provides another therapeutic option of association of techniques in the dermatological practice. Tests using the solutions kept in the fridge and at room temperature, even three months after manufacturing, showing their bactericidal action, offer the safety of a formulation for drug delivery after intervention with IPCA®. It was also important the evaluation of different concentrations (3% and 5%), besides the feature of tinting, confirming the same reliable profile. IPCA® alone leads to platelet and neutrophil release, responsible for making growth factors available, which act on the keratinocytes and fibroblasts, such as transforming growth factors α and β (TGF-α and TGF-β), platelet-derived growth factor (PDGF), protein III activator of the connective tissue and connective tissue growth factor, followed by neutrophils, angiogenesis, fibroblast proliferation and production of collagen type III, elastin, glycosaminoglycans and proteoglycans. In parallel, fibroblast growth factor, TGF-α and TGF-β are secreted by monocytes. Approximately 5 days after the injury, the fibronectin matrix is formed, allowing deposit of collagen underneath the basal layer of the epidermis. This collagen type III is slowly replaced by collagen type I, more longer lasting. In order for this inflammatory cascade to develop, the trauma caused by the microneedle should reach depths of 1mm to 3mm in the skin, resulting in red blood cell columns followed by edema of the treated area and almost immediate homeostasis.
The intensity of these reactions is proportional to the length of the needle used in the procedure. This study proposes a moderate injury associated to the action of tinted 5% retinoic acid for optimization of the results, as well as aiming at masking the small hematic crusts caused by IPCA®. The immediate return to work activities of the whole group studied with only photoprotection provides convenience to the intervention, what favors its application.

CONCLUSIONS

Retinoic acid solution for chemical peels can be safely used after procedures that lead to loss of integrity of the cutaneous barrier.

The absence of side effects and patient’s and assessor’s satisfaction allows for the suggestion of the association of microneedling and retinoic acid peel as an innovative, reproducible and safe proposal.

REFERENCES


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Laboratory evaluation of the samples.
Combining cell therapy with biopolymer films improves wound healing in a juvenile dermatomyositis patient

ABSTRACT

Introduction: Juvenile dermatomyositis (JDM) is a systemic disease that affects children’s proximal musculature and skin. The ulcerated stage of the disease is a therapeutic challenge.

Objective: To evaluate the improvement of ulcerated stage of JDM caused by the use of cell therapy.

Methods: Co-culture of autologous fibroblasts and keratinocytes, application of these cells in ulcers in conjunction with fibrin glue, and placement of chitosan-alginate or chitosan-xanthan membrane on the lesions.

Results: Less than 12 hours after therapy, the patient reported complete cessation of pain and, within 2 days, healing tissue emerged. Some of the ulcers were almost completely healed by the end of the 1st week, and some of the calcinoses disappeared. This technique does not cure the disease, however it improves the patient’s quality of life, and it is possible to cryopreserve healthy cells to treat new lesions. Given the fact that the cells are of autologous origin, the risk of rejection is eliminated. Furthermore, this procedure does not require debridement of the lesions or hospitalization.

Conclusions: The application of autologous cultures of fibroblasts and keratinocytes in ulcers is already considered an effective treatment in patients with burns and other skin wounds, and has now also been proven effective in the treatment of wounds in JDM.

Keywords: Calcinosis; Cell-and tissue-based therapy; Dermatomyositis; Fibroblasts; Keratinocytes; Polysaccharides; Wound healing

RESUMO

Introdução: Dermatomiosite juvenil (DMJ) é doença sistêmica que afeta a musculatura proximal e a pele de crianças. A doença ulcerada é um desafio terapêutico.

Objetivo: Avaliar a melhora da doença ulcerada na DMJ, pelo uso de terapia celular.

Métodos: Realização de cocultura de fibroblastos e queratinócitos autólogos e aplicação dessas células nas úlceras juntamente com cola de fibrina e colocação de membrana de quitosana-alginato ou quitosana-xantana sobre as lesões.

Resultados: Menos de 12 horas após a terapia, o paciente referiu completa eliminação da dor e, dentro de dois dias, estava presente tecido de cicatrização. Algumas das úlceras estavam quase completamente cicatrizadas no final da primeira semana, e algumas das calcinoses desapareceram. Essa técnica não cura a doença, mas melhora a qualidade de vida, sendo possível criopreservar as células saudáveis do paciente para tratar novas lesões. Sendo as células de origem autóloga, elimina-se o risco de rejeição. Além disso, esse procedimento não necessita de debridamento das lesões nem hospitalização.

Conclusões: A aplicação de culturas autólogas de fibroblastos e queratinócitos em úlceras já é considerada tratamento efetivo em pacientes com queimaduras e outras feridas cutâneas e, agora mostrou-se também eficaz no tratamento de feridas na DMJ.

Palavras-chave: Calcinose; Cicatrização; Dermatomiosite; Fibroblastos; Polissacarídios; Queratinócitos; Terapia baseada em transplante de células e tecidos

DOI: http://www.dx.doi.org/10.5935/scd1984-8773.20181011129
INTRODUCTION

Juvenile dermatomyositis (JDM) is a rare and severe systemic condition that affects primarily the proximal muscles and skin in children, with a prevalence of three per million in the population. Its etiology is not completely understood, but it is suggested that it is caused by an autoimmune reaction in individuals genetically susceptible to environmental triggers.\(^1,2\) JDM's cutaneous manifestations can be severe and difficult to treat, with significant long-term morbidity.\(^2,3\)

Calciosis, characterized by the formation of calcium deposits in the skin, usually affects 10% to 70% of pediatric patients with JDM, being generally diagnosed in the first three years of the disease. Together with dermatomyositis itself, calcnosis can negatively impact in the patient's quality of life, resulting in weakness, functional disability, muscular atrophy, cutaneous ulcers and, consequently, local pain and secondary infections.\(^4\) Dystrophic calcifications occur where there is tissue damage, with normal serum levels of calcium and phosphorus. Although they can appear in any part of the body, the areas most commonly affected are elbows, knees, trunk, hands, feet, buttocks and head. Calciosis is usually painless; however, it can be associated to tenderness on palpation and pain with compression, evidencing panniculitis and ulcerations on histopathology. There is deposition of calcium on the skin surface, making it susceptible to infections.\(^5\)

In juvenile dermatomyositis, the severity of the disease can be related to cutaneous calciosis, delay in starting treatment and, potentially, to genetic polymorphisms of TNF-\(\alpha\)-308. Despite the lack of data on the pathogenesis of calcnosis in JDM, a possible mechanism is calcium release from mitochondria of muscle cells affected by the myopathy. Macrophages, pro-inflammatory cytokines and defects in regulatory proteins of calcium were also suggested, as well as vascular ischemia, which also has a role in calcnosis. The deposition of calcium can occur in the subcutaneous tissue or in muscles and then ulcerate, draining a chalky substance. Ulcerated disease is severe and can be fatal, reflecting the importance of cutaneous vascular disorders, with tissue hypoxia and necrosis.\(^1,3,6\)

As calcnosis tends to worsen with the progression of JDM, early and aggressive therapeutic approach has been suggested as an option to reduce cutaneous and muscular sequelae. Multiple medications have been used, such as corticosteroids, methotrexate, bisphosphonates, probenecid, warfarin, aluminum hydroxide, colchicine, diltiazem, infliximab, immunoglobulin, hydroxychloroquine, cyclophosphamide and thalidomide, but there is no consensus on the best therapy.\(^1-5\) Success of these medications is usually limited to disease control, with no extension to cure. Furthermore, side effects associated to their prolonged use can also worsen the patient's health.

Since there is no gold standard nor a therapeutic algorithm to manage JDM and considering that repair of damaged skin is by proliferation and growth of dermal cells (essentially fibroblasts) and/or epidermal cells (keratinocytes and melanocytes), cell therapy can be a relevant alternative for the treatment of JDM associated to calcnosis. Promising results were reported with the use of cell therapy for the treatment of chronic ulcers of different etiologies.\(^7-17\) therefore, a positive result can could also be expected for this disease.

Biopolymers, of the polysaccharide and protein class, have been widely used in the development of interactive and bioactive dressings, being not only useful for protecting the wound, but also playing an important role in promoting healing. Formation of granulation tissue and reepithelization, followed by angiogenesis and continuous deposition of collagen fibers, limiting scar formation and tissue retraction, were seen with the use of chitosan, for example.\(^18,19\) The association of multiple polymers is also relevant, since it allows for the development of dressings with improved properties, such as enhanced fluid absorption. Relevant in vivo studies were described recently on the use of membranes manufactured by the association of chitosan with xanthan\(^20\) and alginate\(^21\) for the treatment of cutaneous ulcers in Wistar rats, with or without the combination or mesenchymal cells, demonstrating a potential that could be used as dressings in JDM lesions.

The objective of this study was to demonstrate an alternative therapy for the treatment of chronic ulcers in JDM patients with calcnosis that are not responsive to conventional treatments, using culture of autologous cells and subsequently covered with membranes of biocompatible polysaccharides.

METHODS

This study was approved by the Committee of Ethics in Research of the Universidade Estadual de Campinas (Unicamp – CEP: 444.726).

Case description and sample collection

A male, 18-year-old patient, diagnosed with JDM since 5 years of age, undergoing conventional treatment with methotrexate and corticosteroids, with universal calciosis and chronic cutaneous ulcers, some with exposure of bone, was submitted to skin biopsy of a non-ulcerated on the right arm after signing a consent form. Fragments of approximately 2cm\(^2\) were collected and stored in saline, antibiotic and antifungal (Anti-Anti 15240, batch 577999, GIBCO/Invitrogen) until being transferred to culture jars. During the whole research, the patient maintained the previous systemic treatment.

Cell culture of skin fragments

The skin fragments were placed into culture jars with a keratinocyte medium (KSFM–GIBCO/ Invitrogen) supplemented with 10% of fetal bovine serum (FBS – LGC Biotechnology) and L-glutamine 0.2mg/ml, penicillin 100UI/mL and streptomycin 0.1mg/ml (GIBCO/Invitrogen), until processing. Subsequently, the fragments were transferred to Petri dishes with trypsin 2.5% and EDTA solution 0.1% (GIBCO/Invitrogen) and incubated at 37°C e 5% CO\(_2\) for 3 hours, when the dermis was separated from the epidermis. Trypsin was neutralized with a KSFM medium supplemented with 10% FBS. The supernatant (containing dermal and epidermal cells) was filtered (40mm Falcon/Corning) and centrifuged for 10 minutes at 400G.
The cell pellet was resuspended in culture medium and transferred to culture jars with a concentration of 1x105 cells/ml in 2ml of specific culture medium for each cell type and then incubated at 37°C and 5% CO2. Keratinocytes were cultivated in KSF medium (KSFM-GIBCO/Invitrogen) supplemented with 10% fetal bovine serum (FBS – LGC Biotechnology) and L-glutamine 0.2mg/ml, penicillin 100UI/ml and streptomycin 0.1mg/ml (GIBCO/Invitrogen). Culture media were changed three times per week. When the cells reached confluence, cultures were trypsinized with trypsin and EDTA solution for 10 minutes at 37°C and 5% CO2. As previously, trypsin was neutralized with fetal bovine serum 10%. This procedure was performed until a minimal amount was obtained for each cell type, approximately 5x106 keratinocytes and 10x106 fibroblasts. The whole process took between 21 and 30 days. Cells were cryopreserved in fetal bovine serum and DMSO solution at -80°C.

The whole process involving cell manipulation was performed in a clean room (class 10.000 ISO 7 – ISO 14644-1).

Membrane preparation

The preparation of membranes followed a technique already established at the Department of Material Engineering and Bioprocesses, Faculdade de Engenharia Química, Universidade Estadual de Campinas.

Membranes were obtained according to adaptations to methods established by Rodrigues et al.,22 Bueno and Moraes23 and adapted by Pires and Moraes,24 in the case of chitosan-alginate devices (C-A), based in procedures developed by Veiga and Moraes25 and Bellini et al.38 for the chitosan-xanthan membranes (C-X).

Chitosan with deacetylation degree of 96% (Sigma-Aldrich, batch number 109K0043V), medium viscosity sodium alginate from Macrocystis pyrifera (Sigma-Aldrich, batch number 058K0126), xanthan gum (Sigma-Aldrich, batch number 109K0038), glacial acetic acid, calcium chloride dehydrate and sodium hydroxide (Merck); besides, the water used was distilled and deionized in a Milli-Q system (Millipore).

Chitosan and alginate membranes were prepared with the addition of quantities of 180ml of 1% chitosan solution (m/v) dissolved in 2% acetic acid (v/v) in 360ml 0.5% alginate aqueous solution (m/v) at the flow of 200ml/h in the stainless steel reactor, maintained at 25°C, under stirring of 500rpm. After mixing the solutions, the stirring intensity was increased to 1000rpm for 10 additional minutes. Afterwards, 26ml of 2M NaOH aqueous solution were added to increase pH to 7, maintaining stirring for 10 more minutes. Afterwards, 7.2ml of 2% CaCl2 aqueous solution (m/v) were added to reticulate alginate carboxyls that did not form complexes with chitosan. The mixture obtained was deaerated for 120 minutes, transferred (in quantities of equal masses) into four polystyrene Petri dishes (diameter of 15cm) and dried in a dryer at 60°C for 6 hours. After drying, the membranes were immersed in 150ml aqueous solution of 2% CaCl2 (m/v) for 30 minutes for reticulation of free carboxyls leftover form alginate and then washed twice for 30 minutes with 200ml of deionized water. The final drying step was performed at room temperature for 24 hours.

In the case of chitosan and xanthan gum membranes, 200ml of aqueous solution of xanthan gum 1.5% (m/v) were added to 200ml of 1.5% chitosan solution (m/v) dissolved in 1.5% acetic acid (v/v) with a flow of 30ml/h, at 25°C and under stirring (1000rpm). After, the suspension was deaerated and the mixture was transferred into a polystyrene dish of 15cm diameter and the material was dried at 37°C for a variable period of 24 to 48 hours. The membrane was washed twice for 30 minutes with 200ml deionized water, once with 250ml of 10mM Hepes buffer (Sigma-Aldrich) to neutralize pH and, finally, with 500ml deionized water. A final drying step was performed at room temperature for 24 hours, securing the edges to prevent shrinking of the membrane.

The membranes were sterilized with OxyFume 30 (30% ethylene oxide and 70% CO2) at 40°C for 8 hours, with relative humidity of 30% to 80% by the Central de Esterilização Comércio e Indústria Ltda – Accell (Campinas, SP).

Transmission electron microscopy was performed to verify behavior of the cells in the membrane (Figure 1).

Patient treatment

The cells were thawed and cultivated for at least 72 hours before application. On the day of application, the cells were trypsinized, washed and counted according to protocols previously described by Rehder et al.,13 Souto et al.,27,15 Bosnardo16 and Dinato et al.17 A culture of fibroblasts of a total of 1x107 cells was prepared. For the application, cells were sprayed with fibrin glue (Beriplast P – CSL-Behring) over the ulcer bed under antiseptic conditions, in an outpatient setting (Figure 2).

After spraying the cells, the polysaccharide membranes previously diluted with saline were placed on the ulcer (Figure 3), in order to protect the area against agents that could remove the graft, aiming at aiding the healing process.

The patient was followed for 20 months, every 7 days in the first month, and every 15 or 30 days thereafter. New applications were performed according to the patient’s response, in a total of 7. Photographic documentation was made with Nikon D5100 camera, using a ruler to determine the total area of the ulcer, delineating its borders. The images were processed with the software Image J2, and the differences in the values of the areas were determined for each ulcer using the software GraphPad Prism5.

A quality of life questionnaire (SF-36) was also used before and at the end of the treatment.

RESULTS

The patient had multiple cutaneous ulcers, ranging from 0.5cm2 to 8cm2, distributed all over the body but mainly on the
lower limbs (Figure 2). The lesions selected for the treatment were the larger and the deeper, which caused more discomfort. The healing process was documented with photographs in weeks zero, 3, 21, 28, 42, 64 (Figure 3) and 79 (Figure 4).

Less than 12 hours after the application of cells and membranes, the patient reported total improvement of the pain and, in two days, the healing process started. Soon thereafter, a shiny film was seen on the surface of the ulcers. Some days later, an intense exudate, attributed to fibrin, granulation tissue and crust was seen in some lesions, with subsequent centripetal closure. Some wounds were completely closed after one week of treatment.

A taxa de cicatrização alcançada foi acima de 95%, com melhora contínua, mesmo após cinco meses da última aplicação (Gráfico 1). O paciente não apresentou mais dores nas áreas tratadas, com importante melhora na qualidade de vida (Gráfico 2). É interessante notar que a calcinose desapareceu mesmo em áreas não tratadas diretamente.

The healing rate achieved was above 95%, with continuous improvement even after 5 months of the last application (Graph 1). The patient did not have any more pain on the treated areas, with significant improvement in quality of life (Graph 2). Interestingly, the calcinosis disappeared even in areas that were not treated directly.

DISCUSSION

JDM is a rare autoimmune disease that affects primarily the muscles and the skin. The main treatment is with high doses of corticosteroids combined to other immunosuppressant drugs. Approximately 30% of patients cannot control the disease, despite multiple interventions, as seen in our patient. The application of stem cells was described as a last resource in the treatment of patients with autoimmune diseases refractory to treatment but persistence of cutaneous disease, including calcinosis and contractures.

Treatment of the ulcerated and refractory disease is very complex, and autoimmune diseases are quite challenging. Tissue engineering focused on autologous keratinocytes and fibroblasts has been used in the treatment of cutaneous ulcers since the 1980s. The technique was initially tested in burn patients, with good results. Then improved healing was seen in vascular and diabetic ulcers. In recent years, tissue engineering significantly advanced with this goal, and one of the trends in dermatology is the use of combined biomaterials with biopolymers and cells fulfilling biosafety requirements and that are active in the type of wound treated.

Positive and relevant results were seen in our JDM patient after the application of autologous fibroblasts and keratinocytes, applied with fibrin glue, followed by the coverage with membranes made of chitosan with xanthan or alginate.

