



Skin penetration and antioxidant effect of cosmeto-textiles with gallic acid



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ABSTRACT

In this work, the antioxidant gallic acid (GA) has been encapsulated in microspheres prepared with poly-ε-caprolactone (PCL) and incorporated into polyamide (PA) obtaining the cosmeto-textile. The topical application of the cosmeto-textile provides a reservoir effect in the skin delivery of GA. The close contact of the cosmeto-textile, containing microsphere-encapsulated GA (ME-GA), with the skin and their corresponding occlusion, may be the main reasons that explain the crossing of active principle (GA) through the skin barrier, located in the stratum corneum, and its penetration into the different compartments of the skin, epidermis and dermis.

An *ex vivo* assessment was performed to evaluate the antioxidant effect of the ME-GA on the stratum corneum (SC) using the thiobarbituric acid-reactive species (TBARS) test. The test is based on a non-invasive *ex vivo* methodology that evaluates lipid peroxides formed in the outermost layers of the SC from human volunteers after UV radiation to determine the effectiveness of an antioxidant. In this case, a ME-GA cosmeto-textile or ME-GA formulation were applied to the skin *in vivo* and lipid peroxidation (LPO) in the horny layer were determined after UV irradiation. This methodology may be used as a quality control tool to determine *ex vivo* the percentage of LPO inhibition on human SC for a variety of antioxidants that are topically applied, in this case GA.

Results show that LPO formation was inhibited in human SC when GA was applied directly or embedded in the cosmeto-textile, demonstrating the effectiveness of both applications. The percentage of LPO inhibition obtained after both topical applications was approximately 10% for the cosmeto-textile and 41% for the direct application of microspheres containing GA. This methodology could be used to determine the effectiveness of topically applied antioxidants encapsulated in cosmeto-textiles on human SC.

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1. Introduction

Biofunctional textiles are materials that exert a biological effect on human skin. Such textiles constitute the basis for the delivery system of cosmetic or pharmaceutical substances when the textiles come into contact with the skin [1]. Active substances are commonly incorporated into the vehicles, which may break when a garment rubs the skin, allowing for the release of the compounds directly to be absorbed by the skin. These cosmeto-textiles, with a slow release of the active compound into the skin, may help people with sensitive skin.

In fact, there are several textile products currently in the market that claim to have properties usually found in cosmetics [2], such as moisturizing, slimming, energizing, refreshing, relaxing, or vitalizing properties, as well as UV protection [3–4] or simply fragrance. Thus, there is a real need to develop test methods to verify the effectiveness and durability of these claimed properties [5].

The specific case of UV protection, sunscreen lotions, clothing, and shade structures provide protection from the deleterious effects of ultraviolet radiations (UV). The formation of reactive oxygen species (ROS) with UV exposure and their effect on lipids have also been extensively studied [6]. ROS have been implicated lipid oxidation [7], which can alter tissue structure via cross-linking, fragmentation, etc. ROS was part of the damage processes in SC. Some studies [8] demonstrated a marked decrease in intercellular delamination energy with increasing UV exposure indicating UV radiation causes a significant decrease in cellular cohesion and thus an alteration of intercellular lipid or corneodesmosome structure.

In case of cloths when UV hits the textile, different types of interactions occur depending upon the substrate and its conditions. The UV protection by textile materials is a function of the chemical characteristics, physico-chemical type of fiber, presence of UV absorbers, construction of fabric, thickness, porosity, extension of the fabric, moisture content of the fabrics, color and the finishing given to the fabric. The UV transmitted through textile fabrics consists of the unchanged waves that pass through the interstices of the fabric as well as scattered waves that have interacted with the fabrics. Another part is absorbed when it penetrates the sample and is converted into a different energy

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form. The portion of radiation that travels through the fabric and reaches the skin is referred to as the transmission component [9]. This portion of radiation could be one of the reasons to study the antioxidant protection effect of a biofunctional textile prepared with an antioxidant active.

Gallic acid (GA) is a polyhydroxyphenolic compound present in leafy vegetables, fruits, and nuts [10–12]. GA exhibits variety of biological activities including antioxidant [13–14], anti-tumor [11,15–17], anti-inflammatory [18], and anti-bacterial [19–20]. Several line of evidence has shown that GA inhibits tumor cell growth, migration, and invasion in vitro [21]. GA has recently been applied in cosmeo-textiles as an active component [22].

In this work, the GA was selected and incorporated into polyamide (PA) through microspheres prepared from poly- ϵ -caprolactone (PCL). PCL is an aliphatic polyester having good chemical resistance to solvents [23]. It is biodegradable and non-toxic to the human body [24]. And, it has the advantage of controlling the release of the active principle over a period of several days to several weeks in contrast to natural hydrophobic polymers, which have a shorter period of release [25]. Therefore PCL microspheres have been used as a vehicle for textile application to study the absorption and desorption properties thereof when incorporated into PA [22].

Polyamide (PA) was used as a textile fabric in this work to obtain biofunctional textiles because of their comfort when in contact with the skin. PA has been used for a number of industrial, apparel, and medical applications, such as wound sutures, artificial tendons, and medical packaging, due to its excellent wear resistance, strength, toughness, elastic recovery, low initial modulus, appearance retention, ease of coloration, and high resistance to rupture [26].

