Research Article

Dr. ProBee: Game Changer for Regenerative and Pain Relief

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Abstract: *Stingless bee Trigona species, or locally known as 'kelulut', has long been exploited as a source of traditional medicine due to presence of various important nutritional and medicinal values. Dr. ProBee is a combination of doctor, propolis and bee; refer to our research-based product introduces in pre-clinical settings, which reference to the needs of an alternative potential. This product is formulated based on natural resources namely propolis from stingless bee Trigona thoracica as one of the main ingredients. Stingless bee propolis and honey are among of the popular complementary and alternative medicine (CAM) extensively used worldwide. The novelty of this product it is a newly formulated of topical gel formulation enriched with polyphenols compounds as antioxidants, anti-inflammatory and analgesic effect, thus potential for the new application in medicine and healthcare especially for the pain and regenerative purposes. The product already tested for its usefulness, and effectiveness via laboratory study. It has undergone few stages include toxicity test in pre-clinical testing and proves non-toxic, and antinociceptive effect of propolis extracts using pain behavioural animal models. Another unique finding from our study is synergistic effect with another pain medication (morphine as a gold standard for opioid) and it enhances the effect and suitable to include in multimodal approaches for pain management. Dr. ProBee may emerge to be commercial ability and good market potential. With the propolis harvested locally, it may support the effort of the agropreneurs in Malaysia. We also promote SDG 3; good health and well-being by developing the products to aid pain management and tissue regeneration.*

Keywords: Analgesic; Anti-inflammatory; Complemetary and alternative medicine; Dr. ProBee; Pain management; Propolis; Stingless bee.

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1. INTRODUCTION

Propolis is a complex resinous substance produced by stingless bee. By lining the inner layer of nest cavities with propolis, stingless bee can reduce the width of hive entrances. It has about 300 biochemical components that have been discovered, the majority of which are polyphenols, flavonoid glycosides, flavonoid aglycones, phenolics, and ketones (Bankova et al., 2014). The chemical makeup of Malaysian propolis is unique, and other propolis kinds have never been documented to contain novel

substances like 20-Hydroxy-24-dammaren-3-one and -panasinsene from sesquiterpene esters (Hossain et al., 2022). According to studies, the Malaysian stingless bee propolis extract exhibits antioxidant and anti-hyperglycaemic properties, cardioprotective characteristics, anti-inflammatory, and healing properties (Ahmed et al., 2017; Asem et al., 2020; Jacob et al., 2015; Nna et al., 2021; Yeoh et al., 2021).

Pain can be treated with invasive or non-invasive methods. Pharmacologic treatment, mostly systemic administration, uses non-invasive methods. For the treatment of somatic and visceral nociceptive pain, non-steroidal anti-inflammatory medications (NSAIDs) and opioids are typically used; anticonvulsants and antidepressants are used for the treatment of neuropathic pain with positive and negative symptoms, respectively. The NSAIDs have anti-inflammatory, antipyretic, and analgesic properties because they inhibit the cyclo-oxygenase pathway, but their therapeutic efficacy is limited. The NSAIDs' adverse responses (ADRs) include gastrointestinal issues, generalised edoema, and an increased propensity to bleed. The opioids only have an analgesic impact when they bind to opioid receptors; they have no anti-inflammatory, antipyretic, or other effects. The opioids' adverse drug reactions (ADRs) range from itchiness and nausea/vomiting to respiratory and cardiovascular depression, as well as constipation (Kim et al., 2020). These problems triggers the researchers to explore another new potential pharmaceutical products to be used synergistically with conventional treatments as multimodal approach in pain management.

