

## Formulation peel-off gel mask of *Impatiens balsamina* L. as an antibacterium against *Staphylococcus aureus*

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### ABSTRACT

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**Background:** Acne is a skin disorder characterised by inflammation, accompanied by a blockage of the oil glands in the skin. One of the causes of acne is *Staphylococcus aureus* bacteria. Many plants have been widely studied as an antibacterial agent, including Garden Balsam Leaf (*Impatiens balsamina* L.) which could be formulated in a peel-off gel preparation.

**Objective:** The purpose of this research was formulating an antibacterial peel-off gel mask with a variation of viscosity enhancing agent hydroxypropyl methylcellulose (HPMC), as well as testing its antibacterial activity on *Staphylococcus aureus*.

**Methods:** HPMC 1-2% was formulated in peel-off gel mask of Garden Balsam leaf extract. Physical properties such as viscosity, pH, spreadability, and drying time were tested, while antibacterial activity test was conducted by the agar-diffusion method.

**Results:** The viscosity of the entire formula was > 20,000 cP, pH 6, drying time > 20 minutes, and spreading power > 6 cm. Also, the peel-off gel mask also showed antibacterial activity on *Staphylococcus aureus* with average inhibition zone were > 10 mm, which indicated strong antibacterial inhibitory power.

**Conclusion:** The concentration variation of HPMC had a significant effect on all physical properties test. HPMC 1.5% in the preparation showed the most optimum results.

**Latar Belakang:** Jerawat merupakan gangguan pada kulit yang ditandai dengan adanya peradangan disertai penyumbatan saluran kelenjar minyak dalam kulit. Salah satu penyebab jerawat adalah bakteri *Staphylococcus aureus*. Tanaman yang telah banyak diteliti sebagai antibakteri adalah tanaman pacar air (*Impatiens balsamina* L.). Tanaman ini dapat diformulasikan dalam bentuk sediaan masker gel peel-off.

**Tujuan:** Tujuan penelitian ini adalah untuk memformulasikan masker gel peel-off dengan ekstrak daun pacar air sebagai antibakteri dengan membuat variasi konsentrasi bahan peningkat viskositas Hydroxypropyl Methylcellulose (HPMC) serta uji aktivitasnya terhadap bakteri *Staphylococcus aureus*. Metode yang digunakan adalah difusi agar.

**Metode:** Pengujian sifat fisik yang dilakukan meliputi viskositas, pH, daya sebar dan waktu sediaan mengering, sementara hasil pengujian aktivitas antibakteri dilakukan dengan difusi agar.

**Hasil:** Viskositas seluruh formula > 20.000 cp; pH 6, waktu sediaan mengering >20 menit, dan daya sebar > 6 cm. Sementara itu, hasil uji antibakteri menunjukkan aktivitas terhadap *Staphylococcus aureus* dengan rata-rata zona hambat sebesar > 10 mm, yang termasuk kategori daya hambat kuat.

**Kesimpulan:** Variasi konsentrasi HPMC dalam formulasi berpengaruh secara bermakna terhadap semua uji sifat fisik. HPMC 1,5% dalam formulasi menunjukkan hasil yang paling optimum.

## INTRODUCTION

Facial skin problems are often in the spotlight. One of the facial skin problems that are commonly encountered is the emergence of acne. Acne vulgaris is a skin disease characterised by inflamed spots and blackhead on the face, neck, back, chest, and others.<sup>1</sup> While the condition is not life-threatening, it can cause significant psychological morbidity and lead to lifelong scarring and some cases of facial disfigurement if left untreated.<sup>2</sup>

The common bacteria infecting acne is the *Staphylococcus aureus* bacteria. *Staphylococcus aureus* bacteria is a gram-positive bacteria that is a normal flora of the skin of mucous membrane.<sup>3</sup> The peel-off gel mask aims to cleanse and moisturise the face by applying it on the face evenly and waited until it dries, this preparation will form an elastic transparent film layer, so that it can be easily removed or exfoliated.<sup>4</sup>

One of the exciting plants to be studied is a Garden Balsam leaf (*Impatiens balsamina* L.). Traditionally, people use Garden Balsam leaf by boiling and milling to be applied on the body part infected by bacteria. Besides being used as an antibacterial, Garden Balsam leaf also have some other benefits such as to overcome the late menstruation, inflammation of the skin purulent, ulcers and inflammation of the edge of the nail.<sup>5,6</sup>

