Aesthetic Medicine / Volume 3 / Nº 3 / July/September 2017

# aesthetic medicine

## Official Journal of the International Union of Aesthetic Medicine UIME

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International Union of Aesthetic Medicine UIME




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# Contents

# Guidelines for Authors

Aesthetic Medicine is a multidisciplinary Journal with the aim of informing readers about the most important developments in the field of Aesthetic Medicine.

## Submission of manuscripts

All articles in their final version - completed with name, surname, affiliation, address, phone number and e- mail address of the author (s) - must be sent in word format to the Editorial Committee at the following e-mail address: aemj@aestheticmedicinejournal.org. Manuscripts must be written in English, and authors are urged to aim for clarity, brevity, and accuracy of information and language. All manuscripts must include a structured abstract. Authors whose first language is not English should have their manuscripts checked for grammar and stylistic accuracy by a native English speaker.

## Manuscript specifications

**Title page**
The title page should include:
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· The affiliation(s) and address(es) of the author(s)
· The e-mail address, telephone and fax numbers of the corresponding author
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· The authors must disclose any commercial interest that they may have in the subject of study and the source of any financial or material support

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The length of the abstract should be no more than 250 words and should include the following hea- dings: Background, Aim, Methods, Results, Conclusions

**Keywords**
Up to six keywords should be listed and separated by a comma (please, verify keywords on MeSH).

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· The manuscript should be organised in the following sections:
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· Introduction
· Materials and Methods
· Results
· Discussion and Conclusions
· Acknowledgments
· Conflict of interest
· Reference list
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This type of article uses Unstructured Abstract. It must not exceed 1500 words and includes figures and tables (max 6), legends, and up to 30 references.

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## Acknowledgments

The authors declare that they have no conflict of interest.
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Conflicts of Interest need to be explicitly defined before any manuscript can be considered for publication.

## References

References must be cited consecutively in the text as superscript numerals and listed on a separate sheet in numerical order at the end of the text. The references must be cited according to the AMERICAN MEDICAL AS- SOCIATION (AMA) CITATION STYLE.
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**Rev. 11/1/2012**

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| **Newspaper article - online** | Pollack A. FDA approves new cystic fibrosis drug. *New York Times.* January 31, 2012. http://www.nytimes.com/2012/02/01/business/fda-approves-cystic-fibrosis-drug.html?ref=health Accessed February 1, 2012. |
| **Websites** | Outbreak notice: Cholera in Haiti. Centers for Disease Control and Prevention Web site. https://www.cdc.gov Published October 22, 2010. Updated January 9, 2012. Accessed February 1, 2012. |
| **Entire book - in print** | Modlin J, Jenkins P. *Decision Analysis in Planning for a Polio Outbreak in the United States.* San Francisco, CA: Pediatric Academic Societies; 2004. |
| **Book chapter - in print** | Solensky R. Drug allergy: desensitization and treatment of reactions to antibiotics and aspirin. In: Lockey P, ed. *Allergens and Allergen Immunotherapy.* 3rd ed. New York, NY: Marcel Dekker; 2004:585-606. |

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AMA Manual of Style: A Guide for Authors and Editors. 10th ed. Oxford: Oxford UP.

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### Example Article

1. Zoellner J, Krzeski E, Harden S, Cook E, Allen K, Estabrooks PA. Qualitative application of the theory of planned behavior to understand beverage consumption behaviors among adults. J Acad Nutr Diet. 2012;112(11):1774-1784. doi: 10.1016/j.jand.2012.06.368.

### In-Text Citation Example

LARGE INCREASES IN AMERICANS' CONSUMPTION OF sugar-sweetened beverages (SSB) have been a topic of concern. Between 1977 and 2002, the intake of "caloric" beverages doubled in the United States, with most recent data showing that children and adults in the United States consume about 172 and 175 kcal daily, respectively, from SSB,[1] it is estimated that SSB account for about 10% of total energy intake in adults[2,3]. High intake of SSB has....

### References Section Example

**References**

1. Duffey KJ. Popkin BM. Shifts in patterns and consumptions of beverages between 1965 and 2002. *Obesity.* 2007:15(11):2739-2747.
2. Nielsen SJ. Popkin BM. Changes in beverage intake between 1977 and 2001. *Am J Prev Med.* 2004;27(3):205-210.
3. Drewnowski A. Bellisle F. Liquid calories, sugar, and body weight. *Am J Clin Nutr.* 2007;85(3):651-661.

Use commas to separate multiple citation numbers in text, like you see between references 2 and 3. Unpublished works and personal communications should be cited in the text (and not on the reference list).1 Superscript numbers are placed outside periods and commas, and inside colons and semicolons. When citing the same source more than once, give the number of the original reference, then include the page number (in parentheses) where the information was found. See pages 41-44 of the AMA Manual of Style for more information.

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Citing AMA guide website http://libguides.stkate.edu/c.php?g=101857&p. Updated April 2011. Accessed October 24, 2012.

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# Publication Ethics and Publication Malpractice Statement

Aesthetic Medicine undertakes to defend the rules of ethical behavior in every stage of the process by adopting and promoting the standards set by Code of Conduct and Best Practice Guidelines for Journal Editors.

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The editor of a peer-reviewed journal is responsible for deciding which of the articles submitted to the journal should be published. The editor will evaluate manuscripts without regard to the authors' race, gender, sexual orientation, reli- gious belief, ethnic origin, citizenship, or political philosophy. The editor may be guided by the policies of the journal's editorial board and constrained by such legal requirements as shall then be in force regarding libel, copyright infringement and plagiarism.

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An editor should take reasonably responsive measures when ethical complaints have been presented concer- ning a submitted manuscript or published paper, in conjunction with the publisher (or society). Such measures will generally include contacting the author of the manuscript or paper and giving due consideration of the respective complaint or claims made, but may also include further communications to the relevant institutions and research bodies, and if the complaint is upheld, the publication of a correction, retraction, expression of concern, or other note, as may be relevant. Every reported act of unethical publishing behaviour must be looked into, even if it is discovered years after publication.

## Duties of Reviewers

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Peer review assists the editor in making editorial decisions and through the editorial communications with the author may also assist the author in improving the paper. Peer review is an essential component of formal scho- larly communication, and lies at the heart of the scientific endeavour. Aesthetic Medicine shares the view of many that all scholars who wish to contribute to publications have an obligation to do a fair share of reviewing.

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Where there are others who have participated in certain substantive aspects of the research project, they should be acknowledged or listed as contributors. The corresponding author should ensure that all co-authors have seen and approved the final version of the paper and have agreed to its submission for publication.

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All authors should disclose in their manuscript any financial or other substantive conflict of interest that might be construed to influence the results or interpretation of their manuscript. All sources of financial sup- port for the project should be disclosed. Examples of potential conflicts of interest which should be disclosed include employment, consultancies, stock ownership, honoraria, paid expert testimony, patent applications/ registrations, and grants or other funding. Potential conflicts of interest should be disclosed at the earliest stage possible.

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# Editorial

In modern years, aesthetics has become quite important in every aspect of everyday life: following the hundreds of journals, magazines, blogs and websites pointing their attention towards this interesting and fascinating topic, the request for aesthetic medicine has increased manifolds.

Aesthetic Medicine is a new field of medicine, in which different specialists share the aim of constructing and reconstructing the physical equilibrium of the individual. Treatment of physical aesthetic alterations and unaesthetic sequel of illnesses or injuries, together with the prevention of aging, are perhaps two of the most iconic areas of intervention for Aesthetic Medicine.

However, in order to prevent frailty in the elderly, a program of education is similarly important.

Furthermore, the line between health and beauty is extremely thin: psychosomatic disorders resulting from low self-esteem due to aesthetic reasons are frequent and can- not be ignored by a clinician.

It is therefore clear that there is no figure in the field of medicine which is not involved in Aesthetic Medicine: endocrinologists, gynecologists, angiologists, psychologists and psychiatrists, plastic surgeons, dermatologists, dieticians, physiotherapists, orthopedists, physical education instructors, massophysiotherapists, podologists, and rehabilitation therapists are just some of the specialists who are sooner or later going to have to answer their patients' needs for aesthetic interventions.

The involvement of all these specialists fits the description of health as defined by the WHO: "a state of complete physical, mental and social well-being and not merely the absence of disease or infirmity" for which, undeniably, a team of different physicians is required.

The number of patients requiring medical consultation for esthetic reasons is rapidly increasing: in order to be able to provide adequate feedback, medical and paramedical specialists should be trained and, more importantly, should be taught how to work together. Existing Societies of Aesthetic Medicine from different countries share the aim of creating such teams and provide constant updates to the literature: the creation of an international network of specialists from all around the world under the flag of Aesthetic Medicine represents a challenge, but at the same time it is the proof of the widespread interest in this topic.

The first issue of this Journal represents the results of the efforts of the many national Societies and of the Union Internationale de Medecine Esthetique, now together as one; it is our hope that in years to come this Journal might improve our knowledge in this field, and provide adequate scientific advancement in the field of Aesthetic Medicine.

***Francesco Romanelli***
*MD Editor-in-chief*
*Associate Professor at "Sapienza" University of Rome*

# Editors' notes

## Aesthetic Medicine, the booming medical activity

Aesthetic Medicine was born in France 40 years ago.
The French Society of Aesthetic Medicine was the first of its kind in the world, followed by Italy, Belgium and Spain. Starts were rather difficult as aesthetic procedures in those early years were only surgical.
At that time aesthetic doctors and cosmetic dermatologists had very few real medical procedures to offer to their patients for treating aesthetic problems on face and body.
At the beginning of the '80s, viable medical procedures started to emerge in Europe for aesthetic and cosmetic purposes. Mostly, at that time, they were imported from the United States: those included collagen injections for wrinkles (Zyderm by Dr. Stegman), and chemical peels (phenol by Dr. Baker, TCA by Dr. Oba- gi). But, subsequently, European research on Aesthetic Medicine gained momentum. Hyaluronic acid appeared on the market, as it was discovered that it could be used as a dermal filler for wrinkles. During the '90s, the use of lasers offered aesthetic doctors and cosmetic dermatologists new possibilities.
The "beam revolution" started with CO2 laser for facial resurfacing.
Today, CO2 resurfacing is not used as much anymore, because of the long and difficult postop. CO2 laser was replaced with the gentler Nd-YAG and Erbium lasers and more recently with non invasive photonic devices for facial rejuvenation, including IPL, US and radiofrequency. These new technologies allow today's aesthetic doctors and cosmetic dermatologists to offer their patients procedures with low risk of post- op complications. Then, Botulinum Toxin has "invaded" both sides of the Atlantic Ocean.
Today, Botox injections are the most popular treatment for facial expressive wrinkles.
Botox injections are now so common everywhere that many cosmetic surgeons have given up their bistouries for syringes. Last but not least, development in Aesthetic Medicine is shown by mesotherapy and adipolipolysis.
About lipolysis, new data and recent publications have explained that radiofrequency, ultrasounds and cryolyse could have positive action to dissolve fat and to improve some unaesthetic disorders like cellulite.
These non invasive procedures intend to replace the surgical liposculpture with success.
Nowadays, Aesthetic Medicine has the necessary tools to address all major disorders within the aesthetic field. After 40 years, Aesthetic Medicine is now active in 27 countries in the world (France, Italy, Spain, Belgium, Morocco, Poland, Russia, Switzerland, Romania, Kazakhstan, Algeria, Brazil, Argentina, Uruguay, Venezuela, Colombia, Chile, Mexico, U.S.A, Canada, South Korea, and recently Ecuador, China, South Africa, Turkey, Ukraine and Georgia).
All 27 national Societies are members of the Union Internationale de M decine Esth tique (U.I.M.E.). Aesthetic Medicine is taught in 8 countries (France, Italy, Spain, Brazil, Argentina, Mexico, Venezuela, Kazakhstan) in universities that deliver UIME's diplomas after 3 to 4 years of studies.

## What is the future of Aesthetic Medicine?

In the last few decades, patients' desires to look and feel younge, have fueled Aesthetic Medicine and Cosmetic Dermatology: many different procedures have been developed to satisfy the demands.
As life-span have increased, patients today are not only asking about aesthetic procedures, they are also asking for a way to stay in good physical conditions in the last decades of their lives. As a direct result, Anti-Aging Medicine, which covers skin aging and general aging, has recently emerged and expanded very quickly. Anti-Aging Medicine can offer senior patients better nutrition, dietary supplementation with vitamins, minerals, antioxidants, and eventually hormone replacement therapy, but only when needed.
Today, and in the near future, both Aesthetic Medicine and Anti-Aging Medicine will offer to our patients, who now live longer, better wellness with aesthetic treatments for skin aging and anti-aging treatments for general aging. Aesthetic Medicine is booming, but all medical practitioners should be correctly trained, so its future will be bright.

***Jean-Jacques Legrand***
*MD General Secretary of UIME*

## Aesthetic Medicine: a bioethic act

When in 1977 the Italian Society of Aesthetic Medicine published the first issue of the magazine “La Medicina Estetica” Carlo Alberto Bartoletti, the Founder, wrote an editorial in which traced the pathway of the discipline and of the Scientific Society, still valid and projected into the future.
Today from that Editorial Board arise an International Journal, which wants to be indexed, in order to give to the doctors practicing Aestehetic Medicine all around the world a solid basis of shared knowledge.
In the late ‘60s, what was called in Italy Aesthetic Medicine, moved its first steps thanks to “remise en forme and anti aging projects” imported from the experience the “Institutul de geriatrie Bucuresti”, directed by Dr. Ana Aslan.
For this reason,there is the bioethical imperative that the Discipline should be first prevention, then return to physiology and finally correction.
The worldwide diffusion and the efforts of Industries born on the wave of the phenomenon have often led to choose the fastest route to achieve and maintain the physical aspect in the myth of beauty at all costs, without considering that aesthetic is not synonymous of beauty, but it is a balance between body and mind, and the role of the doctor is to take care of the Person globally and not only focusing on the correction of “a badly accepted blemish”.
Faithful to the teaching of my Master had almost 50 years ago, this new journal will have the task of elevating the human resources, aligning and validating methodologies, but above all affirming the humanitas of the medical art in its purest sense to pursue the good and the graceful for the person who relies on it.

***Fulvio Tomaselli, MD***
*Honorary President of the Italian Society of Aesthetic Medicine*

## Aesthetic Medicine needs science. All over the world

All Aesthetic Doctors know that science is the basis for safety. Safety is the most important issue in our discipline.
Unfortunately, Aesthetic Medicine is more often surrounded by marketing than by science, despite the hard work done by Scientific Societies all over the World. And, too often doctors working in this field are dealing with sellers that promote products with insufficient scientific studies.
However, they sell it anyway. I think that doctors must learn that the first thing to ask about a medical device is the scientific background regarding that product: patients treated, follow up period, adverse events and, most of all, publications.
With this new International Journal completely dedicated to Aesthetic Medicine, proposed by the Italian Society of Aesthetic Medicine, endorsed by UIME and shared by all the National Societies of Aesthetic Medicine belonging to UIME, World Aesthetic Medicine wants to stimulate scientific production in this discipline to increase safety and quality in aesthetic medical procedures.
Another important goal of the Journal is to catalyze the proposal of new protocols and guidelines in Aesthetic Medicine, with the consensus of the entire Aesthetic Medicine Scientific Community.
What this Journal should achieve in the near future is to improve the number and quality of scientific production in Aesthetic Medicine, in order to allow this discipline to grow in the field of evidence based medicine, not only in the rationale field.
I hope this can be the start of a new era for Aesthetic Medicine, with the commitment of all Scientific Societies all over the world.

