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Perspiration & Odor Testing Methods & New Opportunities for Claims Development

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Perspiration and Odor Testing Methods and New Opportunities for Claims Development

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abstract

Of all the natural functions of the skin, perspiration and its odorous consequences is one of the most lucrative challenges of the cosmetic industry, especially due to its social impact . Body odor and perspiration is deemed offensive in most cultures and cosmetic products to control this phenomenon are in high demand. The reduction of sweat and its resulting odor is normally addressed by antiperspirant formulations containing aluminium salts and standard antimicrobial actives. However concerns over environmental safety, cancer, health and wellbeing are driving innovation in new directions. A wider understanding of the potential positive and negative effects of antiperspirants and deodorants on the skins natural microbiome is becoming more appreciated, alongside a marketing desire for more elaborate claims. This has led to new formulations and different approaches within the confines of legislative requirements for study designs. Here we discuss sweat odor and perspiration, and examine both standard and new developed approaches in clinical testing for claims substantiation within the context of product efficacy, their effect on the skin's microbiome and legislative product claims requirements.

Introduction

Sweating is the body's natural response in body temperature regulation. However, the result of sweating along with skin surface bacterial metabolism of sweat components, leads to an unpleasant body odor. Culturally and socially, the control of perspiration and odor remains a challenge that is uniquely human amongst mammals. Humans generally dislike the appearance of sweat especially when excessive, however it is the accompanying odor that is considered far worse by most individuals, and as such perspiration and odor supports a multibillion-euro industry marketing products to counteract this natural phenomenon.

The human body has two types of sweat glands; eccrine and apocrine sweat glands. Sweat released from the "eccrine" sweat glands is odorless, made up mainly of water and salt. Sweat from the "apocrine" glands (usually located under the arm pits, the genital areas and around the nipples) contains proteins and oily substances that feed skin surface bacteria. Microbial enzyme activity causing smelly molecules and thus body odor is considered a product of bacterial metabolism.

Human psyche is strongly affected by malodor and in some individuals stress levels will increase sweating rates and thus further malodor. Human axillary odor is caused by a complex mixture of volatile organic compounds that produce a characteristic urine/musk-like smell [1,2], and they also contain human primer and modulator pheromones that affect neuroendocrine rhythms and mood respectively [3,4]. It has been shown that axillary odor could be produced by the interaction of odorless apocrine secretions [5-7] by inoculation with gram-positive organisms found on the skin surface [8] and subsequent studies demonstrated the presence of volatile, odorous C-C6-11 normal, branched, hydroxy- and unsaturated acids compounds in the axillae [9,10]. Trace amounts of thio-alcohols [5,11,12] with a high odor impact are also present.

Bromhidrosis or body odor, is a common phenomenon in post-pubertal individuals and is a chronic condition in which excessive unpleasant odor emanates from the skin. Determined largely by apocrine gland secretion, it can substantially impair a person's quality of life [13-16]. In rare cases, bromhidrosis may become pathologic if it is particularly overpowering or if it significantly interferes with individual health and well-being. There are 2 distinct types of bromhidrosis: Apocrine bromhidrosis which is the most prevalent form of bromhidrosis and should be differentiated from the less common Eccrine bromhidrosis. Several factors contribute to the pathogenesis of apocrine bromhidrosis. Bacterial decomposition of apocrine secretion yields ammonia and short-chain fatty acids, with their characteristic strong odors [17-19]. Axillary bacterial florae transform non-odoriferous precursors in sweat to more odoriferous volatile acids [20]. It is the Corynebacterium species that are predominant in males rather than females [21]. In certain circumstances, odorless eccrine secretion, can assume an offensive aroma to cause eccrine bromhidrosis. Eccrine sweat will soften skin keratin in the axillae, and its metabolites of bacterial enzyme activity produce the bromohidrosis foul smell [14]. Metabolic or endogenous disorders [22], some foods, (including garlic, onion, curry, etc.), alcohol, drugs (e.g, penicillin, bromides), may cause eccrine bromhidrosis. The role of excessive sweat, or hyperhidrosis, in the pathogenesis of bromhidrosis is unclear. Hyperhidrosis may promote the spread of apocrine sweat and contribute

