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# Formulation Development and Characterization of Face Masks Containing Natural Pink Clay

# Desenvolvimento de Formulação e Caracterização de Máscaras Faciais Contendo Argila Rosa Natural

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**Abstract**

Pink clay (PC) is a soft, loose, fine-grained earth material composed by a mixture of particles of white and red clay minerals, such as the red hematite, and is less absorbent but a very soft clay. The main goal was to develop facial masks with PC ranging between 10 - 50 (% w/w) and characterize by pH, relative density, apparent viscosity and spreadability. PC powder was subjected to sterilization by dry heating and autoclaving. Both sterilization processes were very efficient to reduce the number of bacteria and fungi to <10 CFU/g, and the pathogens *E. coli*, *P. aeruginosa*, *S. aureus* and *C. albicans* were absent. The face masks produced with the sterilized PC yielded acidic pH (4.1 – 4.7) and similar relative density (1.06 – 1.43 g/mL). The apparent viscosity of the face masks changed with the different rotation speeds employed, decreasing from 11,416 ± 115 cP (at 5 rpm) to 6,964 ± 156 cP (at 10 rpm) and 4,016 ± 98 cP (at 20 rpm), for the 30% PC face mask, which is an indicative of the pseudoplastic behavior. Moreover, the face masks viscosity increased as the clay proportion also increased from 10% to 50% (w/w) and, inversely, spreadability was decreased. Despite that, spreadability was considered good, in the range from 1.55 to 7.73 cm$^2$. Taken together, the results indicate that the intermediate proportion of 20 or 30% (w/w) of this PC in the face masks would be more suitable for the intended cosmetic use.

**Keywords:** Clay Masks. Face Skin. Functional Cosmetics. Geocosmetics. Minerals. Rheology.

**Resumo**

*A argila rosa (AR) é um material terroso macio, solto e de grão fino composto por uma mistura de partículas minerais de argila branca e vermelha, tais como hematita vermelha, sendo menos absorvente, mas uma argila muito macia. O principal objetivo foi desenvolver máscaras faciais com AR na faixa de 10 – 50 (% p/p) e caracterizar por pH, densidade relativa, viscosidade aparente e espalhabilidade. AR em pó foi submetida a esterilização por calor seco e autoclavação. Os processos de esterilização foram muito eficientes para reduzir o número de bactérias e fungos para <10 UFC/g e os patógenos E. coli, P. aeruginosa, S. aureus e C. albicans estiveram ausentes. Máscaras faciais produzidas com a AR esterilizada produziram pH ácido (4,1 - 4,7) e densidade relativa semelhante (1,06 - 1,43 g/mL). A viscosidade aparente das máscaras faciais mudou com as diferentes velocidades de rotação empregadas, diminuindo de 11.416 ± 115 cP (a 5 rpm) para 6.964 ± 156 cP (a 10 rpm) e 4.016 ± 98 cP (a 20 rpm), para a máscara facial com 30% AR, que é um indicativo do comportamento pseudoplástico. Além disso, a viscosidade das máscaras faciais aumentou à medida que a proporção de argila também aumentou de 10% para 50% (p/p) e, inversamente, a espalhabilidade diminuiu. Apesar disso, a espalhabilidade foi considerada boa, na faixa de 1,55 a 7,73 cm$^2$. Em conjunto, os resultados indicam que a proporção intermediária de 20 ou 30% (p/p) desta AR em máscaras faciais seria mais adequada para o uso cosmético pretendido.*

**Palavras-chave:** *Cosméticos Funcionais. Geocosméticos. Máscaras de Argila. Pele Facial. Reologia.*

## 1 Introduction

Throughout human history, clays have been used for religious, artistic, cosmetic and therapeutic purposes, and clay face masks are considered one of the oldest cosmetic preparations. Some reports indicate that the Egyptians and the Greeks already used these preparations for skin cleaning (MOREKHURE-MPHAHLELE *et al*., 2016; ZAGUE *et al*., 2007). Clays are soft, loose, fine-grained, earthy materials consisting of small crystalline particles of clay minerals, which are chemically composed by hydrated aluminum, iron and magnesium silicates, with organic matter, soluble salts, quartz particles, pyrite, calcite, among other minerals (BURITI *et al*., 2019; MATOLA *et al*., 2021).