***Emanuele Bartoletti, MD***
*Managing Editor*
*President of the Italian Society of Aesthetic Medicine*

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aesthetic medicine

Review

# Mediterranean Diet as a tool for achieving successful ageing

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**Abstract**
Targeted interventions to slow or postpone ageing and to favour an active life expectancy represent the new perspectives in ageing investigation. Some mechanisms that delay or prevent the onset of ageing disabilities and pathologies have been identified. In general, maintaining a healthy lifestyle seems to reduce many risk factors. In particular, eating habits represent the most concrete and low-cost method to address ageing. Of all dietary habits analysed, the Mediterranean diet has received much attention since it has consistently proven its beneficial influence on health and longevity. The Mediterranean diet is characterized by low glycaemic index and low animal protein intake, it is rich in nutraceuticals and functional foods thus reducing molecular pathways signalling, as nutrient sensitive pathways which affect the ageing process and lead to unhealthy ageing. This dietary model can be promoted as an “anti-ageing therapy” and can contribute to good health status and therefore to a better quality of life.

**Keywords**
Ageing, functional food, hormesis, longevity, mediterranean diet, nutraceuticals

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## Introduction: healthy foods for healthy ageing

Healthy ageing as well as the occurrence of age-related disabilities and diseases result from several degrees of interaction between genetic, epigenetic and environmental factors[1]. To reach an active and independent end of life and to achieve a healthy life-span, the so called *health-span*, an integrative approach is needed[2]. With the aim of developing preventive and therapeutic measures, it is important to start managing daily habits. Nutrition is probably the most important one[3,4]. Our life, in different ways, turns around food. It can be a positive or negative relationship. It can be a reason for travelling or for working but no one can live without it.

Since ageing is an ineluctable process, the goal is to live longer but in good health rather than just delaying the end of life.

The Mediterranean Diet (MedDiet) is a cultural tradition that contributes to better health and quality of life in Mediterranean countries, where many longevous people exist. In fact, the Elderly Prospective Cohort Study identified a reduced overall mortality among old people who live in the *Mediterranean way*[5,6].

For many years, interventions to slow the rate of ageing and increase life-span have been tested by many scientists. They have tried and succeeded in modulating the nutrient-sensing pathways (NSPs) and in revealing new and interesting research suggestions in the field of anti-ageing medicine[2]. In both animal models and humans dietary intervention can decrease the chronic, generalized pro-inflammatory status, the inflammageing, positively modulating the ageing process[7-9]. The complex relationship between nutrition, the ageing process and healthy ageing is not completely understood. Specific dietary changes however, as recommended by NU-AGE project, such as the reduction or elimination of trans and saturated fats and an increased intake of omega-3 fatty acids, vitamins, minerals and antioxidants can help to minimize inflammation[10].

Similarly, appropriate intake of functional foods may confer health benefits, influencing the maintenance of immune homeostasis and contributing, directly, to the reduction of inflammation and metabolic disorders related to an inadequate diet[11,12].

In this review we evaluate the relationship between adherence to the MedDiet, rich in bioactive compounds, and healthy ageing, underlying its activating or inhibitory role in cellular pathways and the anti-ageing effects of its functional foods.

## Ageing and longevity

Ageing is a complex, unavoidable phenomenon or trait than cannot be exhaustively defined.

In May 2012, a group of scientists and clinicians met in Athens to consider the relevance of ageing and longevity. The workshop led to the creation of a statement to highlight the importance of a common view related to these processes, since they represent phenotypes that are rapidly spreading worldwide.

The panel defined ageing as a process which makes people more vulnerable, leading to death but in a distinct moment of the chronological age (CA) as compared to the biological age (BA)[13].

The United Nations define “elderly” all individuals from the age of 60 years old[14]. Of course it can be considered only a necessary threshold in order to classify the population.

We are however interested in how long people live and remain engaged and physically active so the simplistic classification, based on the CA, is only theoretical and not very useful. CA refers to the date of birth, while BA is a personal feature that depends on how old a person appears to be. These two parameters often do not match. The BA is a dynamic concept, based on the biological health status of tissues, organs and systems[15].

By definition there are two ways to become old, i.e. with success (successful ageing) and without success (unsuccessful or pathological ageing). The ratio between CA and BA is greater than or equal to 1 in successful ageing and less than 1 in unsuccessful ageing.

Unsuccessfull ageing is manifested by people who develop one or more age-related diseases, such as neurodegenerative (Alzheimer’s or Parkinson’s) diseases, metabolic (metabolic syndrome and type 2 diabetes mellitus) and cardiovascular diseases (myocardial infarction and atherosclerosis) and cancer[13,16]. In fact, ageing is due to the loss of molecular fidelity that varies among different subjects, leading to both dangerous and non-dangerous features that can reduce the adaptation to the environment, involving DNA, cells, tissues and the whole system[17]. It results in compromised stress response, greater homeostatic imbalance, elevated risk of disease and, consequently, death[15,17].

Instead, successful ageing implies the avoidance (or the late onset) of diseases and disabilities with a preservation of cognitive and physical functions[13].

Centenarians represent the best model of successful ageing. They live longer than the rest of the population and without any relevant disease, thanks to a favourable genetic background and to a good response to adverse environmental conditions[13]. In 2012, Caruso et al. conducted a study in a small area of Sicily, the Sicani Mountains, where the number of centenarians is high.

These areas showed low mortality rate for cancer and cardiovascular disease. The centenarians recruited tended to be physically active and to have a healthy diet, contributing to inflammation reduction[18,19].

On the other hand, physical inactivity and hypercaloric diet lead to an accumulation of visceral fat and infiltration of pro-inflammatory macrophages and T-cells in adipose tissue.

Moreover, adipose tissue releases adipokines and other pro-inflammatory cytokines, causing inflammation[20].

The centenarians' ability to reach the age of 100 in the Westernized countries and the reduction in overall mortality reflect the improvement of hygienic condition, the reduced exposure to infection and inflammation, the overall improvement of the quality of life, the attention to the diet and the advent of therapeutic and preventive medicine[15,21].

## Mediterranean Diet

MedDiet is one of the most studied healthy dietary patterns. It represents more than an alimentary regimen. It can be considered a life-style that characterizes people, groups, regions and countries with similar but, at the same time, different ways to eat. As many studies demonstrated, MedDiet can positively influence several parameters, such as abdominal obesity, dyslipidaemia, elevated blood pressure and impaired glucose tolerance all being factors that predispose to onset of age-related diseases. Large intervention trials showed that MedDiet could prevent or delay age-related diseases with a great implication for health and social system[22-26]. Since 1995, this dietary pattern is represented by a pyramid updated over the years. The current one suggests a daily consumption of plant foods (fruit, vegetables, nuts, seeds, olives, herbs and spices), whole cereals, extra virgin olive oil (EVOO) as main source of fat, dairy products (principally cheese and yogurt, preferably low fat) and weekly consumption of poultry, fish, eggs (two to four) and legumes. Red and cured meats and sweets can be consumed in very low amount and red wine can be consumed responsibly during meals[27]. So, this diet consists with a low animal protein intake. It is hypocaloric, characterized by a low amount of saturated fats, cholesterol and sugars, and a high content of fibres[25]. Its beneficial effects could also be attributable to the presence of nutraceuticals and functional foods, other than only to the low glycaemic index (GI) and to the low animal protein and caloric intake[25]. Nutraceuticals are *Naturally derived bioactive compounds that are found in foods, dietary supplements and herbal products; they have health-promoting, disease-preventing or medicinal properties*[28,29]. They have antioxidant and anti-inflammatory effects that confer to MedDiet anti-ageing properties. In particular, fruit and vegetables are important sources of nutraceuticals such as polyphenols, as well as fish and red wine, rich in omega-3 fatty acids and resveratrol, respectively[9,30]. All of these belong to the big family of functional foods. Although a universal definition for them does not exist, the Functional Food Center defined them as *Natural or processed foods that contain known or unknown biologically-active compounds which, in defined, effective non-toxic amounts provide a clinically proven and documented health benefit for the prevention, management or treatment of chronic disease*[31].

## Mediterranean diet as a strategy to modulate ageing

As previously stated, Mediterranean functional foods contain bioactive compounds with anti-inflammatory and anti-oxidant effects that can confer health benefits, contributing directly to the reduction of the inflammatory status and of metabolic disorders related to an unhealthy diet[9,25,29].

Thanks to the studies on the positive contribution of MedDiet to the extension of life-span in healthy condition, it can be considered a strategy to delay ageing. In fact, it is characterized by low GI and low animal protein intake and it is able to act directly on specific metabolic pathways, called NSPs[9,25].

The NSPs are signalling cascades activated by the level of nutrients, such as carbohydrates or protein (or aminoacids). The most representative are the insulin/insulin-like growth factor-1 (IGF-1) and the target of rapamicin (TOR) pathways. From yeast to human, they are genetically conserved although, of course, the molecular complexity varies among them[8,32].

Insulin and IGF-1 levels are, respectively, influenced by glucose and growth hormone (GH). During fasting, both GH and insulin decrease with a consequent reduction of IGF-1 circulating levels and a delay in the ageing process, as presented by several model organisms[33].

The insulin/IGF-1 signalling cascade starts from the binding of insulin or IGF-1 to the insulin or IGF-1 receptor (IGF-1R) that triggers, inside the cell, many molecular events, such as the activation of the phosphoinositide second messenger.

It leads to the activation of AKT that can stimulate the nuclear factor kappa-light-chain-enhancer of activated B cells (NF-$\kappa$B) signalling, involved in immune inflammatory mechanisms[34,35]. The translocation of NF-κB to the nucleus and its binding to the DNA triggers the transcription of a number of genes, including pro-inflammatory cytokines, chemokines, adhesion molecules, eicosanoids, growth factors, metallo-proteinases, nitric oxide, etc[34].

The modulation of this pathway, possibly due to low glycaemic and protein intake and, maybe, to some antioxidant and anti-inflammatory molecules, may lower insulin/IGF-1 signal and stimulate the action of different transcription factors (TFs)[36].

Forkhead box O 3A (FOXO3A), extensively studied for its role in longevity, is one of these TFs, involved in the transcription activation of molecules that take part in cellular homeostasis. The gene encoding for this protein belongs to the FOXO family which has a typical DNA-binding forkhead box domain. It is one of the orthologue of DAF-16 in *Caenorhabditis elegans (C. elegans)*, a TF involved in stress resistance and longevity[37,38]. In addition, FOXO3A interacts with sirtuins (SIRTs), a family of histone deacetylase enzymes, identified as anti-ageing molecules in model organisms. SIRT1, one of the seven human sirtuin isoforms, SIRT1-SIRT7, deacetylates FOXO3A modulating its response to oxidative stress[39]. In mammals, downstream of IGF-1R, there is the mammalian TOR (mTOR) composed by mTOR complex 1 (mTORC1) and the mTOR complex 2. mTORC1 is activated by insulin and IGF-1, modulating growth, metabolism, and stress response through regulation of transcription, translation and autophagy, a cytoprotective process. Its inhibition by the protein

kinase AMP, a key sensor of the cellular energy state, activated by low levels of ATP, causes stress resistance and reduced age-related inflammatory status by the NF-κB pathway[40].
Decreased TOR activity extends longevity also in *C. elegans*[41,42] and *Drosophila melanogaster*[43]. In addition, its reduction in a mouse genetic model reduces the incidence of some age-related diseases, including cancer[44].
So, in humans, MedDiet, with its low intake of amino acids and low GI, might directly modulate the insulin/IGF-1 and the TOR pathways trough a down-regulation of the signals that lead to the activation of FOXO and, consequently, to the transcription of homeostatic genes that favour longevity thus decreasing inflammatory status and oxidative stress[25,45,46].

## The Mediterranean hormetic effect

Many components of the MedDiet are known to have healthy properties. The abundance in mono and polyunsaturated fatty acids (MUFAs and PUFAs), fibres, vitamins, minerals and, more generally, nutraceuticals plays a key role in inflammation reduction and oxidative stress response, acting on important risk factors for age-related diseases[25,47,48].
Moreover, nutraceuticals stimulate a hormetic response[49,50].
In 1943, Chester Southam and John Ehrlich described[51], for the first time, the phenomenon of hormesis. The term comes from the Greek word *ὅρμησις* that means *rapid motion.* As it was then defined in 2002, it is a *biphasic dose response phenomenon, characterized by a low dose stimulation and a high dose inhibition*[52].
In biology and medicine, hormetic effect is an adaptive response of cells and organisms to moderate (usually intermittent in terms of intensity) stress, such as a low dose of chemical agent or environmental factor that induces an adaptive beneficial effect. On the contrary, the same stimulus is detrimental at higher doses[53].
In nutrition, hormetins are molecules that can activate the hormetic process; they are produced by plants as a protection against microorganisms, insects and other environmental agents[54]. Some examples of nutritional hormetins are phenolic acids, polyphenols, flavonoids, ferulic acid geranylgeranyl, rosmarinic acid, kinetin, zinc and the extracts of tea, dark chocolate, etc[55].
In human beings, as a reaction to damage, they likely activate stress response pathways, such as the NSPs, in cells as for example after exposure to mild heat stress[56].
The activation of NSPs results in increased production of homeostatic genes and therefore proteins such as heat-shock and others involved in the regulation of cellular energy metabolism or antioxidant enzymes[57].
For example, the isothiocyanates are active compounds obtained from the conversion of glucosinolates by myrosinase, a glucosidase possibly present in human microbiota contained in cruciferous vegetables (cauliflower, broccoli sprouts, cabbage, broccoli and similar green leafy vegetables) typical of the MedDiet. Inside the cell, they stimulate the activation of the nuclear factor erythroid 2-related factor 2 that, consequently, translocates to the nucleus, binding the antioxidant response element, encoding antioxidant enzymes[58].
Polyphenols, classified as phenolic acids, flavonoids, stilbenes and lignans, phytochemicals widely present in fruits and vegetables, including the Mediterranean ones, can regulate the TF NF-κB by reducing the expression of inflammatory cytokines and can activate SIRT1 that may also inhibit NF-κB, reducing the cellular stress response[59-61]. On the other hand, SIRT1 acts on FOXO3A which, as previously stated, modulates genes that encode antioxidant enzymes and other stress-response proteins[25,62].
MedDiet effects on gene expression therefore cause a successful response to environmental changes, allowing better use of nutritional resources and determining epigenetic modifications[25,63].
Phytochemicals, as well as other nutrients, can also influence miRNA expression, a class of small non-coding RNA (19-24 nucleotides) evolutionary conserved that, in turn, regulate post-translational gene expression, suppressing the translation or reducing the mRNA target stability[64]. Moreover, it has been demonstrated that polyphenols can modulate miRNA action on metabolic homeostasis and on chronic diseases but further studies are necessary to identify their targets[64-66].