further to bromhidrosis by creating a moist environment, good for bacterial proliferation [23,24]. Conversely, eccrine hyperhidrosis may cause a decrease in odor because the excess sweat flushes away the more odoriferous secretions.
We are surrounded by individually unique microbial "clouds" which form the body's microbiome [25], and microbiomes of two humans are never identical. This unique microbiome is symbiotic with the body, and is present to help protect the skin and associated tissues [26]. These microbial clouds can be disturbed through internal diseases, hormones, lifestyle choices, pollution, cosmetics and diet, and in consequence can lead to varying types of skin disorders and changes in immune responses [27]. It has also been demonstrated that prevention of sweating and odor control by the use of antiperspirants and deodorants has wider non-beneficial effects on the skins microbiome than intended [28].
In the development of such products, the industry has relied for many years on the use of aluminium based salts to plug the sweat pores, as well as acting in a deodorant capacity. However, due to raising concerns about environmental damage [29] health and safety [29,30], the industry is seeking new and innovative directions in sweat and odor control. Here we evaluate perspiration and body odor, and discuss standard and new approaches in clinical testing for claims substantiation and development, within the context of product efficacy, innovation, and legislative claims support requirements.

Odor & Perspiration Treatments

Clinical Intervention

When excessive sweating is limited to the armpits, surgery to remove the sweat glands is an option for the most extreme cases where all other treatments have failed. However this leads to „compensatory" sweating elsewhere on the body and regular monitoring of kidney function is also advised [31].
Sweat glands can also be removed using liposuction. Endoscopic thoracic sympathectomy, destroys the nerves that control sweating using an electrical current for a given period [32]. Another option is Botulinum toxin – injected into the affected areas of the body, such as the armpits, hands or feet blocking brain signals to the sweat glands, reducing the amount of sweat produced [33]. Anticholinergics, can help reduce sweating by blocking the actions of acetylcholine [34]though effects are not immediate and side effects may outweigh the benefits.

Cosmetic Applications

Cosmetic treatment of perspiration and odor has relied mainly on the incorporation of aluminium salts [35,36] supported by the addition of many types of cosmetic ingredients such as alcohols and plant extracts. Raising concerns over the presence of aluminium found in dementia patients brains [37] [11], breast cancer [38], and effects on the environment [30, 39], the industry is slowly moving away from the use of such salts to more skin and environmentally friendly actives. Furthermore, since deodorants and antiperspirants clearly influence the axillary microbiome [40] alternative formulation strategies are under investigation. In addition to the aluminium salts, other ingredients include octenidine, alcohol,triclosan, pH adjusters, zinc oxide, silver derivatives, and some essential oils, etc.

Microbial Transplantation

Microbial profiles are unique for every human and the use of antiperspirants and deodorants changes the balance though not the stability of these uniquely individual microbial colonies [40,41]. Furthermore, when antiperspirants are applied, the microbiome shows an increase in its diversity. Usage has shown to increase Actinobacteria, favoring body malodor, illustrating that such products modify the microbial colony stimulating odor-producing bacteria [41]. Moreover, replacing a malodor-causing microbiome with a non-odor producing microbiome, through an armpit bacterial "transplantation" or by direct application of probiotics/non-odor-producing bacteria, has been demonstrated, resolving this condition [41]. It is therefore suggested that selective modulation of the microbiome with prebiotics, or plant extracts in cosmetic development could also help underarm as well as foot odor [41,42]. However such an approach would need to carefully consider the uniqueness of each individuals body area related microbiome.

Legislation

When making antiperspirant and deodorant claims, there are global legislative requirements for products of integrity, and in the EU [43] they must meet six specific criteria: legality, truthfulness, honesty, fairness, weight of evidence, and informed decision making at points of purchase. For such claims, unless the natural phenomenon of sweating and odor production is clearly understood by both developer and consumer, meeting these legislative requirements might be confusing.
Therefore developers need to clearly understand:

- the target consumer;
- the target consumers' understanding of sweaty odorous skin and actual need (including hygiene, medical, social and cultural aspects);
- choice of ingredients and bio/chemical stability of those ingredients within a given formulation – breakdown products may be more of a problem than what was initially added;
- importance of evidence requirements based on desired claims concepts;
- regulatory and legal requirements

Once addressed, it is then easier to define which testing strategies could be approached in order to help build the proof of evidence required in order to develop and support claims for these products.