The protection provided by chitosan–alginate and chitosan–xanthan dressings with negative stimuli from the environment plays a role in the healing process. According to Wang et al., the optimal dressing should be flexible and able to control local water loss. It must be resistant to bacterial infection therefore preventing sepsis, have adequate adherence on the ulcer, as well as not being antigenic, non-toxic and easy to apply and

Cell therapy and biopolymers for the treatment of ulcers

From the engineering perspective, the material for the dressing should also have adequate mechanical properties so as to maintain its integrity during use. Rates of water evaporation are also important, both for maintaining adequate humidity in the wound bed and to avoid unwanted accumulation of secretions. Both membranes used in this study achieved this goal, being effective in the contribution to expedite tissue regeneration and promote speedy recovery. Besides, membranes are clear, which allows observation of the wound bed, without needing to be removed.

The complex chitosan-alginate seems to have a positive action in the process of tissue remodeling in scars, increasing the rates of collagen synthesis, while also improving compaction of new fibers and promoting the presence of mature fibroblasts. Moreover, these membranes seem to stimulate and regulate multiple phases of the healing process, being useful in the treatment of cutaneous ulcers. Both collagen synthesis and modulation of wound contraction by chitosan-alginate membranes can result in a fast closure of the lesion. Similar results were seen on chitosan-xanthan membranes associated to mesenchymal cells.

The role of fibrin glue is not clear in this case. The product is a biological adhesive that works by simulating the exudative phase of healing, frequently used in plastic surgery, as well as organ transplants. In this case, our hypothesis of its possible benefit is by the improved cell and membrane adherence to the lesions, and by its hemostatic and antibacterial actions. Under normal circumstances, soon after the injury, fibrin and fibronectin...
tin are deposited in the wound. There, fibrin acts as a hemostatic barrier, adhering tissue to surrounding cells and stimulating the migration of fibroblasts, what probably also occurred in this case. Redher et al.34 demonstrated that the application of fibrin glue alone, without cell culture, was not able to reepithelize the wound, despite crust formation. The authors did not mention reduction of the pain with this application.

Fibrin matrix releases growth factors, such as vascular endothelial growth factor (VEGF), fibroblast growth factor (FGF) and transforming growth factor b (TGF-β), and these proteins can be related to improvement of the pain, soon after beginning of treatment. Besides the antibacterial action, increased endothelial cell migration and proliferation can contribute to improved vascular supply and also provide an ideal environment for fibroblast and keratinocyte migration, proliferation and differentiation, improving healing.31

Improvement of calcinosis is not clear, and Köebener phenomenon was not seen after treatment started, not even in the biopsy site.

Cell therapy with autologous fibroblasts and keratinocytes was used in this study for a more effective treatment of ulcers in JDM patients with better functional and aesthetic results, as well as a faster recovery and elimination of pain, allowing the patient to return to his studies. The development of a strategy based on cell therapy represents a progress in the treatment of ulcers of different etiologies,17 and the use of C-A and C-X membranes associated to autologous cells is very advantageous because membranes can function as a physical barrier, preventing external contamination,25 besides having a possible role in healing. There was no difference of performance in the healing rates with different membranes.

The implants described here were effective when compared to conventional treatments of skin grafting with healthy donor sites,16,17,35 even if some ulcers did not completely heal, probably due to their extension and depth. Cell cultures can be cryopreserved and eventually used in a new application. Autologous cells are great candidates, because with them the risk of rejection is eliminated. Another positive aspect is not needing hospitalization or debridement of the lesions.

CONCLUSION

This was the first case described showing the use of cultures of autologous fibroblasts and keratinocytes associated to chitosan-alginite or chitosan-xanthan membranes for the treatment of cutaneous ulcers associated to juvenile dermatomyositis.

In this study, we demonstrated an effective strategy for the treatment of cutaneous disease caused by juvenile dermatomyositis, even though it was not completely cured. Maybe the combination of stem cell transplant with autologous cutaneous cells could be the cure for a patient like ours, who presents with extensive and debilitating disease. Although it as a sophisticated and restricted technique, it proved to be a valid therapeutic strategy that can be used in JDM and in ulcers with other etiologies.

REFERENCES


Cell therapy and biopolymers for the treatment of ulcers

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Liposculpture as a complementary treatment in rhytidoplasty

A lipoesculutura como tratamento complementar na ritidoplastia

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ABSTRACT

Introduction: Multiple approaches and the use of different surgical strategies seem to contribute to the goal of obtaining more satisfactory outcomes in rhytidoplasty. In light of this, liposculpture has been progressively becoming more relevant as an alternative to improve those outcomes, adding greater final aesthetic value to the procedure.

Objectives: To evaluate outcomes obtained in patients who underwent the association of rhytidoplasty and facial liposculpture.

Methods: Fourteen patients underwent classical Pitanguy rhytidectomy associated with lipoaspiration in specific areas of the face, having been subsequently followed up for 12 months. Seventeen volunteers used the Antell & Orseck method to evaluate the postoperative outcomes.

Results: Of the 14 patients evaluated, 9 (64.2%) were classified as having experienced excellent outcomes, while 3 (21.4%) had moderate outcomes, and 2 (14.2%) had poor outcomes. None of the patients experienced outcomes rated as perfect or undesirable.

Conclusions: Rhytidoplasty associated with facial liposculpture is an important alternative for the treatment of facial aging, leading to satisfactory, safe and long lasting results.

Keywords: Cervicoplasty; Rejuvenation; Rhytidoplasty

RESUMO

Introdução: As múltiplas abordagens e a utilização de diferentes estratégias cirúrgicas contribuem para a obtenção de resultados mais agradáveis nas ritidoplastias. Nesse cenário, a lipoesculutura vem ganhando cada vez mais relevância como alternativa para potencializar esses resultados, agregando maior valor estético final para o procedimento.

Objetivos: Avaliar os resultados obtidos em pacientes submetidas à associação de ritidoplastia e lipoesculutura facial.

Métodos: Quatorze pacientes foram submetidas à ritidoplastia clássica de Pitanguy, associada à lipoesculutura em áreas específicas da face, sendo acompanhadas por 12 meses. Os resultados pós-operatórios foram avaliados por 17 voluntários utilizando o método de Antell & Orseck.

Resultados: Das 14 pacientes avaliadas, nove (64,2%) tiveram o resultado classificado como excelente, três (21,4%) como moderado e dois (14,2%) como fraco. Nenhuma paciente obteve resultados classificados como perfeito ou ruim.

Conclusões: A ritidoplastia associada à lipoesculutura da face se valida como alternativa importante para o tratamento do envelhecimento facial, proporcionando a obtenção de resultados agradáveis, seguros e duradouros.

Palavras-chave: Cervicoplastia; Rejuvenescimento; Ritidoplastia
INTRODUCTION

Treatment of facial ageing is among the conditions that prompts patients to seek aesthetic procedures.1 The demand for new methods aims to fulfill patient’s expectations who still seek complementary procedures after complex surgeries for facial rejuvenation, particularly to overcome loss of volume in certain areas of the face.2

Many studies have been trying to understand the real factors involved in the process of facial ageing, offering theoretical support for technical alternatives to be developed.3 It is believed that facial ageing is the result of a sum of multiple factors, with emphasis on bone resorption and remodeling, relaxation and degeneration of soft tissues and atrophy of the subcutaneous tissue.3

The description of facial anatomical compartments and the new understanding of their relevance for rejuvenation surgery favored the association of liposculpture as a complementary treatment in rhytidoplasties. This combination boosted results and made possible a customization of the technique for each case. The results, progressively more satisfactory, made the number of professionals using the technique increase considerably over the last years.1,2,4,5

The aim of this study is to present the experience of the authors with the association of liposculpture to rhytidoplasties and discuss the quality of the results and advantages of the procedure.

METHODS

This study was conducted in a template of original article, for which theoretical-practical approaches were used, that aimed at achieving conclusive and significant results. A descriptive, prospective study, in which a sample of 14 female patients were followed-up for 12 months, after rhytidoplasty with facial liposuction (cervical region and lower third of the face) and fat insertion on specific areas. The procedures were performed in the authors’ private practices, from January 2013 to March 2016. There was no comparison between the individuals in the sample, with the results obtained with the evaluation of each separately. This study was conducted according to ethical guidelines of the declaration of Helsinki.

A predefined sequence of standard photographs was produced preoperatively, taken by the same photographer, at the same place and with the same image characteristics (Nikon Inc digital camera, Melville, New York).

Surgery was performed according to the classical methodology detailed in a previous study by the authors,2 and liposculpture was then associated with the goal of maximizing results. With the patient sedated, we performed anesthesia in the cervical-facial region with approximately 350ml of a solution containing 0.125% lidocaine with adrenaline 1:200.000. We performed liposuction of the cervical region and then cervicoplasty.7 Before performing Pitanguy’s roundlifting,7 we performed liposuction of the lower third of the face, observing the needs for each case (Figure 1).

The fat collected during liposuction was set aside for sedimentation. After finishing the rhytidoplasty, the fat already washed and prepared using Maricevich et al.4 technique (Figure 2) was injected into specific areas of the face, as shown in figures 3 and 4.

After the procedures, the patients were seen weekly, and then bimonthly until the 12th month, when new photographs were taken (in identical conditions to before the surgery).

Evaluation was made based on classical principles, modified by Antell & Orseck’s method.8 In this analysis resource, the patients included in the study were evaluated subjectively with no defined anatomical patterns or references. The 17 volunteers (non-physicians) analyzed the before and after the surgery photographs of the individual in the sample and, according to their own personal opinion, graded each patient. The grades were attributed in the following fashion: 1 point = no improvement; 2 points = mild improvement; 3 points = moderate improvement; 4 points = marked improvement (excellent); and 5 points = perfect. Each patient’s grades were summed and the results classified the outcome as: poor (17-27 points); weak (28-43 points); moderate (44-57 points); excellent (58-77 points); and perfect (78-85 points).

RESULTS

In the period of the study, 14 female, white patients, with a mean age of 55±9 years underwent the procedure described. The surgical procedure lasted on average 234±33 minutes. No difficulty in performing the association of techniques was
found. All patients were discharged around 24 after the surgery. Post-operative recovery occurred as expected, with only one patient developing hematoma that required drainage in operation room. All other patients evolved with no complications.

The results related to the patients’ assessment (final grades and classification) are shown in table 1. Nine patients (64.2%) achieved an excellent result, three (21.4%), moderate and two (14.2%), weak. There were no results classified as poor or perfect. In figures 5 and 6 the patients who underwent the procedures are represented (pre- and 12-month post-operative photographs, frontal and left and right profile views).

**DISCUSSION**

The emphasis of facial rejuvenation procedures is still in the maintenance of results long-term, and in the minimization of the stigmatized face. The quest for facial harmony, preserving or reaching anatomical aspects considered to be ideal is still a great challenge for surgeons performing rhytidoplasty.6 The association of liposuction in this procedure deals with this aspect and offers possibilities that were not routinely obtained with the classical technique.9,10

Currently, facial liposculpture figures as an excellent alternative to guarantee superior results in the surgical approach of the face, be it repairing or aesthetic.4,5 Liposuction of the lower third of the face and cervical region enables achieving a more triangular face, a desirable feature, particularly in rejuvenation surgery.6 The possibility of sculpting with liposuction of specific amounts of certain regions of the face makes it possible to customize the final aesthetic effects, minimizing the unwanted uniformity of results.2

Many authors widely discussed the qualities of the fat as an adjuvant in facial surgery: it is an excellent subdermal filler, with low rates of complications and low cost.9,10 Because the fat carries stem cells, it has an important role in facial rejuvenation, being considered lately by many authors the gold standard choice.11

However, unfortunately, in many situations fat injection does not feature as the sole and definitive procedure, not being possible to establish with accuracy the level of integration between the content injected and the final result of the procedure.12,13 Thus, after the procedure is finalized, new interventions might be necessary to reach the optimal volume for a certain patient.4,10,14

The evaluation of a plastic surgery result reveals difficulties and the tools available have been questioned by different authors for years.15 Until today, there is no one analysis method of the late results in rhytidoplasty considered to be perfect.13,15-17
Antel & Orseck’s method, here used, is one of the most described resources in the literature and is used by most authors. It is characterized by fast application, adequate understanding, being very practical and having reliable results, regardless of the assessor’s characteristics. These qualities were shown again in our study and the little variation in the grading of each assessor for each patient confirms this.

In the scope of plastic surgery, tactical, methodological and material innovations are frequent. The universe of rhytidoplasty is not different and requires continuous updating of the professionals in the area. Many different techniques continue to be published, and a current trend of international articles is to value ancillary strategy in the trans- and post-operative periods. Among those, fat insertion has been proven to be an alternative that is effective, long-lasting and yields excellent results, positioned as a very interesting option for surgeons. Other volumizers still frequently used are usually absorbable and have a high cost, making it difficult to be used widely and accepted by the patients.

The high result indexes characterized as excellent in our study demonstrate that the technique described can contribute to obtaining a pleasant, natural and harmonic post-operative outcome.

**CONCLUSION**

Liposculpture complementary to rhytidoplasty provided satisfactory results, with a simple technique and low rates of complications. Although more studies about this alternative are needed, particularly on long-term maintenance of results, it is more and more validated as an effective option in facial rejuvenation surgeries.

**TABLE 1: Results according to the post-operative assessment (first year), in grades and classification**

<table>
<thead>
<tr>
<th>Patient</th>
<th>Total grade</th>
<th>Classification of Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>61</td>
<td>Excellent</td>
</tr>
<tr>
<td>B</td>
<td>37</td>
<td>Weak</td>
</tr>
<tr>
<td>C</td>
<td>63</td>
<td>Excellent</td>
</tr>
<tr>
<td>D</td>
<td>64</td>
<td>Excellent</td>
</tr>
<tr>
<td>E</td>
<td>67</td>
<td>Excellent</td>
</tr>
<tr>
<td>F</td>
<td>65</td>
<td>Excellent</td>
</tr>
<tr>
<td>G</td>
<td>75</td>
<td>Excellent</td>
</tr>
<tr>
<td>H</td>
<td>57</td>
<td>Moderate</td>
</tr>
<tr>
<td>I</td>
<td>56</td>
<td>Moderate</td>
</tr>
<tr>
<td>J</td>
<td>58</td>
<td>Excellent</td>
</tr>
<tr>
<td>K</td>
<td>64</td>
<td>Excellent</td>
</tr>
<tr>
<td>L</td>
<td>42</td>
<td>Weak</td>
</tr>
<tr>
<td>M</td>
<td>62</td>
<td>Excellent</td>
</tr>
<tr>
<td>N</td>
<td>50</td>
<td>Moderate</td>
</tr>
</tbody>
</table>

**FIGURE 5: Aspect of patient H submitted to the technique (pre- and 12-month post-operative photographs, frontal, right and left profile views)**

**FIGURE 6: Aspect of patient M submitted to the technique (pre- and 12-month post-operative photographs, frontal, right and left profile views)**
REFERENCES


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Latanoprost and minoxidil: Comparative double-blind, placebo-controlled study for the treatment of hair loss

Latanoprosta e minoxidil: Estudo duplo-cego comparativo, placebo-controlado no tratamento da queda de cabelos

ABSTRACT

Introduction: Latanoprost has been shown to have potential for the treatment of hair loss evidenced by increased thickness and length of eyelashes and hypertrichosis that can be observed when it is used in the periorbital region.

Objective: To evaluate the effectiveness of latanoprost, used isolatedly or in associations, for reducing hair loss and/or stimulating its growth in patients bearers of telogen effluvium or androgenic alopecia.

Methods: A comparative, double-blind study was carried out during 180 days, with 6 groups: G1 - placebo, G2 - 5% minoxidil, G3 - 5% minoxidil + 0.005% latanoprost, G4 - 0.005% latanoprost, G5 - 5% minoxidil + 0.010% latanoprost, G6 - 0.010% latanoprost. The total and percentage count of hair strands was carried out based on phototrichogram in the anagen and telogen phases.

Results: There was improvement in G2 (total number of hair strands, number of anagen strands in D92 and D180), G3 (total number of hair strands and number of anagen hair strands in D242), G4 (total number of strands in D182, and number of anagen hair strands in D92 and D182), and G5 (total number of hair strands in D182, percentage of telogen and anagen hair strands and number of anagen hair strands in D92 and D182). Treatment of G6 did not yield significant difference regarding the placebo.

Conclusions: The treatments with 5% minoxidil, 5% minoxidil + 0.005% latanoprost, 0.005% latanoprost, 5% minoxidil + 0.010% latanoprost were shown effective in controlling hair loss and in increasing the total number of hair strands.

Keywords: Alopecia; Hair; Minoxidil

RESUMO

Introdução: A latanoprosta tem demonstrado potencial para o tratamento de queda de cabelos devido ao aumento da espessura e do comprimento dos cílios e hipertricose, observados com seu uso na área dos olhos.

Objetivo: Avaliar a eficácia da latanoprosta, isolada ou em associações, na redução da queda e/ou estimulando o crescimento de cabelos em pacientes portadores de eflúvio telógeno ou alopecia androgenética.

Métodos: Estudo duplo-cego comparativo, durante 180 dias, entre seis grupos: G1: placebo G2: minoxidil 5%; G3: minoxidil 5% + latanoprost 0,005%; G4: latanoprost 0,005%; G5: minoxidil 5% + latanoprost 0,010%; G6: latanoprost 0,010%. Foi feita a contagem em fototricograma do total e percentual de fios em fase anágnea e telógena.

Resultados: Houve melhora para os grupos G2 (total de fios e número de fios anagénios em D92 e D180), G3 (total de fios e número de fios anagénios em D242), G4 (total de fios em D182; número de fios anagénios em D92 e D182) e G5 (total de fios em D182; percentual de fios telógenos e anagénios e número de fios anagénios em D92 e D182). O Tratamento do G6 não apresentou diferença significativa em relação ao placebo.

Conclusões: Os tratamentos com minoxidil 5%, minoxidil 5% + latanoprost 0,005%, latanoprost 0,005%, minoxidil 5% + latanoprost 0,010% se mostraram eficazes no controle da queda e no aumento total de fios.