2. METHOD & MATERIAL

2.1 Propolis collection and preparation

Geniotrigona thoracica produces fresh propolis, which was procured at Kuala Besut, Terengganu Darul Iman, Malaysia. After being rinsed with tap water to eliminate dirt and foreign objects, the propolis was stored at -20°C. A commercial blender was used to turn the frozen propolis into powder. After that, distilled water was used to dissolve the freeze powder form (40g) for 24 hours. Into a fresh volumetric flask was put the crude extract solution. Three separate extractions of the residue with distilled water were completed. The final total extract was then lyophilized using a freeze dryer and filtered through No. 41 filter paper (Whatmann®). At -4°C, the dried brown APE was kept cool.

2,2 Identification of phenolic acids by HPLC

High Performance Liquid Chromatography-UV Spectrophotometer (HPLC-UV) (Shimadzu Corp., Japan) was used to identify phenolic acids. A LC unit with a UV-Vis spectrophotometer detector (SPD-10A) is part of a chromatographic system.

2.3 In vivo toxicity tests

The experiment was conducted according to the protocols in OECD Guideline 420 (OECD, 2001). The APE extract was dissolved in 1% (v/v) DMSO in aqueous solution and administered orally at doses of 400 (APE 400), 1000 (APE 1000) and 2000 mg/kg of body weight (BW) (APE 2000) (volume of 10 mL/kg BW), whereas the control group received only the normal saline in the same volume.