Using the in vitro methodology of percutaneous absorption, it is possible to detect the amount of the active principle that penetrates each skin layer from a given cosmeo-textile [27]. It is reasonable to assume that the reservoir capacity of a cosmeo-textile, close contact with the skin and the corresponding skin occlusion may be the main factors that determine how an active principle (e.g., GA) crosses the skin barrier, located in the stratum corneum, and penetrates the different compartments of the skin. The in vitro skin delivery of PA containing ME-GA has previously been explored, the results obtained suggested that GA penetrates through the skin detecting at different skin layers (stratum corneum, epidermis, and dermis) [22].

The aim of this work was to assess the antioxidant efficacy of a cosmeo-textile containing microsphere-encapsulated GA. Our study involves a topical antioxidant strategy to prevent damage to skin specially the lipid fraction. The antioxidant capacity was determined using human volunteers after topical cosmeo-textile application. An ex vivo assessment was performed to evaluate the protection effect on the stratum corneum (SC) after cosmeo-textile application. The lipid peroxide formation was determined using the thiobarbituric acid-reactive species test (TBARS). This test is a non-invasive ex vivo method that uses tape strips of the outermost layers of the SC from human volunteers to evaluate the effectiveness of an antioxidant [28].

2. Materials and Methods

2.1. Materials

The textile bandages used were knitted fabrics (plain stitch) of polyamide 78/68/1 (DeFiber, S.A., Spain). Poly(vinyl alcohol) (PVA) (87–89% hydrolyzed, MW 31 000–50 000 Da) was used as a dispersant for microsphere preparation; poly- ϵ -caprolactone (PCL) (MW 45 000 Da) was used as the microsphere polymer. Both PVA and PCL were supplied by Sigma–Aldrich (Madrid, Spain). Gallic acid (GA), as the active ingredient, was supplied by Sigma–Aldrich (Madrid, Spain). All chemicals used were of analytical grade. Methanol (HPLC grade) and phosphoric acid were supplied by Merck (Darmstadt, Germany).

2.2. PCL Microspheres

The solvent evaporation method was used to obtain microspheres by forming microemulsions ($w_1/o/w_2$ double emulsion) [22]. The preparation procedure was carried out twice to obtain a sufficient volume of microspheres for all textile bandage applications.

Briefly, 20 ml of a 2.91% (w/w) dispersion of GA in water was added to 20 ml of 2.91% (w/w) PCL in dichloromethane. A simple emulsion (w_1/o) was generated by mechanical agitation (ULTRA-TURRAX T25, IKA) for 25 min at 24 000 rpm. This simple emulsion was then added to a continuous phase consisting of 200 ml of an aqueous PVA solution (1.96% (w/w)) and was emulsified for 30 more minutes at 20 000 rpm, resulting in a double emulsion ($w_1/o/w_2$). The method used was carried out at 4 °C [29]. The mixture was maintained under agitation at 400 rpm (20 h) at room temperature, leading to solvent evaporation and consequently microsphere formation. The percentage of GA in the formulation is 0.49% (w/v) and its percentage related to microspheres without water was 11.60% (w/w).

PCL microspheres of gallic acid (ME-GA) (mixture of both preparations) were applied to textiles (322 ± 1 cm² area) by bath exhaustion, with a bath ratio of 1/5 (1 g textile per 5 ml of treatment bath), at 50 °C for 60 min with manual stirring performed every 10 min. To quantify the amount of product absorbed onto the fabrics, the dry samples were weighed before and after 24 h of application under standard ambient conditions (23 ± 2 °C and $55 \pm 5\%$ relative humidity).

The GA present in the formulations and absorbed onto the fabrics was extracted with methanol during 15 min under ultrasounds and quantified using a Hitachi-Merck HPLC equipped with an L-6000 Intelligent Pump, AS-4000 Autosampler and L-4250 UV–Vis Detector. The column used was a LiChrocart 250–4/Lichrosorb RP-18 (5 mm) (Darmstadt, Germany). The mobile phase was 90% water (with 0.7% H₃PO₄)/10% methanol flowed at a rate of 1 ml/min. GA detection was conducted at 280 nm with a retention time of 6.9 min. The area below the peak in the chromatogram was used to calculate the concentration of GA using external standards that displayed linearity over a concentration range of 0.25 to 100 mg/ml. The equation yielding the regression line through the experimental values was $ABS = 15,967 \cdot [GA] + 4417.1$, $r^2 = 0.9999$. This analytical methodology was fully validated.

2.3. Size of PCL Microspheres

The size distribution and polydispersity of the PCL microspheres were measured by dynamic light scattering (DLS) (Zetasizer Nano ZS ZEN3600; Malvern Instruments Ltd., Malvern, Worcestershire, UK). The non-invasive backscattering technology was used to minimize multiple scattering effects without the need for sample dilution. The size distribution and polydispersity measurements were performed at room temperature with polystyrol/polystyrene cells (Ref 67.754 Sarstedt). The scattered light was detected at an angle of 173°. Each sample was measured in triplicate. The data were interpreted by considering the size distribution by intensity. All data were collected and analyzed using the program DTS (dispersion technology software) provided by Malvern Instruments Ltd.

2.4. Volunteers

The experimental protocol was conducted with 8 healthy Caucasian volunteers (all women) with phototypes II, III, and IV [30]. The mean

Table 1
Age and phototype of volunteers.

Volunteer	1	2	3	4	5	6	7	8	Mean \pm SD
Age	24	24	28	60	34	36	33	55	36.8 \pm 13.6
Phototype	II	III	IV	II	IV	IV	IV	III	

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