FORMULATION OF SNAIL SLIME (Achatina fulica) ANTI-ACNE EMULGEL USING TWEEN 80-Span 80 AS EMULSIFYING AND HPMC AS GELLING AGENT

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ABSTRACT

Acne is the condition of abnormal skin which is indicated by inflammation caused by the bacterial infection of Propionibacterium acnes. The natural one which can be used for the medical treatment of acne is the snail mucus (Achatina fulica). The achasin protein of it has antibacterial activity. That snail mucus was made to the emulgel form. This research has used an experimental method and the emulgel formulation used the various concentration of emulsifying agents and the gelling agent. They were Tween 80 1.76%, 2.44%, 3.12%, Span 80 2.24%, 4.06%, 5.88% and HPMC 3.5%, 4.5%, 5.5%. Other additives were propylene glycol, methyl paraben, propyl paraben, paraffin liquid, menthol and aquadest. Those formulations were tested in physical evaluation during 4 weeks storage in room temperature, irritation test, hedonic test and cycling test. The organoleptic test showed that the emulgel were milk-white color with distinctive smell. All emulgel were homogen, non-irritant, with emulsion type oil in water (o/w). These emulgel also met the normal skin of pH value and spread ability's range. The emulgel viscosity shift were < 10%, with the viscosity value inversely proportional to spread ability. Formula C with 3.12% of Tween 80, 5.88% of Span 80 and 3.5% of HPMC was claimed as the most stable formula both in room temperature and after cycling test. It was also the most preferred by panelist.

Keywords: Snail slime, Emulgel, Tween 80, Span 80, HPMC

INTRODUCTION

Acne on the face can be caused by bacteria Propionibacterium acnes that convert fat sebum of liquid becomes denser. This can cause infection or swelling of acne (Dewi, 2009). Acne treatment can be administered with antibiotics and other chemicals such as sulfur, salicylic acid. However, these drugs have side effects including irritation, while the long-term use can lead to resistance. Therefore, its needed a natural alternative material that can be used in the treatment of acne. One is the slime of snail.

According to Nugroho (2015) the use of snails for facial treatments was first popularized by the Tokyo Clinical Salon and the treatment was carried out by placing three snails on the face. These snails will run all over the face, removing natural antibiotic substances and hyaluronic acid which fight skin problems while hydrating the skin. Aghina et al (2015) stated that snail slime has a content that can moisturize the skin, namely allantonin compounds and glycosaminoglycan compounds which play an important role in maintaining the connective tissue between cells so that the skin becomes tight.

Snail slime in gel formulation with a concentration of 11% has antibacterial activity against bacteria that cause acne namely P. acnes. Snail slime contains achasin which is considered crucial as antimicrobial peptides. So, the potential to be developed as an active ingredient in an anti-acne preparation (Mardiana et al., 2015). Application of snail slime on the face needs a good topical preparation. Formulation to emulgel selected on the advantages of gel and emulsion can increase the high penetration and usage comfort to the skin.

The emulgel preparations consist of emulsion systems and gel system. Both systems greatly affect the physical properties and stability so that election preparations emulgel emulsifying agent and gel base is very important. This study aimed to see whether the snail slime can be formulated to the stable and qualified emulgel using emulsifying agents Tween 80 and Span 80 and HPMC gel base.

TWEEN 80 is a water-soluble emulsifying agent to form oil in water emulsion type, while Span 80 is a non-ionic emulsifying agent non-ionic emulsifying agent with its dominant lipophilic group. In interfacial films theory, the stable interfacial complex condensed film formed, when a water-soluble emulsifying agent is mixed with a fat-soluble emulsifying agent it is able to form and maintain the emulsion more effectively than the use of a single emulsifying agent (Kim, 2004). In the gel system, base gel will play a role in determining the physical properties and physical stability of the gel. Hydroxy propyl methyl cellulose is a gel base that is not toxic and does not irritate, so safe to use (Rowe et al., 2009).
MATERIAL AND METHODS

Tools and Materials

The materials used in this study were kencur snail slime, Tween 80, Span 80, HPMC, propylene glycol, methyl paraben, propyl paraben, paraffin liquid, menthol, and aquadest. The equipment used includes glasses (Pyrex), porcelain dish, hot plates (Thermo), mortars and stomper, spatula, analytical balance (Fujitsu), container emulgel, viscometer Rion V-06V, and pH meters (Lutron).

Methods

Snails were cleaned first. Mucus were retrieved by touching and pressing the snails body to go into a shell and then mucus collected. Mucus then filtered and put into a sample container.

Massa gel was made by dispersing HPMC in distilled water at a temperature of 80 °C and then crushed to form a gel base. Methyl paraben and propyl paraben dissolved into propylene glycol and menthol in a little ethanol 96%. Both mixtures incorporated into the gel base and crushed to form a homogeneous gel mass.