## Mediterranean functional food: the example of EVOO and table green olives

As previously stated, typical Mediterranean foods contain phytochemicals, defined as chemical compounds produced by plants. They are often characterized by phenolic groups, good source of natural antioxidants that may affect health with their anti-inflammatories and anti-cancer properties[25].
EVOO, the main and common food in the Mediterranean basin, is the principal source of fats in the MedDiet regimen. It has many nutraceutical properties thanks to the complex mixture of bioactive compounds[67].
Several studies have shown that it decreases pro-inflammatory environment induced by oxidized low-density lipoproteins (LDLs) both in experimental models and in humans, reducing the levels of C reactive protein, a powerful marker of inflammation, and the levels of pro-inflammatory cytokine interleukin-6 (IL-6)[68-70].
With its high content in MUFAs and polyphenols, EVOO might exert beneficial effects on the development and progression of age-related diseases. In fact, its non-saponifiable portion (1-2%) contains about 230 bioactive molecules, such as carotenoids, mainly lycopene, sterols, and the phenolic compounds oleoeuropein, oleocanthal, hydroxytyrosol and tyrosol, with antioxidant and anti-inflammatory properties[67,70]. Oleic acid is the main MUFA and is claimed to increase the resistance of LDL to oxidation. It has also been suggested that MUFAs can decrease all-cause (11%) and cardiovascular mortality (12%), cardiovascular events (9%) as well as stroke (17%)[68].
Moreover, in models, oleic acid suppresses cytotoxic function of natural killer cells with a consequent anti-inflammatory effect, although it is simply a possible lower response to microorganism infection[71].
In general, unsaturated fatty acids improve endothelial function, decreasing intercellular adhesion molecule-1 production by endothelial cells and reducing leukocyte adhesion[72]. In fact both in vitro and in vivo studies demonstrated the endothelium-protective properties of

these molecules[68,70,73-76].
Some trials showed that polyphenols intake has been associated with low mortality rates caused by coronary heart disease[77,78].
These compounds are able to bind LDLs, increasing resistance to oxidation and acting as radical scavengers[79].
Moreover, it was observed that the consumption of polyphenol-rich olive oil could decrease blood pressure (BP) and improve endothelial function in young women with high-normal BP[75].
Generally, polyphenols inhibit NF-κB pathway that leads to the expression of pro-inflammatory genes[80-82].
In particular hydroxytyrosol and oleocanthal have ibuprofen-like activity, inhibiting the cyclooxygenases 1 and 2, responsible for prostaglandin production[70,80].
As reported in a recent review, also green table olives are an extremely rich source of polyphenols, especially oleuropein and hydroxytyrosol, comprising 1-3 % of fresh pulp weight.
Despite the high levels of hydroxytyrosol in both table olives and EVOO, in humans its bioavailability was proven only in oil[83]. The amount of polyphenols in olives, as well as in oil, is strongly influenced by the variety and the geographical origin. Greek *Koroneiki* have a very high content, while polyphenol content of the Spanish *Arbequina* is low and that of Sicilian *Nocellara* is medium-high[67], with a conceivable anti-inflammatory and anti-oxidant effect.
In 2013, a new compound was extracted from the wastewaters obtained during olive oil production from *Nocellara del Belice* olives. This compound, known as nocellaralactone, is also present in the leaves and appears to be structurally similar to monoterpenoids, secondary metabolites found in higher plants, with a significant *in vitro* anti-inflammatory activity[84]. Moreover, a pilot study demonstrated that daily consumption of table green olives Nocellara del Belice is likely linked to a decrease in IL-6 and malondialdehyde (MDA) levels[85].
MDA is the main product of PUFAs peroxidation and is an important index of oxidative stress[86]. Noteworthy, this study highlighted a reduction of fat mass with an increase of muscle mass in subjects recruited for nutritional intervention. The possible explanation could be linked to the capacity of conjugated linoleic acid (CLA) to reduce body fat levels, strictly linked to production of adipokines (pro-inflammatory cytokines)[87].
CLA is present both in EVOO and table olives and can also be produced during their digestion.
In experimental models, acting as signalling mediator, CLA inhibits lipogenesis, increases fat oxidation and reduces adipocytes size[88,89].
In fact, such levels significantly decreased at the end of the dietary intervention.

## Conclusion

The extraordinary increase of elderly population in developed countries underscores the importance of studies on ageing and longevity and the need for a prompt spread of knowledge about these topics, in order to satisfactorily decrease medical, economic and social problems associated with old age.
In fact, European public health policy focuses its attention on the achievement of a healthy lifespan which represents an important challenge[16,90].
According to positive biology, an effective approach is needed to understand the causes of positive phenotypes to try and explain the biological mechanisms of healthy ageing, rather than making age-related diseases the central focus of research[91]. Much evidence shows that dietary restriction[2,8] as well as allelic variations in gene encoding proteins that take part in NSPs can increase lifespan[62,92-99]. This phenomenon is evolutionary conserved but the achievement of dietary restriction is not easy in human beings because it implies a considerable caloric restriction. A close daily adherence to MedDiet, including a healthy lifestyle, seems to be one of the more realistic ways to apply dietary restriction. Moreover, the possibility of managing this pattern, based on the combined use of functional foods, should permit to create a new therapeutic strategy. It would be based not only on a specific bioactive molecule or on a specific food but also on an integrated approach. Starting from local dietary habits, it can be extended to a *nutra-functional diet* applicable worldwide.
Oxidative stress and low grade inflammation play a part in the pathogenesis of age-related diseases and consequently in the ageing process. Thus, it is very relevant to study in greater depth the mechanism of action of nutraceuticals contained in functional foods that may constitute a natural remedy with health benefits through the reduction of cellular and tissue damage with the final aim to prevent or fight age-related diseases[100].

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aesthetic medicine

Mini-Review

# Effectiveness and effects of the introduction of ESI collagen in the diet

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## Abstract
Skin ageing has always been at the centre of research in medicine and aesthetic surgery. Patients constantly require non-invasive methods to prevent skin ageing or prolong the effect of injections such as botulinum toxin or hyaluronic acid. Recently the effects of oral intake of collagen extracts have been evaluated. Biocollagenina® ESI is a supplement rich in hydrolysed marine collagen, hyaluronic acid, resveratrol, vitamin C and cranberry fruits extract. We tested it on a 70-patient cohort and evaluated its effectiveness with ultrasound measurements of dermal thickness before and after treatment. Results led us to consider Biocollagenina® ESI as a safe and effective supplement.

## Keywords
Collagen, skin ageing, hyaluronic, body, aloe vera, antioxidant

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## Introduction

Compared with ultraviolet-induced or extrinsic skin ageing, chronological or intrinsic skin ageing is a cumulative process as time progresses. Histologically, chronologically aged skin is characterized by a decreased thickness of the dermal matrix. The principal structural components of extracellular matrix in dermis consist of fibril-forming collagens. In sun-protected young skin, the synthesis and degradation of collagen are balanced to maintain collagen content and structural integrity of the skin. In chronologically aged skin, collagen homeostasis is aberrant and shows molecular features of decreased synthesis of type I and III pro-collagens (the precursors to collagens) and also high-level degradation of mature collagen fibres, which result in general atrophy of the extracellular matrix and impaired collagen fibril organization.

As we know, collagen or gelatin (partially hydrolysed collagen) in dietary supplementation has been credited with promoting extracellular matrix synthesis thus improving joint, nail, and hair condition.

Collagen hydrolysate can traditionally be isolated from the skin of land-based animals, such as cows and pigs. With marine species comprising nearly half of the total global biodiversity, skin, scale and bone of marine life, especially of various fish species, have become a new source of collagen hydrolysate. In recent years, there has been a growing interest in the potentially beneficial effects of marine collagen hydrolysate (MCH) on skin ageing. Several *in vivo* studies have shown that the antioxidative property of collagen hydrolysate from marine fish has a protective effect on UV-induced skin photo-ageing.

Our study has the purpose to evaluate the in vivo effect on the skin of patients on oral Biocollagenina® ESI. The study was based on a voluntary cohort of patients.

## Materials and methods

Biocollagenina® ESI contains:

- **Collagen** (5000 mg): the most abundant structural protein in the human body. There are many kinds of collagen. 75% of skin is Type 1 Collagen.
  It confers tone, elasticity and strength, it contrasts the appearance of wrinkles. Type 1 Hydrolysed marine collagen is derived from fish skin and provides peptides and amino acids (glycine, proline, hydroxyl proline).
  These peptides are quickly absorbed because of their low molecular weight.
- **Hyaluronic** acid (20 mg): one of the main constituents of connective tissue.
- **Resveratrol** (50 mg): a plant-derived molecule often extracted from grape skin. Resveratrol contained in Biocollagenina® ESI is produced through the natural fermentation of corn starch and therefore totally free from solvents and other potential contaminants.
- **Vitamin C** (80 mg): contributes to the formation of collagen for normal skin function.
- **Cranberry Fruits** (150 mg): antioxidant action.
- **Copper** (0.5 mg): contributes to the maintenance of normal connective tissues and participates in protecting cells from oxidative stress.
- **Zinc** (1.5 mg): also contributes to the maintenance of normal skin.

*Subjects and study design*

Our multicenter study enrolled 70 patients in different cities, aged between 45 and 65, non smokers, (Genova, Savona, Imperia, Roma, Bari, Milano, Napoli). Biocollagenina® ESI was administered for 20 days at the dose of 1 vial per day.

To evaluate the efficacy of the supplement the patient cohort was not further subdivided on BMI basis, occupational status, exposure to sunlight, exposure to air-conditioned environments, diet type and other variables.

At the beginning and at the end of the 20 days patients were given a subjective quality assessment form where they were asked to indicate a score on a scale from 1 to 5 for elasticity, hydration, tone and gloss of their skin.

In addition, all patients underwent skin and subcutaneous face tissue ultrasonography for the evaluation of dermal trophism prior to the beginning of treatment and also 3 days after the end of treatment.

## Results

All patients followed the treatment according to instructions. The average value for each parameter examined by our study was then calculated based on the score sheets they compiled *(Tables 1 and 2)*.

| 70 PATIENTS | Before treatment (average value) | After treatment (average value) |
|---|---|---|
| elasticity | 2 | 4 |
| hydratation | 3 | 5 |
| tone | 2 | 5 |
| gloss | 2 | 3 |

*Table 1. The mean value for elasticity before treatment was 2 with an average improvement of 2 points at the end of treatment. The mean value for hydration before treatment was 3 with an average improvement of 2 points at the end of treatment. The mean value for tone prior to treatment was 2 with an average improvement of 3 points at the end of treatment. The mean value for gloss before treatment was 2 with an average improvement of 1 point at the end of treatment.*

All patients were subjected to ultrasound and subcutaneous tissue scanning of the face before and after treatment in order to evaluate the thickness in microns so as to objectify the efficacy of the treatment *(Figure 1)*.

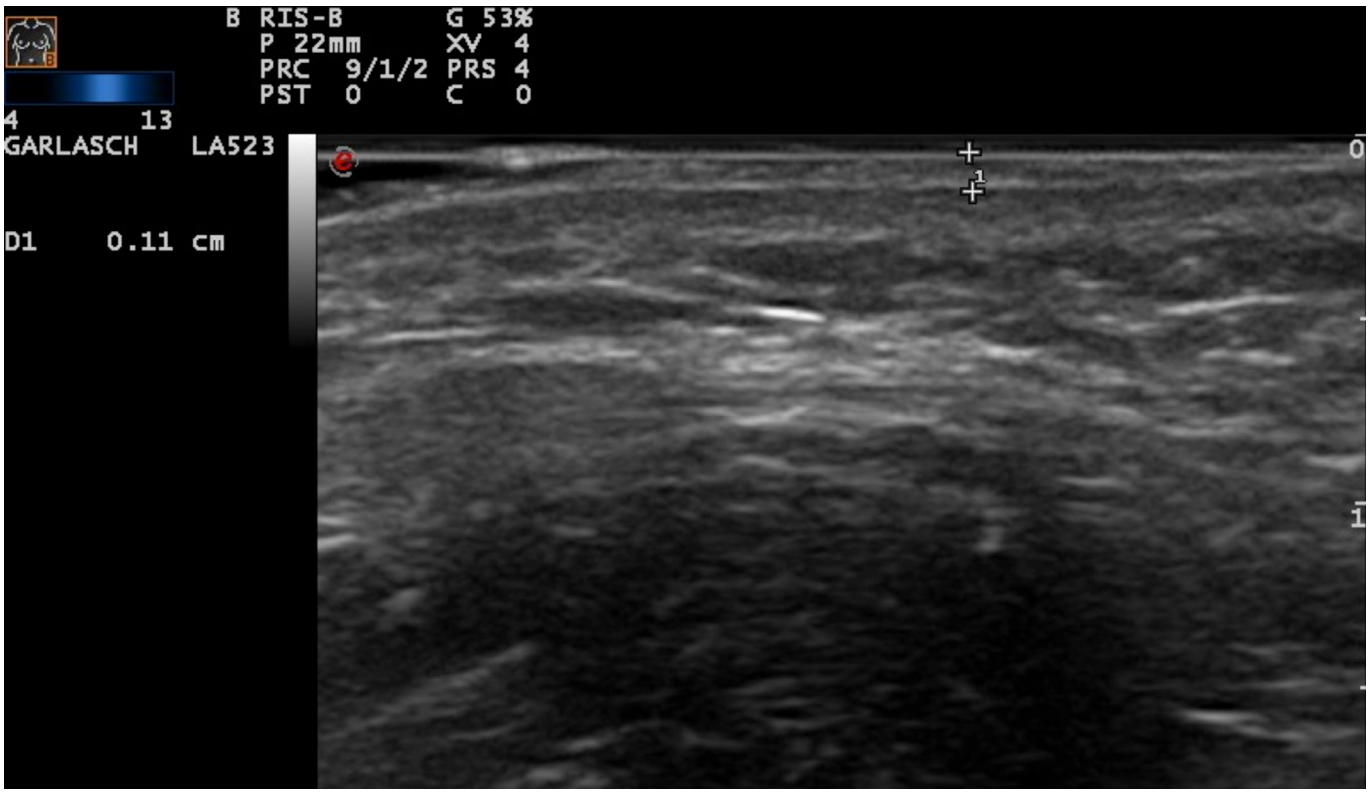


*Figure 1. The median skin thickness calculated on 70 patients before treatment was 0.11 cm.*

| PATIENTS | Before (cm) | After (cm) |
|---|---|---|
| 1 | 0.11 | 0.12 |
| 2 | 0.12 | 0.14 |
| 3 | 0.10 | 0.14 |
| 4 | 0.11 | 0.12 |
| 5 | 0.11 | 0.12 |
| 6 | 0.12 | 0.13 |
| 7 | 0.11 | 0.13 |
| 8 | 0.12 | 0.12 |
| 9 | 0.11 | 0.14 |
| 10 | 0.10 | 0.12 |
| 11 | 0.10 | 0.12 |
| 12 | 0.11 | 0.13 |
| 13 | 0.11 | 0.13 |
| 14 | 0.10 | 0.14 |
| 15 | 0.12 | 0.14 |
| 16 | 0.10 | 0.12 |
| 17 | 0.11 | 0.12 |
| 18 | 0.11 | 0.13 |
| 19 | 0.10 | 0.12 |
| 20 | 0.11 | 0.14 |
| 21 | 0.10 | 0.12 |
| 22 | 0.12 | 0.13 |
| 23 | 0.11 | 0.12 |
| 24 | 0.10 | 0.14 |
| 25 | 0.11 | 0.13 |
| 26 | 0.12 | 0.13 |
| 27 | 0.11 | 0.13 |
| 28 | 0.10 | 0.14 |
| 29 | 0.11 | 0.13 |
| 30 | 0.10 | 0.12 |
| 31 | 0.12 | 0.14 |
| 32 | 0.11 | 0.13 |
| 33 | 0.11 | 0.13 |
| 34 | 0.9 | 0.12 |
| 35 | 0.11 | 0.14 |

| PATIENTS | Before (cm) | After (cm) |
|---|---|---|
| 36 | 0.9 | 0.14 |
| 37 | 0.12 | 0.14 |
| 38 | 0.11 | 0.12 |
| 39 | 0.11 | 0.13 |
| 40 | 0.10 | 0.14 |
| 41 | 0.11 | 0.13 |
| 42 | 0.12 | 0.14 |
| 43 | 0.11 | 0.13 |
| 44 | 0.10 | 0.14 |
| 45 | 0.11 | 0.12 |
| 46 | 0.12 | 0.14 |
| 47 | 0.11 | 0.12 |
| 48 | 0.11 | 0.14 |
| 49 | 0.11 | 0.12 |
| 50 | 0.10 | 0.12 |
| 51 | 0.11 | 0.14 |
| 52 | 0.12 | 0.13 |
| 53 | 0.9 | 0.12 |
| 54 | 0.11 | 0.13 |
| 55 | 0.11 | 0.14 |
| 56 | 0.12 | 0.13 |
| 57 | 0.11 | 0.14 |
| 58 | 0.10 | 0.14 |
| 59 | 0.11 | 0.13 |
| 60 | 0.10 | 0.14 |
| 61 | 0.11 | 0.13 |
| 62 | 0.10 | 0.14 |
| 63 | 0.11 | 0.13 |
| 64 | 0.10 | 0.13 |
| 65 | 0.11 | 0.14 |
| 66 | 0.10 | 0.13 |
| 67 | 0.11 | 0.13 |
| 68 | 0.11 | 0.13 |
| 69 | 0.12 | 0.14 |
| 70 | 0.11 | 0.13 |

*Table 2. The median thickness calculated on 70 patients at the end of treatment was 0.13 cm with a mean significant increase of 0.02 cm.*

## Discussion

Marine proteins are likely to contain a lot of specific and potent bioactive subsequences from a nutritional perspective due to their competitive and aggressive living conditions[2]. With various bioactive properties, collagen hydrolysate has gained increasing popularity as ingredient of functional foods or pharmaceuticals[3].