As abundant minerals found in the nature, clays are inexpensive and sustainable materials with important applications in many fields, including cosmetology. They have the ability of dirt and oils adsorption from the skin surface, while promoting light mechanical exfoliation, with superficial cells remotion and boosting the cell turnover. In addition, clays have moisturizing properties and can be considered anti-aging bioactives (CARRETERO *et al*., 2002; ZAGUE *et al*., 2007).

The clay composition, its layered structure and the colloidal size of its particles are the main driving factors of the cosmetic properties and the rheological characteristics of the final product (BURITI *et al*., 2019; DANELUZ *et al*., 2020;

MATTIOLI *et al*., 2015; MOREKHURE-MPHAHLELE *et al*., 2016). The presence of trace elements in the composition brings important contributions to the cosmetic applications, since iron can act as an antiseptic and booster for cell turnover, silicon helps to rebuild skin tissues, with moisturizing and soothing effects, while zinc and magnesium are essential micronutrients very useful for the skin metabolism (FAVERO *et al*., 2016; GOMES; SILVA, 2007; MACHADO *et al*., 2018; MATIKE *et al*., 2011; MATTIOLI *et al*., 2015).

Clays are found in nature in a wide variety of types, colors and functions. Pink clay (PC) is a mixture of white and red clays that is rich in red hematite, which is usually less absorbent and softer than green clay. Its special composition can include quartz, smectite, illite, kaolinite, and the pink color is usually related with the presence of $Fe^{3+}$ as hematite $\alpha Fe_2O_3$. Thus, PC promotes remarkable antioxidant and soothing effects on the skin and is commonly utilized on dry and sensitive face skin (BURITI *et al*., 2019; LÓPEZ-GALINDO; VISERAS, 2004; RIBEIRO, 2010).

Face masks are cosmetics designed to clean, soften, stimulate or refresh the face skin. These products are applied topically to the face, remaining for a certain period of time, while they get dry and harden. Thus, these products must have appropriate consistency and viscosity, in addition to being smooth, adherent, easy to handle and should cause a pleasant sensation when applied to the skin (MATIKE *et al*., 2011).

The medicinal, therapeutic and aesthetic use of clays must meet several safety requirements, which involves chemical and microbiological stability. Considering its natural source, the control of microbial load becomes an essential requirement for the clays quality control (BURITI *et al*., 2019; MATTIOLI *et al*., 2015). Thus, this study aims to develop and analyze the properties of pink clay in different proportions in the applicability in cosmetic formulations of PC face mask, evaluating its physicochemical characteristics and the PC powder sterilization process by microbiological analysis.

## 2 Material and Methods

Pink clay was obtained in Diamantina, Jequitinhonha Valley, Brazil (18.086398, 43.744017). The mask components were cetostearyl alcohol (Brenntag Quimica, Brazil), cetrimonium chloride (Química Anastacio, Brazil), methylparaben (Isofar, Brazil), propylparaben (EMFAL, Brazil), and glycerin (Isofar, Brazil).

For the microbiological analysis, the following growth media were used: Tryptic *Soy Broth (Kasvi, Spain)*, Sabouraud dextrose broth (HI Media, India), Tryptic *Soy Agar (Kasvi, Spain)*, Sabouraud dextrose agar (*Kasvi, Spain)*. All the other reagents and solvents used were of analytical grade.

### 2.1 Development of the pink clay face mask

In the composition of the PC face mask, cetostearyl alcohol was used as emulsion stabilizer and viscosity-increasing agent; cetrimonium chloride was used as emulsifier and conditioning agent; glycerin was present as humectant and for smoothness improvement; and a combination of methylparaben and propylparaben was the preservative system. Five samples were proposed based on the PC quantitative variations from 10% to 50% (w/w).

