COMPARATIVE STUDIES ON PHYSICOCHEMICAL PROPERTIES OF MORINDA CITRIFOLIA GEL AND OINTMENT FORMULATIONS

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INTRODUCTION
Herbal therapy has been used as traditional medicine as well as in alternative medicine practiced in developing countries. The widespread interest in drugs derived from plants is due to the belief that plants are safe and dependable with fewer side effects. Review of literature revealed that traditional plant drugs are beneficial for several skin-related problems and for wound healing [1-3]. Thailand has been promoting the use of herbs for disease treatment or to be used as a health food supplement. Therefore, the government has made a policy to support research and development of herbal medicines to add value to herbal drug products and to reduce importing drugs from abroad. Furthermore, it is a career option for labor which leads to improving the economy.

Morinda citrifolia commonly known as “Noni” is a small tree native to Southeast Asia. It is also called as Indian Mulberry, Nonu or Nonu, Cheese fruit, and Nhau in various cultures throughout the world. Components isolated from noni include scopoletin, octanoic acid, potassium, Vitamin C, terpenoids, alkaloids, anthraquinones, sotesterol, β-carotene, Vitamin A, flavone glycosides, and linoleic acid. The leaf contains flavonol glycosides, beta-carotene, and iridoid glycosides [5,6]. It is also reported to have a broad range of nutritional and therapeutic values for cancer, infection, arthritis, diabetes, asthma, hypertension, and pain. It also has muscle stimulatory and antistimulative effects along with antibacterial, antiviral, anti-tubercular, antitumor, antihelminthic, analgesic, hypotensive, and immunological effects. Noni has been used as a traditional remedy to treat broken bones, deep cuts, bruises, sores, and wound [7]. The fresh leaf is used for wounds treatment and as a poultice for broken bones in most parts of India [8]. Previous researches have shown that extracts from hexane ethanol and methanol of noni show wound healing capability by adding ligand binding to platelet-derived growth factor and A2b receptors [9]. Previous researches showed 10% of topical Morinda ethanol extract gel had a significant effect on rat skin excisional wound healing compared to 10% of povidone-iodine [10]. At present, products from noni extract have various forms such as capsule, shampoo, soup, and supplement but topical form not a development that is still lagged. The current study is comparative studies on the physico-chemical properties of Morinda citrifolia gel and ointment formulations.

MATERIALS AND METHODS
Collection of plant material
Fruits of Morinda citrifolia were collected from Mahasarakham province, Thailand, in October 2018. The plant material was authenticated by associate researcher Nirun Vipunngeun in the Department of Pharmacognosy, College of Pharmacy, Rangsit University. A voucher specimen was deposited in our laboratory. They were washed with distilled water, air dried, and then made to a fine powder with a mechanical grinder.

Extraction
About 1.6 kg of the powder sample was suspended in 9 L of methanol on a hot plate (30°C) for 24 h. After extraction, the sample was filtered using a fine muslin cloth followed by a filter paper (Whatman No. 1) and concentrated at 45°C using a rotary vacuum evaporator. The preliminary phytochemical screening was carried out by methanolic extract for the presence of phytoconstituents [11,12].

Phytochemical screening methods
Anthraquinone
The extract (10 g) was boiled with 20 ml of 1% hydrochloric acid and 3% hydrogen peroxide 2 ml for 15 min. The extract was filtered and then waited until cooled. The extract was added to dichloromethane 10 ml and ammonium hydroxide 3 ml. The pink color of the base layer indicated the presence of anthraquinone [13].

Cardiac glycosides
The extract (10 g) was boiled with 20 ml ethanol for 15 min. The mixture (5 ml) was placed in an evaporating dish and evaporated to dryness.
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phenols, and flavonoids which corresponds to previous research and shows that the extracted method of this researcher is reliable. The results obtained are summarized in Table 3.

**Evaluation of topical gel and ointment**

**Physical evaluation**

All physical parameters were inspected through visual inspection. Results showed that gel formulation was found only to be changed in color (other physical parameters did not change). However, ointment formulation was found to have a change in color in all of the conditions with a homogeneity change in heating-cooling cycle condition only. The results are shown in Tables 4 and 5.

**Table 4: Physical evaluation of gel formulation**

| Physical evaluation | Initial | 36 days after |
|---------------------|---------|---------------|
|                     | 25±2°C  | 4°C           | Heating-cooling cycle |
| Gel formulation     |         |               |                         |
| Physical appearance | Clear   | Clear         | Clear                   |
| Color               | Light brown | Dark brown   | Brown                  |
| Odor                | Characteristic | Characteristic | Characteristic         |
| Homogeneity         | Good    | Good          | Good                   |
| pH                  | 5.42±0.09 | 5.87±0.14     | 5.91±0.15              |
| Viscosity           | 11980.96±16.5 | 11830.56±19.43 | 12550.80±25.60        |

**Table 5: Physical evaluation of ointment formulation**

| Physical evaluation | Initial | 36 days after |
|---------------------|---------|---------------|
|                     | 25±2°C  | 4°C           | Heating-cooling cycle |
| Ointment formulation|         |               |                         |
| Physical appearance | Semi-solid | Semi-solid | Semi-solid |
| Color               | Light brown | Dark brown | Brown |
| Odor                | Characteristic | Characteristic | Characteristic |
| Homogeneity         | Good    | Good          | Separate layer         |
| pH                  | 5.56±0.11 | 5.62±0.12 | 5.73±0.14 |
| Viscosity           | 58980.21±75.68 | 59240.96±68.50 | 81940.72±85.27 |