As demonstrated by Urushibata et al.[4] long-term oral ingestion of MCH was found to have inhibitory effects on the age-related decrease of dermal thickness, collagen content as well as expressions of collagen type I and III. Moreover, compared with the age controls, the denser and more systematic collagen fibres in the dermis treated with MCH indicated that long-term oral administration of MCH might inhibit the increased degradation of collagen fibres in chronologically aged skin. As we know, both the quantity and quality of collagen fibres are determined by the balance between collagen degradation and synthesis[5]. In this regard, long-term MCH treatment was found to play an important role in the process of collagen metabolism.

According to known literature, our study shows the efficacy of integrating with Biocollagenina ESI even in short-term and not just in long-term administration.

## Conclusions

Our study points to Biocollagenina® ESI as being useful to all people who want to take care of their skin, hair and nails through a process from within the body and not just with the external aid of cosmetics. Nourishment reaches the epidermis through the blood to the deeper layers of the cell as the cells are pushed outward and die. In fact, its action is ideal for those who work indoors and in air-conditioned rooms that dehydrate the skin, for people who do not follow a proper diet, for post-delivery care or after diets, to counteract normal skin ageing and excessive exposure to the sun.

Biocollagenina® ESI has a trophic effect on the subcutaneous tissue by promoting the synthesis of collagen types I and III and at the same time by slowing down skin ageing due to physiological collagen degradation.

From the evidence emerging from our study it appears that Biocollagenina ESI can be proposed as a valuable aid in the prevention of skin ageing and for the improvement of skin trophism.

## Conflict of interest

The authors certify that they have no affiliations with or involvement in any organization or entity with any financial interest (such as honoraria, educational grants, participation in speakers' bureaus, membership, employment, consultancies, stock ownership, or other equity interest and expert testimony or patent-licensing arrangements), or non-financial interest (such as personal or professional relationships, affiliations, knowledge or beliefs) in the subject matter or materials discussed in this manuscript.

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aesthetic medicine

Mini-Review

# Skin abnormalities in people with diabetes mellitus

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**Abstract**
Diabetes mellitus is widely spread all over the world and can be considered an epidemic. Despite the ever increasing number of drugs available against it, diabetes still represents a severe risk for acute and chronic cardiovascular, eye, kidney and peripheral nerve complications.
A number of skin abnormalities are also well known to negatively affect the quality of life of people with diabetes. Nevertheless they are not fully described in specialized textbooks and are therefore mostly ignored by family doctors and diabetologists.
Our paper tries to fill this gap by drawing clinicians' attention onto skin changes in people with diabetes and to be aware of those changes in order to identify the disease early enough to prevent more dangerous complications.

**Keywords**
Diabetes mellitus, complications, skin, lipodystrophy

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## Introduction

Despite its inner nature of chronic non-communicable disease, based on its increasingly high prevalence all over the world, diabetes mellitus (DM) has been recently defined as a pandemic[1]. An ever-growing number of drugs has been made available, during the last decade[2-4], to provide patients with consistently good glycaemic levels. Nevertheless, in most people with diabetes, metabolic control is still inadequate and therefore retinopathy, nephropathy, macroangiopathy and neuropathy occur quite frequently, thus making DM a severe disease[5-8]. This is probably why other less prevalent long-term complications, including different kinds of skin alterations, are often disregarded by textbooks and overlooked by clinicians. In fact, some dermopathies might be the presenting sign of undiagnosed longstanding and ill-controlled DM and help general practitioners alert their patients and prevent more severe consequences of the disease. This paper aims at providing an overview of diabetes-related skin changes.

## Skin abnormalities related to insulin resistance

The underlying condition causing type 2 DM (T2DM) is liver and muscle insulin-resistance, eventually leading to beta-cell failure and consequent insulin deficiency[9-10]. It may be described as a less efficient endocrine signal translation into intra-cellular metabolic effects[11].

To compensate for that, circulating insulin levels markedly increase and thus exert their typical growth promoting effects in a number of organ and systems, including the skin.

A typical expression of skin anabolism is Acanthosis Nigricans *(Figure 1)*.

This is characterized by ill-delimited dark gray, thick skinfold areas (mainly neck, navel, groin and underarm). Commercially available softening creams are commonly used to try and improve it but good results are not expected before insulin levels decrease and the underlying mechanism is reversed by appropriate treatment, including active lifestyle and insulin sensitizers like metformin and/or glitazones[12-14].

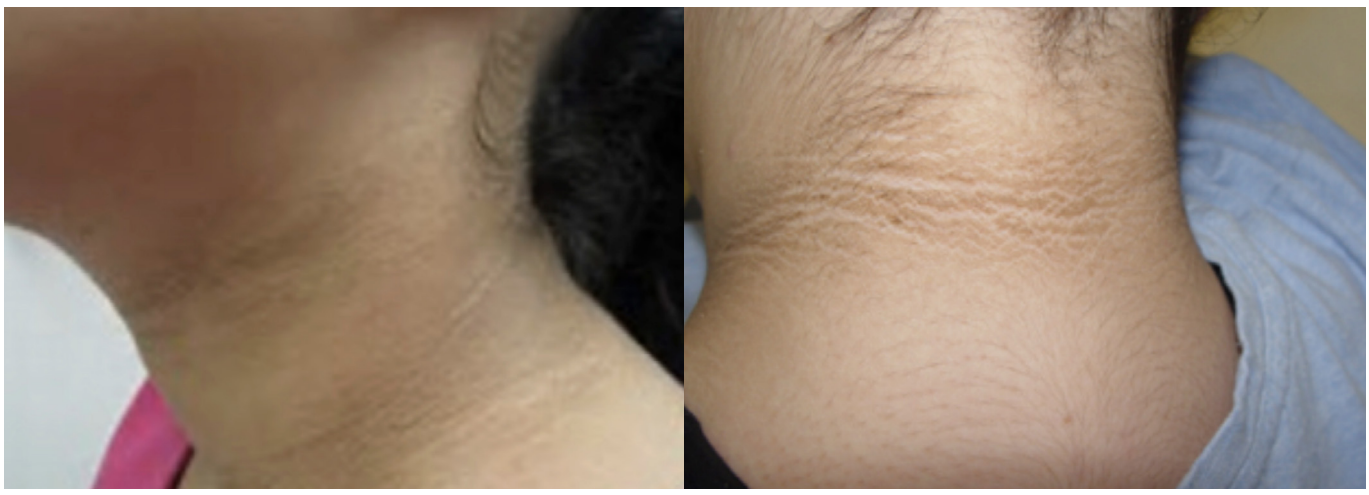

*Figure 1. Acanthosis nigricans.*

## Skin abnormalities related to Diabetes Mellitus *Per Se*

Long standing hyperglycaemia causes oxidative stress and mitochondrial advanced glycation end-products (AGE) accumulation which damages small skin peripheral nerve terminals and consequent firing defects impair capillary function with sebaceous gland dysfunction and skin dehydration. In fact dehydration is an early sign of neuropathy[15]. The latter can be counteracted by orally administered alpha-lipoic acid while topical Sacha Inchi oil may protect the skin by slowly penetrating into it and improving its barrier effect from the inside out. Another abnormality typically occurring in people with DM but not necessarily linked to high glucose levels is granuloma annulare *(Figure 2)*.

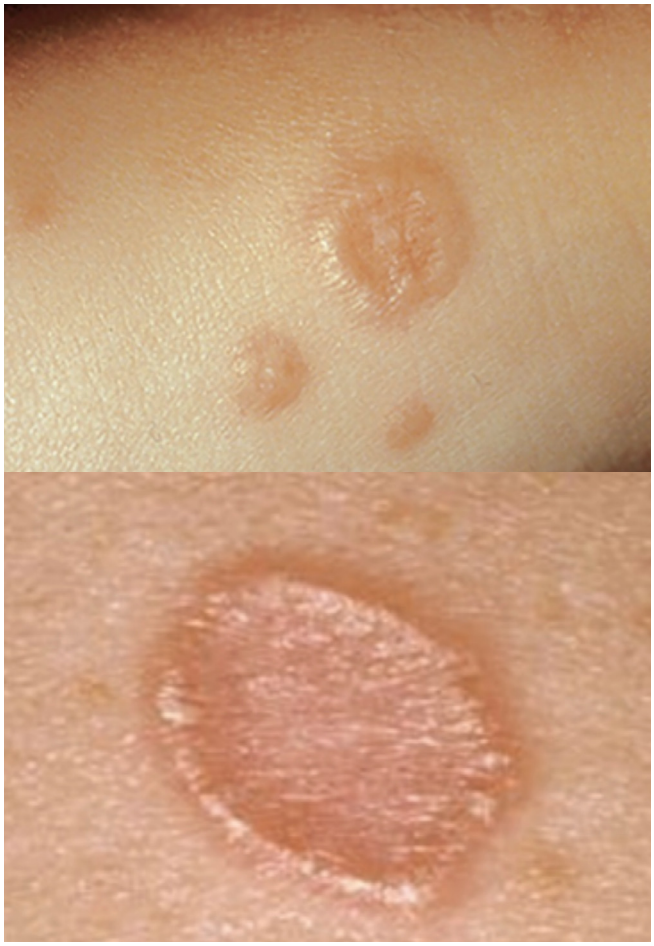

*Figure 2. Typical appearance of granuloma annulare.*

Its inner mechanism has not been identified yet so that, despite some future perspectives, it can't be anticipated or prevented efficiently enough. It most often consists of reddish, ring-like swollen bumps at the hands and feet. They generally cause itching and soothing creams are mostly used for symptom relief[16]. Xantochromia *(Figure 3)* of the palm of the hand and the sole of the foot, as well as peri-ungueal erythema *(Figure 4)* have also often observed as possible markers of undiagnosed DM; the same applies to tibia spots *(Figure 5)* which generally affect mature-age male subjects and unfortunately do not seem to benefit from optimized metabolic control[17].

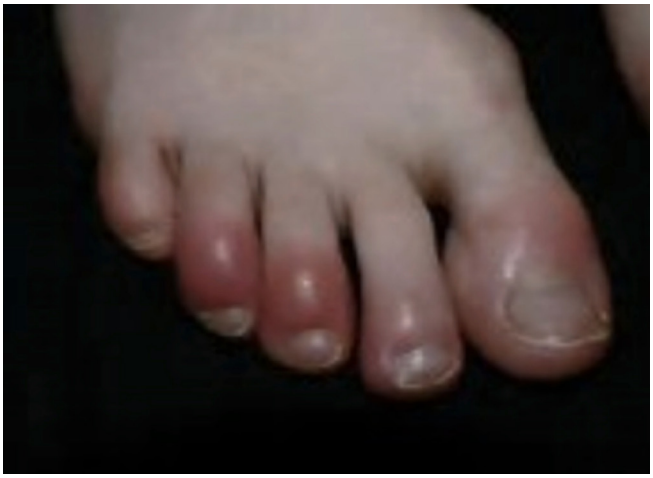

*Figure 3. Xanthocromic lesions.*

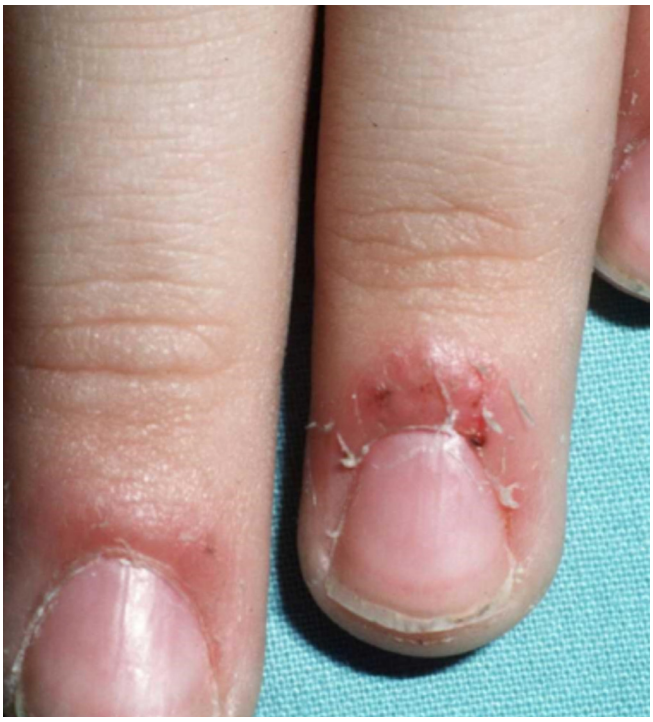

*Figure 4. Peri-ungueal erythema.*

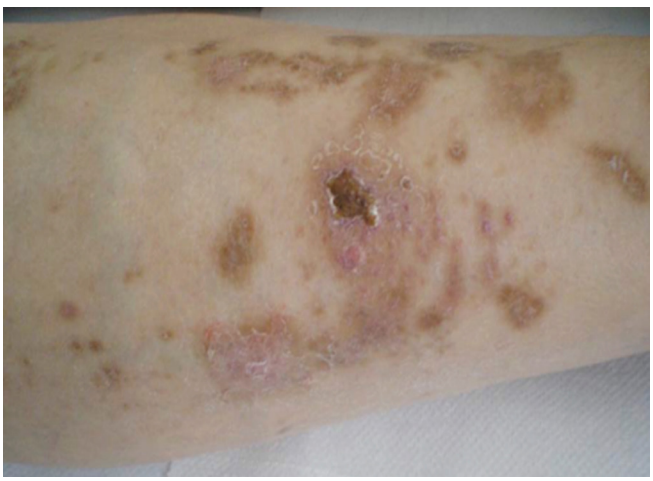

*Figure 5. Tibia spots.*

Bullosis diabeticorum is another skin abnormality suggesting the diagnosis of DM *(Figure 6)*: it consists of spontaneously occurring and relapsing yellowish vesicles, similar to those due to any blunt-force trauma. Fortunately enough, vesicles tend to recover on their own within 2 to 5 weeks[18]. An extremely challenging lesion is Pyoderma Gangrenosum *(Figure 7)*, a well-delimited, purple-red lesion typically characterized by resistance to both debridement and antibiotics, and by sensitivity to systemic treatment with corticosteroid and topical application of advanced medications[19].
Necrobiosis lipoidica (Rosai-Dorfman's disease) represents another rare expression of diabetic dermopathy, occurring in fact not only in diabetes *(Figure 8)*. It is a chronic, granulomatous skin abnormality associated with arguably immune-mediated connective tissue degeneration. It mostly affects the pretibial surface of insulin-dependent women and comes as a series of demarcated, slowly enlarging, yellowish or brownish plaques surrounding an atrophic core. No specific therapeutic strategy has been identified for it so far[20].