Palavras-chave: Alopecia; Cabelo; Minoxidil

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Original Articles

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INTRODUCTION

Both in men and women, androgenetic alopecia and telogen effluvium are among the most common causes of hair loss. Androgenetic alopecia is an androgen-dependent condition characterized by miniaturization of hair follicles, with shortening of the anagen phase (phase of active hair growth) and lengthened telogen phase (“rest” phase) in each development cycle of the hair follicles. Telogen effluvium is characterized by increased number of hairs in the telogen phase and increased number of hairs shed per day. It can be associated to many factors, such as nutritional deficiencies (zinc, iron, essential fatty acids, etc.), endocrine disorders (thyroid, menopause, etc.), stress and drug reactions, among others.

Currently, there are not many options available for the treatment of hair loss, being minoxidil the only treatment approved by the Food and Drug Administration (FDA) for topical use. Minoxidil is a vasodilating agent originally used for the treatment of systemic hypertension and used topically for the treatment of androgenetic alopecia in humans. Latanoprost is analogous to prostaglandin F2, used for the treatment of open-angle glaucoma and ocular hypertension. This substance demonstrated potential for the treatment of hair loss because of the side effects seen with its topical use on the ocular region, mainly thickening and lengthening of eyelashes and hypertrichosis.

The mechanism of action of these substances are not completely understood. Minoxidil seems to act by stimulating hair follicles, particularly those dormant, prolonging the anagen phase, whereas latanoprost seems to act by stimulating the anagen phase, increasing the conversion of vellus hair into terminal hair. The objective of this study was to evaluate the efficacy of minoxidil and latanoprost, in isolation or associated, and confirm if these substances are capable of reducing hair shedding and/or stimulate hair growth on patients with androgenetic alopecia and/or telogen effluvium.

METHODS

Study design

Double-blind, comparative between six treatment groups: G1: placebo = control inactive product; G2: topical lotion containing minoxidil 5%; G3: topical lotion containing 5% minoxidil + 0.005% latanoprost; G4: topical lotion containing 0.005% latanoprost; G5: topical lotion containing 5% minoxidil + 0.010% latanoprost; G6: topical lotion containing 0.010% latanoprost.

The distribution of the participants in the above-mentioned groups was randomized.

Study participants

The study was performed at the Instituto de Pesquisa Clinica Integrada – IPclin (Jundiaí-SP). Before the study commenced, it was approved by the Committee of Ethics in Research of the same institution.

In total, 123 participants were included, with 21 participants in groups G1, G2 and G3, and 20 participants in groups G4, G5 and G6. An initial dermatological assessment was performed at the time of inclusion to verify the absence of initial clinical signs not compatible with the inclusion criteria of the study (D0).

The inclusion criteria were: healthy participants of both genders, phototype I to IV, age between 20 and 55 years, presence of androgenetic alopecia (stages I to III in men, according to the Hamilton scale and I to II in women, according to the Ludwig scale), intact skin on the area to be studied (scalp), habit of washing the hair at least 3 times a week and those who were not using any product for hair growth for at least 4 weeks before the study.

Exclusion criteria were: presence of cicatricial alopecia or concurrent scalp disorders (infections, severe psoriasis and seborrheic dermatitis); active atopic dermatitis; allergy to the products being tested; pregnancy or breastfeeding; candidates with renal, cardiac or liver transplant, immunodeiciencies, use of corticosteroids, antihistamines, immunosuppressants, retinoids or anti-inflammatory agents; previous hair transplant or surgery for scalp reduction; use of minoxidil or finasteride (oral or topical) within 6 months before the study; treatments with low energy light, infrared or laser within 6 months before the study; solar erythema on the area to be studied or predicted intense exposure to sunlight or UV lamps during the study and use of hair extensions, wig or hair straightening during the 3 months prior to the study.

Procedure

The products were used by the participants themselves at home, according to the following instructions: daily topical application on dry and clean scalp, using the fingertips. The participants were instructed to wash their hands soon after using the product.

Total duration of the treatment was 6 months (180 ± 4 days) for all participants, except for the group G3, whose treatment was prolonged for 2 more months (240 ± 4 days). The dermatological safety clinical evaluations were performed in the following experimental periods: D0, D30, D60, D90, D120, D150, D180, D210 and D240 (i.e., on the day of the first visit and 30 ± 2, 60 ± 4, 90 ± 4, 120 ± 4, 150 ± 4, 180 ± 4, 210 ± 4 and 240 ± 4 days after the first visit). On the same days, the participants filled out a questionnaire of sensitization, regarding possible clinical signs and discomfort experienced with the products being investigated.

To visually assess the efficacy of the products in promoting uniformity and coverage of the scalp of the participants, we conducted a macro image registry using a photographic camera (Canon® T3i), with standardized configurations of camera, distance, background (blue) and lighting. The images were captured on the following experimental days: D0, D90 and D180. For the participants using minoxidil + 0.005% latanoprost (G3), the images were also taken on D240.

With the goal of evaluating hair growth and control of shedding provided by the use of the products being studied, according to the percentage increase of hairs in the anagen phase,
reduction of the percentage of hairs in the telogen phase and increase in the total of hairs in the experimental phase, phototrichogram analyses were conducted. For this, images were captured with the device Dermoscope® Dynamic (FotoFinder Systems, Inc, Maryland, USA) in standardized conditions with the software Trichoscale, that performs a semiautomatic count of hairs in each phase of the growth cycle. Data obtained with the equipment: total number of hairs, percentage of hairs in the anagen phase and percentage of hairs in the telogen phase.

Images were captured two days after the participants had a small area of the scalp shaved. An area of the scalp was selected for the shaving (preferably on the right or left frontoparietal region) of approximately 2 cm², which was marked and shaved with an electrical shaver, in the direction of the hair shaft, allowing a maximum length of 1 mm. Images were taken in the following experimental periods: D0, D92 and D182. For the participants in G3, images were also captured on D242. Data obtained with phototrichogram were analyzed statistically with the software SPSS version 17.0. T test was performed with paired data to determine if there was any statistical difference between the periods of evaluation (p-value ≤ 0.05).

RESULTS

Of the 123 participants included in the study, 98 finished it and 25 withdrew for personal reasons not related to the research, being them 3 participants in G1, 4 in G2, 7 in G3, 4 in G4, 3 in G5 and 4 in G6. The high number of withdrawal can be attributed to the long duration of the study. The total of participants finishing the study for each treatment was: G1: 18 participants; G2: 17 participants; G3: 14 participants; G4: 16 participants; G5: 17 participants and G6: 16 participants.

None of the participants reported discomfort related to the treatments and no clinical signs were detected on the scalp after 30 ± 2, 60 ± 4, 90 ± 4, 120 ± 4, 150 ± 4, 180 ± 4, 210 ± 4 and 240 ± 4 days using the products. The evaluation of the results of the sensitization questionnaires showed that the products were well tolerated, with no significant signs of discomfort, pruritus, scaling or erythema on the scalp during the study.

Through comparative visual analysis of the macro images captured at each experimental period, no visual improvement was seen on groups G1 and G6 after 90 ± 4 and 180 ± 4 days of treatment, per treatment (respectively, D90 and D180), in relation to the initial aspect (D0).

For the other treatments, there was a visual improvement for: 35% of the participants in G2 (six), 36% of the participants in G3 (five), 19% of the participants in G4 (three) and 6% of the participants in G5 (one), on D30, D60, D120 and D150, in comparison to D0. Figures 1 and 2 demonstrate the visual improvement seen for the treatment with 5% minoxidil.

Based on comparative analysis, it was observed in the phototrichogram in relation to D0: increased total number of hairs on D92, caused by treatment 2 (p-value = 0.0010) and on D182 caused by treatment 2 (p-value = 0.0026), treatment 4 (p-value = 0.016) and by treatment 5 (p-value=0.005). Treatment 3 caused increased number of total hairs on D242 (p-value = 0.0025).

Regarding anagen hairs, an increased was observed on D92 in the groups receiving treatment 2 (p-value = 0.0005), 4 (p-value = 0.024) and 5 (p-value = 0.004) and also on D182 for the same treatments (treatment 2: p-value = 0.0003; treatment 4: p-value = 0.004; treatment 5: p-value = 0.0001), while treatment 3 increased the amount of anagen hairs after 242 days of use (D242) (p-value = 0.001).

None of the six treatments significantly reduced the amount of telogen hairs.

Graphs 1, 2 and 3 show the total number of hairs, the number of anagen hairs and the number of telogen hairs, respectively, on each period, per treatment.

DISCUSSION

By observing the results obtained, 5% minoxidil (G2) significantly increased the total hairs and total anagen hairs, from the first 90 days of use. A superior result than seen previously, where the increased density of hairs was only seen after 24 weeks of using the product being tested. In another research, the increase was reported after 16 weeks.

The 0.005% latanoprost (G4) yielded slightly inferior results to 5% minoxidil, in the total increase in the number of hairs and in the total number of anagen hairs after 180 days of use. On
the other hand, 0.010% latanoprost (G6) did not yield statistically significant improvement, with comparable results to placebo. When used for a prolonged time and in a higher concentration, more effective results were seen. In the end of a 24-week treatment with 0.1% latanoprost, there was increased percentage of anagen hairs and reduction in the percentage of telogen hairs.6

The combination of both treatments had a better performance for 0.010% latanoprost (G5), with increased total number of hairs after six months of treatment, whereas 0.005% latanoprost associated to 5% minoxidil (G3) only yielded increased number of hairs after eight months of use.

**CONCLUSION**

None of the participants reported discomfort and no clinical signs were detected on the scalp during the study.

From the phototrichogram evaluation, we can conclude that:

Treatment with topical 5% minoxidil increased the total number of hairs and the total number of anagen hairs in the first 3 months of the study (D92).

Treatment with topical lotion containing 5% minoxidil + 0.005% latanoprost only increased the total number of hairs and the total number of anagen hairs after 8 months of study (D242).

Treatment with topical 0.005% latanoprost lotion increased the total number of hairs after 6 months of use (D182).

Treatment with topical 5% minoxidil 5% + 0.010% latanoprost lotion increased the total number of hairs after 6 months of use (D182).

Treatment with topical 0.010% latanoprost lotion showed no statistically significant difference compared to placebo.
REFERENCES
2. KLEINHANS ACS. Stress e raiva em mulheres com Alopecia Androgenética [dissertação]. Campinas: Pontifícia Universidade Católica de Campinas, Centro de Ciências da Vida, Área de Psicologia; 2012.

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Proofreading of the final text
Assessment of university students’ knowledge of photoprotection and exposure to the sun

Avaliação do conhecimento sobre fotoproteção e da exposição solar de estudantes universitários

ABSTRACT

Introduction: The number of cases of cutaneous carcinoma has been steadily increasing, which calls for the necessity of improving cost effective health strategies aimed at preventing that disease and related complications.

Objective: To perform a cross-sectional study with data collected directly from the participants, through the application of a questionnaire.

Methods: Once the data was collected, the bivariate logistic regression was used to calculate the odds ratio (OR) and the 95% confidence interval (CI) between the independent variables and the risk behavior, which corresponded to the non-daily use of sunscreen and/or other photoprotection method. The sample consisted of 200 students – of which 50% studied mathematical sciences and 50% studied biological sciences.

Results: There was no significant difference between the students’ behavior according to the different science field they were studying. Women presented lower risk behavior as compared to men (OR = 0.07; CI 95% = 0.01 - 0.56).

Conclusions: Most university students do not protect themselves adequately when exposed to the sun, with this risk behavior being lower in women. Actions aimed at phot-educating students are important.

Keywords: Disease prevention; Skin; Solar radiation; Students

RESUMO

Introdução: O número de casos de carcinoma cutâneo tem aumentado cada vez mais e vem determinando a necessidade de aprimorar estratégias de saúde economicamente eficazes para prevenir a doença e suas complicações.

Objetivo: Realizar estudo transversal, tendo os dados sido coletados diretamente com os participantes, por meio da aplicação de questionário.

Métodos: Com os dados coletados realizou-se o cálculo da regressão logística bivariada, obtendo-se a razão de chances (OR) e o intervalo de confiança (IC) 95% entre as variáveis independentes e o comportamento de risco, o qual foi determinado pela não utilização diária de filtro solar e/ou outros meios de fotoproteção. A amostra foi composta por 200 estudantes, 50% da área de exatas e 50% da área biológica.

Resultados: Não foi constatada diferença significativa entre o comportamento dos graduação nas diferentes áreas analisadas. Mulheres apresentaram menor comportamento de risco em relação aos homens (OR=0,07; IC95%: 0,01-0,56).

Conclusões: A maior parte dos universitários não se protege da exposição solar adequadamente, sendo esse comportamento de risco inferior nas mulheres. Ações de fotoeducação são importantes entre os estudantes.

Palavras-chave: Estudantes; Pele; Prevenção de doenças; Radiação solar
importance of photoprotection for university students

INTRODUCTION

The cases of cutaneous carcinomas have been steadily increasing all over the world, what represents a high social and economic costs for persons and health care systems.\textsuperscript{1-3} Recognizing this increasingly higher impact has determined the need to improve cost-effective health strategies to prevent the disease and its complications.\textsuperscript{1-3}

The etiology of skin cancer is mainly related to sun exposure, what occurs significantly during childhood.\textsuperscript{3-8} The effects of the exposure to ultraviolet radiation represent a well-established risk factor for skin cancer and photoaging. Sun exposure during the first decades of life increases the vulnerability to harmful effects of the radiation.\textsuperscript{3-8}

Considering a life expectancy of 78 years, 23\% of ultraviolet radiation that one individual is submitted to takes place until in the first 18 years, 46\% up to 40 years of age and 74\% up to 59 years of age.\textsuperscript{9} Guidelines on photo-education should include young people, children and their carers, so they can acquire healthy and sensible habits, therefore reducing the risk of developing a late problem caused by irresponsible sun exposure.\textsuperscript{10,11} Photo-education is important for the maintenance of health in the adult population, mainly by the fact that the damage caused by ultraviolet radiation is cumulative throughout life.\textsuperscript{11}

Besides, young people usually expose excessively to solar radiation, since they spend a lot of time outdoors. Therefore, it is important to understand the behavior of university students in order to increase awareness, because if the exposure is inappropriate it can constitute a risk factor for skin cancer.\textsuperscript{12,13}

The objective of this study was to investigate sun exposure and protection practices and factors associated in university students in Campinas, São Paulo, Brazil.

METHODS

Design

It is a cross-sectional study conducted with undergraduate students of the Universidade Estadual de Campinas (UNICAMP) in 2016.

Context

UNICAMP is a public university in inland São Paulo, Brazil. The participants in the research were undergraduate students of the courses of Pharmacy, Biology, Medicine, Food Engineering, Statistics, integrated elementary and secondary education, Chemistry, Chemical Engineering, Mechanical Engineering and integrated degrees in Chemistry and Physics. Interviews were conducted during the months of April, June, September, October and November 2016 during the mornings and afternoons in the university facilities.

Participants

Students of the exact and biological/health sciences of any age and both genders were considered eligible. Participants were selected by convenience sampling when the interviewer was present at the site of recruitment.

Variables

Demographic and knowledge on photoprotection variables were collected, including: gender (male or female), age (in years, subsequently categorized into 17-18; 19-20 and >21 years), daily use of sunscreen and when intentional exposure occurred (never, sometimes, always), use of other measures of photoprotection daily and during intentional exposure (yes or no), photoprotection measures besides sunscreen (yes or no, and descriptive answer if yes), exposure to artificial tanning booths (never, once or more times), knowledge on how much of the product to apply and on the difference of ultraviolet radiation A and B, opinion whether tanning is healthy (yes or no, and descriptive answer if yes) and use of sunscreen on cloudy days (yes or no).

Source of data, measurements and quality control

Data were collected directly from the participants with a paper questionnaire applied by the interviewer, Pharmacy student. The semi-structured questionnaire was prepared based on previous studies\textsuperscript{4} and made by identification questions (name, university, course, gender and age) and open- and closed-ended questions about photoprotection habits.

The conclusion of the study pointe to a risk behavior in relation to the sun protection of the interviewed, defined as the answers “sometimes” or “no” in the closed-ended questions that evaluated the use of daily sunscreen or other photoprotection measures, intentional exposure and on cloudy days and exposure to tanning booths. The absence of answers on the questions regarding the use and description of other photoprotection measures, besides the photoprotective formulation was considered as “no”. Open-ended questions were used to evaluate the participants’ knowledge about the required amount of photoprotective formulation to be applied, as well as the knowledge about the difference and importance of photoprotection against ultraviolet radiations A and B.

After data collection, the answers obtained were tabulated in Microsoft Excel spreadsheets by the interviewer herself, with posterior typing review.

Size of the study

No calculation of the size of the sample was performed, with an estimation of a minimum of 100 participants from each area of study (exact and biological sciences).

Statistical methods

Descriptive statistics was initially performed, obtaining absolute and relative frequencies of each variable. Bivariate logistic regression was calculated to estimate the odds ratio (OR) and the 95\% confidence interval (95\% CI) between the solar exposure risk behavior and the independent variables. We adopted the level of p < 0.05 to define statistical significance.

Ethical aspects

The project was approved but the Committee of Ethics in Research of the Universidade Federal de São Paulo, under the
number and certificate presented to ethical assessment number 46273415.8.0000.5505.

All participants were instructed and signed the consent form, confirming participation in the study.

RESULTS

Two hundred university students were interviewed, 75.5% female, 50% from exact sciences and 50% from health and biological sciences; more than half (59%) up to 20 years of age.

The prevalence of risky behavior regarding sun protection among these students was of 83% (95% CI: 77.7- 88.3%); no students knew the difference between ultraviolet radiation A and B, 96% did not know the correct amount of sunscreen that should be applied and 23% believed that skin tanning is healthy, most of the subjects associating it to vitamin D production (Table 1).

The use of other photoprotection measures corresponds to 15.5% daily and 47.5% during intentional exposure. Moreover, 80% and 16% did not use sunscreen on the same situations and 67% did not use it on cloudy days (Table 1). None of the participants used artificial tanning booths.

The use of sunglasses was the most mentioned photoprotection measure besides sunscreen (54.3%) for daily protection, followed by opting to stay in the shade or do not expose (11.4%). On occasional situations of intentional exposure, 37.8% reported the use of caps or hats, 32.4% of sunglasses and 18.9% of an umbrella or beach umbrella (Table 2).