In order to prepare the PC face mask (batch 100 g), the oily phase (OP) containing 2 g cetostearyl alcohol and 0.1 g propylparaben, and the aqueous phase (AP) comprising 0.5 g cetrimonium chloride, 7 g glycerin, and 0.15 methylparaben in water, were separately heated at 75 ± 1 °C. Then, the AP was directly poured into the OP with constant mechanical stirring at 200 ± 25 rpm (Quimis Aparelhos Científicos, Brazil), and the PC was immediately incorporated to this hot emulsion. The homogenization was kept until the product reached the room temperature.

### 2.2 Microbiological analysis of the pink clay

#### 2.2.1 Microbial enumeration tests

For the quantitative enumeration of mesophilic bacteria and fungi, a 1:10 dilution of the sample was made in Tryptic *Soy Broth* with 0.1% (w/v) polysorbate 80 and, from this one, successive 10-fold dilutions were made in Tryptic *Soy Broth* until 1:10,000. For bacteria enumeration, 1 mL of each sample dilution was removed and placed on plates (n = 2) with melted and cooled soybean-casein digest agar (depth method). The plates were incubated at 32.5 ± 2.5 °C for 3-5 days and the colony-forming unit (CFU) of bacteria was determined (ANVISA, 2019).

For the fungi enumeration, 1 mL of each sample dilution was placed on plates (n = 2) with melted and cooled Sabouraud dextrose agar (depth method) was added. The plates were incubated at 22.5 ± 2.5 °C for 5-7 days and the fungi CFU was counted. All the results were expressed in CFU/g of sample and negative controls of the culture media were properly performed (ANVISA, 2019).

#### 2.2.2 Determination of the absence of specified microorganisms

Tests for the pathogens *Escherichia coli* (*E. coli*), *Pseudomonas aeruginosa* (*P. aeruginosa*), *Staphylococcus aureus* (*S. aureus*) and *Candida albicans* (*C. albicans*) were carried out (ANVISA, 2019; SCCS, 2021). A 1:10 dilution was performed in Tryptic *Soy Broth* for the bacteria and in Sabouraud dextrose broth for *C. albicans*. From this dilution, inoculation was performed in selective media for detection of each microorganism species, which were then incubated, and identification tests were performed.

According to the European Union’s Scientific Committee on Consumer Safety (SCCS, 2021), these microorganisms must be absent in 1 g of cosmetic product.

### 2.2.3 Sterilization process

Considering the natural source of the PC powder, sterilization methods by dry heating and moist heating were proposed. Thus, approximately 5 g of PC were subjected to sterilization in the following conditions: dry heating at 250 °C for 2 hours (sterilization oven model 3; ICAMO, Brazil) and autoclaving at 121 °C for 15 minutes (autoclave Phoenix Luferco, SP, Brazil). The efficiency of these processes was determined by microbiological analysis: tests for microbial enumeration and absence of pathogens (ANVISA, 2019).

## 2.3 Characterization of the pink clay face masks

### 2.3.1 Determination of pH

The pH of the samples of PC face masks was determined by dispersing each sample in distilled water pH 7.0 (10% w/w) and direct determination at 25 ºC in a pre-calibrated Micronal pHmeter B474 (ANVISA, 2008).

### 2.3.2 Relative density

The relative density (specific gravity) was determined using a clean and dry pycnometer with a constant volume of 25 mL. The pycnometer was weighed empty, then full of water, and finally filled with the face mask samples, at 25 °C. The relative density was calculated by the net weight ratio of the samples to the water net weight (ANVISA, 2008).

### 2.3.3 Apparent viscosity

The PC face masks were analyzed in the Quimis Rotary Viscometer Q860M (spindle 3), at 5, 10 and 20 rpm. The percentage of adjustment was kept in the range from 55–75%. The apparent viscosity values (cP, centipoise) were registered after 2 minutes of agitation at room temperature (ANVISA, 2008).