Antiperspirant Requirements: Since antiperspirants are designed to inhibit sweat production, several study protocol approaches are available and include those to comply with FDA (USA) and Clearcast (UK) requirements, as well as a screening method on the back of volunteers. The imprint method [44] is rarely used, and the starch-iodine semi-quantitative method [45] preferred by some clinicians for evaluating hyperhydrosis, are not discussed here. In the USA, antiperspirants are classified as over-the-counter (OTC) drugs [46] and therefore their efficacy testing must follow well-defined FDA procedures.

Deodorant Requirements: Deodorant efficacy evaluation is performed with the Sniff Test (based on the Standard Guide for Sensory Evaluation of Axillary Deodorancy, ASTM E1207-14) [47]. An expert panel of odor judges or Sniffers trained according to DIN EN 13725: 2003 [48,49] carry out these assessments.

Antiperspirant Efficacy Studies

FDA Method (Fig. 1)

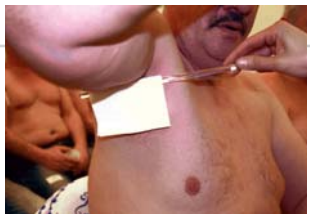
After a wash-out period of at least 17 days volunteers remain in a hot room (approximately 38°C (100F), at 30-40 % r.h. (relative humidity)), for two successive sweat collections (cotton/Webril® pads placed under their axilla for gravimetrical baseline measurements). Following volunteer qualification the test product is applied under one axilla for typically 4 consecutive days. The other axilla acts as an untreated control. Hot room sweat challenge is repeated typically 24h, 48h, or 72h after application. Sweat reduction is calculated by weighing the cotton pads prior and post sweat induction. In order to claim "antiperspirant efficacy" a product must reduce axilla sweat production by more than 20 % compared to the untreated control axilla. If a decrease of more than 30 % is reached extra efficacy could also be claimed.

Screening Method (Fig. 2)

The FDA protocol only permits the testing of one product. To enhance this restriction a rapid screening method was developed by one of the co-authors [50, 51] and further improved at proDERM [52], where up to eight formulations are tested simultaneously on the back of volunteers. For each product there is an untreated control area located on the contralateral area. The semi-occlusive nature of the axilla is mimicked by two hours of occlusion after product application. The hot room challenge is replaced by an exposure of the volunteers to a sauna at approximately 80°C for about 15 minutes to induce sweating. Applied pads for sweat collection on the

Test Procedure FDA	Days					
	1-17	18	19	20	21	22/ 23/ 24/ 25/ 26
• Discontinuation of use of antiperspirants	x	x	x	x	x	x
• Sweat collections in hotroom		x				x*
• Washing of test areas and application of test material		x	x	x	x	

* One hour after the last application of test material, at least two further collections spanning the period of the claim for enhanced claim duration



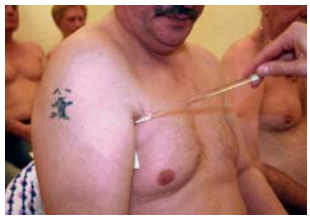
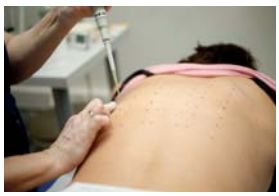


Fig. 1 FDA procedure for sweat collection during hot room exposure.

Test Procedure	Days				
	1	2	3	4	5/6/7/8
• Application	x	x	x	x	
• Removal of patch	x	x	x	x	
• Sauna and gravimetrical measurement of sweat secretion (24h, 48h, 72h or 96 post last admin.)					x



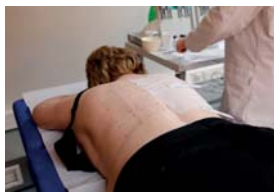


Fig. 2 Screening method for intra-individual comparison of multiple antiperspirant formulations.

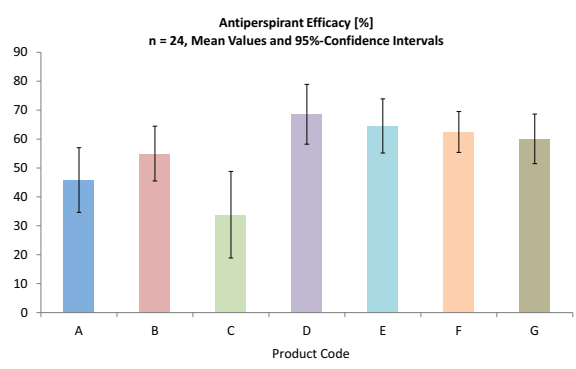


Fig. 3 Comparison of efficacy of aluminium salts in different formulation using the antiperspirant screening method

