Trop J Nat Prod Res, June 2023; 7(6):3153-3157 ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Original Research Article

Evaluation of Pain-Relieving Effect of The Herbal Remedy "Hoang Ky Que Chi Ngu Vat Thang" in Animal Model

Hoang M. Le[1]*, Nuong T.K. Huynh[2], Tuan T. Vo[3], Tien T.B. Nguyen[4], Quan M. Nguyen[5]

[1]*Department of Traditional Medicine, Can Tho University of Medicine and Pharmacy, Can Tho, Vietnam*
[2]*Kien Giang Traditional Medicine Hospital, Kien Giang, Vietnam*
[3]*Department of Traditional Medicine, Ho Chi Minh City Medicine and Pharmacy University, Ho Chi Minh, Vietnam*
[4]*Department of Traditional Medicine, Can Tho University of Medicine and Pharmacy, Can Tho, Vietnam*
[5]*Department of Drugs quality control – Analytical chemistry – Toxicology, Can Tho University of Medicine and Pharmacy, Can Tho, Vietnam.*

ARTICLE INFO

Article history:
Received 13 April 2023
Revised 16 May 2023
Accepted 31 May 2023
Published online 01 July 2023

ABSTRACT

Musculoskeletal pain is a significant burden on individuals and society. "Hoang ky que chi ngu vat thang" is a traditional Vietnamese remedy used to treat this condition, but there is a lack of clinical evidence regarding its pain relief mechanism. The objective of this study is to evaluate the analgesic effect of "Hoang ky que chi ngu vat thang" in Swiss mice. The central analgesic effect was evaluated using the hot plate latency test, and the peripheral analgesic effect was evaluated using the writhing test with acetic acid. The results of this study showed that statistically significant differences were observed between the treated groups with "Hoang ky que chi ngu vat thang" and the standard group 1 and control group ($p < 0.05$). Furthermore, the number of writhing pain during 25 minutes after acetic acid injection in both groups with dose 14.112 and dose 28.224 was significantly lower ($p < 0.01$) compared to the reference group (Lysine acetylsalicylate). Conclusively, oral doses of 14.112 g/kg and 28.224 g/kg of "Hoang ky que chi ngu vat thang" have a good analgesic effect via both central and peripheral analgesic mechanisms.

Keywords: Pain relief, Hoang ky que chi ngu vat thang, traditional medicine, mice

Introduction

The theory proposed by Hai Thuong Lan Ong in the 18th century emphasizes the importance of nourishing blood for the treatment of leprosy, improving fire for curing colds, and stimulating the spleen for healing rheumatism. While rheumatic drugs are commonly used, the theory suggests the use of qi and blood tonics to primarily control the evil in the two meridians of the kidney and supplement the essence to affect the tendons and bones. In line with this theory, Hai Thuong Lan Ong used medicinal herbs such as *Morinda officinalis* How., *Polygonum cuspidatum*, and *Clematis chinesis* Osbeck to treat musculoskeletal diseases[1]. The Hoang ky que chi ngu vat thang, an ancient remedy in the Truong Trong Canh's Kim quy yeu luoc, includes herbs such as *Astragalus membranaceus* Bge, *Cinnamomum cassia* Presl, *Radix Paeoniae lactiflorae*, *Rhizoma Zingiberis*, *Zizyphus jujubae* Mill, and *Glycyrrhiza uralensis* Fisch, which are believed to enhance aerodynamics, regulate meridians, and promote harmony in Oriental medicine[2]. Medical literature and studies have shown the anti-inflammatory and analgesic effects of these remedies in peripheral neuropathic pain[3,4,5,6,7,8]. Further research is needed to better understand the underlying mechanisms of these medicinal herbs and their potential use in the treatment of various ailments.