Gender was associated to risk behavior and was significantly lower in females OR = 0.07 (95% CI: 0.01-0.56). Age, study area, knowledge of the difference between ultraviolet radiation A and B, considering tanning healthy and knowledge on how much sunscreen to apply were not associated to risky sun exposure behavior (Table 3).

DISCUSSION

In this study, 80% of the undergraduates interviewed had risky behavior regarding sun exposure. Among males, this behavior was significantly worse. Body worship, outdoor physical activities and the aesthetic value of tanning can lead young people to a prolonged sun exposure and, many times, without the adequate protection.

Limitations of this study include a convenience sample, lack of socioeconomic data and use of a non-validated questionnaire, what impairs data extrapolation.

Other studies also report low adherence to daily use of sunscreen among undergraduates. In a cross-sectional study with 2,622 people in Saudi Arabia in 2010 and 2011, AlGhamdi et al.³ reported that 23.7% (of the total of 2,566) used sunscreen daily.³ In the analytical cross-sectional study in a Brazilian university of the Midwest region, Castilho, Sousa and Leite¹³ reported that around 25% of the 308 students interviewed used sunscreen daily.

In contrast with these results, in a study conducted in the metropolitan region of Porto Alegre, 85% of the 1,030 individuals interviewed confirmed using sunscreen in 2001.¹² In Italy, questionnaires were applied to students between 11 and 16 years of age, observing that 91% of the 379 teenagers confirmed using sunscreen, but only 50.4% reapplied it. The need for sunscreen use when the exposure is intentional, except for when at the beach was questioned and 52% believed that it would not be necessary.⁷

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<tr>
<td>No</td>
<td>192</td>
<td>96</td>
</tr>
<tr>
<td>Yes</td>
<td>8</td>
<td>4</td>
</tr>
<tr>
<td>Considers tanning healthy</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>154</td>
<td>77</td>
</tr>
<tr>
<td>Yes</td>
<td>46</td>
<td>23</td>
</tr>
</tbody>
</table>
The influence of gender in risk behavior was also reported in studies conducted in Brazil (Porto Alegre, Piauí, Curitiba) and in other countries (Australia, Saudi Arabia, Germany and China), where women had better photoprotection habits and the non-use of sunscreen was higher in men.\textsuperscript{2,3,8,12,14-16}

Few participants could tell the correct amount of sunscreen that should be applied. A study performed in the city of Natal, Rio Grande do Norte, Brazil, with 200 volunteers demonstrated that the amount of sunscreen applied affects its efficacy. The smaller amount applied, the lower is the protection, what demonstrates the need of knowing how much to be used.\textsuperscript{17}

None of the undergraduates interviewed knew the difference between ultraviolet radiation A and B. Sun protection factor (SPF number) displayed in the packaging of photoprotection products refers only to protection against ultraviolet B, that can cause erythema, pain, dyspigmentation and peeling.\textsuperscript{1,16} It is important that the user also seeks protection against ultraviolet A, that contribute more significantly to immunosuppression and early ageing, drying the skin and damaging its elasticity.\textsuperscript{1,16}

A good part of undergraduates who consider healthy to have a tanned skin mentioned the importance of the sun for vitamin D synthesis, which occurs by photolysis in the epidermis with the action of ultraviolet B on ergosterol and cholesterol.\textsuperscript{18} Lack of sun exposure can cause deficiency in the synthesis of vitamin D,\textsuperscript{19} however, the exposure must be sensible, i.e., photoprotection measures should be used so as to take advantage of the benefits from this exposure, minimizing harm. In a study conducted in China, the importance of sun for vitamin D production was discussed, observing that men believed that tanned skin is healthier and more attractive, whereas women believed that tanned people look older.\textsuperscript{14}

None of the participants used artificial tanning booths. This result can be explained by the awareness of the dangers of this method and the difficulty of access due to regulatory measures. In Taguatinga (Distrito Federal, Brazil), this habit was reported by 3.5% of 202 female students in 2007, close to 4.5% of the 379 teenagers interviewed in Italy in 2010, who confirmed using tanning booths and 7.4% in this study judged safe having artificial tanning before sun exposure.\textsuperscript{7,13}

The area of study did not interfere with the risk behavior of undergraduates, similarly to a previous study conducted in Curitiba with 398 medical school students in 2012, who had already had the discipline of dermatology or not.\textsuperscript{16} In contrast to these results, a cross-sectional study also with 398 Brazilian undergraduates in Teresina, Piauí in 2011 noticed that undergraduates of health sciences adopted better photoprotection measures.\textsuperscript{15}

Australian studies with 101,449 teenagers interviewed in 1993, 1996, 1999 and 2002 demonstrated that care related to sun exposure got poorer over the years.\textsuperscript{6} One study in Germany also observed that the risk behavior is more pronounced among younger people.\textsuperscript{8} In Saudi Arabia, it was seen that young employees or students younger than 30 years of age used more sunscreen than those unemployed and older than 30 years.\textsuperscript{3}

In this study, adoption of other photoprotection measures was of 15.5% daily and 47.5% during intentional exposure. In contrast, in Saudi Arabia, 81.5% reported using sunscreen, 95% clothing coverage, 90% caps or hats and 97.9% sought shade to protect from the sun.\textsuperscript{3} In a study performed in 2015 over the phone in Germany with teenagers and adults regarding sun protection behavior, it was found that the most used photoprotection measure was the use of long-sleeved clothing, mentioned by 54% of the 3,000 interviewed, and the least used was hats, only used by 18% of all interviewed.\textsuperscript{8} In China, it was observed that women preferred using parasols and sunscreen for protection, whereas men opted to use long-sleeved clothing and to reduce sun exposure; both genders were not as concerned regarding eye protection.\textsuperscript{14}

### Table 2: Description of photoprotection measures daily and during intentional exposure used, except for sunscreen

<table>
<thead>
<tr>
<th>Measures mentioned</th>
<th>Daily use</th>
<th></th>
<th>Occasional use</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N</td>
<td>%</td>
<td>N</td>
<td>%</td>
</tr>
<tr>
<td>Cap/hat</td>
<td>3</td>
<td>8.6</td>
<td>56</td>
<td>37.8</td>
</tr>
<tr>
<td>Umbrella/sun umbrella</td>
<td>3</td>
<td>8.6</td>
<td>28</td>
<td>18.9</td>
</tr>
<tr>
<td>Make up</td>
<td>3</td>
<td>8.6</td>
<td>1</td>
<td>0.7</td>
</tr>
<tr>
<td>Clothing</td>
<td>3</td>
<td>8.6</td>
<td>9</td>
<td>6.1</td>
</tr>
<tr>
<td>Shade/no exposure</td>
<td>4</td>
<td>11.4</td>
<td>4</td>
<td>2.7</td>
</tr>
<tr>
<td>Sunglasses</td>
<td>19</td>
<td>54.3</td>
<td>48</td>
<td>32.4</td>
</tr>
<tr>
<td>Lip balm</td>
<td>0</td>
<td>-</td>
<td>2</td>
<td>1.4</td>
</tr>
</tbody>
</table>

### Table 3: Odds ratio (OR) and 95% confidence interval (95% CI) of risk behavior according to the variables in the study

<table>
<thead>
<tr>
<th>Variable</th>
<th>OR (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Demographic characteristics</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>0.07 (0.01-0.56)</td>
<td>0.012</td>
</tr>
<tr>
<td>Age (years)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>17-18</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>19-20</td>
<td>1.84 (0.67-5.06)</td>
<td>0.235</td>
</tr>
<tr>
<td>&gt;21</td>
<td>1.28 (0.49-3.35)</td>
<td>0.620</td>
</tr>
<tr>
<td>Area</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Exact sciences</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Biological sciences</td>
<td>0.75 (0.36-1.58)</td>
<td>0.452</td>
</tr>
<tr>
<td>Knowledge on photoprotection</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Considers tanning healthy</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>1.90 (0.69-5.24)</td>
<td>0.213</td>
</tr>
<tr>
<td>Knows how much sunscreen to apply</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>0.60 (0.12-3.11)</td>
<td>0.543</td>
</tr>
</tbody>
</table>
The participants in this study showed a high risk behavior, demonstrating the need for promotion of photoprotection. In the United States, it was also raised the need for adoption of sun protection, because it was observed that sun protection was not common in all levels of school, particularly for those in high school. A successful example of the dissemination of knowledge on the subject was the intervention done in Australia between 1992 and 1996, regarding daily sunscreen use for the prevention of skin cancer. In a subsequent study from 1997 to 2002, the participants showed improved compliance to the application of sunscreens and a safer behavior regarding sun exposure.

Due to geographical characteristics and cultural trends, Brazilians are among the people who expose more to the sun. Therefore, photoeducation measures should be encouraged and broadcasted in Brazil, in order to prevent the development of acute and chronic actinic damage, particularly because epidemiological data point towards a steady increase in the incidence of skin cancer.

CONCLUSION

Risk behavior regarding sun exposure is high among undergraduates and more common among males. The students have poor knowledge on photoprotection. Photoeducation activities are important for the young.

REFERENCES


DECLARATION OF PARTICIPATION:

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Tais Freire Galvão | b ORCID 0000-0003-2072-4834 Statistical analysis, assistance in writing, critical review and approval of the final version.

Priscila Gava Mazzola | b ORCID 0000-0002-3795-8189 Literature and text review.

Gislaine Ricci Leonardi | b ORCID 0000-0002-7126-1326 Approval of the final version, study design and planning; preparation and writing of the text, active participation in the research process, critical review of the literature and of the text.
Comparative study between two topical anesthetics in dermatological procedures

Estudo comparativo entre dois anestésicos tópicos em procedimentos dermatológicos

ABSTRACT

Introduction: Managing pain during the performance of dermatological procedures is a challenge. Several topical anesthetics are available for use prior to carrying out minimally invasive treatments.

Objective: To evaluate the efficacy of topical anesthetics for reducing the pain during dermatological procedures and to compare the effectiveness of two different anesthetics using the visual numeric pain scale.

Methods: Twenty-five patients underwent facial dermatological procedures after the application of two commercial preparations: one containing 7% lidocaine and 7% tetracaine (applied in one hemiface) and the other containing 4% lidocaine. Each of the preparations was applied in one hemiface. Pain intensity was assessed using the Visual Numeric Pain Scale (VNS) at the end of the procedure.

Results: Eighty-four percent of the patients reported a lower pain score in the hemiface that received the commercial preparation containing 7% lidocaine and 7% tetracaine. The mean pain score in the hemifaces that received the 4% lidocaine preparation was 7.3, while that computed for the hemifaces that received the 7% lidocaine and 7% tetracaine preparation was 5.3.

Conclusions: Topical anesthetics are effective and safe for pain reduction in dermatological procedures, with the commercial preparation containing 7% lidocaine and 7% tetracaine was more effective than the one containing 4% lidocaine.

Keywords: Anesthesia; Anesthesia, local; Dermatologic surgical procedures; Minimally invasive surgical procedures; Pain perception; Pretreatment

RESUMO

Introdução: A dor durante a execução dos procedimentos dermatológicos é um desafio na prática médica. Vários anestésicos tópicos estão disponíveis para ser utilizados previamente aos tratamentos minimamente invasivos.

Objetivo: Avaliar a eficácia do uso do anestésico tópico na redução da dor durante a execução de procedimentos dermatológicos e comparar a eficácia de dois deles, utilizando a escala visual numérica de dor.

Métodos: 25 pacientes foram submetidos a procedimentos dermatológicos faciais após a aplicação da preparação comercial de lidocaína 7% e tetracaína 7% em uma hemiface e preparação comercial de lidocaína 4% em outra hemiface. A intensidade da dor foi avaliada por escala visual numérica de dor (EVN) no final do procedimento.

Resultados: 84% dos pacientes referiram escore de dor menor na hemiface que recebeu preparação comercial de lidocaína e tetracaína a 7%. A média dos escores de dor nas hemifaces que receberam preparação comercial de lidocaína 4% foi de 7,3, enquanto naquelas que receberam preparação comercial de lidocaína 7% e tetracaína 7% foi de 5,3.

Conclusões: Anestésicos tópicos são eficazes e seguros para diminuição da dor em procedimentos dermatológicos, sendo que a preparação comercial de lidocaína e tetracaína a 7% foi mais eficaz na redução da dor do que a preparação comercial de lidocaína 4%.

Palavras-chave: Anestesia; Anestesia local; Percepção da dor; Procedimentos cirúrgicos dermatológicos; Procedimentos cirúrgicos minimamente invasivos; Pré-tratamento
INTRODUCTION

The increased demand for aesthetic dermatological procedures is remarkable; however, one of their challenges is pain. This lead to an increased need for the use of effective and safe topical anesthetics, providing more comfort for the patient and guaranteeing better results.1,2 There are many topical anesthetics available in the Brazilian market, such as commercial preparations of 2.5% lidocaine and 2.5% prilocaine (Emla® AstraZeneca do Brasil Ltda, São Paulo, Brazil),3 commercial preparation of 4% lidocaine (Dermomax® Ache laboratórios farmacêuticos, São Paulo, Brazil)4 and commercial preparation of 7% lidocaine and 7% tetracaine (Pliaglis® Galderma Brasil Ltda, São Paulo, Brazil).5 The objective of this study was to compare the efficacy between two topical anesthetics, the commercial preparation of 4% lidocaine (Dermomax®) and the commercial preparation of 7% lidocaine and 7% tetracaine (Pliaglis®), evaluating pain scores in patients undergoing different types of dermatological procedures.

METHODS

Prospective and comparative pilot study, in the same patient.

Twenty-five patients from a private practice in Campinas (SP) were submitted to different dermatological preparation – fractional CO2 laser (SmartXide®, Deka Medical Lasers, Florence, Italy), intense pulsed light (all wavelengths – Harmony XL®, LBT Lasers, São Paulo, Brazil), infrared (Cutera xeo®, Titan® tip, California, USA), radiofrequency (Reaction®, Viora Medical Solutions, Israel), q-switched laser for the removal of pigmented from the eyebrows and intradermal injection of hyaluronic acid.

All patients were female, aged between 25 and 71 years (mean of 48.6 years). Thirty minutes before the procedure, a thick layer of the commercial preparation containing 4% lidocaine (Dermomax® Ache laboratórios farmacêuticos, São Paulo, Brazil) was applied to one side of the face, chosen randomly, and on the other side the commercial preparation of 7% lidocaine and 7% tetracaine (Pliaglis®) was applied to one side of the face, chosen randomly, and on the other side the commercial preparation of 7% lidocaine and 7% tetracaine (Pliaglis® Galderma Brasil Ltda, São Paulo, Brazil) was applied. The study was blind for the patients. The parameters used in each procedure were the same for both sides of the face. Pain intensity was evaluated by the Visual Numeric Pain Scale (VNS) at the end of the procedure (Figure 1), considering zero absence of pain and 10, maximum pain.6,7

The ethical guidelines from the declaration of Helsinki were adopted in this study.

RESULTS

Eighty percent (20/25) of the patients reported better analgesia on the side where the commercial preparation of 7% lidocaine and 7% tetracaine was applied; 12% (3/25) of the patients reported similar pain scores; and 8% (2/25) reported slightly less pain on the side of the face pre-treated with 4% lidocaine, as shown in table 1.

The mean score of the side of the face where the commercial preparation of 4% lidocaine (Dermomax®) was applied was of 7.4, while the mean of the side of the face where the commercial preparation of 7% lidocaine and 7% tetracaine (Pliaglis®) was applied was of 5.3, as shown in table 2.

<table>
<thead>
<tr>
<th>TABLE 1: Number and percentage of patients with equal or lower pain score with each anesthetic evaluated</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
</tr>
<tr>
<td>-------</td>
</tr>
<tr>
<td>Total number of patients evaluated</td>
</tr>
<tr>
<td>Patients whose pain score was lower on the side of the face</td>
</tr>
<tr>
<td>face with 7% lidocaine and 7% tetracaine</td>
</tr>
<tr>
<td>Patients whose pain score was the same on both sides of the</td>
</tr>
<tr>
<td>face with 4% lidocaine</td>
</tr>
<tr>
<td>Patients whose pain score was lower on the side of the face</td>
</tr>
<tr>
<td>face with 7% lidocaine and 7% tetracaine</td>
</tr>
</tbody>
</table>

The two patients who reported equal values on the pain scale (patients 12 and 24, according to Table 1) reported high scores in the pain scale, respectively 8 and 9 on a scale with maximum pain of 10. Also, in relation to the comparison between both anesthetics, there were cases like patients 15 and 22, who reported a large difference regarding the evaluation of analgesia when comparing both sides of the face (Table 1). We would also like to call attention to patient 16, who reported maximum pain (score 10) during fractional CO2 laser on the side of the face where lidocaine was applied and 7.5 on the other side, where the anesthetic containing lidocaine in higher concentration associated to tetracaine was applied, what allows us to infer an important effect in analgesia provided by the product containing the combination of anesthetics. Only two patients reported more pain on the side of the face where the commercial preparation of 7% lidocaine and tetracaine was applied, however, we should mention that the difference observed was small compared to the side of the face where the anesthetic containing 4% lidocaine was applied (patients 11 and 18, 1 point difference).

There were no significant side effects.