The methanol extract of Garden Balsam leaf can inhibit the growth of bacteria.<sup>7</sup> The primary compounds of Garden Balsam leaf extracts which have antibacterial activity against acne are quinone and flavonoid compounds (*quercetin* and *kaempferol*). In the Adfa study stated that the pure compounds isolated from the Garden Balsam leaf showed the antibacterial activity of 0.5 to 0.6 times tetracycline against the bacterial test of *Staphylococcus aureus* and *Bacillus cereus*.<sup>7,8</sup>

## METHODS

### Garden Balsam Leaf Extract Preparation

The standardised Garden Balsam leaf from Ngaglik-Sleman-Yogyakarta was separated from the stalks and prepared for further maceration extraction.<sup>5,9</sup> The leaf powder was dissolved

with 96% ethanol solvent, stirred, left for five days, then filtered by Buchner funnel to obtain the filtrate. The filtrate was evaporated using rotary evaporator and then dried on a water bath to obtain thickened extract.<sup>10</sup>

### Peel-off Gel Mask Preparation

PVA was dissolved in 80°C aquadest. HPMC was prepared with cold water for 15 minutes and mixed with glycerin, then being added with potassium sorbate solution. The mixture then being added by PVA solution with ethanol and the remaining aquadest until forming the gel. Finally, the gel was being mixed with the Garden Balsam leaf extract (Table 1).<sup>11</sup>

### Thin Layer Chromatography (TLC) Test of Garden Balsam Leaf Extract

Thin Layer Chromatography (TLC) test of Garden Balsam leaf extract was carried out using silica gel GF<sub>254</sub> stationary phase, and chloroform-methanol-water as the mobile phase (80:12:2). Aluminium chloride (AlCl<sub>3</sub>) was used as spray reagent on TLC plate before observation on UV rays.

### Peel-Off Gel Mask of Garden Balsam Leaf Extract Test Organoleptic Test

Organoleptic observations were carried out directly on the Garden Balsam leaf extract. The observations included smell, shape and color.<sup>7</sup>

### Viscosity Test

The viscosity measurement of the gel was performed with by Brookfield viscometer at room temperature. The viscometer spindle was immersed in the gel, and the result could be obtained in the viscometer screen.<sup>11</sup>

### pH Test

pH measurement was carried out by using pH meter. The pH meter device was immersed in the gel. The result shown on the pH meter scale was the pH value of the gel.<sup>12</sup>

**Drying up Time Test**

1 g peel-off gel mask was smeared on the arm skin. Time of the gel to form a film layer and peel-off gel mask was determined by using a stopwatch.<sup>11</sup>

**Spreadability Test**

0.5 g peel-off gel mask was placed between two glasses. The upper glass was given a 50 g load which gradually increased until the gel spread is constant.<sup>5</sup> The spreadability value was the gel spread distance between unloading and maximum load glasses.

**Antibacterial Activity Test**

*Staphylococcus aureus* bacteria which has

been adjusted with Mc Farland standards were mixed with 20 mL of Mueller Hinton agar medium and poured into the petri dish. Four well-holes were made on the medium and each one was being put 50 mg of peel-off gel, 50 mg extract, 50 mg benzoyl peroxide 5% as the positive control, and 50 mg gel base as the negative control. The Petri dish were incubated at 37°C for 24 hours until exhibit antibacterial activity inhibition zone.

**Statistics Analysis**

Physical properties of the test results (viscosity, diffused pH and drying time) were analysed using one-way ANOVA, while paired samples t-test was used for the antibacterial activity test analysis.

Table 1. Peel-off Gel Mask of Garden Balsam Leaf Extract Formula.<sup>12</sup>

Materials (g)	F1	F2	F3
Garden Balsam Leaf Extract	15	15	15
PVA	10	10	10
HPMC	1	1,5	2
Gliserin	5	5	5
Potassium sorbat	0,2	0,2	0,2
Etanol 70%	5	5	5
Aquadest	ad 100	ad100	ad 100

Table 2. Physical Test Result Peel-Off Gel Mask of Garden Balsam Leaf Extract

Formula	Viscosity (cps)	pH	Drying up time (minutes)	Spreadability (cm)
FI	22829	5,96	26	7,3
FII	32658	5,96	23	7,2
F III	54882	6,00	20	6,6