Massa emulsion was made by mixing the oil phase (the melted of Span 80 and liquid paraffin) to the water phase (the heated of Tween 80 and distilled water) at a temperature of 70 °C. Furthermore, the mass of emulsion incorporated into the gel mass while the rest of snail slime and the distilled water added gradually. Emulgel crushed to form a homogeneous preparation. Composition of snail slime emulgel is shown in Table 1.

Evaluation was conducted on the physical evaluation during 4 weeks of storage at room temperature (organoleptic, homogeneity, emulsion type, pH measurement, viscosity, the shifting viscosity, the spreading power), irritation test, preference test, and cycling test.

Table 1. Composition of snail slime emulgel

| Ingredients        | Concentration (%) | Function|
|--------------------|-------------------|---------|
| Active Substance   |                   |         |
| Snail Slime        | 11                | 11      | 11      | Active |
| Gel Materials      |                   |         |
| HPMC               | 3.5               | 4.5     | 5.5     | Substance |
| Propylene glycol   | 10                | 10      | 10      | Gel Base |
| Propyl paraben     | 0.2               | 0.2     | 0.2     | Preservatives |
| Methyl paraben     | 0.1               | 0.1     | 0.1     | Preservatives |
| Emulsions Materials|                   |         |
| Tween 80           | 0.05              | 0.05    | 0.05    | Flavoring Agent |
| Span80             | 1.76              | 2.44    | 3.12    | Emollient |
| Aquadest           | 2.24              | 4.06    | 5.88    | Emulsifier |
|                     | 100               | 100     | 100     | Solvent |

RESULTS AND DISCUSSION

Organoleptic Test

The snail slime obtained has a clear white color and rather sticky with thick consistency. This mucus was then formulated into an emulgel preparation with other additives. The results of organoleptic test in Table 2 show that formula C produces stable emulgel for 4 weeks of storage at room temperature. The low concentrations of Tween 80, Span 80 and HPMC caused the consistency of formula A and B to remain dilute. Formula A even tends to be unstable, because it experiences creaming. Creaming on the emulgel was marked with the emulsion phase at the top and the gel phase at the bottom. The complete emulsion separation occurs because of the formation of larger droplets by combining smaller droplets (Syukri et al., 2009). Variations concentration of Tween 80, Span 80 and HPMC did not affect the color and aroma of the emulgel.

Table 2. Evaluation results of emulgel

| Parameters   | Formula | 1st Week | 4th Week | Shifting Viscosity (%) |
|--------------|---------|----------|----------|------------------------|
| Organoleptic | A       | Watery,  | Creaming,|
|              | B       | distinctive| distinctive|                        |
|              | C       | Emulgel, | Watery,  |
|              |         | distinctive| distinctive|                        |
| Homogeneity  | A, B & C| Homogeneous| Homogeneous|                        |
| pH           | A       | 6.30     | 5.07     | -                      |
|              | B       | 6.13     | 5.09     | -                      |
|              | C       | 5.85     | 5.15     | -                      |
| The Spreading | A      | 5.91     | 5.96     | -                      |
| Power (cm)   | B       | 5.55     | 5.53     | -                      |
|              | C       | 4.76     | 4.91     | -                      |
| Viscosity    | A       | 103.33   | 110.00   | 6.45                   |
| (dPa.s)      | B       | 143.33   | 136.66   | 4.65                   |
|              | C       | 153.33   | 143.33   | 6.32                   |

Homogeneity and Emulsion Type Test

The three formulas produced were homogeneous which characterized by evenly distributed colors and the absence of lumps or coarse particles in the emulgel. The emulsion type was oil in water (O/W), which was done by the dilution method. This emulsion type of O/W were easy to wash and not sticky when used.

pH Measurement

Table 2, shows the pH of emulgel ranging from 5.07 to 6.30. Although the pH value of the emulgel varies, but still meets the requirements of normal skin pH of 4.5-6.5. The pH value will affect the comfort and safety of using emulgel.
The pH values that are too acidic can cause irritated skin, whereas if too alkaline can cause dry skin.

**Viscosity Test and The Shifting Viscosity**

Viscosity test is needed by a semisolid preparation to see the flow properties of the preparation when applied to the skin (Garg et al., 2002). Table 2 shows that the higher the concentration of Tween 80, Span 80 and HPMC, the higher the viscosity value. Laverius (2011) stated that high viscosity will provide stability of the emulsion system in emulgel preparations, because it will minimize the dispersion phase droplet movement, so that the droplet size changing to a larger size can be avoided and the possibility of coalescence can be prevented. Conversely, low viscosity will cause dispersion phase droplets to move more easily, so that the tendency of dispersion phase droplets to join is greater than that of high viscosity emulsions. Formula C with the highest viscosity value, organoleptically was the most stable preparation.