## 2.4 *In vitro* spreadability

For the spreadability determination of the PC face masks, the method adapted from Lange *et al.* (2009) was utilized. The equipment consisted of a circular glass mold plate (diameter = 20 cm; thickness = 0.3 cm), with a central hole of 1.2 cm diameter, placed on a support glass plate (20 cm x 20 cm), which was positioned over a sheet of graph paper. The hole in the mold plate was completely filled with the sample, the surface was leveled and then the mold plate was removed. A 5-g glass plate was carefully placed on the sample and the diameter was registered in two opposite positions, using the scale of the graph paper, to calculate the average diameter (d) (LANGE *et al*., 2009).

The spreadability (S) at the temperature of 25 ± 1 °C was then calculated using the equation:

## 2.5 Statistical analysis

All determinations were performed in triplicate and the results were expressed as average value ± standard deviation. One-way analysis of variance (ANOVA) followed by Tukey's test was used to determine the statistical differences among the average values ($\alpha$ = 0.05), using the GraphPad Prism 6.0 software.

# 3 Results and Discussion

## 3. 1 Microbiological analysis of pink clay

Initially, the mesophilic microorganisms in the fresh PC powder were enumerated before and after being incorporated into the face mask, and the absence of some pathogens were also verified (Table 1). High numbers of both microorganisms were found: 3.9 x $10^4$ CFU/g for bacteria and 4.7 x $10^4$ CFU/g for fungi, probably due to the natural source of this raw material, which was collected in nature and was not subjected to pre-treatments to reduce the number of microorganisms. After incorporating the PC powder into the face mask, a reduction in the number of microorganisms was observed (1.3 x $10^4$ CFU/g and 0.9 x $10^4$ CFU/g for bacteria and fungi, respectively), probably due to the preservative system and the heating process. The presence of the pathogens *E. coli*, *P. aeruginosa*, *S. aureus* and *C. albicans* was not detected in the fresh PC samples and in the PC face mask.

**Table 1 -** Microbiological examination: microbial enumeration and tests for pathogens

| Microorganism examination | Without pre-sterilization | | After dry heating | | After moist heating | |
|---|---|---|---|---|---|---|
| | PC | Face mask | PC | Face mask | PC | Face mask |
| **Bacteria (UFC/g)** | 3.9 x $10^4$ | 1.3 x $10^4$ | <10 | <10 | <10 | <10 |
| **Fungi (UFC/g)** | 4.7 x $10^4$ | 0.9 x $10^4$ | <10 | <10 | <10 | <10 |
| ***E. coli*** | Absent | Absent | Absent | Absent | Absent | Absent |
| ***P. aeruginosa*** | Absent | Absent | Absent | Absent | Absent | Absent |
| ***S. aureus*** | Absent | Absent | Absent | Absent | Absent | Absent |
| ***C. albicans*** | Absent | Absent | Absent | Absent | Absent | Absent |

**Source:** Research data.

However, the very high number of mesophilic microorganisms (bacteria and fungi) found in both samples (fresh PC powder and face mask) was above the maximum limit of 1 x $10^3$ CFU/g allowed by the European legislation (SCCS, 2021) or 5 x $10^3$ UFC/g allowed by the the Brazilian legislation (ANVISA, 1999). Therefore, the microbial enumeration was performed after the sterilization treatments by dry heating and moist heating, proposed to reduce the number of microorganisms present in the fresh PC powder (Table 1).

Both treatments were efficient to reduce the number of mesophilic bacteria and fungi of PC to <10 UFC/g, which remained low even after incorporation in the face mask, below the maximum limit established by the European and Brazilian legislation. None of the pathogens were also detected in these samples. Therefore, both heat sterilization processes can be used to reduce the number of microorganisms found in the fresh PC powder, in accordance with previous reports that used the same methods (ANVISA, 2019; ANVISA, 1999; PETERLE *et al*., 2014).

