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RESISTANCE OF PROBIOTIC LACTOBACILLI, BIFIDOBACTERIA
AND PROPIONIC ACID BACTERIA STRAINS TO PRESERVATIVES
APPLIED IN COSMETIC FORMULATIONS

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Resistance of probiotic lactobacilli, bifidobacteria and propionic acid bacteria strains to preservatives applied in cosmetic formulations: The resistance of probiotic strains of the genus *Lactobacillus*, the genus *Bifidobacterium* and the genus *Propionibacterium* to preservatives (GMS SE, Lanette O, Arlacel165, glyceryl oleat, evimulWO, cetareth-20, glyceryl monostearate, IPM, silicone oil) applied in cosmetic formulations was examined. It has been shown that with a few exceptions (*Lactobacillus delbrueckii ssp. bulgaricus* GB) all the studied strains were resistant to the cosmetic preservatives. This opens up possibilities for application of probiotic bacterial strains in the production of cosmetics with a functional purpose.

Key words: *Lactobacillus*, *Propionibacterium*, *Bifidobacterium*, cosmetic, preservative.

INTRODUCTION

In recent years technology for the production of cosmetic preparations, influencing the physiological processes of the skin, is applied in cosmetic production. This effect is achieved through the application of functional additives providing new properties to cosmetic preparations for the prevention of skin diseases, skin protection from external influences, thus preventing its premature aging. As alternative supplements are applied vitamins and micro- and macroelements, phospholipids, plant extracts, vegetable oils and probiotics. Lactobacilli and bifidobacteria are widely applied for biological preservation of cosmetic products and for the preparation of face masks. In cosmetic preparations with functional purpose, such as face masks, creams and conditioners are included probiotic bacteria (lactobacilli or bifidobacteria or propionic acid bacteria and their lysates) that influence the skin condition on one hand, and protect the cosmetic formulations from microbial spoilage on the other, by the produced bioactive substances without the addition of chemical preservatives (bronopol and parabens) [1, 2, 3].

Not all lactobacilli and bifidobacteria may be used in cosmetic formulations but only those that can survive under the conditions of cosmetic production and storage. This requires selection of strains of lactobacilli, bifidobacteria and propionic acid bacteria resistant to the preservatives most commonly applied in cosmetic formulations [1, 2, 3].

The purpose of the present work was to study the resistance of probiotic strains of lactobacilli, bifidobacteria and propionic acid bacteria to the preservatives most commonly used in cosmetic formulations.

MATERIALS AND METHODS

Microorganisms

genus *Lactobacillus* - *Lactobacillus delbrueckii* ssp. *bulgaricus* GB, *Lactobacillus acidophilus* Ac, *Lactobacillus acidophilus* 5R, *Lactobacillus plantarum* P1.

genus *Bifidobacterium* - *Bifidobacterium bifidum* BB1.

genus *Propionibacterium* - *Propionibacterium freudenreichii* ssp. *shermanii* NBIMCC 328.

Media

LAPTg10 - agar medium. Composition (g/dm³): peptone - 15; yeast extract - 10; tryptone - 10; glucose - 10; Tween 80 - 1 cm³/dm³, agar - 15. pH = 6.6 - 6.8. Sterilization - 20 minutes at 121°C.

Selective agar medium for *Propionibacterium* sp. Composition (g/dm³): tryptone - 10; yeast extract - 10; Na-lactate (fresh) - 10; KH₂PO₄ - 2.5; MnSO₄ - 0,005; agar - 15; pH=6.8. Sterilization - 20 minutes at 121°C.

Selective agar medium for *Bifidobacterium* sp. Composition (g/dm³): peptone - 10; yeast extract - 10; lactose - 10; MnSO₄ - 1; casein hydrolyzate - 8; NaCl - 3,2; CH₃COONa - 1; agar - 15. pH=6,6 - 6,8. Sterilization - 20 minutes at 121°C.

Preservatives - GMS SE, Lanette O, Arlacel165, glyceryl oleat, evimulWO, cetareth-20 and glyceryl monostearate, IPM, silicone oil.

Determination of the resistance to preservatives - disc-diffusion method

Sterile melted LAPTg10-agar medium was poured in Petri dishes and after the solidification of the agar, the dishes were spread plated with 24-hour suspensions of the corresponding strain of the genus *Lactobacillus*. *Propionibacterium freudenreichii* ssp. *shermanii* was pour plated in selective agar medium for *Propionibacterium* sp. *Bifidobacterium bifidum* BB1 was pour plated in selective agar medium for *Bifidobacterium* sp. The experiments were conducted with the following solutions of each of the substances tested: 5%, 10% and 15%. The used paper discs were 6 mm in diameter. 6 µL of the corresponding solution were pipetted on the corresponding paper disc. Paper discs soaked in distilled water were used as blanks. The results were recorded as diameters of the clear zones around the paper discs, in millimeters, after 24–48 hours of incubation of the Petri dishes at 37°C.