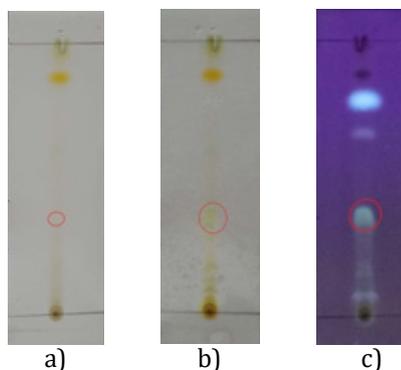


Figure 1. TLC test of Garden Balsam leaf extract; a) before spraying AlCl3 reagent; b) after spraying AlCl3 reagent; c) under UV366 light which showed quercetin spot at Rf 0.35

**RESULTS**

**Garden Balsam Leaf Extract Properties**

As much as 75.02 g thick, black, and typical smell extract were obtained with 6.25% rendement. The TLC on figure 1 showed flavonoid quercetin content at Rf 0.35. There was a change in spot color of TLC into greenish yellow after AlCl<sub>3</sub> reagent spraying, and glowing under UV light which also indicated flavonoid content existence.

Organoleptic properties were determined by direct observation of the gel including smell, shape and colour. The result came out as the blackish brown concentrated extract with a

specific scent. The extract was stable with no change in colour, smell or shape after long time storage.

**Peel-Off Gel Mask Properties**

The viscosity test at 2 rpm showed that the smallest viscosity was found in the formula 1 followed by the formula II and III. The viscosity of the entire formula was > 20,000 cp. The pH test result obtained from all formula (FI, FII, and FIII) was 5,96; 5.96 and 6.0 respectively (Table 2). The three formulations produced proper drying up time around 20-26 minutes. The formula I created the greatest spreadability due

Table 3. Inhibition Zone Diameter (mm) of Antibacterial Activity Test

	FI	FII	FIII
Garden Balsam leaf extract	13,6	14,8	13,567
Control (+)	12,0	13,3	12,5
Peel-off gel mask	10,9	12,1	11,2
Control (-)	0,0	0,0	0,0

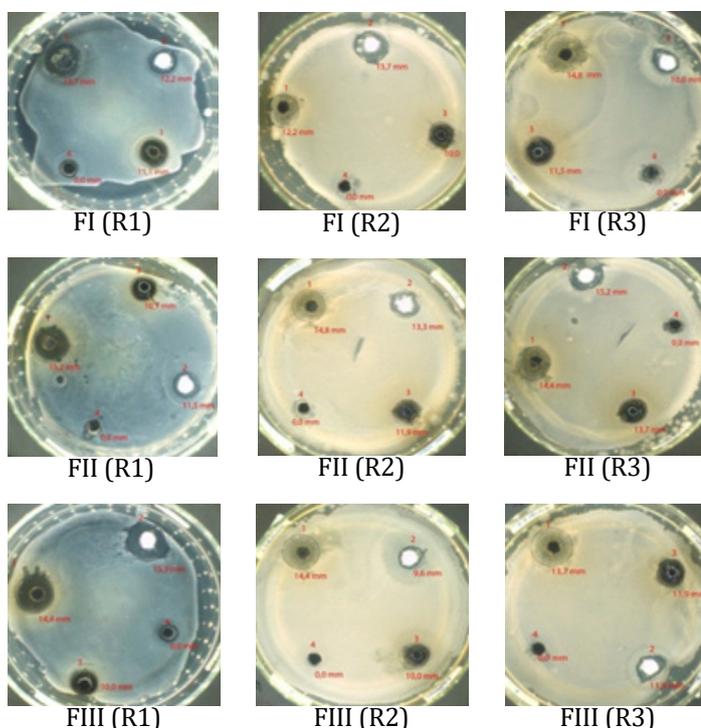


Figure 3. Antibacterial activity test of FI-III (triplicate R1-3); caption (red font, inline figure) 1. Garden Balsam leaf extract; 2. Benzoyl peroxide 5% as positive control; 3. Peel-off gel mask of Garden Balsam leaf extract; 4. Base of the peel-off gel mask as negative control

to the smallest number of HPMC among other formulations. The result of the inhibition zone diameter was described in table 3. The peel-off gel mask also showed antibacterial activity on *Staphylococcus aureus* with average inhibition zone were  $> 10$  mm, which indicated strong antibacterial inhibitory power. Based on the data in table 3, the Garden Balsam leaf extract formulated in the peel-off gel mask produced inhibition zone of *Staphylococcus aureus* bacteria, indicating the presence of antibacterial activity.