## 3.2 Characterization of the clay face masks

### 3.2.1 pH and relative density

Determination of pH values of the cosmetic products under development is an important initial analysis, since these values are directly related with the compatibility of the formulation components, stability over time, and compatibility with the skin, thus influencing the product efficacy and safety for use. The pH values of all samples of face mask were in the range between 4.1 and 4.7, regardless the PC concentration (Table 2), which is compatible with the slightly acidic pH value of the face skin (4.6 – 5.8) (LEONARDI *et al*., 2002).

**Table 2 -** Characterization of the face masks containing different proportion of pink clay in the composition

| PC proportion (w/w) | pH | Relative density |
|---|---|---|
| **10%** | 4.7 ± 0.10 | 1.0643 ± 0.0014 |
| **20%** | 4.5 ± 0.07 | 1.1273 ± 0.0008 |
| **30%** | 4.3 ± 0.01 | 1.1175 ± 0.0005 |
| **40%** | 4.1 ± 0.02 | 1.1518 ± 0.0051 |
| **50%** | 4.4 ± 0.37 | 1.4322 ± 0.0026 |

**Source:** Research data.

Slightly acidic pH values also have contribution in skin protection against bacteria and fungus proliferation, yielding the adequate conditions for the resident bacterial flora to stay more strongly attached to the skin. Determination and control of cutaneous pH after application of cosmetic products is very useful to avoid the use of inappropriate topical products, which may alter the natural skin pH, as the surface acidic pH is decisive for the conditions of growth balance between the resident microflora and the transient opportunistic microflora that can be potentially pathogenic (FLUHR; ELIAS, 2002; LAMBERS *et al*., 2006; LEONARDI *et al*., 2002; MELO; CAMPOS, 2016). Furthermore, the study of pH changes on the skin surface during exposure to the cosmetic ingredients is also relevant in clinical research, since the application of the developed product on the skin should not decrease its buffering capacity, for the skin may become locally more vulnerable to aggressive agents, especially microorganisms (LEONARDI *et al*., 2002; MELO; CAMPOS, 2016).

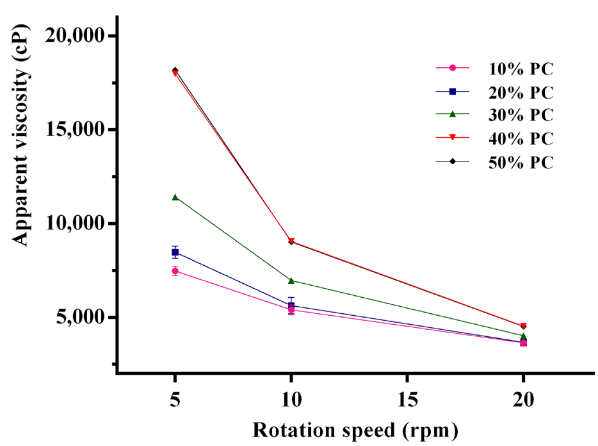
Relative density can also be an important specification to be established, since this parameter can even be used to verify the potential for stability of the face masks. For instance, the excessive incorporation of air into the product along the production process can be detected by significant decrease of the expected density (SILVA *et al*., 2019). Thus, the values of relative density were found to be in the range of 1.06 – 1.43 g/mL, slightly increasing ($p < 0.05$) as the proportion of PC was increased (Table 2).

### 3.2.2 Apparent viscosity

Using a rotary viscometer, the apparent viscosity is measured from the force required to rotate the spindle inserted in the sample. Besides the physicochemical characteristics of the product, the apparent viscosity also depends on the rotation speed of the spindle and on the temperature of the material. The rheological studies may then indicate different flow and deformation properties of the materials, such as elasticity, viscosity, plasticity, deformation and fluidity (ANVISA, 2008; MODIGELL *et al*., 2018; VISERAS *et al*., 2007).

Overall, the clay masks were observed to be non-Newtonian fluids, since the viscosity varied according to the shear rate (Figure 1). For instance, the apparent viscosity of the face mask containing 30% PC reduced from 11,416 ± 115 cP to 6,964 ± 156 cP and 4,016 ± 98 cP ($p < 0.05$), as the rotation speed increased from 5 rpm to 10 and 20 rpm, respectively. Thus, the reduction in the apparent viscosity as the rotation speed increases is an evidence of the pseudoplastic behavior of these clay masks, which is appropriated since it favors the application on the skin. The slight reduction in the product viscosity from its application on the skin can assist in spreading the product and in forming a uniform layer over the skin surface (DANELUZ *et al*., 2020; DARÉ *et al*., 2015; SILVA *et al*., 2019; VISEIRAS *et al*., 2007).