*Corresponding author. E mail: lmhoang@ctump.edu.vn
Tel: +84-973431666

Citation: Le HM, Huynh NTK, Vo TT, Nguyen TTB, Nguyen QM. Evaluation of Pain-Relieving Effect of The Herbal Remedy "Hoang Ky Que Chi Ngu Vat Thang" in Animal Model. Trop J Nat Prod Res. 2023; 7(6): 3153-3157 http://www.doi.org/10.26538/tjnpr/v7i6.14

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

Osteoarthritis is a degenerative joint disease that is commonly associated with pain and inflammation. Although cartilage loss, synovial bursitis, joint space constriction, and joint deformity damage are known contributors to pain, inflammation can also be a significant factor. Inflammation can exacerbate the pain experienced by individuals with osteoarthritis by sensitizing pain receptors and producing pro-inflammatory mediators. To address the issue of pain, various treatments have been developed, including the use of Hoang ky que chi ngu vat thang. This traditional remedy has been tested in models of pain caused by various agents, including temperature and chemical agents such as acetic acid[9, 10,11, 12, 13, 14]. While the results of these studies are promising, further research is necessary to fully understand the mechanisms of action of Hoang ky que chi ngu vat thang and its potential as a treatment option for osteoarthritis-related pain.

Hoang ky que chi ngu vat thang is a popular treatment option for a variety of diseases, including peripheral neuropathic pain caused by conditions such as diabetes, inflammatory rheumatoid arthritis, shoulder-arm syndrome, and disc herniation. At the Kien Giang Traditional Medicine and Pharmacy Hospital, Vietnam, it is currently the clinicians' first choice for treating these conditions due to its significant analgesic and anti-inflammatory effects. To further investigate the analgesic mechanism of Hoang ky que chi ngu vat thang, we designed a study to evaluate the analgesic effect of the remedy in Swiss mice. The aim of this study is to provide scientific evidence supporting the widespread use of Hoang ky que chi ngu vat thang in clinical practice. The findings of this study may contribute to the development of new treatments for pain management and provide insights into the mechanisms underlying the therapeutic effects of traditional remedies.

Materials and Methods

Hoang ky que chi ngu vat thang remedy

Table 1 presents the specific ingredients employed in the formulation of the remedy. The dosages of these ingredients, expressed in grams of dried herbs, were determined based on a total remedy weight of 60

grams, which is intended for daily consumption by a single individual. For human dosing, a reference body weight of 50 kg was used, resulting in a recommended dose of 1.2 g/kg. Using this dosage, the expected adequate amount for Swiss mice was calculated to be 14.112 g/kg, based on the assumption that the conversion factor from humans to mice is approximately 11.76. Similarly, for Swiss rats, the tentative effective dosage was estimated to be 7.764 g/kg, calculated by multiplying the human dose of 1.2 g/kg by the conversion factor of 6.47[15]. The experimental remedy was administered orally to the Swiss mice using a specialized curved needle designed to facilitate insertion into the stomach[16].

Animal experiments

Ethical approval for the study was obtained from the Medical Ethics Council of Can Tho University of Medicine and Pharmacy, Can Tho city, Vietnam (approval number 22.004.HV/PCT-HDDD). Swiss adult mice, with a weight range of 18-22 grams, were used in accordance with experimental standards. Animals were sourced from the Laboratory Animal Breeding Board, Military Medical University, Vietnam, and were raised in the laboratory for at least one week prior to the commencement of the study[17]. The animals were fed a standard diet and provided with boiled and cooled clean water to drink ad libitum[18]. Daily observations were made and experimental results were routinely monitored and recorded. After adaptation to the standard laboratory conditions, the experimental mice were divided randomly into four groups (n = 10) based on two models of evaluation. The control group (Con), codeine phosphate group (CDP, 20 mg/kg), and different HCQCNV dose treatment groups (14.112 g/kg, 28.224 g/kg) were established for the analgesic activity experiment. The second experiment followed a similar allocation, with the CDP group replaced by an Lysine acetylsalicylate dose (ASG 180 mg/kg) group. All drugs were administered intragastrically to each group for five consecutive days. The Con group mice received distilled water in the same volume (10 mL/kg) as the drugs, administered via the oral route.

Analgesic activity evaluation

The hot plate latency test, developed by G. Woolfe and A. D. Macdonald, is a widely-used research method based on thermal stimuli principles for evaluating analgesic effects. In this test, mice were exposed to pain by placing their paws on a hot plate maintained at a constant temperature of 56°C[19,20]. The reaction time to the thermal stimulation was measured as the time taken for the mice to reflexively lick their hind paw. Mice that reacted too quickly (less than 8 seconds) or too slowly (after 30 seconds) were excluded from the study. The response times to thermal stimulation were compared before and after the administration of the reagent, and between groups of mice.