DISCUSSION

Pain is still a limiting factor for many dermatological procedures. With the advent of topical anesthetics, these procedures became more reasonable in clinical practice without the need of resorting to infiltrative anesthesia, blocks or sedation.1,2,8

Topical anesthetics are effective and safe for many dermatological procedures, such as ablative and non-ablative lasers, injection of fillers and botulinum toxin, radiofrequency, infrared and intense pulsed light, with a low risk for adverse events.8,10

These agents act in dermal nerve endings, reducing the transmission of afferent nerve impulses.11,12 The topical anesthetic interacts with sodium channels in the nerve endings, blocking their influx. The threshold of excitation increases and gradually reduces the action potential onset and conduction of the nerve impulse. However, for that to occur, local anesthetics should propagate through the stratum corneum and reach the interior of nerve fibers. Ability of propagation, anesthetic potency, pharmacokinetics characteristics and adverse events are intrinsically related to their chemical structure and physical-chemical properties.1,2,9,10,12
<table>
<thead>
<tr>
<th>Patient</th>
<th>Procedure performed</th>
<th>Score, according to pain scale, on the side of the face where the anesthetic containing 7% lidocaine and 7% tetracaine was applied</th>
<th>Score, according to pain scale, on the side of the face where the anesthetic containing 4% lidocaine was applied</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient 1</td>
<td>Skinbooster</td>
<td>20</td>
<td>80</td>
</tr>
<tr>
<td>Patient 2</td>
<td>Infrared</td>
<td>3</td>
<td>12</td>
</tr>
<tr>
<td>Patient 3</td>
<td>Radiofrequency</td>
<td>6.5</td>
<td>8</td>
</tr>
<tr>
<td>Patient 4</td>
<td>Fractional CO₂ laser</td>
<td>5</td>
<td>9</td>
</tr>
<tr>
<td>Patient 5</td>
<td>Fractional CO₂ laser</td>
<td>4</td>
<td>8</td>
</tr>
<tr>
<td>Patient 6</td>
<td>Fractional CO₂ laser</td>
<td>3</td>
<td>8</td>
</tr>
<tr>
<td>Patient 7</td>
<td>Fractional CO₂ laser</td>
<td>6</td>
<td>7</td>
</tr>
<tr>
<td>Patient 8</td>
<td>Fractional CO₂ laser</td>
<td>6</td>
<td>7</td>
</tr>
<tr>
<td>Patient 9</td>
<td>Fractional CO₂ laser</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Patient 10</td>
<td>Fractional CO₂ laser</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>Patient 11</td>
<td>Fractional CO₂ laser</td>
<td>8</td>
<td>9</td>
</tr>
<tr>
<td>Patient 12</td>
<td>Fractional CO₂ laser</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>Patient 13</td>
<td>Fractional CO₂ laser</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>Patient 14</td>
<td>Fractional CO₂ laser</td>
<td>6</td>
<td>8</td>
</tr>
<tr>
<td>Patient 15</td>
<td>Fractional CO₂ laser</td>
<td>1</td>
<td>8</td>
</tr>
<tr>
<td>Patient 16</td>
<td>Fractional CO₂ laser</td>
<td>7.5</td>
<td>10</td>
</tr>
<tr>
<td>Patient 17</td>
<td>Fractional CO₂ laser</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>Patient 18</td>
<td>Fractional CO₂ laser</td>
<td>7</td>
<td>6</td>
</tr>
<tr>
<td>Patient 19</td>
<td>Fractional CO₂ light</td>
<td>5</td>
<td>7</td>
</tr>
<tr>
<td>Patient 20</td>
<td>Fractional CO₂ laser</td>
<td>6</td>
<td>8</td>
</tr>
<tr>
<td>Patient 21</td>
<td>Fractional CO₂ laser</td>
<td>6.5</td>
<td>8</td>
</tr>
<tr>
<td>Patient 22</td>
<td>Fractional CO₂ laser</td>
<td>2</td>
<td>8</td>
</tr>
<tr>
<td>Patient 23</td>
<td>Fractional CO₂ laser</td>
<td>7</td>
<td>9</td>
</tr>
<tr>
<td>Patient 24</td>
<td>Fractional CO₂ laser</td>
<td>9</td>
<td>9</td>
</tr>
<tr>
<td>Patient 25</td>
<td>Fractional CO₂ laser</td>
<td>4</td>
<td>7</td>
</tr>
<tr>
<td>MÉDIA</td>
<td></td>
<td>5.3</td>
<td>7.4</td>
</tr>
</tbody>
</table>
indicated in the Visual Numeric Pain Scale (Figure 1); with a two-point difference when compared to the other formulation containing only 4% lidocaine. Three patients reported similar scores for both products. It is also worth highlighting that for the only two patients who reported higher pain with the association of lidocaine and tetracaine, the difference was small when compared to the side where only 4% lidocaine was used.

CONCLUSION

It was possible to observe in this study that the cream with 7% lidocaine and 7% tetracaine was more effective in most patients assessed, with more effective analgesia of the face during different dermatological treatment than the product containing 4% lidocaine.

REFERENCES


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Data collection, text review and submission of the article.

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Bibliographical review and preparation of the manuscript.

Even though pain is a subjective symptom, difficult to measure, the Visual Numeric Pain Scale (VNS) is already considered a good parameter for the quantification of pain and is widely used in many areas of medicine.6,7

This study compared the efficacy in the control of pain of two topical anesthetics: a commercial preparation of 7% lidocaine and 7% tetracaine and a commercial preparation of 4% lidocaine. Pain scores were lower for most patients pre-treated with the commercial preparation of 7% lidocaine and 7% tetracaine (20 patients – 80%) and higher in patients pre-treated with the commercial preparation of 4% lidocaine.

The first commercial preparation evaluated was more effective in 80% of 25 patients participating in this study, in relation to the pain reported during a dermatological procedure, indicated in the Visual Numeric Pain Scale (Figure 1); with a two-point difference when compared to the other formulation containing only 4% lidocaine. Three patients reported similar scores for both products. It is also worth highlighting that for the only two patients who reported higher pain with the association of lidocaine and tetracaine, the difference was small when compared to the side where only 4% lidocaine was used.

CONCLUSION

It was possible to observe in this study that the cream with 7% lidocaine and 7% tetracaine was more effective in most patients assessed, with more effective analgesia of the face during different dermatological treatment than the product containing 4% lidocaine.

REFERENCES

Nutraceutical compound for the treatment of telogen effluvium associated with brittle nails syndrome

Composto nutracêutico no tratamento do eflúvio telógeno associado à síndrome das unhas fracas

DOI: http://www.dx.doi.org/10.5935/scd1984-8773.2018101954

ABSTRACT

Introduction: The use of nutraceutical products in the treatment of chronic telogen effluvium of non-specific cause – associated or not with signs and symptoms of nail plate frailty – has been proposed in the literature. Supplementation of nutrients and trace elements that are crucial to follicle’s metabolism seems to be linked to this effect.

Objective: To evaluate the efficacy of a nutraceutical containing vitamins A, C, E and B complex, folic acid, iron, niacin, biotin, zinc and calcium pantenolate, among others, in the treatment of telogen effluvium associated with the brittle nails syndrome.

Methods: A clinical, prospective, open and monocentric study was carried out with the evaluation of 62 volunteers. Clinical evaluation measurements – based on digital phototrichogram and imaging – were performed at the baseline, and at 45 and 90 days after the use of the studied product.

Results: At the end of the study, a statistically significant (p <0.05) reduction of 31.56% in the number of telogen strands could be observed, coupled with a significant improvement in clinical parameters related to hair and nails. The instrumental evaluation based on image analysis showed a 36.63% reduction in nail desquamation, which, although not statistically significant, has shown benefits in the absolute outcome.

Conclusions: The use of nutraceuticals was proven effective in the treatment of telogen effluvium associated with the brittle nails syndrome.

Keywords: Alopecia; Dietary supplements; Nails

RESUMO

Introdução: O uso de produtos nutracêuticos no tratamento do eflúvio telógeno crônico de causa inespecífica, associado ou não a sinais e sintomas de fragilidade da lâmina ungueal, tem sido proposto na literatura. A suplementação de nutrientes e oligoelementos essenciais ao metabolismo do folículo parece ser o mecanismo relacionado a esse efeito.

Objetivo: Avaliar a eficácia de nutracêutico contendo, entre outros elementos, vitaminas A, C, E, complexo B, ácido fólico, ferro, niacina, biotina, zinco e pantenolato de cálcio, no tratamento do eflúvio telógeno associado à síndrome das unhas fracas.

Métodos: Estudo clínico, prospectivo, aberto e unicêntrico, com avaliação de 62 voluntárias. Medidas de avaliação clínica, por fototrigíramo digital e por imagem, foram realizadas na visita inicial, 45 e 90 dias após o uso do produto estudado.

Resultados: Ao final do estudo, observou-se redução estatisticamente significativa (p < 0.05) de 31,56% do número de fios telógenos, acompanhado de melhora significativa nos parâmetros clínicos relacionados aos cabelos e às unhas. A avaliação instrumental por análise de imagens demonstrou redução de 36,63% da descamação das unhas, que, embora não estatisticamente significativa, demonstrou benefícios no resultado absoluto.

Conclusões: O uso de produtos nutracêuticos demonstrou eficácia no tratamento do eflúvio telógeno associado à síndrome das unhas frágeis.

Palavras-chave: Alopecia; Suplementos nutricionais; Unhas
INTRODUCTION

We can define hair disorders as any condition in which the visible hair coverage does not represent the normal parameters of growth. Normal parameters of hair growth vary according to gender, ethnic group, age and/or local culture.

Diffuse hair loss is a condition as common as challenging for the dermatologist. There are numerous causes for the disorder, being telogen effluvium (TE) one of the most common.

TE is an abnormality of hair cycle that results in excessive shedding of telogen hair. It’s actual incidence is uncertain, since many cases are sub-clinical. Common causes of TE include the use of medications, thyroid disorders and post-partum period. However, in a large number of cases it is not possible to find a cause.

The activity of the hair follicle is cyclical, and scalp hairs have 10 to 30 cycles throughout life. In healthy individuals, hair grows approximately 0.35mm/day. Scalp hair density varies, with most people having between 100,000 and 150,000 hairs. We can shed 40 to 100 hairs on an average day and 200 to 300 hairs when the hair is washed.

Hair growth occurs during the anagen phase; involution in catagen phase; and rest in telogen phase. Shedding of “dead” hair through the follicle (exogenous phase) occurs at the end of telogen phase of beginning of anagen. At the beginning of each anagen phase, a new hair is produced. On the scalp, 86% of hair is in the anagen phase (that lasts two to six years) and up to 13% in the telogen phase (that lasts three to six months). Catagen phase is transient, lasting three to six weeks, and 1% of hairs are in this phase.

In TE there is a disturbance in the hair cycle, where a trigger precipitates the simultaneous transition of a large amount of anagen hairs into telogen hairs, which shed at the same time.

The main complaint is hair loss. In view of the shedding, a complete and detailed history should be taken, inquiring about family history, use of medications, previous illnesses, recent surgeries and restrictive diets. We should request a full blood count, urinalysis, ferritin, thyroid hormones, levels of vitamin D and other vitamins, according to the history. Of the ancillary diagnostic follow-up methods for TE, the one considered most appropriate is phototrichogram performed by the device Trichoscan (TRICHIOLOG GmbH, Freiburg, GER).

It is a non-invasive method that combines epiluminescence microscopy with the analysis of automatic digital imaging for the study of important hair parameters. It can analyze: hair density (n per cm²), hair shaft diameter (µm), growth rate (mm per day) and anagen/telogen ratio. It is a very useful option in cases of diffuse alopecia, when we need to identify and quantify the response to the treatments used.

For the treatment of TE, besides eliminating the cause, supplementation of substances involved in the production of the hair shaft is particularly important. Since the hair is among the most metabolically active tissues in the human body, stimulation of the follicular metabolism can increase hair resistance to external harmful agents, besides stimulating hair growth.

Vitamin and mineral supplementation for hair disorders has been used for a long time and has many reports in the literature. Special attention is given to micronutrients, a term that encompasses trace elements, minerals, vitamins and amino acids. Since hair shafts are formed by 98% of proteins, it is assumed that a diet rich in proteins is essential for their health. Indeed, in nutritional states when there is a deficiency in protein intake, hair shafts are clearly affected.

The use of vitamins, particularly of the B complex, is also described and there are many reports in the literature, with evidences of benefit in hair growth. Thiamine (vitamin B1) deficiency leads to beriberi, and one of the consequences is the production of fine hair. Vitamin B5 (pantothenic acid) is considered to be an ingredient that gives strength and flexibility to hair. It is suggested that zinc is one of the minerals responsible for the repercussion of hair fragility, subsequent to changes with protein deficiency, being also essential for the growth of anexitexes (hair and nails) since hair follicles maintain a significant metabolism with higher demand of nutrients. Biotin, or vitamin H, is a cofactor for many metabolic enzymes and has been used successfully for the treatment of animals with hair growth deficiencies. Even though the daily recommended dose is still unknown, there are reports that its supplementation provides benefits for the treatment of brittle nails and hair loss.

Different studies evaluated the benefit of the use of vitamin supplementation in the treatment of telogen effluvium, with positive results.

On the other hand, brittle nail syndrome (BNS) is a heterogeneous disorder, characterized by increased fragility of the nail plate. Nearly 20% of the population is affected, women twice as men. Association between TE and BNS is frequent, and it usually requires a combined approach from the dermatologist.

Most patients experience brittle nails as a significant cosmetic problem and a significant number report that these disorders are painful, affect daily activities and can have a negative impact on professional abilities.

Hydration and delamination of the nail plate can change according to the seasons and play a significant role in certain occupations such as housewives, nursing and hairdressers, for whom repetitive immersion and drying of hands result in contraction and expansion of the nail, leading to splitting. In particular, occupational exposure to chemical products such as thiglycolates, cement, solvents, acids, alkalis, amines, salt and sugar solution can dissolve intercellular lipids and damage intercellular adhesion, leading to splitting. Besides, cosmetic products, particularly nail polish and cuticle removers and procedures such as excessive use of manicure instruments can cause intercellular fractures. The interactive trauma of the nails (for example typing, dial phones, inadequate nail clipping and long duration of nails) can damage the nail plate and cause fractures.

BNS treatment should take into consideration removal of the causal factors and use of nutraceuticals, that have been frequently advocated with good results.

This study evaluated the benefits of the use of a nutritional supplement (nutraceutical) for the treatment and TE and nail fortification.
METHODS

Objective of the study
The objective of this study was to evaluate in an open, non-comparative fashion the efficacy and safety of a nutraceutical compound for the treatment of chronic TE associated to BNS.

Population studied
After approval by the Committee of Ethics in Research (CER), from May to October 2016, 68 female volunteers, aged between 18 and 45 years, with the complaint of diffuse and non-specific hair loss associated to brittle nails were recruited and included. All volunteers expressed their desire of participating in the study by signing a consent form before undergoing any of the procedures envisaged in the clinical protocol.

To assure the eligibility of the volunteers, besides populational characteristics they should also present, as a primary criterion for inclusion, rates of telogen hair on the centroparietal region higher than or equal to 20% determined by phototrichogram and could not meet any of the following criteria: pregnancy or risk of pregnancy, lactation, presence of alopecia areata or androgenetic alopecia, signs of specific nail disorders, history of nutritional deficiencies or hormonal disorders or any other systemic condition that could affect the hair and nails. No restrictive diet could have been commenced up to three months before the study. Hair treatments such as hair straightening and coloring in the previous three months would also be considered exclusion criteria.

Methodological procedures

Clinical evaluation
Clinical evaluations were performed in the beginning (T0), in 45 days after beginning (T45) and in 90 days after beginning (T90), through a standardized scale to evaluate hair loss, density (scalp coverage), hair quality (flexibility, resistance to traction, tip breakage) and signs of nail fragility (plate splitting, diagonal splitting, ridges and furrows, longitudinal splitting and nail plate thinning). A four-point scale was used, where zero represented absence of abnormality and three maximum abnormality in each criterion.

Phototrichogram (Trichoscan®)
The evaluation with phototrichogram is non-invasive and consists in capturing standardized images for the evaluation of hair growth.

Once the experimental area is defined, the device allows capturing micro images, with a standardized lens magnification of 20x of a 2cm diameter area, with the hair shaved. Reading of the micro image is performed by the software Trichoscan® Fotofinder (TRICHOLOG GmbH, Freiburg, GER), device that issues reports with the following information:

- Hair density: number of hairs per millimeter square;
- Anagen hairs: indicates the percentage of growing hairs;
- Telogen hairs: indicates the percentage of shedding hairs.

Evaluation of ungual desquamation by image analysis
Nail images were registered using a specific support. Each image contains nails from the 3rd, 2nd and 4th fingers of both hands. Only one nail was chosen for the analysis.

Images were captured on the initial visit (before using the product being investigated) and after 90 days of the study (T90), from which the software Image Pro Plus® version 6 (Media Cybernetics, Rockville, EUA) analyzed the nail images, quantifying clear pixels and correlating them with the degree of desquamation.

The software calculated the accumulated histogram of the image with desquamation. The subsequent step was calculating the inflection point of the histogram, determined by the point of tangency of the lines at 45°. All pixels above the inflection point were considered desquamation areas.

The desquamation percentage was calculated according to the formula:
\[
\% \text{ desquamation} = 100 \times \frac{P_x}{P_t},
\]
where:

- \( P_x \) = number of pixels above the inflection point.
- \( P_t \) = number of total pixels of the image.

Product studied
The nutraceutical product (Tacitá® Cristália, Itapira, SP, Brazil) is formed by vitamins A, B complex, C, E, folic acid, iron, niacin, biotin, zinc, calcium pantothenate, magnesium, selenium and lutein, among other micronutrients.

Table 1 shows the detailed formulation of the product studied.

They were provided by the sponsor for the study, 90 capsules by volunteers included in the study.

Each volunteer was instructed to take 1 capsule per day, during the 90 days of the study.

RESULTS
The age group of the volunteers included was from 19 to 45, with a mean age of 36 years.

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Concentration</th>
<th>%DV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vitamin A</td>
<td>2000IU</td>
<td>100</td>
</tr>
<tr>
<td>Vitamin E</td>
<td>14.9UI</td>
<td>100</td>
</tr>
<tr>
<td>Folic acid</td>
<td>240mcg</td>
<td>100</td>
</tr>
<tr>
<td>Biotin</td>
<td>30mcg</td>
<td>100</td>
</tr>
<tr>
<td>Niacinamide</td>
<td>16mg</td>
<td>100</td>
</tr>
<tr>
<td>Vitamin B1</td>
<td>1.2mg</td>
<td>100</td>
</tr>
<tr>
<td>Vitamin B12</td>
<td>2.4mg</td>
<td>100</td>
</tr>
<tr>
<td>Vitamin B2</td>
<td>1.3mg</td>
<td>100</td>
</tr>
<tr>
<td>Vitamin B6</td>
<td>1.3mg</td>
<td>100</td>
</tr>
<tr>
<td>Vitamin C</td>
<td>45mg</td>
<td>100</td>
</tr>
<tr>
<td>Pantothenic acid</td>
<td>5mg</td>
<td>100</td>
</tr>
<tr>
<td>Iron</td>
<td>14mg</td>
<td>100</td>
</tr>
<tr>
<td>Magnesium</td>
<td>130mg</td>
<td>50</td>
</tr>
<tr>
<td>Selenium</td>
<td>34mcg</td>
<td>100</td>
</tr>
<tr>
<td>Zinc</td>
<td>7mg</td>
<td>100</td>
</tr>
</tbody>
</table>
Three volunteers did not return for the final assessment, what was considered loss to follow up. Four volunteers had an adverse event during the study, which was mild and difficult to link to the use of the product. Three of these volunteers discontinued and their data were not considered in the efficacy evaluations. Data of 62 volunteers were considered population Per Protocol.