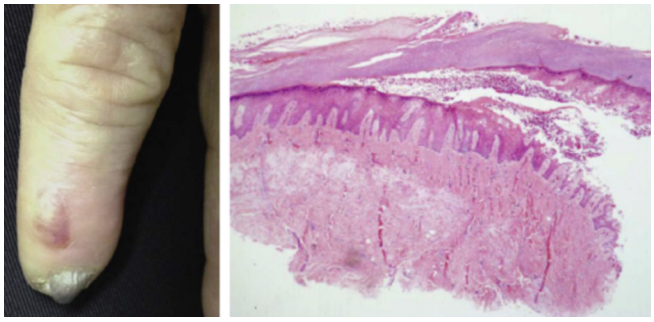

*Figure 6. Bullosis diabeticorum: clinical appearance and histologic features.*

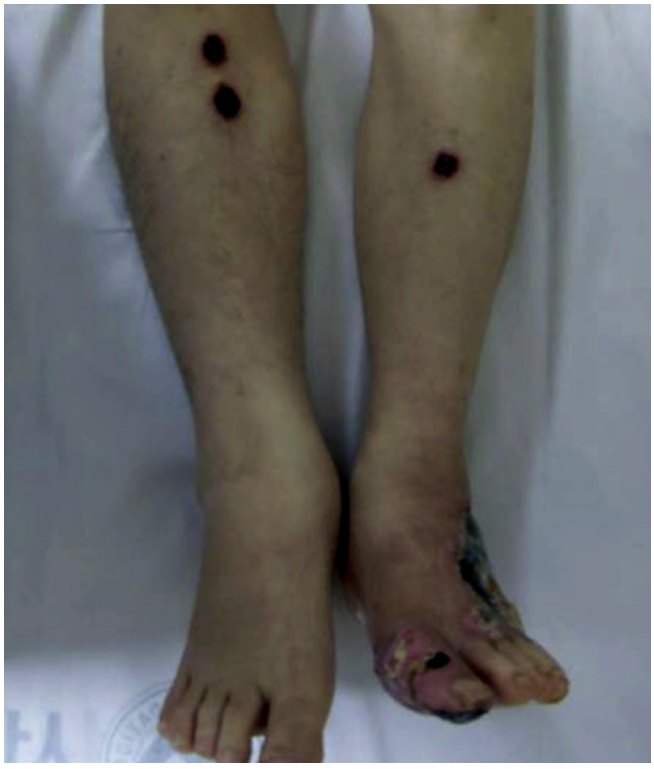

*Figure 7. Pyoderma Gangrenosum.*

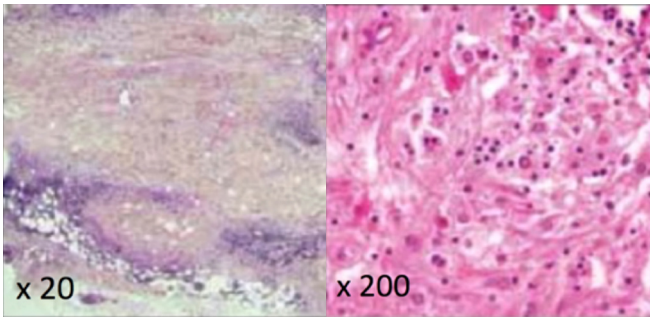


*Figure 8. Necrobiosis lipoidica (Rosai-Dorfman's disease).*

## Skin abnormalities related to poor metabolic control

Scleredema (scleredema adultorum of Buschke) is a rare sclerodermiform fibromucinosis that primarily affects the upper body and is caused by excessive collagen and mucin concretions in the dermis *(Figure 9)*. It is typically associated with infections (usually streptococcal), monoclonal gammopathy and diabetes mellitus. Its typical presentation is a firm, non-pitting, symmetrical induration of the posterior neck and upper back with overlying erythema in a woman with diabetes; extremities are usually not involved. Treatment of primary disease including strict glycemic control should always be considered but UVA1-phototherapy may be the first choice for patients with disabling manifestations. Due to adverse effect concerns, systemic corticosteroids and immunosuppressive drugs should be reserved for those with persistent, debilitating disease only[21]. A very common finding is also psoriasis *(Figure 10)*. In fact it is often associated with diabetes both *per se* and as a component of the metabolic syndrome therefore pointing to high cardiovascular risk[22].

*Figure 11* summarizes all associations shown so far between psoriasis and a series of inflammatory diseases. Due to that, it is recommended, as a minimum measure, to screen all affected people for the metabolic syndrome and/or diabetes in order to put effective cardiovascular disease prevention programs into practice[23]. While treating these lesions it is also recommended to try and improve metabolic control. Corticosteroids should be avoided if possible due to their negative impact upon glucose levels.

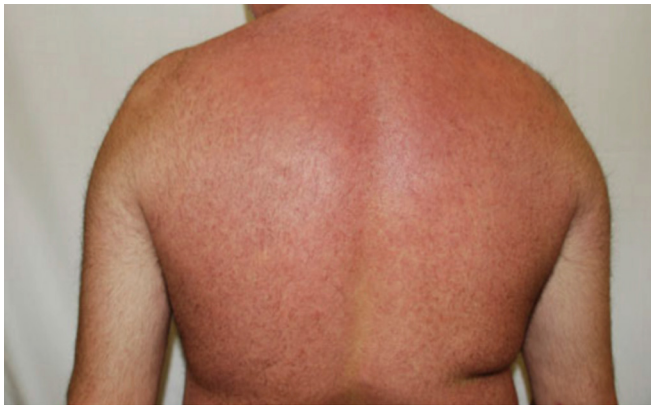

*Figure 9. Scleredema.*

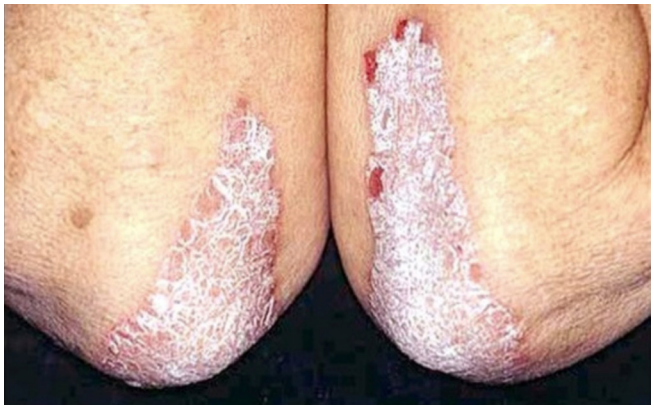

*Figure 10. Psoriatic lesions.*

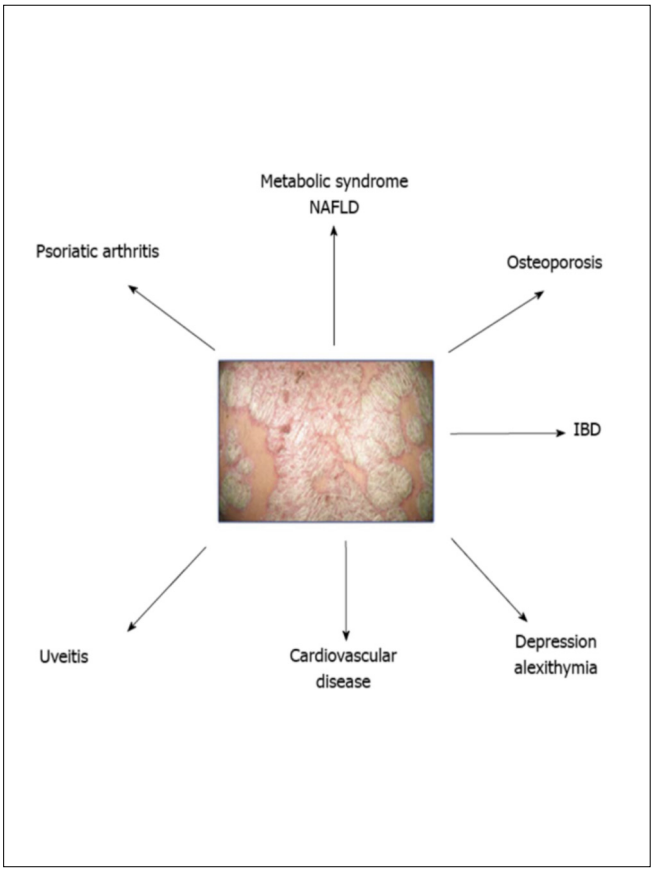


*Figure 11. Association between psoriasis and a series of inflammatory diseases.*

## Skin abnormalities displaying infection susceptibility

Hidradenitis suppurativa has many similarities with psoriasis in terms of underlying pathogenetic mechanisms.
It is also strongly linked with diabetes and the metabolic syndrome. This skin disease comes in the form of pimple-like bumps mostly affecting underarms and groins *(Figure 12)*.
They resemble deep-acne like cysts, folliculitis lesions (looking like swollen pimples with a hair in the center) and boils and are generally responsive to antibiotics when diagnosed and treated early enough. If allowed instead to grow, lesions go on healing and scarring, thus generating tunnel-like tracts eventually thickening the overlying skin.
Hypertrophic underarm scars restrict arm movements and scars in the groin area can make walking difficult[24]. That's why in 2015 the costly, yet effective, drug called adalimumab was approved for it by EMA (European Medicines Agency).

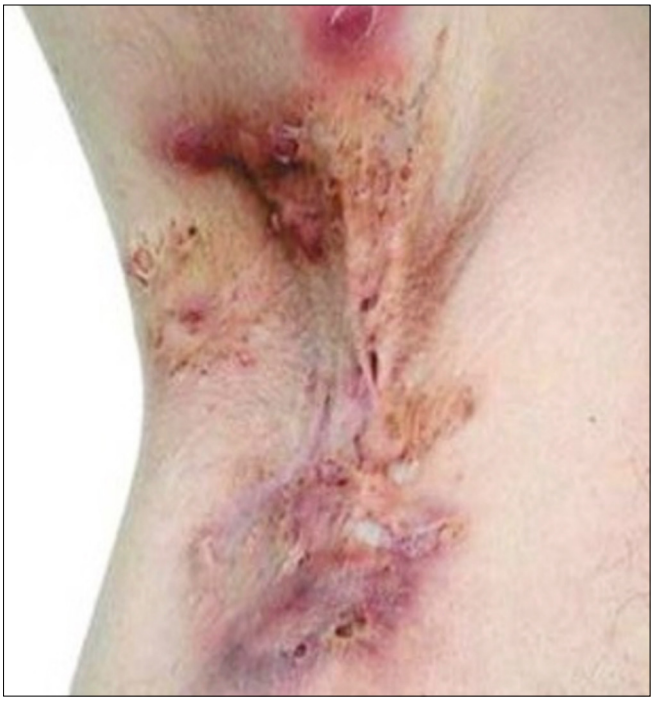

*Figure 12. Hidradenitis suppurativa.*

## Skin lesions due to inappropriate insulin administration habits

An often overlooked aspect of insulin treatment is the presence of lipodystrophic lesions, mostly due to repeated injections with re-used needles into a limited skin area. It follows trauma caused by a less and less sharp needle into the same skin area and by a sort of overlapping subcutaneous "foreign body" reactions due to the relatively large molecules penetrating the skin within a few seconds *(Figure 13)*.

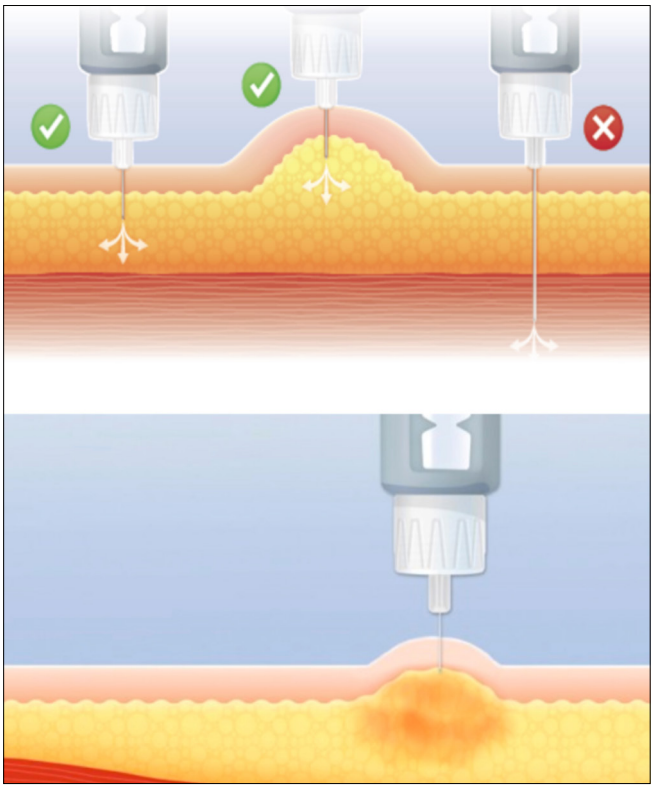

*Figure 13. Mechanisms underlying lipodystrophic lesions in insulin treated people.*
*The upper panel shows how injections with needles of different length may discharge insulin either into the subcutaneous tissue or intramuscularly. The lower panel shows how insulin molecules spread all around after being injected into the subcutaneous tissue.*

Aesthetic consequences are often only minor in the beginning and in fact most of the cases have to be looked for carefully by experienced diabetes team members but the real problem is represented by the extremely wide variation of glucose levels associated with lipodystrophy, often leading to a totally inappropriate diagnosis of "intrinsically brittle diabetes". This would keep patients for years under poor metabolic control with severe health consequences despite the possibility to have the problem solved by simply following good clinical practice rules, i.e. regular injection site rotation and used needle disposal. A detailed guideline on this subject was recently published and is freely available to any interested colleagues[25-26].

## Conclusions

Nowadays there are many treatment options for diabetes and personalized medicine is the recommended option for all cases[27]. Our paper does not intend to go too deeply into details of diabetic skin abnormalities. However we are convinced that just paying some attention to them is the cornerstone of a broad and effective preventative strategy. In fact we can do a lot for our patients by just carefully inspecting and palpating their skin, namely the basic rules any doctor should follow every day to get the most out of his/her clinical practice.

## Acknowledgements

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aesthetic medicine

Original Article

# Up-to-date combined therapy of stable vitiligo. Personal experience

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## Abstract

**Background**: The disfiguring aesthetic features of vitiligo, especially facial, may result in significant negative psychosocial effects. Its high rate of occurrences, considerable influence of patient psychological status as well as the absence of reliable treatment methods dictates the current need to look for new ways of vitiligo treatment.
**Aim**: Considering advantages and disadvantages of existing methods of skin repigmentation, we developed complex approaches of stable vitiligo treatment. The aim is to study the efficacy and long-term effectiveness of the developed complex for stable vitiligo treatment with the use of cell technologies compared with traditional treatment methods.
**Methods**: All patients were divided into 4 groups depending on the skin phototype and performed therapy. The 1st group (Fitzpatrick I and II skin phototype) and 2nd group patients (III and IV skin phototype), they were both treated according to the treatment protocol developed by us. 3rd control group (I and II skin phototype) and 4th group (III and IV skin phototype) – both were treated by standard vitiligo treatment (narrow-band UVB 311 nm, external treatment).
**Results**: In the 1st and 2nd groups we observed remarkable reduction of the length of treatment comparing to the control groups and increased percentage of patients with excellent results. The duration of follow-up was 2 years consistent with stable results.
**Conclusions**: The method has demonstrated its high efficacy, safety (as compared to surgery), shorter treatment course, possibility to treat larger vitiligo areas by means of small donor site, way of long term storage of cell material in cryobank for future use.