Test Procedure	Days			
	1-7	8	9	10/ 11/ 12/ 13
Wash-out phase	x			
Standardized washing procedure		x		
Sniffing assessment (Baseline)			x	
Product application			x	
Sniffing assessment (24h, 48h, 72h or 96 post last admin.)				x

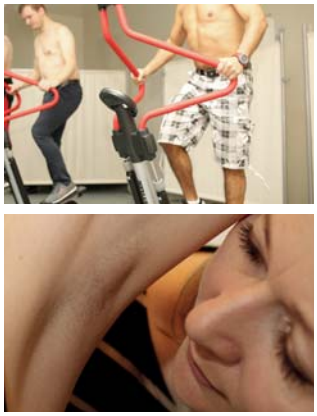


Fig. 4 Sniff test procedure using trained experts according to EN 13725:2003-07 and physical activity on steppers for wider claims.

respective test areas enable the gravimetric measurement and comparison of sweat production. **Fig. 3** shows different antiperspirant formulations applied and the respective efficacy assessed. A differentiation between products then enables a more precise choice for further development of product formulations and further efficacy studies.

Deodorant Efficacy Studies

A deodorant controls malodor through absorption, odor masking and/or antibacterial activity. Its main function is to reduce and prevent odor, and not sweat production. As highlighted previously, deodorant efficacy evaluation is performed with the Sniff Test [47]. This test is carried out after a standardized wash-out period of approximately 7 days (**Fig. 4**). Sniffers, an expert panel of trained odor judges (according to DIN EN 13725:2003), assess the malodor/sweat odor directly in the axilla at baseline and a defined period after product application on a 5 point scale or on a visual analogue scale. There are other designs where pads are applied to the axilla and the sweat odor of the pads is collected and determined in vials. Efficacy claims can be made according to the length of time the malodor is significantly lower compared to untreated under these controlled conditions. Study protocols with the aim to compare 2 products – or in addition to – a multi-product comparison of several test formulations can be also performed.

Microbiome Evaluation Studies

Each individual's microbiome is unique and is susceptible to changes when using cosmetic products especially deodorants and antiperspirants [15,42]. Although knowledge of the skin's microbiome is not new, with the rapid advances in gene sequencing, the microbiome has become a focal point for both the pharmaceutical and cosmetic industries. Previously, only single species such as e.g. *Staphylococcus aureus* could be analyzed using elaborate procedures. It is now much easier and possible to determine the entire microbiome from simple skin rinsing solutions. Moreover, innovative parameters like the Shannon Index can be simultaneously used to quantify biodiversity (**Fig. 5**).

Study Principle

We have developed in conjunction with a partner laboratory an efficacy study to support microbiome friendliness or probiotic claims for antiperspirants and deodorants (as well as other cosmetic products):

Sampling: Samples of microbial sweat are obtained by swabbing axillae skin 24 hours after a given washing proce-

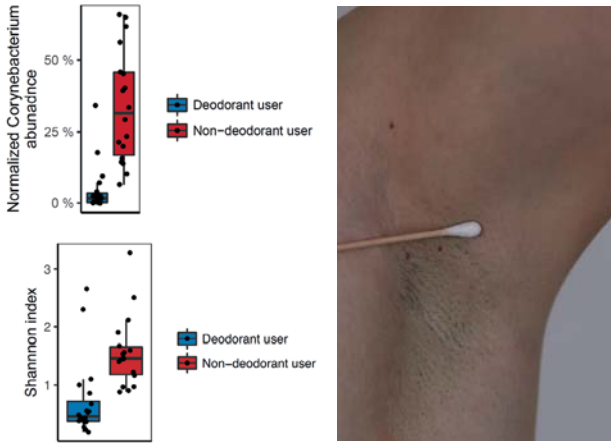


Fig. 5 Sample swabbing of axillae for microbial analysis and determination of effects of antiperspirant and deodorants on the skins microbiome (on the right picture). Graphs on the left side: Study on 2 panels of approx. 20 users of antimicrobial deodorant compared to 20 non-deodorant users shows a dramatic decrease of Corynebacterium abundance and a clear of the biodiversity (Shannon Index)

dure with a cotton swab soaked in phosphate buffered saline (PBS) for a defined number of times (**Fig. 5**). Each swab is collected separately and stored on ice prior to freezing at -80°C.