**Efficacy evaluation**

The primary criterion of efficacy was defined as being the reduction of the percentage of telogen hairs in the phototrichogram evaluation with Trichoscan®.

Secondary efficacy criteria were:

- Increased percentage of anagen hairs in the phototrichogram
- Increased hair density in the phototrichogram
- Reduction of ungual desquamation by image analysis
- Improved clinical parameters for nail evaluation
- Improved clinical parameters for hair evaluation

Graph 1 shows the mean evolution of the parameters: density, anagen and telogen hairs, initial evaluation (D0) and 90 days after using the product investigated (D90), in the phototrichogram.

Table 2 shows the numerical values regarding the variation in percentage of the parameters evaluated in the phototrichogram, as well as statistical analysis performed using the Student T test. Regarding the primary efficacy criterion, we could observe that there was a statistically significant reduction (p ≤ 0.05) in 31.56% of telogen hairs after continuous use of the product for 90 days.

Regarding secondary efficacy criteria evaluated by the phototrichogram, we observed that there was a 6.74% increase of hair density and 19.64% increase in anagen hairs, both also statistically significant (p < 0.05).

In the clinical evaluation and regarding hair loss, we observed a statistically significant improvement in three parameters evaluated (hair loss, quality and density) at 90 days. In the intermediate evaluation (45 days), only the parameter “hair loss” showed statistically significant improvement.

Table 3 shows the results of the clinical evaluation of parameters related to hair.

Regarding evaluation of nail desquamation using the imaging technique, a 36.63% reduction in median desquamation values was observed in the sample analyzed, between the initial and final date (T90).

**TABLE 2: Percentage of variation for the experimental dates**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>% variation</th>
<th>P value</th>
<th>Conclusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Density</td>
<td>6.74</td>
<td>0.047</td>
<td>Rejects the hypothesis</td>
</tr>
<tr>
<td>Anagen</td>
<td>19.64</td>
<td>&lt; 0.001</td>
<td>Rejects the hypothesis</td>
</tr>
<tr>
<td>Telogen</td>
<td>-31.56</td>
<td>&lt; 0.001</td>
<td>Rejects the hypothesis</td>
</tr>
</tbody>
</table>

* Significance level: 5%
**Hypothesis: there is no difference between the experimental dates

**TABLE 3: Comparison test (Student T) of the experimental dates in relation to the initial visit**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Time</th>
<th>Variation</th>
<th>P value</th>
<th>Conclusion**</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hair loss</td>
<td>D45-D0</td>
<td>54%</td>
<td>&lt; 0.001</td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>D88-D0</td>
<td>50%</td>
<td>&lt; 0.001</td>
<td></td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>Densidade dos cabelos</td>
<td>D45-D0</td>
<td>17%</td>
<td>0.182</td>
<td>Does not reject the hypothesis*</td>
</tr>
<tr>
<td>D88-D0</td>
<td>27%</td>
<td>0.017</td>
<td></td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>Qualidade dos fios</td>
<td>D45-D0</td>
<td>8%</td>
<td>0.373</td>
<td>Does not reject the hypothesis*</td>
</tr>
<tr>
<td>D88-D0</td>
<td>23%</td>
<td>0.005</td>
<td></td>
<td>Rejects the hypothesis*</td>
</tr>
</tbody>
</table>

* Hypothesis: there is no difference between the experimental dates
** Significance level of 5%
Despite the representative values, due to sample dispersion, the statistical analysis with Wilcoxon test did not show significant differences between the initial and final date (p > 0.05).

However, when we observe the results obtained by the clinical assessment performed by the medical investigators, we notice that the improvement was statistically significant (p < 0.05) in all parameters and all experimental dates, as seen in table 4:

Figures 1 and 2 demonstrate examples of the results found in two volunteers with phototrichogram and nail photography, exemplifying how the efficacy criteria were evaluated.

**DISCUSSION**

Hair loss and brittle nails is a very common complaint in dermatological practice, many times presented in combination and resulting in psychological disturbances in the patients, most of them young women who are concerned about their cosmetic appearance.

When specific causes are ruled out, chronic, non-specific telogen effluvium and brittle nail syndrome can be responsible for the larger number of cases.

After ruling out systemic conditions, treatment is mandatory. In these cases, oral supplementation with specific nutraceuticals for hair and nails is quite common among dermatologists, with a positive feedback to the patient.

The association of vitamin and micronutrients is the most adequate choice in these circumstances, promoting restoration of depleted elements due to inappropriate diet that still has no obvious signs of clinically manifested deficiency.

Nutraceutical compounds especially developed for the treatment of hair and nails are able to offer doses close to the dietary reference intakes (DRI) of essential vitamins, micronutrients and trace elements for the metabolism of hair and nails.

The nutraceutical evaluated in this study has the goal of providing an adequate intake of these elements, preventing and treating the first signs of telogen effluvium associated to brittle nail syndrome.

The results obtained after 90 days of treatment were positive, demonstrating that the product tested was capable of statistically reducing the percentage of telogen hairs and, in the same way, increase the percentage of anagen hairs. Reversal of TE is characterized by this effect, with increased anagen/telogen ratio.

Besides, clinical evaluations followed the same pattern, with statistically positive results for the three parameters evaluated.

Regarding the treatment for nail fortification, the results obtained were also positive. The evaluation with image analysis

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**TABLE 4: Comparison test (Student T) of the experimental dates in relation to the initial visit**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Time</th>
<th>Variation</th>
<th>P value</th>
<th>Conclusion**</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nail splitting - onychoschizia</td>
<td>D45-Do</td>
<td>39%</td>
<td>&lt; 0.001</td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>D88-Do</td>
<td>49%</td>
<td>&lt; 0.001</td>
<td></td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>Diagonal splitting</td>
<td>D45-Do</td>
<td>69%</td>
<td>0.018</td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>D88-Do</td>
<td>66%</td>
<td>0.036</td>
<td></td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>Ridges and furrows</td>
<td>D45-Do</td>
<td>9%</td>
<td>0.034</td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>D88-Do</td>
<td>36%</td>
<td>0.006</td>
<td></td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>Longitudinal splitting</td>
<td>D45-Do</td>
<td>65%</td>
<td>0.060</td>
<td>Does not reject the hypothesis*</td>
</tr>
<tr>
<td>D88-Do</td>
<td>87%</td>
<td>0.022</td>
<td></td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>Ungual splitting and thinning</td>
<td>D45-Do</td>
<td>53%</td>
<td>&lt; 0.001</td>
<td>Rejects the hypothesis*</td>
</tr>
<tr>
<td>D88-Do</td>
<td>69%</td>
<td>&lt; 0.001</td>
<td></td>
<td>Rejects the hypothesis*</td>
</tr>
</tbody>
</table>

* Hypothesis: there is no difference between the experimental dates
** Significance level of 5%
demonstrated a 36.63% reduction of desquamation of the nails evaluated and, even though not statistically significant due to data dispersion, the absolute result leaves no doubts of the benefit obtained. To confirm this positive action, clinical evaluations followed, with marked improvement for the items “diagonal splitting” and “longitudinal splitting”.

CONCLUSION
The use of the combination containing vitamins A, B complex, C, E, folic acid, iron, niacin, biotin, zinc, calcium pantothenate, magnesium, selenium and lutein demonstrated the ability of treating chronic TE associated to brittle nail syndrome, increasing the anagen/telogen ratio and improving the aspect of the affected nails, proving to be an adequate therapeutic option for the treatment of hair and nails.

REFERENCES
The utility of confocal microscopy in lentigo maligna lesions: a case report

A utilidade da microscopia confocal nas lesões de lentigo maligna

DOI: http://www.dx.doi.org/10.5935/scd1984-8773.201810105

ABSTRACT
The diagnosis of pigmented facial lesions is considered challenging since benign and malignant lesions might have similar clinical and dermoscopic features – especially in the early stages of the lesion – entailing that it is often difficult to identify lentigo maligna lesions in the face. In this way, confocal reflectance microscopy has the potential to become a useful tool both in the diagnosis and surgical planning of lentigo maligna.

Keywords: Dermoscopy; Hutchinson’s melanotic freckle; Keratosis, actinic; Melanoma; Microscopy, confocal

INTRODUCTION
Lentigo maligna (LM) is a form of in situ melanoma that occurs in photodamaged skin of elderly patients. Due to the fact it most often arises as a facial pigmented lesion, its clinical diagnosis is challenging, for it may resemble benign pigmented lesions.

Facial dermoscopy presents some peculiarities linked to this anatomical area. It is a region with intense exposure to ultraviolet radiation, which causes a certain degree of epidermal atrophy, solar elastosis and epithelial cones rectification, leading to the absence of pigmentary networks (used to classify lesions as melanocytic or non-melanocytic in other regions of the skin) and presence of the so-called pseudo network. The latter is formed by the interruption of the melanin pigment in the follicular ostia and the presence of adnexal structures in melanocytic and non-melanocytic lesions of the face.

In 2000, Schiffner et al. proposed a dermoscopic diagnostic model for LM lesions and lentigo maligna melanoma (LMM) with 93% accuracy, 89% sensitivity and 96% specificity. According to these authors, the dermoscopic features corresponding to the combi-
nation of grayish spots and globules, dark rhomboidal structures and asymmetric pigmentation of the follicular ostia are the most commonly observed in these lesions and correlate with the histological characteristics typical of this type of melanoma – folliculotropism in particular.2

Although the dermoscopic findings described for the diagnosis of LM have a high degree of accuracy, differential diagnosis with pigmented actinic keratosis (PAK) is still challenging, since these latter may present some of the dermoscopic characteristics typical of LM. Only black blotches are specific for the diagnosis of LM, however their occurrence is delayed, rarely emerging in early lesions.3

In this context, Nascimento et al. described a new relevant dermoscopic feature, the inner gray halo (IGH), whose presence proved to be considerably effective in differentiating LM from PAK. The presence of three or more of these new structures (IGH) would characterize a PAK, with a 91.4% sensitivity, 71.4% specificity and positive predictive value of 89.8%. The IGH’s histological substrate would correspond to the umbrella pattern described by Pinkus.4

Dermoscopy has particular limitations in incipient pigmented lesions of the face, and confocal microscopy becomes a promising test in when considering a greater degree of diagnostic reliability.1

CASE REPORT

A 42-year-old male patient, Fitzpatrick’s type II skin, with light brown hair and eyes, with history of sunburn in childhood and adolescence, family history of cutaneous melanoma (father and paternal grandfather, confirmed by histological examination) and personal background of surgically excised atypical nevi, with histological confirmation. In the fifth assessment follow-up (total body mapping and digital dermoscopy examination due to multiple melanocytic nevi), the patient presented a new pigmented lesion on the upper left lip, measuring roughly 3mm in diameter (Figure 1A). The dermoscopic examination evidenced a lesion with brownish pseudo network with a discrete area containing an annular-granular pattern, asymmetric pigmentation in the follicular openings, and two structures compatible with peripheral IGH (Figure 1B). The diagnostic hypotheses of PAK and LM were considered. A confocal microscopy examination showed the presence of bright dendritic cells in large numbers in the epidermis with absence of buds due to typical epidermal rectification of the face. Also, it evidenced the presence of bright dendritic cells around and in the adnexal openings in the dermal-epidermal junction (DEJ), as well as bright dendritic cells in the epidermal appendages in the papillary dermis (Figure 2).

Based on the clinical and dermoscopic findings, in addition to those obtained from confocal microscopy, the diagnostic hypothesis of LM in a photodamaged area was established. Histological examination (H&E) of the excisional biopsy suggested the presence of atypical melanocytic proliferation in the junctional layer of the epidermis, extending up until the follicular infundibulum. Atypical melanocytes had epithelioid and fusiform cell pattern. The presence of melanin, mitotic figures, and intracytoplasmic melanosomes was observed. The dermal component showed a chronic inflammatory infiltrate. The definitive diagnosis was lentigo maligna melanoma (LMM) with junctional and dermal involvement.

Figure 1: A - Clinical photograph of the pigmented lesion in the left upper lip (red circle). B - Dermoscopy (10x magnification) showing the pseudo-network with follicular asymmetric pigmentation (yellow arrow) and the two IGH structures in the lesion’s periphery (white arrows).

Figure 2: A - Confocal microscopy of the epidermis (3.5x3.0 mm): presence of atypical honeycomb (yellow dotted frame). B - Confocal microscopy / Figure 2A’s zoom area (1.0x2.5 mm): presence of perifollicular dendritic cells. C - Confocal microscopy of the DEJ / Dermis (4.0x4.5mm): presence of dendritic cells in and around the appendages’ openings. D - Confocal microscopy / Figure 2C’s zoom area (0.5x0.5mm): presence of dendritic cells in the follicular ostium (yellow dots) and the presence of dendritic cells resembling palisade in the epidermal adnexum (red dot).
form patterns. The nuclei were hyperchromatic, and sometimes multilobular. The melanocytes were distributed continuously and contiguously. Some cells were arranged perpendicular to the epidermis. The cytoplasm had a variable amount of moderate to intense melanin pigment. There were scarce atypical mitoses in junctional melanocytes. There was no proliferation of melanocytes in the dermis, but only melanophages in the papillary dermis. Some dendritic cell clusters with mitotic figures and atypical multinucleated melanocytes were present at the dermo-epidermal junction and adjacent follicular sheath (Figures 3A and 3B). The immunohistochemical study with double expression of MITF1 / P63 antibodies demonstrated the presence of lentiginous proliferation of atypical melanocytes in the basal layer of the epidermis. The melanocytes' nuclei stain the antibody MITF1 in brown while the keratinocytes' nuclei stain the antibody P63 in red (Figures 3C and 3D).

Fifteen days after the biopsy was carried out, a new confocal microscopy examination was performed aimed at refining the surgical planning (to decrease the chance of local recurrence and achieve better esthetical results), and evidenced the persistence of the lesion in one of the scar's margins (Figure 4).

DISCUSSION

Confocal microscopy is useful in the early diagnosis of LM and LMM. Although these entities are cellularly different in nature from PAK, differential diagnosis between them is sometimes difficult, from both the clinical and the dermoscopic points of view. In this context, confocal microscopy can be of great help in the diagnostic definition.5

The finding related to bright dendritic cells invading the follicular ostium strongly suggests the diagnosis of LM and corresponds to the asymmetrically pigmented follicular openings observed in dermoscopy. Although these cells located within the follicle can be mistaken with the basal cell carcinoma's palisade in the dermoepidermal junction, images of the stratum spinosum are key to the diagnosis with the presence of large numbers of dendritic cells.

The dermoscopic findings of the IGH structures can be explained by the presence of small bright cells that do not invade the follicles at the lesion's periphery, which in tandem with the atypical honeycomb arrangement, and the presence of photodamage, suggest the diagnosis of PAK. According to the histopathology described by Pinkus, non-invasion by tumor cells of the perifollicular epithelium in actinic keratosis is a possible correlation model with dermoscopy and with confocal microscopy, and may be an supporting factor in the differential diagnosis with LM.4

In this clinical case, confocal microscopy was important not only for elucidating the clinical diagnosis – which was confirmed by histology – but also for being significantly useful in the surgical planning, identifying the lesion's presence in the margins of the excisional biopsy.
REFERENCES


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Innovative technique for the treatment of cervical laxity with mono nylon thread for trans-mastoid support (TMS)

Técnica inovadora para tratamento de flacidez cervical com fio mononylon para sustentação transmastoide

DOI: http://www.dx.doi.org/10.5935/scd1984-8773.20181011084

ABSTRACT

There are many procedures for the rejuvenation of the cervical region. Nevertheless, the most invasive ones – such as the rhytidectomy – can lead to relatively large unaesthetic scars. In contrast, noninvasive procedures – such as the classic absorbable lifting threads – have no significant effect on the treatment of the most evident platysmal bands. Aiming at solving the problem of short durability linked to the lifting threads and eliminating the undesirable submental scar caused by rhytidoplasty, a less invasive technique is described for the treatment of cervical laxity. This technique can be used isolatedly in patients with small amounts of skin redundancy, or associated to mini face lifts in patients with considerable sagginess.

Keywords: Cosmetic techniques; Dermatologic surgical procedures; Esthetics; Lifting; Mandible; Neck muscles; Rejuvenation; Rhytidoplasty

INTRODUCTION

Bone resorption, combined with sagging, determines the unsightly appearance of the cervical region.\textsuperscript{1,2} Over time, there is loss of the mandibular contour and accumulation of submentonian fat, with a consequent increase in the cervical-mentonian angle, muscular hypotonia, prominence of platysmal bands, ptosis of the submandibular glands, and changes in texture and pigmentation due to photoexposure.\textsuperscript{1,2} There are several classification systems aimed at quantifying the severity of cervical aging, including the McKinney’s (Table 1).\textsuperscript{3}

Although cervical lifting techniques have gone through remarkable advance, the stigma caused by visible scars or the appearance of “stretched face”, causes many patients to seek alternative techniques to rhytidectomy.\textsuperscript{4,5}

Minilifting and its variations are considered very effective treatments, nevertheless very invasive. Most recently, suspension threads have popularized the treatment of this region.\textsuperscript{5,6} Polylactic acid or polydioxanone support threads are generally absorbable and are pledged to cause a skin lift effect, and promote collagen stimu-
Table 1: McKinney classification for cervical aging

<table>
<thead>
<tr>
<th>Grade</th>
<th>Clinical Findings</th>
<th>Treatment usually indicated</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Barely visible platysmal bands, minimal sagginess</td>
<td>Lifting of the superficial musculoaponeurotic system (SMAS)</td>
</tr>
<tr>
<td>II</td>
<td>Moderately visible platysmal bands</td>
<td>Plication of the platysmal bands in the mid-cervical line</td>
</tr>
<tr>
<td>III</td>
<td>Platysmal bands very visible, sagginess</td>
<td>Plication of the bands in the medial-platysmal line, and resection of redundant skin</td>
</tr>
<tr>
<td>IV</td>
<td>Very visible platysmal bands and excess sagging</td>
<td>Plication of the medial-platysmal line and lateral traction of the SMAS</td>
</tr>
</tbody>
</table>

In order to solve the problem of the little durability linked to the suspension threads and the undesirable submental scar caused by rhytidectomy, one of the authors of the present paper has developed a technique for treating cervical laxity, aimed at providing greater durability with minimal visible scarring.