## Keywords

stable vitiligo, cell technology, suspensions of melanocytes and keratinocytes, repigmentation, phototype, narrow-band

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## Topicality

Traditionally dyschromias are of particular importance in modern dermatology and aesthetic medicine. Vitiligo is a frequent acquired disorder of skin pigmentation characterized by distinctly outlined de-pigmented areas; its etiopathogenesis is still not fully understood. The undesirable aesthetic properties of vitiligo, especially facial, may result in significant negative psychosocial effects, particularly a rate of depression twice that of the general population. Histologically we can observe either absence or abrupt decrease of melanin content in melanocytes within vitiligo patches. Its high rate of occurrence among many ethnic groups and in many regions, the considerable impact on patients' psychological status as well as the absence of reliable treatment methods dictate the current need to look for new ways to treat vitiligo. Standard vitiligo therapy (external: NB UVB and PUVA) is safe, however it requires a long-lasting treatment course, taking as a rule between 1.5 to 2 years to complete. Among its significant disadvantages it should be noted that only half of patients achieve 75% repigmentation or more with long-term, consistent therapy[1]. Recently we have been observing a widely followed combination of standard and surgical methods for vitiligo treatment. Dermatosurgical techniques such as tissue therapy (epidermal blister grafting, follicular grafting) and cell therapy (non-cultured cell suspensions of melanocytes and keratinocytes) are widely used to treat stable forms of vitiligo, resistant to standard therapy[2,3].

## Purposes and objectives

Long-lasting standard-type therapy, unstable results and a high number of complications occurring with the combination of regular and surgical techniques determined the need to look for new methods of stable vitiligo treatment.

New possibilities in this area have become available with cell technologies implementation[4].

Biotech lab "SmartCell" was founded in 2012 in Odessa, Ukraine. The laboratory facilities and equipment comply with European and world norms of GMP standards.

The laboratory specialists follow relevant methods of PCR and conduct cytogenetic studies while adhering to microbiological protocols of continuous biotechnological process for quality control purposes.

Considering advantages and disadvantages of existing methods of skin repigmentation as well as the possibility to use cultured melanocytes and keratinocytes, we developed a series of approaches to the treatment of persistent vitiligo types[5,6].

## Materials and methods

The study, according to the presented algorithm efficiency, included 27 volunteers ranging in age from 9 to 63 of which 9 males and 18 females *(Figures 1 and 2)*.

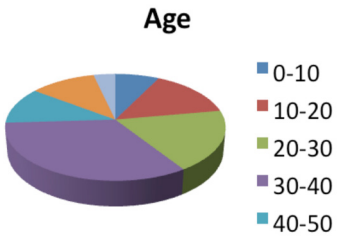


*Figure 1. Main group patients' distribution by age.*

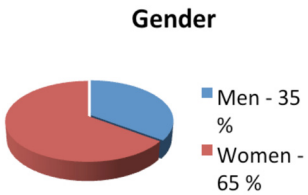


*Figure 2. Main group patients' distribution by gender*

Prior to treatment, all volunteers signed informed consent to participate in the study and to the use of achieved results for scientific purposes.

Quality control of the administered cell suspensions was monitored by means of ductal cytofluorometry using specific melanocytes antibodies.

## Study methods

Clinical efficiency of the suggested algorithm was evaluated:

- visually by a dermatologist;
- using objective methods of digital dermatoscopy and digital photo-imaging;
- subjectively by the patient him- or herself.

All patients were divided into 4 groups depending on skin phototype and received therapy.

The 1st group consisted of patients of Fitzpatrick I and II skin phototype. They were treated according to the protocol developed by us. The 2nd group with III and IV skin phototype was also treated according to our protocol. The 3rd group (control) of patients with I and II skin phototype received standard vitiligo treatment (narrow-band UVB 311 nm, external treatment). The 4th group, consisting of patients with III and IV skin phototype, also received standard vitiligo treatment.

Our algorithm of the complex technique for stable vitiligo treatment consists of 3 stages. At the first stage we have pigmentation induction at the donor site of healthy skin (NB UVB 311 nm Nº 5, 3 times a week, first dose is 70% of MED, every next dose is increased by 20 mJoules/cm2) in aesthetically insignificant or naturally hyperpigmented areas (labia majora) followed by skin punch biopsy, then transferred to the biotechnical laboratory. Once a week patients also receive intradermal injections of plasma enriched platelets (PRP) into the vitiligo patches to create a high concentration of growth factors including Epidermal Growth Factor (EGF) - Nº 3; local anesthesia is given, injection density - 1.5-5 mm and concentration of platelets 1 ml per 10 cm2 of vitiligo-affected skin.

At the second stage, 3-5 weeks later, we performed intradermal administration of cell suspension diluted in PRP solution with concentration of 1mln melanocytic-keratinocyte suspension per 1 cm² depigmented skin area. 2-3 days later we began a local photo-therapy course NB UVB 311 nm, 3 times a week, Nº 30, the first dose is 70% of MED, every further dose is increased by 20 mJoules/cm$^2$.

Patient L., 26 y.o., Diagnosis: Stable vitiligo, (12 years) focal type, treated by transplantation of cultured melanocytic – keratinocyte suspension according to protocol, 1 session *(Figures 3 and 4)*.

The third stage consists of repeated administration of cell suspension but only after a 2 months interval if the percentage of reconstructed pigmentation is less than 50%.

Patient A., 28 y.o., Diagnosis: Stable vitiligo (7 years) generalized pain, treated by transplantation of cultured melanocytic – keratinocyte suspension according to protocol, 2 sessions *(Figure 5)*.

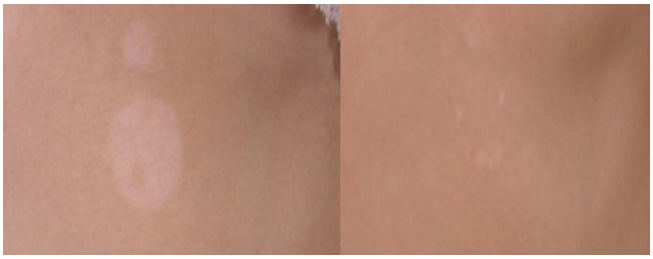

*Figure 3. The result of patient L treatment before and 3 months after a single transplantation of cultured melanocytic – keratinocyte suspension according to protocol.*

*Figure 4. The result of patient L treatment before and 3 months after a single transplantation of cultured melanocytic – keratinocyte suspension according to protocol (dermoscopy).*

*Figure 5. The result of patient A treatment before and 5 months after a double transplantation of cultured melanocytic – keratinocyte suspension according to protocol.*

## Results and discussion

The first group comprises 11 patients (7 females, 4 males), the second group – 16 (11 females and 5 males). The control group has 17 patients (8 females, 9 males) and 21 patients (13 females and 8 males) formed the fourth group *(Table 1)*.

| Protocol | | | | Control | | | |
|---|---|---|---|---|---|---|---|
| 1 group | | 2 group | | 3 group | | 4 group | |
| 11 pts. | | 16 pts. | | 17 pts. | | 21 pts. | |
| 7f. | 4m. | 11f. | 5m. | 8f. | 9m. | 13f. | 8m. |

*Table 1. Number of patients in the study groups.*

Within the first group, treated according to protocol, we achieved excellent repigmentation (75-100%) in 15 patients (56%), good repigmentation (50-75%) in 10 patients (37%) and unsatisfactory results were observed in 2 cases (7%). Besides, in the first group excellent results were achieved in 6 cases, and in 9 cases in the second group. Good repigmentation was observed in 4 patients from the first group and 6 from the second; unsatisfactory repigmentation was recorded for 1 patient in the first and second group each. It is important to note that repeated administration was required for 6 patients in the second group with excellent results *(Table 2)*.
The treatment period in 1st and 2nd group lasted 10-14 weeks.

| Group | Number of patients | Excellent repigmentation (75 - 100%) | | Good repigmentation (50 - 75%) | | Unsatisfactory repigmentation (less than 50%) | |
|---|---|---|---|---|---|---|---|
| | | Absolute number of patients | % | Absolute number of patients | % | Absolute number of patients | % |
| 1 | 11 | 6 | 55 | 4 | 36 | 1 | 9 |
| 2 | 16 | 9 | 56 | 6 | 38 | 1 | 6 |

*Table 2. Treatment results using melanocyte-keratinocyte suspension.*

In the 3rd and 4th control groups on standard treatment without cell technologies, excellent results (75-100%) were achieved in 7 patients of the 3rd group and 9 patients of the 4th group. Good rate of pigmentation reconstruction (50-75%) was observed in 8 patients of the 3rd group and 9 patients of the 4th group.
Unsatisfactory pigmentation (less than 50%) was observed in 2 patients from the 3rd group and 3 from 4th group *(Table 3)*.
The treatment period for the 3rd and 4th groups lasted between 4 and 8 months.

| Group | Number of patients | Excellent repigmentation (75 - 100%) | | Good repigmentation (50 - 75%) | | Unsatisfactory repigmentation (less than 50%) | |
|---|---|---|---|---|---|---|---|
| | | Absolute number of patients | % | Absolute number of patients | % | Absolute number of patients | % |
| 3 | 17 | 7 | 41 | 8 | 47 | 2 | 12 |
| 4 | 21 | 9 | 43 | 9 | 43 | 3 | 14 |

*Table 3. Treatment results in the control group.*

It is important to note that in the 1st and 2nd groups we observed considerable reduction of the length of treatment compared to the control groups and an increased percentage of patients had excellent results. Approximately 40% of 3rd group cases (with III and IV Fitzpatrick phototype) required repeated administration of cells, which should be taken into consideration when composing programs for this category of patients. Presumably, it is necessary to use higher cell dosages for these patients.
The duration of follow-up was 2 years, the results were stable.

## Conclusions

Widely used standard methods of vitiligo treatment are often very time consuming and do not deliver guaranteed results[7-9].

To achieve optimal results in treating the stable form of vitiligo we suggest a complex algorithm based on autologous melanocytes and keratinocytes administration. The method is about donor site preparation (induction of pigmentation or the use of naturally hyperpigmented areas), preparation of the recipient vitiligo site (intradermal platelet-rich plasma injections to create high concentration of growth factors) along with intradermal administration of melanocytic-keratinocyte suspension, diluted in PRP solution, into depigmented skin sites followed by local NB UVB 311 nm photo-therapy.

The method has demonstrated its high efficacy, safety (as compared to surgery), shorter treatment course, possibility to treat larger vitiligo areas by means of small donor site and possibility of long-term storage of cell material in cryobank for future use.

This protocol offers excellent results in treating stable vitiligo after single application.

The next perspective research will focus on determining optimal doses of individualized cell product in the pursuit of better results[10,11].

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aesthetic medicine

Review

# Morphological description of hypothetical cellulite starting unit. The first step to explain cellulite development

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## Abstract

**Introduction:** Cellulite is a complex pathology that affects about 85% of women. The origin of cellulite is poorly understood, even if some studies describe different hypotheses based on the observation of subcutaneous architecture, vascular and lymphatic patterns.

**Material and methods:** in this work an ultra-structural description of cellulite specimens, harvested from fifteen healthy women affected by first degree cellulite, was provided. Specimens were harvested from trochanter, after local anaesthesia using punch biopsy. Samples were fixated, immediately after biopsy, in glutaraldehyde 2% and processed for electron microscopy.

**Results:** with scanning and transmission electron microscopy it was possible to describe the hypothetical cellulite starting unit, consisting of mature unilocular adipocytes strictly connected with sweat glands. The peculiar structure of this complex was not described in other adipose depots, even if this formations resembled the mammary glands during the pubertal phase in which there is a hormonal influence, leading to breast augmentation. With the ultra-structural and morphological analysis of cellulite specimens it was possible to define the possible role of sweat glands associated with mature adipocytes in the dermis of patients. Sweat glands probably stimulate mature adipocytes and mesenchymal stem cells to cause an increase in adipose tissue. In the case of cellulite the increase in adipose tissue was associated also with the stimulation of fibroblasts with the effect of collagen fibres deposition and with the consequent formation of the typical cellulite morphology.

**Discussion:** understanding cellulite origin might be conducive to developing more specific and accurate treatments and to hypothesize novel prevention strategies.

## Keywords

Cellulite, mature adipocyte, sweat gland, TEM, SEM, cellulite starting unit

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## Introduction

Cellulite's definition, aetiology, anatomy, and diagnostic approaches are the subject of continued debate[1-3]. In the majority of post-pubertal women, cellulite represents one of the most common topographic alterations to cutaneous surfaces in the thighs and buttocks. The skin acquires a spectrum of findings ranging from an orange-peel appearance to mattress-like undulations of transverse dimpling, nodularity and folds[4]. Despite this, only a scarce number of publications in the literature has focused on cellulite origin. Traditionally, there are three main theories for cellulite etiology1. The first is based on the architectural subcutaneous tissue differences between genders, and on alterations in the connective tissue[5,6]. The second regards vascular changes and the presence of oedema in the inter-cellular matrix of affected areas as the possible causes[7,8]. The third, which has currently been reformulated, considers the existence of a chronic inflammatory process secondary to the hormonal activity of the menstrual cycle as the main cause[9]. All these theories are based on topographical observation of the adipose tissues affected by cellulite and on histological analysis. Until now cellulite has been described as a pathology regarding subcutaneous adipose tissue, but in previous published studies there was only marginal mention of adipose tissue histology. Some authors mainly focused their attention on the connective septa and on the formation of typical cellulite structure: papillae adiposae, that protrudes into the dermis breaking into the reticular layer[10]. Papillae adiposae organize forming oblique polygonal lobules in which adipocytes modify their shape, without changing their volume, depending on the direction of pressure of collagen septa. This causes the papillae to extrude into the dermis–hypodermis interface, thus modifying the appearance of the skin surface in the so-called "mattress phenomenon"[10,11]. Other authors focused their attention on the vascular pattern of tissues affected by cellulite[11]. During cellulite occurrence lymphatic stasis could promote pathology development, but does not represent the only cause of cellulite development. In fact in the adipose tissue, in particular in the adipose compartment of cellulite, the lymphatic vessels are relatively scarce[12]. Other authors described defects in lymphatic vessels wall accompanied by fibroblast activation responsible for collagen septa formation[13,14]. In all these studies the adipose tissue, within the papillae adiposae, was not deeply described and in our opinion it is involved in cellulite development. The adipose tissue of other fat pads, located in different regions of the body, are well characterized at histological and ultrastructural level[16-18].

The study of subcutaneous adipose tissue has underlined the presence of different classes of adipose tissue depending on the distribution of adipocytes and collagen fibres and also on the adipose tissue localisation[16]. Particularly adipose tissue is often studied and observed in physiological conditions, but recently the attention of researchers have been focused on the adipose tissue harvested from injured tissues or influenced by pathological conditions as recently by Sbarbati and co-workers[18].

In the literature there is a paucity of ultrastructural and morphological characterization of adipose compartments of subcutaneous tissue after cellulite development, probably due to the belief in the role played by connective septa or by vascularisation changes.

This study, by employing electron microscopy, aims to verify the hypothesis on the origin of cellulite related with modifications in the adipose compartment. In fact the adipose tissue is to be considered as a paracrine organ that, with the secretion of hormones, influences the distribution of adipocytes in the cellulite lobules, their increase in size and their function[19]. Moreover, a deep analysis of the adipose compartment of cellulite led to identifying a particular complex form which cellulite could originate from named Cellulite Starting Unit (CSU).