Microbial Analysis: The microbiome is then characterized using the 16SrDNA method (Clinical Microbiomics) amplified by PCR. The library molarity of DNA is sequenced with an Illumina MiSeq. The microbiome is analyzed by the 16S phylogenetic profiling method based on sequencing the 16SrRNA gene. It is a common and widely accepted method for studying bacterial phylogeny and taxonomy [53]. Since this gene consists of both – highly conserved as well as hypervariable regions – it probably is the most established genetic marker used for bacterial identification and classification. Since many sequences of the gene are available in public databases, this method is considered by many as the gold standard for microbial analysis.

Data Analysis: The relative abundance and biodiversity of the microbiome in the controlled and treated skin areas (such as *Corynebacterium*, *Staphylococcus* etc) are then calculated and identified (**Fig. 5**). Microbes with a similarity of at least 97 % regarding 16s rRNA-gene sequence are clustered into separate operational taxonomic units (OTUs). The relative abundance of all OTUs and bacterial taxa are presented from phyla (e.g. *Actinobacteria, Bacteroidetes, Firmicutes*, etc.) to genus (e.g. *Bifidobacterium, Lactobacillus, Clostridium*, etc.).

Advantages of this method include:

- Characterization of non-cultivable bacteria
- Profiling of hundreds of microorganisms from a single analysis
- Semi-quantification of the relative abundance of microbiome members
- Studies of complex microbiomes
- Provides faster and more accurate classification than traditional identification methods
- Identification of low-abundance bacteria

Addressing Wider Claims

To meet desires for wider/enhanced performance claim desires, including duration of efficacy and resistance to physical activity, adaptation to standard study designs is possible (**Tab. 1**). Under certain conditions, additional testing methods for claims e.g. inclusion of fabric care, skin cooling, skin care benefits, etc., have also been developed. Other test areas

Objectives	Study Overview	Potential Claim
Duration of Efficacy	Consumers prefer a long lasting efficacy as it implies better efficacy. Standard study design with wash-out phase and sweat amount or odor assessment	Number of hours (e.g. 24h, 48h) performance, enhanced duration or extra efficacy
Physical activity	After product application a defined sport unit e.g. 15 minutes on ergometer/stepper (**Fig. 4**) is included into standard protocols prior to/or instead of the common heat induction. Standardized showering after sports can also be integrated into the study procedure.	Sport resistance
Stress resistance	Psychological stress is a major issue when it comes to sweating, and there is an increased demand for claims to address this. The Trier Social Stress Test includes an interview with a psychologist with a stressful presentation topic, the videotaped presentation in front of an auditorium in a tense atmosphere and a difficult mathematic task.	Stress resistance Stress protect
Extreme climatic conditions	Global travels to tropical regions demand products which are also efficient under high humidity or under high temperatures. Thermal challenge is achieved by increasing the temperature of the hot room to higher temperatures from 38 up to 52°C and/or increased relative humidity e.g. 60 % at 35°C.	Heat resistance Humidity resistance Xtreme heat resistance
Effect on the Microbiome	The influence on the skin microbiome is assessed by swab analysis (single bacteria strains or microbiome) and compared to baseline or untreated.	Microbiome friendly, Helps maintain a healthy Microbiome
Cooling effects	Cooling of the axilla after application could easily be shown by measurement of skin temperature with a thermography camera	Cooling effect; helps keep you cool
Fabric care	Yellow stains might be caused by sweat mixing with antiperspirants or deodorants and hard water. Removal is nearly impossible. Subjects wear T-Shirts after product application overnight. Each day the T-Shirt is returned and washed for up to 15 days. Afterwards an image of the axillae area of the shirt is captured for assessment by quantitative image analysis. Only subjects who can prove that they tend to have yellow dots on T-Shirts are included in the study. A similar process is followed for white spots on black fabrics.	Yellowing minimized, Yellow stain resistance Non-whitening Spotless
Tolerance	Skin irritation is common after shaving and can be worse when antiperspirants or deodorants are applied. Standard irritation and stinging methods can be utilized.	Skin friendly after shaving

Tab. 1 Overview of additional claims development following standard study design adaptations.

Fig. 6 Determination of sweat production and reduction of hands and feet.

such as the hands and feet can also be assessed using a simple adaptation to standard designs (**Fig. 6**).