The experiment was conducted at two different time points - before and after administering the reagent for five days. On the 5th day, 1 hour after the mice received the drug, they were placed on the hot plate with a stable temperature of 56°C. The pain tolerance time of the mice was calculated from the time they placed their paw on the hot plate until they licked their hind paw. The pain relief effect was evaluated by calculating the percentage prolongation of pain tolerance time in the mice. The criteria for elimination were mice that responded before 8 seconds or after 30 seconds during the initial pre-treatment evaluation conducted by the researchers.

Table 1: Ingredients of Remedy

Materia medica	Science name	Quantity
Hoang ky	*Astragalus membranaceus* Bge	12
Que chi	*Cinnamomum cassia* Presl	09
Bach thuoc	*Radix Paeoniae lactiflorae*	09
Sinh khuong	*Rhizoma Zingiberis*	12
Cam thao	*Zizyphus jujubae* Mill	06
Dai tao	*Glycyrrhiza uralensis* Fisch	12

Peripheral reduction evaluation

Writhing tests

The mice were administered the remedy or distilled water orally for five consecutive days

On the 5th day, 60 minutes after oral administration of the remedy, the mice were injected with a 0.6% acetic acid solution at a dose of 0.1 mL/10 g of body weight into the peritoneum. After the intra-abdominal injection of acetic acid, all the mice exhibited cramping pain characterized by abdominal retraction, pressure to the floor, and lengthening of the trunk and hind legs. The onset of pain (defined as the time from acetic acid injection to the first cramping episode) and the number of abdominal writhes in each 5-minute interval until the end of 25 minutes after acetic acid injection were recorded for each mouse in each batch[21].

The analgesic effect of the remedy was calculated by determining the percentage inhibition of abdominal cramps, using the following formula:

$$A\% = \frac{Dc - Dt}{Dc} \times 100$$

where: A (%) – the rate of inhibition of abdominal writhing of the drug trial batch;

Dc – the mean of cramps of the animals in the control group;

Dt – the mean of cramps of the animals in the treated group.

Statistical analysis

The mean ± standard error of the mean was used to express all values obtained from the experiments. The variance of the data was evaluated using the Student's t-test. The significance level was set at $p < 0.05$, which allowed for the assessment of statistical significance between the control and treated groups.

Results and Discussion

Hot plate latency test

Table 2 shows the statistical evaluation of the time of onset of pain response in mice across the four groups prior to treatment, and no significant differences were observed ($p > 0.05$). Compared to the control group, the time to pain response in mice in both doses of HKQCNVT was significantly longer ($p < 0.05$ and $p < 0.01$). HKQCNVT at doses of 14.112 g/kg/day and 28.224 g/kg/day showed significant pain-reducing effects in the hotplate test, which were equivalent to codeine at a dose of 20 mg/kg ($p > 0.05$). Based on the results shown in Table 3, the pain response time in the 28.224 g/kg/day group was longer than in the 14.112 g/kg/day group, indicating a dose-dependent response to HKQCNVT's pain-relieving effects on the hotplate test, although this difference was not statistically significant ($p > 0.05$). Within each group, the pain response time in mice in both doses of HKQCNVT was significantly longer at the time point after drug administration compared to the time point before drug administration ($p < 0.05$ and $p < 0.01$). These results further confirm the pain-reducing effects of HKQCNVT in the hotplate test.

In each group, our research found that the pain response time of mice in the groups using Hoang Ky Que Chi Ngu Vi Thang (HKQCNV) at both dose levels was significantly longer after taking the drug compared to before treatment ($p < 0.05$ and $p < 0.01$). The results of the self-directed comparison confirm the pain-relieving effect of HKQCNV when tested using the "hot plate" method. The observed effect could be attributed to the inhibitory effect of HKQCNV on nerve conduction reflexes from the periphery to the brain. In contrast, the findings of Dinh Thi Lam (2017) did not reveal a significant effect on the lengthening of the response time when evaluating the efficacy of Bach Xa ointment administered 30 minutes before measuring the pain response compared to biological controls[16]. Our research findings indicate a more promising outcome compared to Nguyen Thi Thanh Tu's study in 2015. Their evaluation of the analgesic effect of Hoang Kinh capsules did not demonstrate any significant pain-relieving effect on the nerve mechanism through thermal impact on the skin[19].