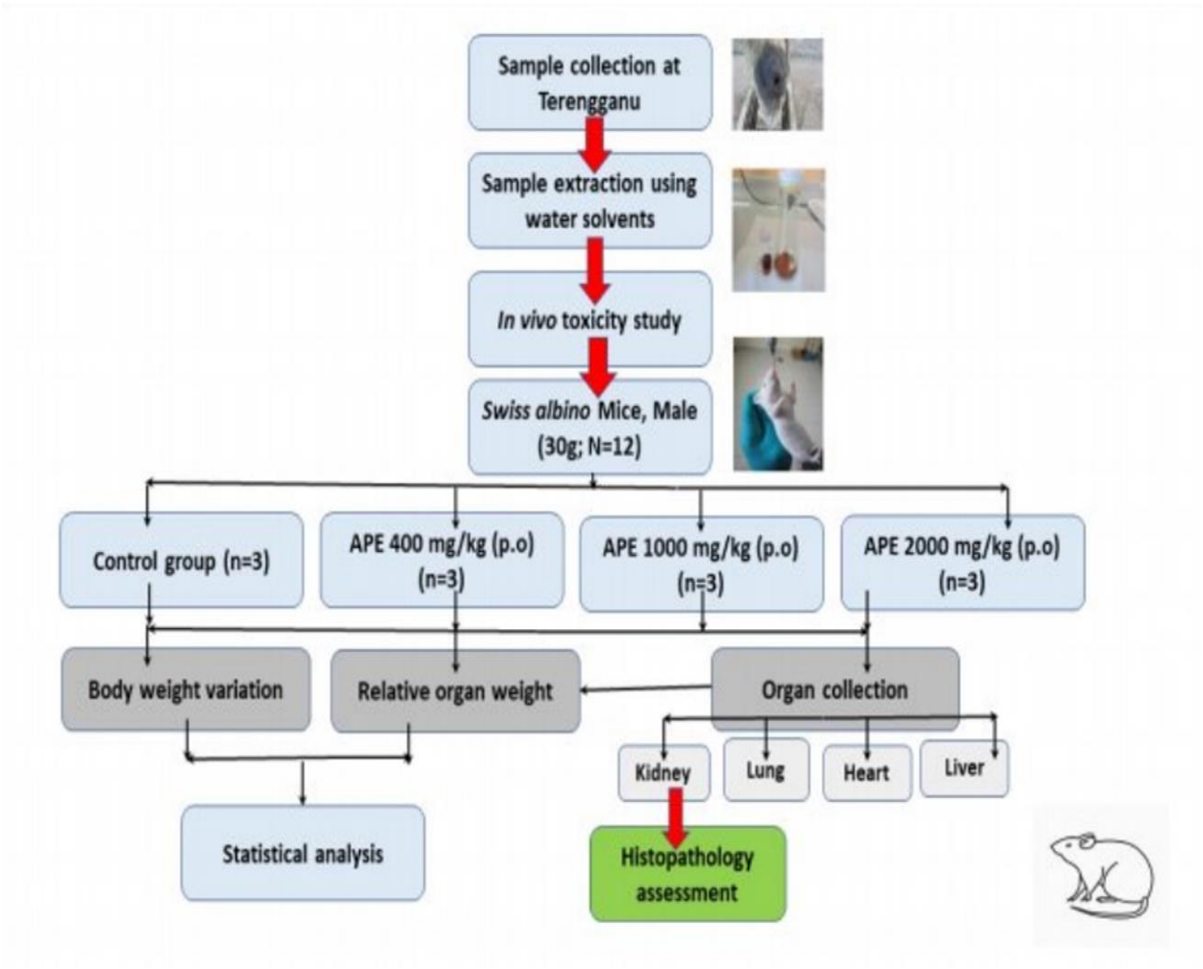

Figure 1. This is a figure of in vivo toxicity tests.

2.3 Animal pain models

To assess the antinociceptive effect, male Albino mice were administered orally with APE (400 mg/kg, 1000 mg/kg, and 2000 mg/kg) before pain induction. The antinociceptive properties of the APE using 0.6% (v/v) acetic acid-induced abdominal constriction via intraperitoneally, hot plate test (52.5°C + 0.5°C), and 20 μL of 5% (v/v) formalin-induced paw licking test were evaluated.

2.4 Development of Dr. ProBee

Extracted propolis was transformed into topical gel formulation to meet our protocol criteria and ready to be used as a topical application to the skin.

Figure 1. Final product of Dr. ProBee.

3. FINDINGS

3.1 High Performance Liquid Chromatography (HPLC) Analysis

HPLC analysis revealed the samples of APE contain p-coumaric acid (PCA) and caffeic acid (CA).

3.2 In vivo toxicity tests

The absence of abnormalities was demonstrated in the current investigation by the treated mice's physical and general behaviours, which did not change after receiving oral dosages of APE ranging from 400 to 2000 mg/kg of body weight (Table 2). Throughout the course of 14 days of APE supplementation, male mice in the control and treatment groups exhibited no outward symptoms of toxicity. After receiving APE for 14 days, male mice in the control and treatment groups displayed no observable toxicity.

3.3 Animal pain models

The animal model used to test the analgesic effects of irritating substances intraperitoneally in demonstrating peripherally mediated nociceptive responses. When tested utilising the acetic acid-induced abdominal constriction test, the current study's findings showed that APE successfully lowers nociceptive response.

3.4 Dr. ProBee

Dr. ProBee is a combination from words Doctor, Propolis and Bee. It is hoped to become a game changer in complimentary and alternative medicine (CAM) for regenerative and pain relief. It is enriched with polyphenols compound.

4. DISCUSSION

Dr. ProBee is a problem-solving initiative from one of the healthcare issues globally. Acute and chronic pain present a serious clinical problem. Inevitable musculoskeletal (muscle and joint) pain problems increase expenditure, loss of productivity and decrease patient's quality of life. It is a product with new exploratory scientific data and testing beneficial pharmaceutically active compounds for regenerative and pain relief process.

Dr. ProBee is a novel product formulated of topical gel formulation, enriched with polyphenols compounds and analyzed with high performance liquid chromatography (HPLC). The product is not only relief the pain, but previous study showed it effectiveness in wound healing and tissue regenerative. The impact of this product not for experimental study only, but it promotes and support Sustainable Development Goals (SDG); SDG 3 for good health and well-being, and SDG 15 for conserving life on land. Another impact of this project is for talent development; produce postgraduate student with scientific research experience, and for economy development; support economy cycle from starting to get the resources of propolis until producing the topical gel of propolis, Dr. ProBee.