**Figure 1 -** Apparent viscosity of face masks containing different proportion of pink clay in composition (10 to 50% w/w) and measured at 5, 10 and 20 rpm




**Source:** Research data.

In addition, an increase in viscosity from 7,480 ± 248 cP to 18,184 ± 41 cP (5 rpm) was observed ($p < 0.05$) as the clay proportion was increased from 10% to 50% (w/w). Some types of clays, especially those containing kaolinite, can generate increase of the viscosity in aqueous dispersions due to the expansion flow caused by the interactions among the dispersed particles during the viscometer rotation (BURITI *et al*., 2019; DANELUZ *et al*., 2020; VISEIRAS *et al*., 2007; YUAN; MURRAY, 1997). The PC particles dispersion is influenced by their size and shape, thereby modifying the final formulation rheology, while other factors can also influence the final viscosity, including the temperature, the presence of other formulation components, and the preparation procedure (VISEIRAS *et al*., 2007).

### 3.2.3 *In vitro* spreadability performance

The spreadability performance can be defined as the expansion of a semi-solid formulation onto a surface after being pressed by a determined weight. This is one of the essential characteristics of cosmetic products such as face masks, as they are intended for topical application and spreadability is closely related to the contact area with the skin. The larger the diameter measured after the application of this technique, the greater the sample spreadability (BORGHETTI; KNORST, 2006).

All the face masks showed good spreadability, in the range from 1.55 to 7.73 $cm^2$ (Table 3). The formulation containing 10% (w/w) PC showed the highest spreadability, which was reduced as the PC proportion in the composition increased ($p < 0.05$), so that the formulation containing 50% (w/w) was the one with the lowest spreadability. This fact is directly associated with the increase in viscosity observed as the PC proportion in the product increases, as very viscous products have less spreadability and vice versa (DANELUZ *et al*., 2020; DEUSCHLE *et al*., 2015; ESTANQUEIRO *et al*., 2016).

**Table 3 -** *In vitro* spreadability of the face masks in function of the PC proportion in the composition

| PC proportion (w/w) | d (cm) | S ($cm^2$) |
|---|---|---|
| **10%** | 3.13 ± 0.19 | 7.73 ± 0.94 |
| **20%** | 2.7 ± 0.16 | 5.74 ± 0.69 |
| **30%** | 2.23 ± 0.09 | 3.92 ± 0.32 |
| **40%** | 2.0 ± 0.08 | 3.14 ± 0.26 |
| **50%** | 1.43 ± 0.12 | 1.55 ± 0.35 |

**Source:** Research data.

Good spreadability is essential for the performance of a cosmetic product and the consequences are easy application, with homogenous distribution over the skin, and high product effectiveness, also resulting in higher consumer acceptance (BORGHETTI; KNORST, 2006; DANELUZ *et al*., 2020; DEUSCHLE *et al*., 2015).

## 4 Conclusion

A cosmetic product based on a natural pink clay was successfully obtained by using the appropriated procedure method. Both autoclaving and dry heating were efficient sterilization processes for PC, ensuring that a face mask produced with this raw material has a number of microorganisms within the specifications, attesting to its safe use. From the face masks obtained, pH and density slightly varied with the different PC concentration in the cosmetic products, and their values were compatible for skin application. Variations on the PC concentration directly influenced the apparent viscosity of the PC face masks, which also showed pseudoplastic behavior. The higher the PC concentration, the higher the values of viscosity obtained, but with less spreadability of the cosmetic product. Therefore, the intermediate proportion of 20 or 30% (w/w) of this pink clay in the face masks would be more suitable for the intended cosmetic use.

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