**Table 6: Chemical evaluation of gel and ointment formulation**

| Formulations       | Scopoletin (mg/100 g sample) | 36 days after | Heating-cooling cycle |
|--------------------|-------------------------------|---------------|------------------------|
|                     | Initial | 25±2°C | 4°C |                          |
| Gel formulation     | 16.38±0.56 | 15.46±0.19 | 15.81±0.09 | 12.87±0.22* |
| Ointment formulation| 4.89±0.34* | 2.58±0.06† | 3.55±0.06† | 0.90±0.02† |

*p<0.05 when compared to gel formulation on the initial day. †p<0.05 when compared to ointment formulation the initial day.

**Measurement of pH**

pH of the gel and ointment was measured using digital pH meter. The gel and ointment formulations average value was about 5.77±0.15 and 5.68±0.14, respectively. The results are given in Tables 4 and 5.

**Viscosity**

The viscosities of gel and ointment formulations were determined using Brookfield Viscometer. Measuring with all temperature conditions after 36 days, viscosities in the gel formulation did not change, but it was increased in the ointment formulation at the heating-cooling cycle condition only. The viscosities of the formulations were reported in Tables 4 and 5.
Chemical evaluation

The chemical stability of the formulations was evaluated by detecting and compared the amount of scopoletin at various times and temperature conditions. Results show that on the initial day, scopoletin in a gel formulation was found to be three folds higher than in ointment formulation. On day 36, scopoletin in gel formulation decreased without significant compared to the initial day at both room and cold temperature, while it had significant decrease in heating-cooling cycle conditions. Scopoletin in the ointment formulation had a significant decrease in all tested conditions. The results are shown in Figs. 1-3 and Table 6.

DISCUSSION

*M. citrifolia* is a widely used herb and commonly found in Southeast Asian countries. It has been used in medicine for a long time. It has
various medicinal properties such as helping to eliminate toxins in the body, stimulate the immune system, antioxidants, anti-inflammatory antibacterial, antiviral anti-tuberculosis, antitumor, anthelmintic, analgesic, hypotensive, and wound healing effects. In vivo studies, it supports the wound healing effect due to the found active substances in M. citrifolia extracts, namely proxeronine, scopoletin, anthraquinone, vitamins, and amino acids [19,20]. Those active substances play a role in reducing inflammation and wound healing [21-23]. This study found that M. citrifolia extract contains active substances such as anthraquinone, cardiac glycosides, coumarins, tannins, alkaloids, phenols, and flavonoids which corresponds to previous research and shows that the extracted method of this researcher is reliable.

At the present, products from noni extract have various forms but the topical form, gel, or ointment, detailed information regarding its wound healing capability are not completely documented. Thus, the studies were first reported to comparative studies on the physico-chemical properties of noni formulation in various conditions. Development and evaluation of M. citrifolia extract in gel and ointment formulations contain 10% extract. Physical stability test of the formulations was carried out for 36 days by measuring in the initial (0 days) and the second time (36 days after) at various temperature conditions. The physical stability of formulations was evaluated using physical parameters such as physical appearance, color, odor, and homogeneity. All physical parameters were inspected through visual inspection. Results showed that gel formulation was found to be better than ointment. All physical parameters did not change the only color to be changed. However, ointment formulation was found to have a change in color in all of the conditions with a homogeneity change in heating-cooling cycle condition only which change in color of gel and ointment formulation maybe caused by the oxidizing reaction. Chemical stability gets evaluated by detecting scopoletin on the initial day at various temperature conditions. The scopoletin was found to be three folds more in gel formulation than in ointment formulation. Moreover, after 36 days only at heating-cooling cycle condition found scopoletin significant decrease, but ointment formulation found scopoletin significant decrease in all conditions. The ointment formulation found scopoletin less than gel formulation maybe the formulation was a separated layer and physical appearance very hard due to the formulation not dissolve well when measured using the HPLC, its found scopoletin less. Furthermore, ointment formulation has properties of drug release lower than gel formulation [24-26]. Therefore, the detection of scopoletin found less which according to the experimental results. At heating-cooling cycle condition is not an appropriate store of this gel and ointment formulations.

CONCLUSION
Development and evaluation of M. citrifolia extract in gel and ointment formulations; based on the results of all experiments, it can be said that gel formulation is suitable for development due to its physical and chemical stability results which were better than ointment formulation results. Thus, gel formulation could become a media to be used for its medicinal properties. Results found in this research can be used as the base for further development of the gel formulation to obtain a formulation that can be used in drugs or in the form of topical treatment for wound healing or anti-inflammation effect.

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AUTHORS' CONTRIBUTIONS
PataweeKorn Ketkomol conceived and designed the analysis, collected the data and performed the analysis. Tadsane Punjanon supervised the findings of this work. All authors discussed the results and contributed to the final manuscript.

CONFLICTS OF INTEREST
The authors declare that they have no conflicts of interest.

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