This product is readily to use and potential to commercialize in the market. The formula and solution have potential to develop into other products such as ready-made wound dressing or plaster enriched with polyphenols. The therapeutic properties of propolis from stingless bee are not only limited to regenerative and pain relief but also for other several therapeutic properties such as antibacterial, antifungal, anticancer and anti-inflammatory agent.

5. CONCLUSION

The current product, Dr. ProBee, provides evidence by establishing the antinociceptive profile of aqueous propolis extracts from Geniotrigona thoracica species of Malaysian stingless bees and by identifying the chemical components of the samples, and turned it into topical gel formulated with enriched polyphenols as a game changer for regenerative and pain relief.

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References

Ahmed, R., Tanvir, E.M., Hossen, M.S., Afroz, R., Ahmmed, I., Rumpa, N.-E.-N., Paul, S., Gan, S.H., Sulaiman, S.A. & Khalil, M.I. (2017). Antioxidant properties and cardioprotective mechanism of Malaysian propolis in rats. Evidence-Based Complementary and Alternative Medicine, 1–11. https://doi.org/10.1155/2017/5370545

Asem, N., Abdul Gapar, N.A., Abd Hapit, N.H. & Omar, E.A. (2020). Correlation between total phenolic and flavonoid contents with antioxidant activity of Malaysian stingless bee propolis extract. Journal of Apicultural Research, 59(4), 437–442. https://doi.org/10.1080/00218839.2019.1684050

Bankova, V., Popova, M. & Trusheva, B. (2014). Propolis volatile compounds: Chemical diversity and biological activity: A review. Chemistry Central Journal, 8(1), 1–8. https://doi.org/10.1186/1752- 153X-8-28

Hossain, R., Quispe, C., Khan, R.A., Saikat, A.S.M., Ray, P., Ongalbek, D., Yeskaliyeva, B., Jain, D., Smeriglio, A., Trombetta, D., Kiani, R., Kobarfard, F., Mojgani, N., Saffarian, P., Ayatollahi, S.A., Sarkar, C., Islam, M.T., Keriman, D., Uçar, A., Martorell, M., Sureda, A., Pintus, G., Butnariu, M., Sharifi-Rad, J. & Cho, W.C. (2022). Propolis: An update on its chemistry and pharmacological applications. Chinese Medicine, 17:100, 1–60. BioMed Central Ltd. https://doi.org/10.1186/s13020-022-00651-2

Jacob, A., Parolia, A., Pau, A. & Davamani Amalraj, F. (2015). The effects of Malaysian propolis and Brazilian red propolis on connective tissue fibroblasts in the wound healing process. BMC Complementary and Alternative Medicine, 15(294), 1–11. https://doi.org/10.1186/s12906-015- 0814-1

Kim KH, Seo HJ, Abdi S, Huh B. (2020). All about pain pharmacology: what pain physicians should know. Korean J Pain. Apr 1;33(2):108-120. doi: 10.3344/kjp.2020.33.2.108. PMID: 32235011; PMCID: PMC7136290.

Nna, V.U., Abu Bakar, A.B., Md Lazin, M.R.M.L. & Mohamed, M. (2018). Antioxidant, anti-inflammatory and synergistic anti-hyperglycemic effects of Malaysian propolis and metformin in streptozotocin–induced diabetic rats. Food and Chemical Toxicology, 120, 305–320. https://doi.org/10.1016/j.fct.2018.07.028

Yeoh, B.S., Omar, N., Mohammad, M., Mokhtar, S.S. & Ahmad, R. (2021). Antioxidative propolis from stingless bees (Heterotrigona itama) preserves endothelium-dependent aortic relaxation of diabetic rats: The role of nitric oxide and cyclic guanosine monophosphate. Brazilian Journal of Pharmaceutical Sciences, 57, 1–9. https://doi.org/10.1590/s2175-97902020000419187