The objective of the present study is to describe a minimally invasive technique for treating cervical laxity, termed transmastoid support (TMS) by the authors.

**METHOD**

Lean female patients with prominent platysmal bands and loss of mandible angle definition (McKinney grade II and III) are the ones that benefit most from this technique. Patients with a large accumulation of submental fat may require extensive liposuction of the submental area, and if there is a large amount of skin sagginess (of McKinney grade IV), association with mini lifting should be considered (Figures 1 and 2).

After the cleansing of the skin, surgical marking is performed with the patient in sitting position, and the head rectified in an anatomical position so that the central point of the cervical-mental angle, as well as the area to be liposorbed, can be identified and marked (Figure 3). Next, injection of tumescent anesthetic solution with 0.5% lidocaine and 1/100,000 adrenaline is performed.

**Surgical technique description**

1) Incision with n. 11 blade, enough for the passage of the 2mm liposuction cannula, bilaterally in the retroauricular region, in the mastoid bone's topography
2) Incision with n. 11 blade in the submental fold for the passage of the cannula
3) Tumescent liposuction is performed in the submental region according to each patient's need. For patients who do not
have significant accumulation of fat in this region, liposuction is aimed at effecting a blunt dissection, partially detaching the skin, facilitating the passage of the thread.

4) The retroauricular excisions are enlarged up until the mastoid bone’s periosteum, bilaterally

5) Two 3.0 mononylon sutures are respectively attached to the mastoid bone’s periosteum on each side (Figure 4)

6) After the fixation of the thread in the periosteum, the circular needle is discarded. The free end of the thread is passed through a small hole in the Casagrande needle, which is then introduced by the incision in the retroauricular region, passing along the mandibular contour, in the direction of the cervical-mental angle. A small incision performed with a n. 11 blade in the center of this angle is sufficient to allow exiting with a Casagrande® needle and the suture. The process is repeated on the contralateral side (Figure 5).

7) The threads meet in the ventral region of the neck, in the incision carried out in the center of the cervical-mental angle. A surgical knot is performed in order to join and pull the two ends of the thread up until the point the desired retraction of the platysmal bands is achieved. Next, the tie/knot is placed into the incision so that it does not remain visible (Figure 6).

8) Surgical closure of skin incisions. Some patients with a great volume of sagginess will experience skin redundancy in the pre-auricular region after the threads have been tractionned. This should be corrected with the assistance of miniliftings, which will position the scars in the pre and post-auricular regions.

RESULTS

The outcome can be seen immediately after the procedure. In the case described, it was necessary to remove the redundant skin in the pre-auricular region after the threads were tractionned (Figures 7 and 8). The obtained effect was long lasting, with great improvement of cervical sagginess and devoid of

![Figure 4: Mononylon thread anchorage. Fixation of the suture in the mastoid bone's periosteum. Notice that the thread has already been passed on the contralateral side and lies with its end on the anterior portion of the neck.](image)

![Figure 5: Passing of the thread. The Casagrande® needle functions as a guide, the thread is on the needle's tip. The other end is already attached to the mastoid.](image)

![Figure 6: Immediate postoperative. Immediate postoperative, with the suture located where the Casagrande® needle passed the thread, and the suture in the submentonian fold, where the liposuction cannula has passed.](image)

![Figure 7: Late postoperative (1 year). Late post-operative 12 months after the procedure. Notice the improvement of the platysmal bands, which have practically disappeared, and the improved contour of the mandibular border.](image)
complications. In the postoperative period it was already possible to notice improvement in the neck’s and mandible’s contours (Figure 9).

DISCUSSION

A younger silhouette is characterized by well-defined cervical-mandibular and mandible branch contours, cervical-mental angle of roughly 90°, little local sagging and adiposity.

Knowledge of the anatomy allows greater precision in the treatment of the senile neck, reducing the incidence of postoperative complications. Beneath of the dermis are the subcutaneous fat and the superficial cervical fascia, which is intimately connected with the platysma muscle to the SMAS. The TMS technique is carried out in the plane located between the superficial cervical fascia and the dermis, with the suction of the submental fat and subcutaneous fat from the lateral aspect of the neck, with the subsequent pass of the support thread in the described plane.

The facial nerve mandible’s marginal branch deserves close attention. It is located beneath the platysma, along the mandible’s body (in 80% of cases) – or 2cm below – surfacing on the anterior border of the masseter muscle. If injured, paralysis of the depressor muscles of the angle of the mouth and lower lip may occur, causing asymmetry during the smile.10,11

For patients affected by micrognathism, surgical mentoplasty or filling procedures may be associated with the TMS technique. Exaggeratedly prominent platysmal bands and redundant skin (classified as McKinney degree IV) may require associated minilifting. Special attention should be given to extremely thin women, in whom the lift effect caused by the TMS technique may anteriorize and project the thyroid’s cartilage, lending a masculinized appearance to the neck.2,12

Several types of thread have already been used in cervical rejuvenation, among them, polypropylene, spiculated polypropylene, spiculated mononylon among others. Giampapa and Di Bernardo were pre-cursors in the use of threads for cervical support as an alternative to traditional rhytidectomy.13 Later on, the use of spiculated polypropylene threads became popular (with a technique known as “Russian thread”).9 More recently, a new technique was published employing a colorless modified spiculated 2.0 mononylon, specifically developed for the cervical lift, being marketed under the commercial name I-lift ®. It is introduced with an 18G epidural 18cm needle, and may be an alternative in cervical treatment.14

CONCLUSION

The authors deem the TMS technique as a good option due to the fact it does not requiring a specific thread, being car-
Carried out with a simple 3.0 mononylon suture thread. In addition to its low cost, it is easy to perform, and does not cause extensive submental scars.

The technique can directly address the main factors involved in cervical aging: the accumulation of submental fat and the sagging of the platysma muscle.

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Purse string-suture combined with second intention healing for temporal region repair

Sutura purse-string combinada com cicatrização por segunda intenção para reparo de defeito cirúrgico temporal

DOI: http://www.dx.doi.org/10.5935/scd1984-8773.20181011100

ABSTRACT
The purse-string suture is one of the several repair options in dermatologic surgery. Its main indications are round or oval wounds on the trunk and limbs.

The authors describe the combination of purse string suture and second intention healing for a surgical defect after removal of a basal cell carcinoma on the temple.

The purse-string suture is an important closure method that may yield good results in selected cases. Although it is more often indicated on the trunk and limbs, it may be performed on the temple to avoid more complex repair.

Keywords: Suture techniques; Wound closure techniques; Wounds and injuries

INTRODUCTION
The purse-string suture is one of several surgical defect repair techniques used in dermatologic surgery. It is considered a primary closure method, which can be used for complete or partial closure of round or oval wounds.1,2 Since it was described roughly 60 years ago, the technique has gone through variations over time, and can be combined with other methods of reconstruction.1,4 The authors report the use of the purse-string suture associated with second intention healing for repairing surgical defects in the temporal region.

CASE REPORT
A 72-year-old male patient presented with a well-defined nodular basal cell carcinoma in the right temporal region measuring 2 x 1.5 cm (Figure 1). The patient underwent excision with 5mm margins, which lead to an operative wound of 2.7 x 2.5cm, above the superficial temporal fascia (Figures 2 and 3). Other repair options, such as a flap, a graft and partial linear closure were considered; the authors, however, decided for par-
Purse-string suture for temporal repair

Partial closure with intradermal *purse-string* aimed at preventing and minimizing the possibility of injury to the temporal nerve and optimizing the healing time (as compared to isolated healing by secondary intention). The suture was performed with the insertion of the needle horizontally in the dermal plane, advancing 5-10 mm with each pass, and with needle exiting towards the center of the wound. The needle was then consecutively reinserted at a point approximately 2-5 mm distant from the exit site, up until the entire circumference of the wound was covered by the suture (Figure 4). Finally, the thread was tractionned and tied, with the knot remaining buried inside the dermis (Figure 5). The suture used was 4-0 polyglecaprone 25, and after the *purse-string* was carried out, the defect size was reduced to 1.4 x 1.1 cm (78% reduction) (Figure 6). No detachment of the borders was performed, and the remaining surgical defect was left to heal by second intention (Figure 7), with complete closure having been achieved after 4 weeks. Figure 8 shows the 8th week follow-up, with resolution of the induced wrinkling, without damage to the temporal nerve.
DISCUSSION

The purse-string suture consists of a uniform movement of the entire edge of the cutaneous wound towards the center (centripetal direction). It can be performed by introducing the thread horizontally into the dermis (intradermal purse-string) or vertically, crossing all skin layers (cuticular purse-string). The main indications for the technique are oval or round shaped wounds located on the trunk and limbs; nevertheless, it may be used in selected cases of surgical defects located on the face, especially in areas with potential for good healing by second intention, such as the temporal region. The purse-string suture can be performed isolatedly for the complete closure of small or medium-sized wounds, as well as for partial closure of larger wounds, leaving the remaining defect to heal by second intention. In addition, it can be performed in association with other repair techniques, such as grafts or linear closures. It is a quick and straightforward procedure that optimizes healing time by significantly reducing the wound’s size. In addition, it aids in hemostasis and spares adjacent viable tissue (dog ears).

Ideal patients are the elderly, with loose skin, photodamage or when it is convenient to avoid complex reconstructions due to greater surgical risk or better cancer follow-up. The main disadvantage is the initial rough appearance of the scar, as well as the patient’s concern about dealing with an open wound. In this manner, patient orientation is key. Studies have shown that long-term aesthetic and functional outcomes of purse-string suture are similar or even better than other repairing methods for trunk and limb wounds, which tend to widen even when linear suture is performed in planes. The choice of undermining or not the edges of the wound before the purse-string suture is controversial. Some authors do not recommend the detachment, justifying that this minimizes the procedure’s morbidity. Others recommend performing it in order to increase tissue mobility by decreasing the tension of the edges. Most of the times, however, adequate tissue movement can be obtained with little or no undermining. The authors believe that the decision should be individualized.
according to the defect, adjacent skin sagging and proximity of
nearby structures, which may be damaged or distorted (e.g. free
margins, motor nerves). Different suture threads can be used in
the purse-string technique, the most frequently used being ab-
sorbable monofilaments (3-0 or greater).1-3

CONCLUSION
The purse-string suture is a skin repair technique with ac-
ceptable – and in selected cases, surprising – aesthetic outcomes.
Although more appropriate for the trunk and limbs, it may be
indicated for application in the temporal region when avoiding
more complex repair methods is desirable. ●

REFERENCES
1. Cohen PR, Martinelli PT, Schulze KE, Nelson BR. The purse-string su-
ture revised: a useful technique for the closure of cutaneous surgical
2. Cohen PR, Martinelli PT, Schulze KE, Nelson BR. The cuticular purse
string suture: a modified purse string suture for the partial closure of
3. Weisberg NK, Greenbaum SS. Revisiting the purse-string closure: some
4. Hughes MP, Kalajian AH, Brown TS. Purse-string-assisted full-thickness
skin graft: an underutilized technique to reduce graft size and improve
5. Brady JG, Grande DJ, Katz AE. The purse-string suture in facial recon-
6. Ciatti S, Greenbaum SS. Modified purse-string closure for reconstruc-
7. Yuen JC. Versatility of the subcuticular purse-string suture in wound
8. Dzubow LM. Patient contribution to reconstructive Decision: The
purse-string closure. JAMA Dermatol. 2015;151(10):1142-3

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Reconstruction of extensive lesion in the ear with a "saloon door" flap

Reconstrução de extensa lesão de orelha com retalho em “porta de Saloon”

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ABSTRACT
Basal cell carcinoma (BCC), also known as basal cell epithelioma, is the most frequent epithelial neoplasm in the dermatological practice, being more common in men. The authors report the case of a 75-year-old female patient with a BCC in the concha, anti-helix and triangular fossa regions of the right ear. The lesion was completely excised, including the cartilage. The surgical defect was repaired by means of a “saloon door” flap, which yields good aesthetic and functional outcomes. The posterior auricular flap is a versatile option for partial reconstruction of defects in the ear.

Keywords: Carcinoma, basal cell; Skin neoplasms; Surgical flaps

INTRODUCTION
Basal cell carcinoma (BCC), also known as basal cell epithelioma, is the most frequent epithelial neoplasm in Brazil, being more common in men. Its incidence increases in higher age groups, and it is estimated that in the last 30 years it has increased from 20% to 80%. The mean age at diagnosis is 68.1

Masson introduced the retroauricular island flap – also known as “saloon door” flap, revolving door flap, or flip flop flap – in 1972. Several authors later on modified the technique, and the indication for its use was extended for extensive auricular defects.

CASE REPORT
A 75-year-old female patient sought care describing the emergence of a lesion in the right ear’s concha roughly one year before. Physical examination showed an erythematous, infiltrated plaque in the central portion adhered to the cartilage. Dermoscopic examination revealed arboriform vessels and hypopigmic leaflet-like structures (Figure 1). A previous biopsy indicated the presence of BCC, nodular subtype. During surgery planning the lesion was marked using dermoscopy, with a 5mm margin. The flap was also marked in the retroauricular region. Local infiltrative anesthesia was used. The exeresis of the lesion and underlying cartilage were performed, followed by the cre-
Reconstruction of extensive lesion in the ear

it is necessary to attempt to preserve facial aesthetics. Alternatives to the reconstruction of the defects are: second intention closure, skin grafts, and wedge excision – all of which entail the reduction of the auricular height.

Some studies have shown that when dermoscopy is used for demarcating the margins, there is a high rate of complete excision of the BCC (95%-98.5%). Thus, it is critically important to demarcate the lesion’s margins via dermoscopy when micrographic Mohs surgery is not available.

The posterior auricular flap is a versatile option for the partial reconstruction of the ear defect. Planning, choosing an appropriate and individualized technique depending on the type of tumor, the location of the lesion and conditions of the patient are key to achieving a good aesthetic outcome.

DISCUSSION

To reconstruct partial defects of the ear while preserving decreasing its size and changing its natural contour and shape has always been a challenge. In this location, in addition to the cure, it is necessary to attempt to preserve facial aesthetics. Alternatives to the reconstruction of the defects are: second intention closure, skin grafts, and wedge excision – all of which entail the reduction of the auricular height.

Some studies have shown that when dermoscopy is used for demarcating the margins, there is a high rate of complete excision of the BCC (95%-98.5%). Thus, it is critically important to demarcate the lesion’s margins via dermoscopy when micrographic Mohs surgery is not available.

The posterior auricular flap is a versatile option for the partial reconstruction of the ear defect. Planning, choosing an appropriate and individualized technique depending on the type of tumor, the location of the lesion and conditions of the patient are key to achieving a good aesthetic outcome.

FIGURE 1:
A - erythematous infiltrated plaque in the central portion of the auricular pavilion compromising part of the concha, anti-helix, and the whole of the triangular fossa; marking of 5mm margins; B - Dermoscopy evidencing arboriform vessels and hyperchromic leaflet-like structures

FIGURE 2: A - Exeresis of the lesion and underlying cartilage; B - Preparation of the island flap in the retroauricular portion; C - Island flap with the base attached to the mastoid, passing through the defect and into the anterior face of the ear*; D - Flap positioned on the anterior face of the auricular pavilion*

FIGURE 3: A - Flap sutured to the anterior skin, completely closing the anterior face of the auricular pavilion; B - Primary closure of the posterior defect by suturing the posterior skin of the ear to the skin of the mastoid

FIGURE 4: Final outcome after 15 days
REFERENCES


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The importance of the early use of hyaluronidase in the treatment of arterial occlusion resulting from hyaluronic acid based cutaneous filling

A importância do uso precoce de hialuronidase no tratamento de oclusão arterial por preenchimento de ácido hialurônico

DOI: http://www.dx.doi.org/10.5935/scd1984-8773.20181011110

ABSTRACT

Cutaneous ischemia, caused by arterial embolism, is one of the vascular adverse events resulting from the application of hyaluronic acid. Early application of hyaluronidase will lead to the successful degradation and reversal of the condition — which might be severe and cause harm to the patient. The authors report cases with suspected arterial occlusion in which degradation was performed with hyaluronidase at different timepoints. This analysis has evidenced that the degradation time and the procedure’s effectiveness are correlated to the intervention timepoint.

Keywords: Dermatology; Ischemia; Reperfusion

RESUMO

A isquemia cutânea é um dos eventos adversos vasculares decorrentes da aplicação de ácido hialurônico, causada por embolia arterial. A aplicação precoce de hialuronidase representa o sucesso da degradação e reversão do quadro que pode ser grave e causar danos ao paciente. Relatam-se casos com suspeita de oclusão arterial no qual foi realizada degradação com hialuronidase em diferentes períodos. Tal análise permitiu observar que o tempo de degradação e a eficácia do procedimento relacionam-se com tempo de intervenção.