In the polymer dispersion of cellulose derivatives, polymer molecules enter cavities formed by water molecules, causing hydrogen bonds between hydroxyl groups (-OH) of polymers and water molecules. This hydrogen bond plays a role in hydration in the swelling process of a polymer. Therefore, increasing the concentration of HPMC, causing the -OH group to bind more so that the viscosity of the preparation increases (Erawati et al., 2005).

 Tween 80 along with span 80 plays a role in stabilizing the emulsion system, so that the more tween 80 will make the dispersion medium more rigid. The more rigid the dispersion medium will result in an increase in the viscosity of the emulsion system. Emulsion is part of this emulgel. Therefore, when the viscosity of the emulsion system increases it will affect the viscosity of the emulgel which will also increase (Laverius, 2011).

In addition, in Table 2 can also be seen the percentage shift in the third viscosity formula were <10%. The viscosity shift value represents the instability of the emulgel during storage for a period of one month. The desired viscosity shift from emulgel is ≤ 10%. Emulgel is expected to have the lowest possible viscosity shift because the viscosity shift describes the physical stability of the emulgel. A large shift in the emulgel viscosity within a certain period of time during storage shows the instability of emulgel during storage within this period (Laverius, 2011).

**The Spreading Power Test**

The expected spreading power for emulgel preparations ranges from 3-5 cm. Because with this value emulgel can be used properly (Laverius, 2011). Table 2 shows that the C and B formulas met the expected spread value, while the formula A approaches the semifluid category. Formula A had the lowest consistency and the lowest viscosity value compared to formula B and C. These results were in accordance with Garg et al. (2002) that scattering is inversely proportional to the viscosity of the preparation. The higher the viscosity value of a preparation, the lower the spreading power of the preparation, and vice versa.

**Irritation Test and Preference Test**

The results of the irritation test using the patch test method, showed that all three formulas were safe to use. They were characterized by the absence of irritation symptoms such as erythema, edema, itching and pain when used. As for the preference test results, Figure 2 shows the formula C was the most preferred formula in terms of texture, color and smell.

**Cycling Test**

This test was carried out for 6 cycles and the most stable formula C was produced, compared to 2 other formulas. This was indicated by the unchanged preparation after passing 6 cycles.
emulsifying agents. The most stable and best formula was formula C, with HPMC concentration 5.5%, Tween 80 3.12%, and Span 80 5.88%.

ACKNOWLEDGEMENT

This work was supported by a grant from Directorate of Research and Community Service of the Ministry of Research and Technology, for the Research Grants of Beginning Lecturers for Fiscal Year 2018. We also thank Polytechnic Bina Husada Kendari Foundation for the assistance given in completing this research.

REFERENCES

Aghina, Y., Amila, G. dan Dina, M. 2015. Formulasi Masker Gel Pell Off Lendir Bekicot (Achatina fulica) dengan Variasi Konsentrasi Bahan Pembentuk Gel. Prosiding Penelitian Spesia UNISBA Farmasi Gelombang 2, Bandung, pp 246.

Dewi, S.A. 2009. Cara Ampuh Mengobati Jerawat Secara Alami dan Medis. Buana Pustaka, Yogyakarta.

Erawati, T., Rosita, N., Hendropriyono, W., Juwita, D. R. 2005. Pengaruh Jenis Basis Gel Dan Penambahan NaCl (0.5% -b/b) Terhadap Intensitas Echo Gelombang Ultrasonik Sediaan Gel Untuk Pemeriksaan USG (Acoustic Coupling Agent). Airlangga Journal of Pharmacy. 5(2).

Garg, A., Aggrawal, D., Garg, S., Singla, A.K. 2002. Spreading of Semisolid Formulations: An Update. Pharmaceutical Technology. pp 84-105.

Kim, C. 2004. Advanced Pharmaceutics Physicochemical Principles. CRC PRESS, London.

Laverius, M.F. 2011. Optimasi Tween 80 Dan Span 80 Sebagai Emulsifying Agent Serta Carbopol Sebagai Gelling Agent Dalam Sediaan Emulgel Photoprotector Ekstrak Teh Hijau (Camellia sinensis L.): Aplikasi Desain Faktorial. Skripsi. Universitas Sanata Dharma, Yogyakarta.

Mardiana, Z.H., Amila, G. dan Lanny, M. 2015. Formulasi Gel yang Mengandung Lendir Bekicot (Achatina fulica) Serta Uji Aktivitas Antibakteri Terhadap Propionibacterium acne. Prosiding Penelitian Spesia UNISBA Farmasi Gelombang 2 (2014-2015), pp 223-224.

Rowe, R.C., Paul, J.S., Sian, C.O. 2009. Handbook of Pharmaceutical Excipients 6th Edition. Pharmaceutical Press, Washington D.C.

Syukri, Y., Sari, F., Zahliyatul, S. 2009. Stabilitas Fisik Emulsi Ganda Virgin Coconut Oil (VCO) Menggunakan Emulgator Span 80 dan Tween 40. Jurnal Ilmiah Farmasi. 5(1):4.