The experiments were performed in quadruplicate. The mean values and the standard deviations were calculated using MS Office Excel 2010.

RESULTS AND DISCUSSION

In a series of experiments was investigated the resistance of probiotic lactobacilli, bifidobacteria and propionic acid bacteria to the preservatives most commonly used in cosmetics (GMS SE, Lanette O, Arlacel165, glyceryl oleat, evimulWO, cetareth-20, glyceryl monostearate, IPM, silicone oil). The results of these experimental studies are shown in Table 1 and Table 2. Experimental data indicated that with the exception of *Lactobacillus delbrueckii* ssp. *bulgaricus* GB, the growth of which was affected by the emulsifiers evimul WO and Cetareth-20, the other preservatives did not inhibit the growth of the probiotic lactobacilli, bifidobacteria and propionic acid bacteria strains (Table 1 and Table 2). This in turn indicated that the selected probiotic strains may be used in cosmetic formulations, together with the examined.

Table 1.

Resistance of probiotic lactobacilli, bifidobacteria and propionic acid bacteria to preservatives applied in cosmetic formulations

Substance	Glyceryl oleat			IPM			Evimul WO			Cetareth-20			Glyceryl monostearate		
	5	10	15	5	10	15	5	10	15	5	10	15	5	10	15
<i>Lactobacillus delbrueckii</i> ssp. <i>bulgaricus</i> GB	-	-	-	-	-	-	11,00 ±1,00	13,00 ±1,73	12,00 ±1,73	12,00 ±0,50	13,00 ±0,50	12,00 ±0,50	-	-	-
<i>Lactobacillus acidophilus</i> Ac	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Lactobacillus acidophilus</i> 5R	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Lactobacillus plantarum</i> P1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Bifidobacterium bifidum</i> BB1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
<i>Propionibacterium freudenreichii</i> ssp. <i>shermanii</i> NBIMCC 328	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

Table 2.

Resistance of probiotic lactobacilli, bifidobacteria and propionic acid bacteria to preservatives applied in cosmetic formulations

Substance	GMS SE			Lanette O			Arlacel 165			Silicone oil		
	5	10	15	5	10	15	5	10	15	5	10	15
<i>Lactobacillus delbrueckii</i> ssp. <i>bulgaricus</i> GB	-	-	-	-	-	-	-	-	-	-	-	-
<i>Lactobacillus acidophilus</i> Ac	-	-	-	-	-	-	-	-	-	-	-	-
<i>Lactobacillus acidophilus</i> 5R	-	-	-	-	-	-	-	-	-	-	-	-
<i>Lactobacillus plantarum</i> P1	-	-	-	-	-	-	-	-	-	-	-	-
<i>Bifidobacterium bifidum</i> BB1	-	-	-	-	-	-	-	-	-	-	-	-
<i>Propionibacterium freudenreichii</i> ssp. <i>shermanii</i> NBIMCC 328	-	-	-	-	-	-	-	-	-	-	-	-

CONCLUSION

With the exception of *Lactobacillus delbrueckii* ssp. *bulgaricus* GB, all other probiotic lactobacilli, bifidobacteria and propionic acid bacteria were resistant to the preservatives, most commonly applied in cosmetic preparations. This opened up opportunities for their inclusion in cosmetics with functional purpose. Thus the included lactobacilli, bifidobacteria or propionic acid bacteria strains would influence the skin condition and protect the cosmetic formulations from microbial spoilage by the produced bioactive substances with or without the addition of chemical preservatives.

REFERENCES

- [1] Baki G., K. Alexander, *Introduction to cosmetic formulation and technology*. Hoboken, New Jersey: John Wiley & Sons, Inc., 2015
- [2] Tsai C.C., C. F. Chan, W. Y. Huang, J. S. Lin, P. Chan, H. Y. Liu, Y. S. Lin, Applications of *Lactobacillus rhamnosus* Spent Culture Supernatant in Cosmetic Antioxidation, Whitening and Moisture Retention Applications. *Molecules*, 2013, 18, 14161-14171.
- [3] Viera R., A. Fernandes, T. Kaneko, V. Consiglieri, C. Pinto, C. Pereira, A. Baby, M. Velasco, Physical and physicochemical stability evaluation of cosmetic formulations containing soybean extract fermented by *Bifidobacterium animalis*; *Brazilian J. of Pharmaceutical Sciences*, 2009, 45(3)

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