Palavras-chave: Dermatologia; Isquemia; Reperfusão

INTRODUCTION

Cutaneous ischemia is one of the most serious and undesirable complications following application of hyaluronic acid in aesthetic rejuvenation and facial volumization procedures. It is triggered by arterial embolism caused by hyaluronic acid or occlusion caused by compression, often with immediate manifestations. Its diagnosis is clinical. Changes range from livedo reticularis, erythema to pallor and, more rarely, necrosis. The use of hyaluronidase in the right concentration and early application to treat occlusion will most certainly lead to the successful degradation and reestablishment of local blood flow.¹

This adverse effect constitutes a potential aesthetic discomfort and risk of irreversible damage to the patient if early diagnosis and early degradation are not carried out. The authors of the present paper report two cases of ischemia after use of hyaluronic acid in aesthetic procedures, reversed with hyaluronidase at different time points, for the reversion of the picture, demonstrating that early degradation led to a considerably faster resolution of the picture.

Indications and contraindications for the use of hyaluronidase

The indications listed by the US Food and Drugs Administration (FDA) for the use of hyaluronidase are classified into three situa-
tions: 1) facilitation of absorption and dispersion of other injectable drugs; 2) aid in the infusion of subcutaneous fluids; and 3) use in subcutaneous urography. In dermatology, this drug has been indicated for hair transplant and for tumescent liposuction. More recently, it has also been used in cutaneous filling procedures followed by ischemia, aiming at reducing the time of tissue revascularization and aiding in the healing process, leading to a favorable prognosis. Side effects of hyaluronidase use have a low incidence – among them is pruritus due to local application, described by Sopakar et al. in only two patients within a sample of 100 individuals. Another side effect is related to home-made bovine origin hyaluronidase, associated with the occurrence of spongiform encephalopathy.

CASE REPORT

CASE 1

MBF, a 27-year-old female patient, sought medical attention due to dissatisfaction with the aesthetics of her nasal region, even though she had previously undergone rhinoplasty. After evaluation, 0.8 ml of hyaluronic acid (Emervel Deep®, Galderma, Otten, Switzerland) was applied in the columella and nasal tip region with a needle, aspirating before the injection, with no alteration being observed at the moment of the procedure. The patient returned to the practice 36 hours after, with intense local pain, pallor interspersed with livido reticularis areas, compatible with arterial occlusion. Hyaluronidase Biometil® (Laboratório Biometil, São Bento do Sul, SC, Brazil) was used for reverting the picture in a single 1,600 IU application, associated with the following oral drugs: 100mg/day Aspirin® (Bayer, Barmen, Germany), 100mg 12/12 hours cilostazol (Eurofarma, São Paulo, Brazil), 40 mg/day prednisone (Eurofarma, São Paulo, Brazil), 20 mg/day rivaroxaban (Bayer, Barmen, Germany), 500mg 12/12 hours ciprofloxacin (Eurofarma, São Paulo, Brazil), and 300mg 12/12 hours clindamycin (Teuto-Brasileiro S/A, Anápolis, GO, Brazil), for 15 days. Doppler ultrasonography of the site was performed, showing normal arterial and venous blood flow. Three hyperbaric chamber sessions were carried out in the first three days, and warm compresses were applied several times a day for 7 days. The patient evolved with progressive improvement and complete resolution of the condition after 3 weeks.

CASE 2

MSM, a 31-year-old female patient, sought medical attention aimed at undergoing facial harmonization treatment. Hyaluronic acid was applied to several sites of the face, among them in the deep proximal nasogenian sulcus (piriform fossa) – 0.1ml Emervel Deep® (Galderma, Otten, Switzerland), with a needle positioned at 90º and aspiration prior to the injection. An erythematous-purplish area emerged instantly, extending from the left lateral region of the nasal wing to the medial region of the ipsilateral nasogenian sulcus. In face of the persistence of the picture and hypothesis of arterial occlusion for 12 hours, the authors decided for the early degradation using 400 IU of hyaluronidase (Biometil®) and 100 mg/day sildenafil, 40 mg/day Clexane (Sanofi–Aventis Farmacêutica Ltda, Suzano, São Paulo, Brazil), 40 mg/day prednisone (Eurofarma, São Paulo, Brazil), 500mg 12/12 hours ciprofloxacin (Eurofarma, São Paulo, Brazil), and 500mg 12/12 hours clarithromycin (Eurofarma, São Paulo, Brazil) for 15 days. Doppler ultrasonography of the site was also performed, showing normal blood flow. Improvement
in the coloration was observed 2 days after the degradation, with complete resolution after 4 days.

DISCUSSION

With the increased numbers of hyaluronic acid based esthetic procedures, knowledge of the possible adverse reactions and respective handling is necessary. The pathophysiology of the embolic process caused by these procedures has not yet been elucidated, nevertheless the decrease in blood flow may be linked to the volume injected and the site of injection of high viscosity fillers. In both cases reported, alterations in the blood flow resulting from application of hyaluronic acid were observed, with typical manifestations at different time points. The chronology of the dermatological manifestations of embolic processes usually follows the sequence bleeding, emergence of vesicles, bedsores and necrosis, meaning it is crucial for the practitioner to recognize them early. The use of hyaluronidase is recommended for the degradation of the material, which leads to the normalization of the blood flow and prevents sequelae. The supportive therapy is based on increased perfusion, decreased inflammatory process and prophylaxis of associated infections, and may rely on vasodilators, corticosteroids, antimicrobials and antiaggregants.

Comparing the cases reported and observing their development in Figures 1 and 2, it is possible to observe that the time for degrading hyaluronic acid had a significant impact on the prognosis and normalization of the local blood flow. In addition, it was possible to see that hyaluronidase only degrades injectable hyaluronic acid and does not interfere with what exists in the body, given that a large amount was used in Case 1 with absence of alterations whatsoever in the patient’s previous physiognomy.

REFERENCES


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Treatment of psoriasis vulgaris with cyclosporine and methotrexate injections using the MMP® technique

Tratamento de psoríase vulgar pela microinfusão de medicamentos na pele (MMP®) usando ciclosporina e metotrexato

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ABSTRACT

Systemic oral medications – such as cyclosporine (CYA) or methotrexate (MTX) – for the treatment of psoriasis have limited bioavailability due to incomplete gastrointestinal absorption and first-pass hepatic metabolism. Moreover, they are associated with adverse effects. The application of CYA or MTX using the microinfusion of drugs into the skin method (MMP®) for the treatment of psoriasis vulgaris yielded a therapeutic response with significant reduction of lesions, and absence of side effects. In the present report, the authors describe 4 cases of psoriasis vulgaris treated using the MMP® method, with the application of CYA or MTX.

Keywords: Cyclosporine; Methotrexate; Psoriasis

INTRODUCTION

Psoriasis is a chronic inflammatory and recurrent skin disease with a worldwide prevalence of approximately 2%. It is associated with a high degree of morbidity and impact on quality of life and the most common clinical presentation is vulgar psoriasis, which represents 85% to 90% of cases.1-3 Due to its diverse clinical presentations, treatment should be individualized, taking into account the location of the lesions, the impact on the patient’s quality of life, and the coexistence of psoriatic arthritis.

Three modalities are mainly used, isolatedly or in combination: 1) topical agents, 2) phototherapy and 3) systemic medications. Both phototherapy and systemic medications are used in moderate to severe cases, which are defined as those with 10% or more of involvement of the body area (BSA Index - Body Surface Area); or a score equal or greater than 10 points in the Psoriasis Area Severity Index (PASI) or in the Dermatology Quality of Life Index (DQLI). The most commonly used systemic medications are methotrexate (MTX), acitretin and cyclosporine (CYA).4,5

Methotrexate is a teratogenic substance used orally, subcutaneously or intramuscularly. In addition to being associated...
with gastrointestinal intolerance, it presents a risk of myelosuppression and hepatotoxicity. Its bioavailability is limited by oral route due to incomplete absorption by the gastrointestinal tract (GIT) in addition to the hepatic first-pass metabolism, releasing only small concentrations of the drug in the systemic circulation despite the ingestion of high doses. The reduced folate carrier 1 (RFC1), a ubiquitous transmembrane carrier protein, may limit oral absorption of MTX when saturated, which occurs from 15mg MTX.7,8

Cyclosporine, a calcineurin inhibitor available for oral or intravenous use, has a rapid response as a treatment for psoriasis, however it can be nephrotoxic and increase blood pressure levels, in addition to being associated with side effects such as hydroelectrolytic disorders, hyperlipidemia, among others. Its bioavailability is also limited in the oral form due to low absorption and hepatic first-pass metabolism.9-14

In order to maintain and possibly improve therapeutic efficacy and at the same time decrease the side effects of systemic treatments, the authors propose the treatment of patients bearers of psoriasis vulgaris via the microinfusion of drugs through the skin (MMP®), which was described by Arbache15,16 with the use of MTX (25mg/ml sterile vial) (Thevametho®, Pharmachemie BV, Haarlem, The Netherlands) (imported by Teva Farmacêutica Ltda., São Paulo, Brazil) and CYA (50 mg/ml sterile vial) (Sandimmun®, Novartis, São Paulo, Brazil).

CASE REPORTS

Case 1: Thirty-eight-year-old male patient, frequent consumption of alcoholic drink, bearer of psoriasis vulgaris with lesions in the upper limbs and trunk for nine years. The patient reported previous use of acitretin and MTX, which were suspended due to side effects. The patient received 4 applications of MMP® with 12.5 mg/ml CYA in the lesions located in the right forearm, with a two-week interval. Serum cyclosporinemia was not detectable at 8 hours after the intervention, and laboratory tests and blood pressure levels were kept within normal range. There was no complaint of adverse effects by the patient. All the lesions, both those treated on the right forearm and those not treated on the left arm and on the back, receded (Figure 1).

Case 2: Twenty-seven-year-old male patient, bearer of psoriasis plaques for 5 years. Lesions located in the anterior and posterior regions of the right leg were resistant to topical therapy. The patient underwent 3 MMP® sessions with 25 mg/ml MTX distributed among the lesions. There was a decrease in desquamation and thickness, and remission of pruritus 2 weeks after the first application (Figure 2). After a series of 3 applications every 2 weeks, there was complete remission of the lesions. No methotrexate serum levels were detected the day after the application, nor there were complaints of side effects.

Case 3: Fifty-eight-year-old male patient, hypertensive, history of stroke with left hemiparesis for 12 years, under continuous use of captopril, aspirin, atenolol and amiodipine. Bearer of psoriasis in localized plaques for 17 years, resistant to topical treatment. The patient underwent 5 MMP® sessions with 12.5 mg/ml CYA every 2 weeks in the lesion located in the dorsum, with improvement of approximately 90% of the lesion (Figure 3). The patient had good tolerance to the procedure, without need for topical analgesia. Serum cyclosporinemia after 6 hours was undetectable. Laboratory tests and blood pressure levels were unchanged. The patient is still under treatment.

Case 4: Twenty-two-year-old male patient, with plaques of psoriasis for 2 years, having previously improved with oral MTX, having nevertheless not adhered to the treatment, experiencing recurrence of the lesions. Initial PASI: 10.4. An application of MMP® with MTX was performed in the lesions located on the dorsum and left arm (Figure 4). After 1 week, the lesions became less thick and scaly, and only macules were observed at the site of the treated lesions after 2 weeks, improving the appearance of the lesions when looked at from distance (Figures 5 and 6). The patient remains under treatment.

The patients were duly informed about the treatment and signed a Free and Informed Form of Consent.
DISCUSSION

The treatment of moderate to severe psoriasis involves drugs that might lead to possible side effects. Some studies have investigated the possibility of using topical MTX as an alternative, which would reduce the occurrence of such effects. Due to the fact it is a water-soluble molecule, it has limited ability to permeate the stratum corneum, and its use in untouched skin is ineffective. Therefore, techniques such as electroporation, iontophoresis and ablative lasers were used aimed at increasing its permeation through the skin, with therapeutic response in the treatment of psoriasis.

Similarly, studies on topical use of CYA suggested ineffectiveness on untouched skin. Griffiths et al. obtained therapeutic response following intralesional injection of 2 ml of 17 mg/ml CYA in psoriasis lesions 3 times per week with undetectable levels of serum cyclosporinemia. Despite the application of considerable amount of medication, due to the CYA's lipophilic characteristic, it would rapidly redistribute through the tissues, becoming undetectable at plasma levels. The main side effect observed in the study was pain caused by the procedure. Using the MMP® technique it was possible to infuse the drug directly in the lesions with excellent patient tolerance after topical anesthesia. The percutaneous injection of the drug has a powerful local effect and is capable of avoiding hepatic first pass metabolism, reaching the systemic circulation in a low and undetectable concentration, nevertheless enough to obtain therapeutic response, meaning that treatment of patients with moderate to severe psoriasis, as well as of lesions resistant to other therapies can be carried out without side effects.

CONCLUSION

Treatment with the MMP® technique using MTX or CYA solution demonstrated good tolerability, lack of adverse effects, rapid response (already noticed within 2 weeks), and effectiveness both in lesions that have been treated isolatedly and in distant lesions that did not undergo the application.
MMP® performed with CYA or MTX can be a novel treatment modality for psoriasis that arise in resistant plaques, in difficult-to-treat locations, and in patients at risk of system-

ic treatment related complications. Further studies are needed aimed at defining protocols and randomized clinical trials in order to determine the effectiveness and efficiency of MMP® in the treatment of psoriasis with MTX and CYA.

REFERENCES

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Laser versus microneedling for the treatment of facial scar – a case report

ABSTRACT

Both microneedling and ablative fractional laser can be used for the treatment of scars. The present article reports the case of a female patient who had the first half of a perioral scar treated with ablative fractional CO2 laser, and the second half treated with microneedling, using a 2mm roller. Outcomes were superior in the half treated with laser.

Keywords: Cicatrix; Collagen; Lasers

INTRODUCTION

Both microneedling and fractional ablative laser are beneficial for the treatment of scars. A number of conditions involving fibrosis, contractures and scarring can be treated with CO2 ablative fractional laser.1 Similarly, microneedling can also be used to stimulate skin remodeling.2 In the present case report, the authors compare the use of CO2 laser with that of microneedling for the treatment of a traumatic scar located in the perioral region.

CASE REPORT

A 35-year-old female patient sought medical care related to a scar in the perioral region, caused in an automobile accident during childhood. The patient had undergone three previous reconstructive plastic surgeries at the site aimed at improving the appearance of the scar, nonetheless was still unsatisfied. Moreover, there were limitations in the mouth opening and smiling movements (Figure 1). The scar was divided into two areas, with one of them (located in the right hemiface) being treated with CO2 ablative fractional laser, and the other (located in the left hemiface) being treated with microneedling. Topical anesthetic (lidocaine hydrochloride – 40 mg/g Dermomax® cream, Biósintética, Guarulhos, SP, Brazil) was applied 30 minutes before the procedure and supplemented with injectable anesthesia containing 1% lidocaine hydrochloride with 1:200,000 epinephrine (Xylestesin®, Cristália Produtos Químicos Farmacêuticos Ltda, São Paulo, SP, Brazil). Fractional CO2 laser (Sculptor, Vydence Medical) was performed with a 120nm tip, first pass: random
mode, 140mJ energy, 75 MTZ/cm² density; second pass: brush mode, 30mJ energy, 200 MTZ/cm² density. Microneedling was conducted with Dr.Roller® needled cylinders (Mooham Enterprise Co. Gyeonggi-do, South Korea / ANVISA 80669600001). Using 2mm long needles, the rolling movement was performed from 10 to 15 times in the four directions so that 250-300 punctures/cm² were inflicted, causing uniform punctate bleeding (Figure 2). A drug delivery formulation containing 4% Hydroxyprolisilane, 5% Omega active, 2% Regestril, 2% Matrixil 3000 and 1.5% IGF fluid anhydrous serum (Dermogral Dispensing Pharmacy, Porto Alegre, RS, Brazil) was applied immediately after the procedure. The selected active principles, such as Hydroxyprolisilane and Matrixil 3000, act in the synthesis of collagen and elastin, while others, such as Regestril and Omega active, inhibit collagenase. Two sessions of the procedure were performed with a 20-day interval between them.

The improvement in scarring was evaluated using the Manchester Scar Scale (scores from 5 to 28, with higher scores indicating worse pictures). The right hand side of the scar, treated with CO2 laser, presented better outcomes (Figures 3A and B). The initial Manchester Scar Scale score for both sides was 13 points (Table 1). After the treatment, the area treated with laser improved by 5 points, while the area treated with microneedling improved by only 1 point (Table 2).

**DISCUSSION**

Both microneedling – also termed percutaneous collagen induction – and ablative fractional lasers are effective therapeutic options for the treatment of scars. In addition, both methods allow drug delivery processes, which optimize topical delivery of active principles.

The remodeling that occurs after the use of CO2 laser is caused by the photomechanical effect, which removes the scle-rotic scar bands containing thickened collagen and releases the contracted tissue through dermal remodeling. In the literature, the use of CO2 laser for the treatment of scars demonstrates the presence of improvement in texture and pigmentation. In the case described in the present report, improvement was observed in texture, pigmentation, contour, brightness and distortion. In addition, studies that have used higher densities led to better outcomes, which justifies the parameters chosen in the present case. Particularly for hypertrophic scars, positive outcomes can be observed in the reduction of scar's firmness, and in the improvement of the surface and contractures.
Likewise, microneedling stimulates the healing cascade, culminating with the formation of collagen type I. This purpose is achieved as long as the needles penetrate from 1–3 mm, in order to reach the dermis. Since only 50–70% of the total length of the needle penetrates during the rolling process, they should be at least 1.5 mm long.² Studies demonstrate that epidermal thickness can be increased by up to 205% by performing serial microneedling sessions. In addition, the dermal connective tissue becomes denser.⁷,⁸ The needle used in the present case (2 mm long) was chosen aiming at inflicting deep injury to the dermis, which is indicated for the effective treatment of scars.² Despite the length of needles was deemed adequate, the authors observed superior outcomes in the area treated with ablative fractional laser.

CONCLUSION

In the present case report, the authors observed superiority of outcomes with the use of CO₂ fractional laser as compared with that of microneedling (2 mm long needles) for treatment of a perioral traumatic scar. Nevertheless, this result requires further studies with a greater number of patients (randomized and blinded), as well as a great number of sessions, in order to ratify the findings. The authors also emphasize the possibility of performing drug delivery in the immediate post-procedure of both therapeutic modalities, provided that appropriate active principles and adequate formulation for the purpose are employed.

REFERENCES


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