Writhing tests

Table 4 shows both HKQCNVT and Lysine acetylsalicylate significantly delayed the onset of colicky pain compared to the control

group ($p < 0.05$). Thus, both HKQCNVT and Lysine acetylsalicylate demonstrated an effect of delaying the onset of colicky pain compared to the control group. The onset time of colicky pain in the HKQCNVT dose 2 group appeared to be longer than in the dose 1 group, but this difference was not statistically significant ($p > 0.05$). Compared to the reference group using Lysine acetylsalicylate, the onset time of colicky pain in the HKQCNVT groups was equivalent, with no statistically significant differences ($p > 0.05$).

Figure 1 illustrates that the number of abdominal writhes in the groups receiving HKQCNV and the standard group using Lysine acetylsalicylate was significantly lower than in the control group during the 5-10 minute and 20-25 minute measurement periods, with p-values less than 0.01. Similarly, during the 10-15 minute and 15-20 minute measurement periods, the number of abdominal writhes in the groups receiving HKQCNV and the standard group using Lysine acetylsalicylate was significantly lower than in the control group, with p-values less than 0.05. However, no statistically significant difference was found in the number of abdominal writhes during the 0-5 minute measurement period between the groups receiving HKQCNV, the standard group using Lysine acetylsalicylate, and the control group ($p > 0.05$).

Table 2: Immersion time before treatment (n = 10)

Group	Immersion time (second, mean ± SD)	P-value (compared to control group)	P-value (compared to standard group)	P-value (compared to dose 14.112 g/kg group)
Control	14.83 ± 3.30	-	-	-
Standard	14.66 ± 2.04	> 0.05	-	-
Dose 14.112 g/kg	14.56 ± 2.98	> 0.05	> 0.05	-
Dose 28.224 g/kg	15.39 ± 2.11	> 0.05	> 0.05	> 0.05

Table 3: Effect of HKQCNV on the pain response time (n = 10)

Group	• Immersion time (second, mean ± SD)	Pain inhibition (%)	P-value (compared to control group)	P-value (compared to standard group)	P-value (compared to dose 14.112 g/kg group)
Control	14.39 ± 2.48	-	-	-	-
Standard	19.95 ± 4.40	38.64	< 0.01	-	-
Dose 14.112 g/kg	17.47 ± 3.15	21.40	< 0.05	> 0.05	-
Dose 28.224 g/kg	19.33 ± 4.11	34.33	< 0.01	> 0.05	> 0.05

Table 4: Effect of HKQCNV on the time of onset of colicky pain

Group	Time of onset of colic pain	P-value (compared to control group)	P-value (compared to standard group)	P-value (compared to dose 14.112 g/kg group)
Control	260.50 ± 66.88	-	-	-
Standard	353.00 ± 102.13	< 0.05	-	-
Dose 14.112 g/kg	345.60 ± 101.61	< 0.05	> 0.05	-
Dose 28.224 g/kg	350.90 ± 93.04	< 0.05	> 0.05	> 0.05

Table 5 shows that compared to the control group, both HKQCNVT groups at 14.112 g/kg and 28.224, as well as the standard group using Lysine acetylsalicylate, exhibited a significant decrease in the total number of abdominal writhes during the 25 minutes following acetic acid injection ($p < 0.01$). Within this 25-minute period, the percentage reductions in the number of abdominal writhes were 32.77%, 28.36%, and 33.40% for the group receiving Lysine acetylsalicylate and the groups receiving HKQCNVT at 14.112 g/kg and 28.224, respectively. No significant difference was found in the number of abdominal writhes between the HKQCNVT groups at both dose levels and the standard group using Lysine acetylsalicylate ($p > 0.05$). Although the group receiving HKQCNVT at 28.224 had fewer colicky pains during the 25 minutes after acetic acid injection compared to the group receiving HKQCNVT at 14.112 g/kg, the difference was not statistically significant ($p > 0.05$).

In addition to the findings on the pain-relieving effect of Hoang Ky Que Chi Ngu Vat Thang, our study also investigated the mechanism of pain induction in mice. We found that peritoneal injection of acetic acid in the abdominal cavity induced an inflammatory response that resulted in pain. This pain was manifested as colicky pain, which caused the mice to flex their bodies, contract their abdomens, press their abdomens against the floor, and lengthen their trunk and hind legs. Medications with analgesic effects were observed to increase the pain threshold, resulting in a delay in the onset of colicky pain and a decrease in the number of episodes. These findings suggest that the mechanism of pain induction in mice can be used as a model for studying the analgesic effects of different medications.