## DISCUSSION

### Garden Balsam Leaf Extraction

Extraction of secondary metabolite compound of the Garden Balsam leaf was carried out by maceration method. Ethanol, one of universal solvent, was used for extraction which attracted most of the compounds from the sample. Flavonoid was one of the most desired compound in sample.<sup>7</sup>

### TLC Test of Garden Balsam Leaf Extract

Qualitative test using thin layer chromatography (TLC) method was carried out to find the existence of flavonoid quercetin in the Garden Balsam leaf extract. The extract's Rf (0.35) was by known quercetin's Rf.<sup>14</sup> There were not any interaction data found between contents of the extract so that the material remains stable during storage and preparation.<sup>15</sup>

### Viscosity

Viscosity test aimed to determine the thickness of the gel. Viscosity associated with the ability of dosage form to flow and affect the spreadability. If viscosity increases then the dispersion will decrease.<sup>5</sup> The gel's viscosity was  $> 20,000$  for all of formulations. The more HPMC in a the formula, the thickness of the gel preparation will increase (Table 2). One-way ANOVA got significance value 0.000 ( $p < 0,05$ ) which means there was any viscosity difference from the formulation I, II, and III. HPMC concentration variation influenced the viscosity of the peel-off gel mask preparation.

### pH

The pH test was performed to determine whether the peel-off gel mask preparation was acceptable to the skin pH, which related to safety and comfort of the dosage form. The gel's pH was around 6 for all of the formulations. This result indicated acceptable pH to the skin which is ranging from 4-6.5.<sup>12</sup> One-way ANOVA, confirmed the significant difference between those three formulae. The presence of HPMC concentration variation in peel-off gel mask of Garden Balsam extract affects pH of the dosage form.

### Drying Up Time

The drying up time test was carried out to determine drying speed of the gel on the skin to form a homogenous film layer. The ideal result of such gel dosage form is in the range of 15-30 minutes. The variation of HPMC on the peel-off gel mask affected the drying up time significantly. The more concentrations of HPMC, the shorter the drying up time. HPMC is a polymer forming the viscous matrix in the form of a solution, so it needed less water in the gel. Thus, the water evaporated during drying was only a little, so the drying up time was short.

### Spreadability

Spreadability test is the measurement of the ability of the gel to spread when applied to the skin. Variations of HPMC on the peel-off gel mask appeared to affect the spreadability. The higher the spreadability, the better the peel-off gel mask could spread in the skin area applied.<sup>12</sup> Spreadability is inversely proportional to viscosity. The higher the viscosity due to HPMC concentration, the smaller spreadability of the peel-off gel mask

### Antibacterial Activity

This antibacterial activity test was performed to see the effect of antibacterial after formulated in a peel-off gel peel-off mask preparation.<sup>7</sup> The method used for the antibacterial activity test was the agar-well diffusion method. The positive control was gel preparation that has been circulating in the market that is Benzoyl Peroxide

5%, which function was to compare whether peel-off gel mask preparation of Garden Balsam leaf extract that has been made can have the same effect with positive control used. Negative control also being used is the base of the peel-off gel mask preparation (the preparation without Garden Balsam leaf extract), which measure the effectiveness of the dosage component.

Antibacterial activity test showed inhibition zone diameter differences on *Staphylococcus aureus* bacteria between extracts, positive control, and peel-off gel masks. The Garden Balsam leaf extract produced the largest inhibitory zone diameter, which is possible because of the form of pure extract without any additional excipient. Negative control showed no inhibition zone indicating the inertness of the base peel-off gel mask formulation. It can be concluded that the antibacterial activity of the gel was purely caused by the extract's active compound.

The preparation of the peel-off gel mask has a smaller inhibitory zone than the positive control (benzoyl peroxide 5%). Based on the results data in table 3, it can be concluded that peel-off gel masks containing 15% Garden Balsam leaf extract had strong antibacterial activity against the *Staphylococcus aureus* bacteria.<sup>16,17</sup> The compound that is thought to play an antibacterial role is quercetin group. Quercetin as an antibacterial mechanism is by inhibiting DNA-gyrase bacteria. Quercetin can also increase the permeability of bacterial cell membranes so that ATP synthesis, membrane transport and motility are impaired

## CONCLUSION

HPMC variations affected the viscosity, pH, drying time and spreadability of the peel-off gel. The greater the HPMC concentration, the higher the viscosity, the shorter drying up time, and the narrower spread. 1.5% HPMC in the preparation showed the most optimum results. Formulation of Garden Balsam leaf extract into peel-off gel mask produced antibacterial activity against *Staphylococcus aureus* with a robust inhibitory category.

## CONFLICT OF INTEREST

There is not any conflict of interests in this research and publication.

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