## Materials and methods

A sample of 60 volunteers, both female and Caucasian, was recruited in apparent good health.

The selected sample is aged between 22 and 61 (average age of 41.5 years).

The choice was weighted to have a greater representation of the various age ranges of patients, in the case of a fairly uniform outcome.

The experimental group was selected from a larger team of 250 people, and a special questionnaire was submitted.

The choice fell among those who, in response to a specific question, reported suffering from cellulitis.

Patients were then subjected to a clinical evaluation in sequence:

Examination performed on patients standing upright: front, posterior and both sides, with particular attention to the femoral-trochanter, gluteal, super-lateral, super-medial and posterior regions of the thighs and knees, abdominal, supra-pubic, and subscapular In seemingly exempted subjects, the pinch-test manoeuvrer was performed, able to highlight subclinical conditions of excessive tension of the interlobular subcutaneous septum. Locoregional palpation has been used to detect the presence of hypodermic nodules and to evaluate their painfulness.

The subject of interest was also the recognition of skin stretch marks (striae distensae).

Particular attention has been paid to the search for symptoms and signs indicative of venolymphatic circulation conditions. Substantial traits (sense of heaviness, paraesthesia, diurnal and/or nocturnal cramps) were investigated and objectivised.

Among them, first of all, the colour of the skin: blotches of excessive pallor, indications of small local haemorrhage, swelling (alternating skin patches of irregular shape with different chromatic tone due to local circulation imbalance), hyperglycaemia by haemoglobin pigmentation.

The presence of areas of atrophy of the skin and of the attachments and/or stasis ulcers, positivity of the fovea sign (index of declining oedema), and the development of small-size ectasia of superficial veins (telangiectases) and varicose dilations of the major vein trunks; anthropometric evaluation, the following anthropometric data were collected: weight, height, BMI, circumferences; Particular attention was paid to the

precision in the measurement, performed with patient in upright position and with the use of 2 operators, so as to check the correct position of the meter on both sides. The following circumferences were taken:
- waist: minimum circumference between the last rib and the iliac ridges;
- belly: maximum circumference between the last rib and the iliac ridges;
- hips: maximum circumference for the buttocks;
- upper or proximal thigh: circumference immediately below the gluteal groove;
- medium thigh: circumference at the midpoint of a line drawn between the inguinal fold and the proximal margin of the crown;
- knee: circumference passing through the middle of the knee.

The relationship between the body mass consisting of fat tissue (Fat Mass) and the remaining components (Lean Mass) was defined by impedance measurements made with the BC-420MA High Capacity Body Composition Analyser (Tanita Corporation Tokyo) impedance.
The instrument allows us to evaluate: fat mass (in kg and in percentage terms), visceral fat, free fat, lean mass (in kg and in percentage terms), amount of body water (in kg and in percentage terms), mass bone, basic metabolism and metabolic age; Postural evaluation, patients were examined with the postural analyser Lux (Chinesport SpA Udine, Italy) for the detection of torso asymmetries: the alignment and symmetry of shoulders, margin lower jawbones, lacy ridges, gluteal and popliteal folds. Examination of the spine in orthostatism and during bending test allowed to detect any deviations on the frontal and sagittal plane (abnormal straightening of the physiological curves, scoliosis, dorsal hyperacidosis, lumbar hyperlogy). Knees have been evaluated to identify variance or valgism. The presence of cavity or flatness of the feet has been sought through the aid of an odoscope; colour Doppler ultrasound, performed with Esaote My Lab (Esaote S.p.a. Genoa). The aim of the study was to evaluate the efficiency of the arteriovenous circulation of the lower limbs, in particular looking for any signs of incontinence of the valve systems, inversion of the flow in the perforating veins, and slowing or interrupting the flow; subcutaneous ultrasound, realized with Esaote My Lab ultrasound and 12 MHz linear probe (Esaote S.p.a. Genoa).
The subcutaneous tissue has a thickness varying widely between 5 and 40 mm, depending on the habit and the body region; its echo-structure is normally characterized by the hypoechogenicity of adipose lobules, interlaced by hyperechogenic connective branches, predominantly parallel to the skin surface (perpendicular ones are more difficult to highlight). The superficial band, placed between the subcutaneous and the muscular tissue, appears as a linear hyperechogenic structure parallel to the skin plane. The aim of the ultrasound study is to evaluate the thickness of the subcutaneous tissue, the study of its general architecture (which is deeply altered in cellulite, with fragmented and irregular branches) and the identification of nodular images. The typical cellulite nodule has a rather characteristic ultrasound pattern, consisting of a “collated” hypoechogenic structure with multiple hyperechogenic strokes. Cellulite nodules have, by definition, over-fascial seats; if, on the other hand, the image is sub-fascial, it corresponds to a lipoma;
Laboratory surveys at the TEST Laboratory of Modena have analysed the biological samples obtained from the subjects in order to evaluate:
- haematochemical parameters of routine: the following indexes have been evaluated: complete haemogram, leukocyte formula, transaminase, gamma gamma, alkaline phosphatase, creatinine, glycaemia, azotemia, total cholesterol, HDL cholesterol, LDL cholesterol, triglycerides, whole bilirubin fractional, complete urine examination.
- Hormone structure: evaluations related to plasma 17-β-estradiol, FSH, LH, progesterone, prolactin, FT3, FT4, TSH. Because sex steroids and gonadotropins are produced differently depending on the stage of the ovarian cycle of females, three separate blood samples have been taken in different doses, subject by subject, at follicular, ovulatory and luteinic phenomena, respectively.

One sample was, however, sufficient for menopausal women. The blood sample for the determination of TSH was acquired in the morning, so as to capture the physiological peak time of the maximum plasma concentration of the hormone. Prolactinaemia values follow a circadian rhythm: they are highest after REM sleep and minimum on awakening.
Under normal conditions they do not exceed 25 ng / ml; increase in pregnancy, starting from the 8th week and reach the peak (40-200 ng / ml) toward the 30th week.
The plasma rate was evaluated on three samples, taken at a distance of 15 minutes, leaving in situ the needle of the withdrawal, since hormone secretion may be affected by the puncture stress.
The main adipokines: the plasma rate of Leptin, Adiponectin and Resistin was evaluated. The main site of leptin synthesis is adipose tissue. Serum levels are higher in obese subjects, where they show a positive correlation with BMI. The production and secretion of leptin are regulated by the calorie intake, reaching a peak within 12 hours after the meal but also by various substances: sex hormones, insulin, glucocorticoids, TNF-α and IL -6 promote them while catecholamines, thyroid hormones, GH and testosterone inhibit them. At cerebral level, particularly in the hypothalamic arched nucleus, leptin has the task of inducing satiety and increasing energy expenditure, causing the release of anorexia neurotransmitters, such as α-melanocyte-stimulating hormone (α-MSH) and CART (cocaine and amphetamine-regulated transcript) and inhibit the secretion of hunger-stimulating neurotransmitters such as NPY (neuropeptide Y). Obese people are likely to have a leptin-resistance at the cerebral stage. Leptin receptors, however, are also found in many other sites: liver, adipose, muscle, pancreatic, splenic, pulmonary, ovarian and adrenal tissue, immune and endothelial cells. This is to indicate that leptin is a pleiotropic molecule, not a simple hormone of satiety; in fact, in many leptin-deficient murine models, there are, in addition to hyperphagia and severe obesity, also abnormalities in reproductive, immune, hormonal and nervous function. Resin activities are not yet completely known; most of the information comes from studies done on rats, which appear to have phlogogenic and diabetic effects. In man it is secreted by white adipocytes

and inflammatory cells (macrophages and neutrophils). Resin diminishes tissue responses to insulin: Muscle reduces glucose uptake. At liver level, gluconeogenesis increases; it also stimulates the synthesis of LDL and inhibits its recapping, degrading specific hepatocyte receptors. This results in an increase in circulating LDL and consequent atherogenesis. Adiponectin is a protein belonging to the complement C1q family and is secreted exclusively by adipocytes.
Its plasma concentration is significantly reduced in obese and diabetic subjects. In humans it sensitizes tissues to the action of insulin. In addition, it has anti-inflammatory and anti-atherogenic properties: it suppresses the production of pro-inflammatory cytokines by macrophages, while at endothelial level it reduces the expression of adhesion molecules (VCAM 1, selectin E, ICAM 1).
- the level of endogenous antioxidant defences: the D-Rom Test was performed according to the Carratelli method and the BAP Test. The first identifies the signs of oxidative stress by evaluating the presence of lipoproxidation products, in particular of alkoxy and hydroperoxyl radicals, through photometric measurement of the oxidising capacity of a plasma sample relative to a chromogenic indicator. The normal value is between 250 and 300 U-CARR (corresponding to the oxidative capacity expressed by an equal volume of a solution containing between 20.00 and 24.00 mg / dl of H2O2) regardless of sex and age. However, values tend to be considerably lower in infants and higher in pregnant women. BAP Test - the Biological Antioxidant Potential Biological Test - allows the total concentration of antioxidant factors (bilirubin, uric acid, C and E vitamins, proteins, etc.) to be determined in a sample of plasma, depending on its ability to reduce iron from ferrous to ferrous. Under normal conditions, 1 ml of plasma has an antioxidant potential to reduce 1 ml of a ferric ion solution 2.2 µEq / l. Lower values are indicative of an oxidative stress condition due to insufficient antioxidant defences or vice versa of a saturation of the latter due to a persistent hyper-production of radicals. Again, the interpretation of the result is based on the photometric reading of a chromogen colouring. The most important liposoluble food-producing antioxidants: the plasma Co-enzyme Q10, Vitamin E, Lipoic Acid rate was measured. Co-enzyme Q10 is a liposoluble chinone found in mammalian cell mitochondria: normal blood rates are 0.6-1.6 µg / ml. Seven varieties of tocopherols (alpha, beta, delta, epsilon, eta, gamma and zeta) are found in nature under the common denominator of Vitamin E. Typically, only the tocopherol alpha plasma concentrations are evaluated, whose normal values are 578-1679 µg / dl. Lipoic acid is also known as thioctic acid and can be classified between liposoluble vitamins; it promotes the synthesis of glutathione (one of the most important natural antioxidants). In addition, it can act as a carrier of electrons and acetyl groups (or other acyls). For this reason, alpha lipoic acid acts as a cofactor for numerous enzymes (e.g. pyruvate dehydrogenase, alpha-ketoglutarate dehydrogenase) involved in the oxidation process of glucose, fatty acids and other substrates in order to produce adenosine triphosphate (ATP).
From the subdivision by decades the composition of the sample is as follows:

- 16 women for 20-30 age group;
- 18 women for the 30-40 decade;
- 15 women for the 40-50 decade;
- 11 women for the 50-60 decade.

Criteria for exclusion in the selection of the sample were: acute disease in progress, dysmetabolic states, dysendocrine or immunopathological assays, significant organ pathologies, severe obesity (BMI required no more than 30), ongoing pharmacological treatments against cellulite. Patients volunteered for the study and signed a specific informed consent, including biopsy and release in anonymous form.

***Histology and transmission electron microscopy (TEM)***
Sample of cellulite were fixed with glutaraldehyde 2% and were post-fixed in 1% osmium tetraoxide ($O_sO_4$) in aqueous solution for 2h, dehydrated in graded concentrations of acetone and embedded in Epon-Araldite mixture (Electron Microscopy Sciences, Fort Washington, PA, USA). The semi-thin sections (1mm thickness) were examined by light microscopy and stained with toluidine blue. The ultra-thin sections were cut at 70 nm thickness and placed on Cu/Rh grids with Ultracut E (Reichert, Wien, Austria), stained with lead citrate and observed using an FEI Morgagni 268D electron microscope (FEI Company, Eindhoven, Netherlands).

***Scanning electron microscopy (SEM)***
Specimens of cellulite were fixed with glutaraldehyde 2% in 0.1 M PB, post-fixed in 1% osmium tetraoxide ($O_sO_4$) in the same buffer for 1h, dehydrated in concentrations of acetone, critical point dried (CPD 030, Balzers, Vaduz, Liechtenstein), fixed to stubs with colloidal silver, sputtered with gold by an MED 010 coater (Balzers), and examined with a FEI XL30 scanning electron microscope (FEI Company, Eindhoven, Netherlands).

## Results

Histological and ultrastructural analysis of cellulite samples from different patients provided clear and very important information about the organization of subcutaneous, dermal compartments and of connective septa. In histological slices stained with toluidine blue, the organization of adipocytes into a peculiar functional structure was evident and it was called Cellulite Starting Unit (CSU). It was characterized by clustered mature cells of 60 µm *(Figure 1a)* surrounding sweat glands *(Figure 1 sg)*. This functional unit was embedded in a stroma consisting of large collagen fibres bundles *(Figure 1 cf)*. Also some blood vessels were observable *(Figure 1 bv)*. The ultrastructure of the specimens evidenced the particular composition of CSU, underlining the strong interaction between adipocytes and sweat glands *(Figures 2A-C)*. TEM showed that adipocytes and sweat glands are strictly connected and it is possible to suppose that there is a paracrine communication. The paracrine communication between sweat gland and adipocytes was performed through scarcely electron dense vesicles *(Figures 3 AB)* well detectable in the sweat glands or in contact with adipocytes membranes *(Figures AB black arrows)*. Adipocytes were unilocular and characterized by an ovoid shape. At higher magnification it is possible to recognize fibroblasts close to unilocular mature adipocytes *(Figure 3 C)*. SEM showed the organization in small clusters of adipocytes retrievable in the cellulite specimens *(Figure 4)*. Clusters were embedded in a dense collagen network in which thick fibres, that enveloped entirely the clusters, were detectable. Moreover adipocytes were embedded in a very thin collagen basket, typical of structural adipose tissue[16]. Adipocytes were characterized by typical morphology, appeared unilocular and having a diameter of about 40-50 µm. In some specimens of cellulite, observed at SEM at low magnification, the morphology of papillae adiposae is appreciable, with apical layer, corresponding to the skin, in which the orange-peel formation is clearly visible. The subcutaneous layer is characterized by the deposition of very thick collagen fibres interposed to rare adipocytes organized in clusters. Dermal layer appeared characterized by a strong recruitment of lymphocyte and erythrocytes. The recruitment of lymphocytes is related to the inflammation status induced by pathology of dermal compartment.