Psychological stress is a major issue when it comes to sweating and can trigger immediate formation of malodor. There is an increased demand for claims to address this. The Trier Social Stress Test design [54, 55] includes an interview with a psychologist with a stressful presentation topic, the videotaped presentation in front of an auditorium in a tense atmosphere and a difficult mathematic task. Stress levels are monitored by measuring cortisol levels in saliva, heart rate (assessed with a pulse rate meter on index finger), and subject self-assessment of the induced stress. Identified high responders are evaluated in a Sniff study immediately after stress. Sweating can be measured by application of sweat pads during the stress challenge replacing the hot room.

Care is advised when creating more advanced claims, in that developers need to: understand the uniqueness of human individuality when it comes to body odor and perspiration; care must be taken that they do not exceed claims legislation requirements; and that claims remain absolutely relevant to the average consumer need. For example whilst antiperspirant claims of 48 hours could be considered valid, a 96 hour claim which can be substantiated in the clinical setting might have less relevance to the average consumer.

The responsibility is with the study sponsor to determine the relevance of their claims within the context of the average consumer and legislative compliance, especially since clinically generated data needs to be supported by end user relevant studies. Claims and product development will require a clearer differentiation between consumer expectations, understanding, and marketing desire.

Concluding Remarks

Since sweating is the body's natural response in regulating body temperature, educating both the consumer and product developer on the requisite nature and validity of body perspiration is of clear importance. Moreover, blocking the sweat process for prolonged periods will cause compensatory sweating elsewhere and the potential for safety concerns. Internationally accepted standards have been developed to evaluate the efficacy of antiperspirants and deodorants on both, sweat and malodor intensity. Since deodorants and antiperspirants clearly influence axillary microbiome further research and development is an exciting opportunity to generate new strategies and test methodologies. Investigations into microbiome changes during adolescence and menopause, the influence of diet, lifestyle, and environment, are all valid approaches in the pursuit of uniquely tailored perspiration and odor management. The evolution of clinical testing methods alongside legislative demands will provide the opportunity to meet these challenges.

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References

[1] *Labows J, McGinley K, Kligman A.* Perspectives on axillary odor. J Soc Cosmet Chem 1982; 34:193-202.

[2] *Zeng X-N, Leyden J, Spielman A, et al.* Analysis of the characteristic human female axillary odors: qualitative comparison to males. J Chem Ecol 1996a; 22:237-57.

[3] *Preti G, Wysocki C, Barnhart K, et al.* Male axillary extracts contain pheromones that affect pulsatile secretion of luteinizing hormone and mood in female recipients. Biol Reprod 2003; 68:2107-13.

[4] *Wysocki C, Preti G* (2009). Human pheromones: what's purported, what's supported. A Sense of Smell Institute White Paper <http://senseofsmell. org/papers/Human_Pheromones_Final %207-15-09.pdf>.

[5] *Zeng C, Spielman A, Vowels B, et al.* A human axillary odorant is carried by apolipoprotein D. Proc Natl Acad Sci USA 1996b; 93: 6626-30.

[6] *Flower D.* The lipocalin protein family: structure and function. Biochem J. 1996; 318:1-14.

[7] *Spielman A, Zeng X-N, Leyden J, et al.* Proteinaceous precursors of human axillary odor: isolation of two novel odor binding proteins. Experientia 1995; 51:40-47.

[8] *Shehadeh N, Kligman A.* Effect of topical antibacterial agents on the bacterial flora of the axillae. J Invest Dermatol 1963a; 40:61-71.

[9] R*ennie P, Gower D, Holland K. In vitro* and *in vivo* studies of human axillary odour and the cutaneous microflora. Br J Dermatol 1991; 124:596-602.

[10] *Gower D, Ruparelia B.* Olfaction in humans with special reference to odorous 16- androstenes: their occurrence, perception and possible social psychological and sexual impact. J Endocrinol 1993; 137:167-87.

[11] *Natsch A, Gfeller H, Gygax P et al.* A specific bacterial aminoacylase cleaves odorant precursors secreted in the human axilla. J Biol Chem 2003; 278:5718-27.

[12] *Natsch A, Schmid J, Flachsmann F.* Identification of odoriferous sulfanylalkanols in human axilla secretions and their formation through cleavage of cysteine precursors by a C-S lyase isolated from axilla bacteria. Chem Biodiversity 2004; 1:1058-72.