Nguyen Thi Thanh Tu's study evaluated the efficacy of Hoang Kinh capsules at doses of 9.6 g/kg and 28.8 g/kg for three consecutive days, which significantly reduced the number of abdominal cramps at all time points compared to the control group ($p < 0.05$, $p < 0.01$, and $p < 0.001$)[13]. This finding is consistent with our study results. The chemical composition of the herbs in the remedy contains flavonoids that have potent antioxidant effects. These effects reduce free radicals, oxidative stress on the cell membrane phospholipid layer, and the release of inflammatory mediators, which ultimately reduce pain. Our findings are also consistent with several clinical studies demonstrating the effectiveness of the remedy for pain relief in neurological and musculoskeletal conditions[3,4,22]. Overall, our study further supports the

analgesic properties of the herbal remedy and provides a possible explanation for its mechanisms of action.

Limitations of the study should be acknowledged. Firstly, the conversion factor used to calculate the effective dosage for mice and rats was estimated and may not accurately reflect the actual conversion factor. Additionally, the study was conducted on a small sample size, which limits the generalizability of the findings to a larger population. In terms of animal experiments, Swiss adult mice were used in accordance with experimental standards. However, it is important to note that animal models do not always perfectly replicate human physiology, and therefore the results of this study may not be directly applicable to humans. For the analgesic activity evaluation, the hot plate latency test was utilized to evaluate the analgesic effects of the different treatment groups. While this is a widely used research method, it should be noted that thermal stimuli principles may not fully capture the complexity of pain experience in humans. Finally, in the evaluation of peripheral reduction, the writhing test was utilized. This test involves the administration of acetic acid to induce cramping pain in mice. While this is a commonly used method for evaluating the analgesic effects of drugs, it should be noted that the pain experienced by mice in this test may not fully capture the complexity of pain experience in humans.

Conclusion

This study found that the oral administration of Hoang ky que chi ngu vat thang at doses of 14.112 g/kg and 28.224 g/kg had significant analgesic effects both by the Hotplate method and Writhing Tests. These findings suggest that Hoang ky que chi ngu vat thang has the potential to be a useful treatment option for pain management. However, further research is needed to fully understand the mechanisms of action and potential side effects of this remedy.

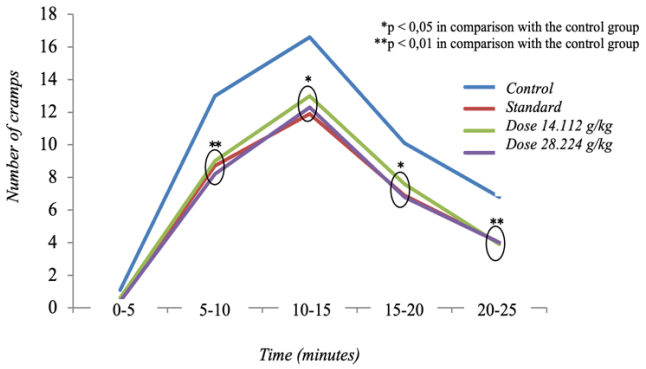

Figure 1: The number of abdominal writhes was measured in each 5-minute interval following the injection of acetic acid in the study groups.

Table 5: Effect of HKQCNV on the total number of abdominal writhes in a group of animals during the 25-minute period following acetic acid injection (n = 10)

Group	Number of abdominal writhes in 25 minutes after acetic acid injection	P-value (compared to control group)	P-value (compared to standard group)	P-value (compared to dose 14.112 g/kg group)	The percentage (%) reduction in the number abdominal writhes compared with the control group
Control	47.60 ± 10.22	-	-	-	-
Standard	32.00 ± 8.56	< 0.01	-	-	32.77
Dose 14.112 g/kg	34.10 ± 8.16	< 0.01	> 0.05	-	28.36
Dose 28.224 g/kg	31.17 ± 8.31	< 0.01	> 0.05	> 0.05	33.40

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgements

The authors express their gratitude to members of the Can Tho University of Medicine and Pharmacy, the Ministry of National Defense of Vietnam and the Drug Research and Production Center, Military Medical Academy, Vietnam by providing laboratory materials and equipment used in executing the research.