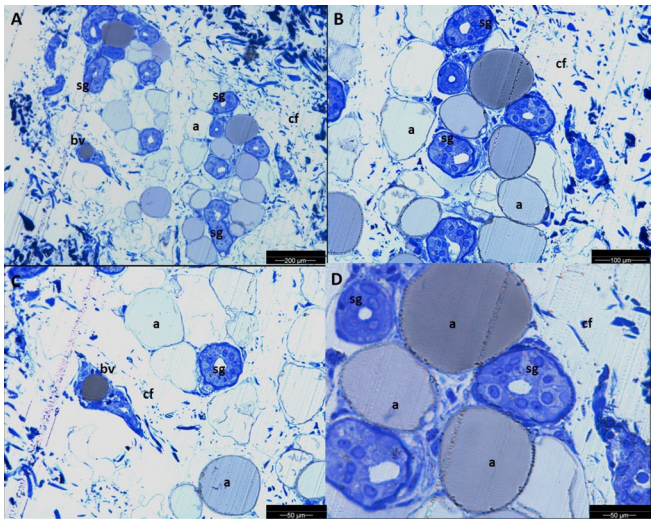


*Figure 1. Histological evaluation of representative specimen of cellulite (toluidine blue). The figure shows the structure and organization of the dermal compartment in which cellulite originates. In panels A and B at low magnification it is possible to recognize small and clearly defined cellulite starting units, consisting of adipocytes (a) and sweat glands (sg) and embedded in a fibrous stroma. At higher magnifications, shown in panels C and D it is possible to appreciate a blood vessel (bv), collagen fibres (cf), and in particular the strict correlation between adipocytes and sweat glands (panel D).*

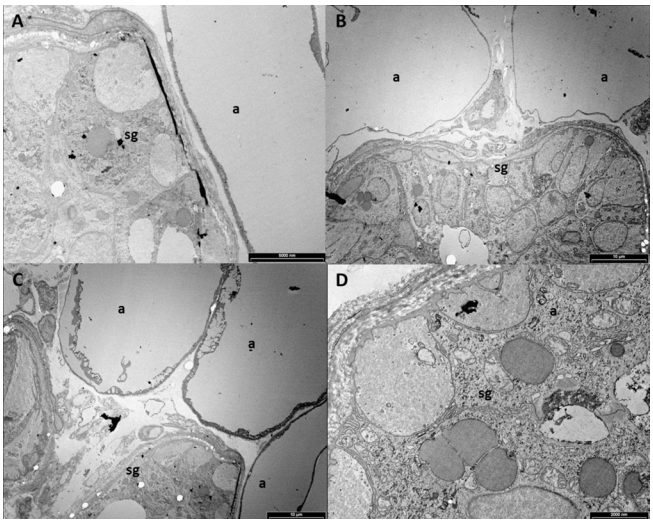


*Figure 2. Transmission electron microscopy of CSU. The figure shows the ultrastructure of CSU evidencing, particularly, the relationship between adipocytes (a) and sweat gland (sg). The adipocyte appears as unilocular cell with very thin membrane close to the membrane of sweat glands that are characterized by the presence of numerous mitochondria and abundant Golgi. In the thin space between sweat glands and adipocytes it is possible to see the collagen spacer with its characteristic feature and some stem cells and fibroblasts.*

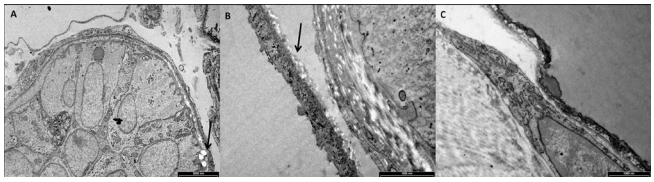


*Figure 3. High magnification TEM of CSU. The observation of sweat glands, at higher magnification, highlights the presence of collagen fibres deposed on glands membranes. Moreover on the adipocyte membrane close to the sweat glands it is possible to observe a deposition of scarcely electrondense vesicles (panels AB black arrows), also often observable on the sweat glands membrane. In panel C of the figure it is possible to see a fibroblast located near collagen deposits.*

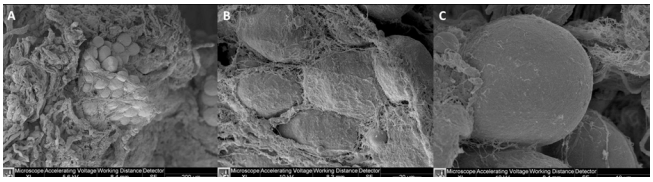


*Figure 4. SEM of cellulite specimens. The figure shows the morphology of cellulite lobules at low and high magnification. In panel A it is possible to see mature unilocular adipocytes and the stroma surrounding the clustered mature cells. The fibres in the stroma are very thick and well structured. In panel B the collagen component that envelops mature adipocytes is well visible. There is a basket-like cover that surrounds completely the unilocular cells. Panel C shows a mature unilocular adipocyte and it is well preserved, without membrane alterations and embedded in a dense collagen matrix.*

## Discussion

During the last decade cellulite was described as lipodystrophy, lipo-oedematous pathology, vascular pathology, basing on the observation of subcutaneous connective fibres, the distribution of elastic fibres and the distribution of adipocytes[2,20,21]. These theories focused on the changes in the organization and polarization of adipocytes distribution during cellulite development, comparing male and female and noted that in men, cellulite development was not as frequent and supposing a development related to the effect of sex hormones[22].

These findings were described and continuously confirmed by numerous authors that describe the principal component of cellulite: the papillae adiposae, formed by mature adipocytes protruding into the skin and collagen fibres that surround adipocytes, forming typical lobules[13]. These data were based on the histological, ultrasonographic observation; in some papers the use of Magnetic Resonance Imaging (MRI) was described[22,23].

Imaging techniques, even if very useful to diagnose vascular pathologies and the presence of oedema, provided information on the macro-structure of cellulite. Light microscopy could be not sufficient to clarify the real structure of the papillae adiposae and to explain the role played by adipocytes, collagen fibres, vessels, and other components. The ultrastructural approach appears the most useful tool to comprehend the relationship among the adipocytes, collagen fibres, elastic fibres and vessels. In particular, this study had the aim to clarify the organization of CSU and its function in dermal adipose tissue. Recently, in previous publications we had used the ultrastructural approach to describe different types of subcutaneous white adipose tissue (sWAT) in the abdominal area, in trochanters, in the internal face of the knee, in the heel and in the face[16,1718,23,24]. Every fat pad was characterized by different organization of adipocytes and collagen fibres. Moreover we have studied the ultrastructure and morphology of sWAT in injured tissues: scars after radiotherapy[24], sWAT from obese patients[18] and finally the cellulite development. Cellulite is a pathology strictly related to the increment of body weight and of sWAT deposits[2]. As mentioned before, also others authors have described the functional unit of cellulite and the organization of collagen fibres, but the origin of orange-peel phenotype visible in the skin is not completely understood. In this study the histological finding had led us to suppose a strict correlation between adipocytes and sweat glands that form a peculiar structure visible in all specimens of cellulite: CSU. The phenotype of CSU resembles the mammary glands. In fact mammary adipose tissue originates from sweat glands sensitive to hormonal effects during the pubertal age that cause an increase in dermal adipose tissue and the consequent augmentation of the breast[25]. We suppose that the development of cellulite follows the same pattern. Sweat glands become more sensitive to hormones and numerous chemical factors, leading to reprogramming adipocytes and probably the stem component of dermal compartment and, at the same time, causing an increase in inflammatory stimuli detected by fibroblasts that, in turn, cause new deposition of collagen. The increase in adipocytes in these areas leads to the protrusion of papillae adiposae with the subsequent acquisition of orange-peel shape. This theory has been described for hair formation[25].

The influence of hormones is quite irrefutable: cellulite is principally localized in thigh and its development starts in pubertal age and progresses during fertile period of the woman. In male cellulite it is absent, and this finding strengthens the theory of oestrogen stimulation. Ultrastructure performed on our cellulite specimens demonstrates a strict relationship between adipocytes of the papilla and sweat glands, like in mammary glands and in hair follicle induction[26]. Their membranes are adjacent and this aspect led us to suppose the paracrine role of sweat glands on adipose tissue, similar to that registered in the breast. Mature unilocular adipocytes within the CSU are also characterized by smaller size in comparison with the adipocytes located far from the CSU and far from sweat glands, leading to suppose that these adipocytes are newly formed. It is possible to suppose that this mechanism also involves adipose mesenchymal stem cells that differentiate in mature adipocytes, similarly to that occurring in hair follicle induction, where the involvement of adipose mesenchymal stem cells has been proved[26]. Another important aspect evidenced in this work is the location of CSU. The formation of cellulite starts in dermal compartment and not in subcutaneous adipose tissue. The next step could be the investigation of hormones, cytokines and adipokines expressed in the dermal compartment during cellulite development and, particularly, at different stages of pathology development.

## Conclusions

This work locates the origin of cellulite in the dermal compartment of the epidermis, describing the cellulite starting unit as the characteristic structure visible in all the specimens harvested from affected areas. Understanding the origin of cellulite might make it possible to improve therapy, in order to have a standardized treatment that contributes to newly establishing the physiological architecture of the tissue with the loss of inflammation and of the orange-peel appearance.

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## Conflict of interest

The authors of the manuscript declare that no conflict of interest exists

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# Comment to:
# "Height enhancement using hyaluronic acid and minimally invasive technique"

by Martusciello D. published on the Journal Aesthetic Medicine Volume 3 · Number 2 · April - June 2017; pages 57-64.

### Martusciello pouch

There is no reason to doubt this space exists, as described by Dr. Martusciello, but beyond the ultrasound findings, in truth far from conclusive, it would be very useful to have anatomical validation at the dissection table. To date, this space has not been described in the main anatomy books, not even under a different name.

### Photographic depictions

The photos shown do not clearly indicate the height increase described in the text: it would be very useful, not with standing the incorrect caption of photo 7, to take photographs with centimetre grid paper in the background in order to appreciate the actual result.

### Pain

The Author does not dwell on the painfulness of the implant, which according to us must be far from negligible. On the other hand, despite not disclosing the therapy cycle, the Author reports they have prescribed opioids and sometimes NSAIDs to control it. By inference it seems therefore clear that the manoeuvre must be significantly painful and, it should be noted, opioids often have severe side effects that may even prevent social activities. Finally, one could at least have used one of the multidimensional scales (Mc. Gill Pain Questionnaire, Wisconsin-Madison, Brief Pain Inventory etc.) or uni-dimensional scales (VAS, NRS, VRS) or VAS, Visual Analogue Scale, to provide more correct information on the pain.

### Ilizarov

The Author seems to have drawn inspiration from the Ilizarov technique to justify their work. However, Ilizarov, as well as the most recent work reported, designed his technique for certain types of fractures, as well as for lengthening dwarfs with severe achondroplasia, nothing to do with lengthening of few centimetres for aesthetic purposes. Therefore the comparison with the Ilizarov technique does not appear to be justifiable.

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# Courses and Congresses

## 2017

**12-14 May - Rome (Italy)**
**38th National Congress of the Italian Society of Aesthetic Medicine**
**12th National Congress of the Italian Academy of Aesthetic Medicine**
Venue: Congress Centre Rome Cavalieri
President: E. Bartoletti
E-mail: sime@lamedicinaestetica.it - congresso@lamedicinaestetica.it
Web: www.lamedicinaestetica.it

**19 August - Montevideo (Uruguay)**
**16th Congress of the Uruguayan Society of Aesthetic Medicine**
Regency Way Montevideo Hotel
President: A. Elbaum
E-mail: info@sume.com.uy - medicinaesteticacongreso@gmail.com
Web: www.sume.com.uy

**8-9 September - Paris (France)**
**38th National Congress of Aesthetic Medicine and Dermatologic Surgery**
French Society of Aesthetic Medicine
French Association of Morpho-Aesthetic and Anti-Aging Medicine
National Institute of education in aging prevention
President: J.J. Legrand
Web: www.sfme.info

**22-24 September - Almaty (Kazakhstan)**
**9th National Congress of Aesthetic Medicine and Plastic Surgery**
**Kazakhstan Association of Aesthetic Medicine and Plastic Surgery**
President: G. Zhumatova
E-mail: info@estetic.kz
Web: www.estetic.kz

**6-8 October - Warsaw (Poland)**
**17th International Congress of Aesthetic and Anti-Aging Medicine**
Polish Society of Aesthetic Medicine and Anti-Aging
Hilton Warsaw Hotel and Convention Center
President: A. Ignaciuk
E-mail: sekretariat@ptmeiaa.pl
Web: www.icaam.pl

**27-29 October - Istanbul (Turkey)**
**21th World Congress of Aesthetic Medicine**
Turkish Society of Aesthetic Medicine
Rumeli Caddesi Durak Apt N° 2, D.7 - Nisantasi, Istanbul - Turkey
President: H. Subasi
E-mail: subasihasanm@superonline.com
Web: www.estetiktipdernegi.org.tr

**3-4 November - Lausanne (Switzerland)**
**15th Congrès De La Société Suisse De Médecine Esthétique**
**5th Congrès De La Société Suisse De Chirurgie Esthétique**
Le Beau-Rivage Palace à Lausanne
President: S. Le Huu
Venue: Le Beau-Rivage Palace à Lausanne
Web: www.ssme.ch/congres-ssme-et-ssce-2017

**10-11 November - Santiago (Chile)**
**11th Chilean Congress of Aesthetic Medicine**
Chilean Association of Aesthetic Medicine
Santiago - Chile
Hotel Intercontinental - Las Condes
President: G. Marzullo
E-mail: contacto@creativaproducciones.cl
Web: sochme.cl/congresomedicinaestetica

**10-12 November - Miami (Florida – USA)**
**14th Annual AAAMC**
**American Academy of Aesthetic Medicine Congress**
**Aesthetic Medicine from Research to Practise**
JW Marriott Miami
President: M. Delune
E-mail: delegate@aaamed.org
Web: www.aaamed.org/congress2017

**24-25 November - Toronto (Canada)**
**CAAM 14th Annual Conference**
**American Academy of Aesthetic Medicine Congress**
Hilton Toronto
President: R. Van Aardt
E-mail: info@caam.ca
Web: www.caam.ca

**30 November - 1th December – Algeri (Algeria)**
**16th National Congress of Aesthetic Medicine and Surgery**
**Algerian Society of Aesthetic Medicine**
Hotel Mercure Alger
President: M. Oughanem
E-mail: Oughanem_m@hotmail.com
Web: www.same-dz.com

**7-9 December - Estoril, Lisbon (Portugal)**
**2nd National Congress of Aesthetic Medicine**
**Aesthetic and Anti-Aging Medicine Society of Portugal**
Hotel Palacio Do Estoril
President: J.P. Vale
E-mail: congressonacional@spme.pt
Web: www.spme.pt

**2018**

**22-24 February - Malaga (Spain)**
**33th National Congress SEME**
**Spanish Society of Aesthetic Medicine**
Palacio de Ferias y Congresos
President: P. Vega
E-mail: seme2018@pacifico-meetings.com
Web: www.seme2018.org

**2-3 March - Mexico City (Mexico)**
**15th Mexican Scientific Congresso of**
**Aesthetic Medicine and Antiaging**
**15th Venezuelan Congress of Aesthetic Medicine**
Mexican Scientific Society of Aesthetic Medicine
Aesthetic Medicine Society of Venezuela
Presidents: J-B. Miller Kobisher and V. García Guevara
Venue: Pepsi Center - World Trade Center, Mexico City
E-mail: congresoacademico@ippc.mx

**4-6 April - Buenos Aires (Argentina)**
**12th Pan-American Congress of Aesthetic Medicine**
**28th Argentinian Congress of Aesthetic Medicine**
Argentinian Society of Aesthetic Medicine - SOARME
President: R. Pinto
Venue: Auditorio de la Universidad Católica Argentina
Av. Alicia Moreau de Justo 1680
Puerto Madero - Buenos Aires
Web: www.soarme.com

**19-21 April - Brussels (Belgium)**
**12th European congress UIME**
**National congress SBME-BVEG**
**Belgian Society of Aesthetic Medicine and Lasers**
Radisson Blu Royal Hotel
President: J. Hebrant, H. Cartier
E-mail: info@aesthetic-medicine.be
Web: sbmebveg.be

**10-12 May - Pretoria (South Africa)**
**12th Aesthetic Medicine Congress South Africa**
**AMCSA 2018**
**Aesthetic and Anti-Aging Medicine Society of South Africa**
CSIR Convention Centre, Pretoria
President: J. Van Niekerk
E-mail: info@aesthmed.co.za
Web: www.aesthmed.co.za

**18-20 May - Rome (Italy)**
**39th National Congress of the Italian Society of Aesthetic Medicine**
**13th National Congress of the Italian Academy of Aesthetic Medicine**
Venue: Congress Centre Rome Cavalieri
President: E. Bartoletti
E-mail: sime@lamedicinaestetica.it - congresso@lamedicinaestetica.it
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