[13] *He J, Wang T, Dong J.* A close positive correlation between malodor and sweating as a marker for the treatment of axillary bromhidrosis with botulinum toxin A. J Dermatolog Treat 2012; 23(6): 461-64.

[14] *Guillet G, Zampetti A, Aballain-Colloc M.* Correlation between bacterial population and axillary and plantar bromidrosis: study of 30 patients. Eur J Dermatol 2000; 10(1):41-42.

[15] *Leyden J, McGinley K, Holzle E, Labows J, Kligman A.* The microbiology of the human axilla and its relationship to axillary odor. J Invest Dermatol 1981; 77(5):413-16.

[16] *Lindsay S, Holmes S, Corbett A, Harker M, Bovell D.* Innervation and receptor profiles of the human apocrine (epitrichial) sweat gland: routes for intervention in bromhidrosis. Br J Dermatol 2008; 159(3):653-60.

[17] *Natsch A, Derrer S, Flachsmann F, Schmid J.* A broad diversity of volatile carboxylic acids, released by a bacterial aminoacylase from axilla secretions, as candidate molecules for the determination of human-body odor type. Chem Biodivers 2006; 3(1):1-20.

[18] *Spielman AI, Sunavala G, Harmony J, et al.* Identification and immunohistochemical localization of protein precursors to human axillary odors in apocrine glands and secretions. Arch Dermatol 1998; 134(7):813-8.

[19] *Zeng C, Spielman A, Vowels B, Leyden J, Biemann K, Preti G.* A human axillary odorant is carried by apolipoprotein D. Proc Natl Acad Sci U S A 1996; 93(13):6626-30.

[20] *Sato T, Sonoda T, Itami S, Takayasu S.* Predominance of type I 5alpha-reductase in apocrine sweat glands of patients with excessive or abnormal odour derived from apocrine sweat (osmidrosis). Br J Dermatol 1998; 139(5):806-10.

[21] *Troccaz M, Gaïa N, Beccucci S, et al.* Mapping axillary microbiota responsible for body odours using a culture-independent approach. Microbiome 2015; 3:3.

[22] *Parera E, Sinclair R.* Hyperhidrosis and bromhidrosis - A guide to assessment and management. Australian Family Physician 2013; 42(5):266-299.

[23] *Mao G, Yang S, Zheng J.* Etiology and management of axillary bromidrosis: a brief review. Int J Dermatol 2008; 47(10):1063-68.

[24] *Mao GY Yang S, Zheng J.* Cause of axillary bromidrosis. Plast Reconstr Surg 2009; 123(2):81e-82e.

[25] *Schommer N, Gallo R.* Structure and function of the human skin microbiome. Trends Microbiol 2013; 21(12):660-68.

[26] *Eloe-Fadrosh E, Rasko D.* The human microbiome: from symbiosis to pathogenesis. Ann Rev Med 2013; 64:145-63.

[27] *Abdallah F, Mijouin L, Pichon.* Skin Immune Landscape: Inside and Outside the Organism. Mediators Inflamm 2017; 2017:5095293.

[28] *Stenzaly-Achtert S, Schölermann A, Schreiber J, Diec K, et al.* Axillary pH and influence of deodorants. Skin Research and Technology 2000; 6:87-91.

[29] *Rosseland B, Eldhuset T, Staurnes M.* Environmental effects of aluminium. Environ Geochem Health 1990; 12(1-2):17-27.

[30] *Exley C.* Human exposure to aluminium. Environ. Sci.: Processes Impacts 2013; 15(10):1807-1816.

[31] *Kataoka A.* Surgical treatment of bromhidrosis. Rev. Bras. Cir. Plast 2017; 32(3):377-382

[32] *Herbst F, Plas E, Fugger R, et al.* Endoscopic thoracic sympathectomy for primary hyperhidrosis of the upper limbs: a critical analysis and long-term results of 480 operations. Ann Surg 1994; 220:86-90.

[33] *Swartling C, Naver H, Lindberg M.* Botulinum A toxin improves life quality in severe primary focal hyperhidrosis. Eur J Neurol 2001; 8(3):247-52.

[34] *Wolosker N, Milanez de Campos J, Kauffman P, et al.* The use of oxybutynin for treating axillary hyperhidrosis. Ann Vasc Surg 2011; 25:1057–62.

[35] *Bretagne A, Cotot F, Arnaud-Roux M, et al.* The mechanism of eccrine sweat pore plugging by aluminium salts using microfluidics combined with small angle X-ray scattering. Soft Matter 2017; 13(20):3812-21.

[36] *Shehadeh N, Kligman A.* The effect of topical antibacterial agents on the bacterial flora of the axillae. J Invest Dermatol 1963a; 40:61-71.

[37] *S. Bhattacharjee, Zhao Y, Hill J M, et al.* Selective accumulation of aluminum in cerebral arteries in Alzheimer's disease (AD) J Inorg Biochem 2013; 126:35-37.

[38] *Darbre P, Mannello F, Exley C.* Aluminium and breast cancer: Sources of exposure, tissue measurements and mechanisms of toxicological actions on breast biology. J. Inorg Biochem 2013; 128:257-61.

[39] *Klotz K, Weistenhöfer W, Neff F, et al.* The health effects of aluminum exposure. Dtsch Arztebl Int 2017; 114:653-59.

[40] *Urban J, Fergus D, Savage A, et al.* (2016). The effect of habitual and experimental antiperspirant and deodorant product use on the armpit microbiome. PeerJ 4:e1605; DOI 10.7717/peerj.1605

[41] *Callewaert C, Lambert J, Van de Wiele T.* Towards a bacterial treatment for armpit malodour Exp Dermatol 2017; 26(5):388-91.

[42] *Callewaert C, Hutapea P, Van de Wiele T, Boon N.* Deodorants and antiperspirants affect the axillary bacterial community. Arch Dermatol Res 2014; 306(8):701-10.

[43] Commission Regulation European Union: (EU) No 655/2013. 10 July 2013. Laying down common criteria for the justification of claims used in relation to cosmetic products.

[44] *Keyhani R, Scheede S, Thielecke I, et al.* Qualification of a precise and easy to handle sweat casting imprint method for the prediction and quantification of anti-perspirant efficacy. International journal of cosmetic science. 2009; 31(3):183-92.

[45] *Muller S A, Kierland R R.* The Use of a Modified Starch-Iodine Test for Investigating Local Sweating Responses to Intradermal Injection of Methacholine1. Journal of Investigative Dermatology 1959; 32(2):126-128.

[46] Antiperspirant Drug Products For Over-the-Counter Human Use; Final Monograph. 21 CFR Parts 310, 350, and 369. Federal Register/Vol. 68, No. 110/2003/Rules and Regulations 34273-34293.

[47] Standard Guide for Sensory Evaluation of Axillary Deodorancy ASTM E1207-14. American Society for Testing Materials – https://www.astm.org/Standards/E1207.htm.

[48] *Maxeiner B, Ennen J, Rützel-Grünberg S, Traupe B, Wittern K.-P., Schmucker R, Keyhani R.* Design and application of a screening and training protocol for odour testers in the field of personal care products. International Journal of Cosmetic Science 2009; 31: 193-199.

[49] DIN EN 13725:2003-07 Luftbeschaffenheit – Bestimmung der Geruchsstoffkonzentration mit dynamischer Olfaktometrie; Deutsche Fassung EN 13725:2003 (Air quality – Determination of odour concentration by dynamic olfactometry).

[50] *Piérard G, Elsner P, Marks R, Masson P, Paye M*; EEMCO Group. EEMCO guidance for the efficacy assessment of antiperspirants and deodorants. Skin Pharmacol Appl Skin Physiol 2003; 16(5):324-42.

[51] *Bielfeldt S, Frase T, Gassmueller J.* New sensitive method for assessment of antiperspirants with intraindividual comparison of up to eight formulations. SOFW 1997; 123(10):639-642.

[52] *Brandt M, Bielfeldt S, Springmann G, Wilhelm K.-P.* Influence of climatic conditions on antiperspirant efficacy determined at different test areas. Skin Research and Technology 2008; 14:213-219.

[53] *Langille M, Zaneveld J, Caporaso J, et al.* Predictive functional profiling of microbial communities using 16S rRNA marker gene sequences. Nature biotechnology 2013; 31(9):814-821. doi:10.1038/nbt.2676.

[54] *Martin A, Hellhammer J, Hero T, et al.* Effective prevention of stress-induced sweating and axillary malodour formation in teenagers. Int J Cosmet Sci 2011; 33:90-97.

[55] *Kirschbaum C, Pirke K, Hellhammer D.* The ‚Trier Social Stress Test'-a tool for investigating psychobiological stress responses in a laboratory setting. Neuropsychobiology 1993; 28:76-81.

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