References

1. Hai TN, Canh LG. Chinese nomenclature of holistic remedy (Trung Quốc danh phương toàn tập). 2004.
2. Tan DP, Phi L. Epidemiology (Phương tễ học). 2010.
3. Pang B, Zhao TY, Zhao LH. Huangqiguizhi Wuwu Decoction for treating diabetic peripheral neuropathy: a meta- analysis of 16 randomized controlled trials. Neural Regen Res. 2016; 11:1347-1358.
4. Liang L, Wei X, Feng M, Zhu L, Yu J, Yang G, Yin X, Zhou S, Li K, Yang M, Wang X. Huangqi Guizhi Wuwu Decoction for treating cervical radiculopathy: A systematic review and meta- analysis of randomized controlled trials. Med. (Baltimore). 2020; 99:e19137.
5. Wang L, Fan Y, Xin P. The efficacy and safety of Huangqi Guizhi Wuwu decoction for rheumatoid arthritis: A protocol for systematic review and meta-analysis. Med. 2020; 99(36):e2201.
6. Deng B, Jia L, Cheng Z. Radix Astragali-based Chinese herbal medicine for oxaliplatin-induced peripheral neuropathy: a systematic review and meta-analysis. Evid Based Complement Alternat Med. 2016; 2016:2421876.
7. Wang T, Jia Q, Chen T, Yin H, Liang Q. Alleviation of synovial inflammation of juanbi-tang on collagen-induced arthritis and TNF-tg mice model. Front Pharmacol. 2020; 11:45.
8. Chien M, Chuang, C, Lin Y, Yang C, Chen C. Analgesic and anti-inflammatory effects of herbal mixture extracts in mice. Pharmacol Pharm. 2018; 9:1-9.
9. Koster R, Anderson M, Debeer F. Acetic acid for analgetic screening. Fed Proc. 1959; 18:412-417.
10. Woolfe G, Macdonald AD. The evaluation of the analgesic action of pethidine hydrochloride (demerol). J. Pharmacol Exp Ther. 1944; 80:300-307.
11. Rezq S. Anti-Inflammatory, antipyretic, and analgesic properties of *Potamogeton perfoliatus* Extract: in vitro and in vivo study. Molecules 26. 2021; 26:4826.
12. Li Y, Cui HJ, Huang JC, Wu XQ. Clinical study of Jiawei Huangqi Guizhi Wuwu Decoction in preventing and treating

peripheral neuro-sensory toxicity caused by oxaliplatin. Chin J. Integr Med. 2006; 12:19-23.
13. Wang Y, Liu Y, Xi Z, Yu Y, Liu L. A multicenter, randomized, double-blind, placebo-controlled trial evaluating the efficacy and safety of Huangqi Guizhi Wuwutang granule in patients with rheumatoid arthritis. Med. 2019; 98:e14888.
14. An Y. The usage of biological dmards and clinical remission of rheumatoid arthritis in China: A real-world large scale study. Clin Rheumatol. 2017; 36:35-43.
15. Administration of Science Technology and Training (Vietnam). Decision No. 141/QD-K2ĐT on guidelines for pre-clinical and clinical trials of traditional and herbal medicines (Tài liệu chuyên môn "hướng dẫn thử nghiệm tiền lâm sàng và lâm sàng thuốc đông y, thuốc từ dược liệu"). 2015.
16. Lam DT. Research on safety, anti-inflammatory and analgesic effects of cypress extract on experimental and clinical rheumatoid arthritis. 2017.
17. Wang Y, Chen T, Yang C. Huangqi Guizhi Wuwu decoction improves arthritis and pathological damage of heart and lung in TNF-Tg Mice. Front Pharmacol. 2022; 13:e14888.
18. Tran YH, Nguyen TTT, Nguyen PT, Nguyen KT, Duong CX, Tran HM. Effects of *Ganoderma Lucidum* extract on morphineinduced addiction and memory impairment in mice. Biointerface Res Appl Chem. 2022; 12(1):1076-1084.
19. Abdullahi I D, Yaro A H, Nazifi A B. Preliminary Studies on the Anti-Inflammatory and Analgesic Effects of Methanol Leaf Extract of Ficus asperifolia Miq. Trop J. Nat Prod Res. 2020; 4(3):85-90.
20. Thu Thuy L, Anh Tuan P, Hoang Ngan N, Minh Hoang L. The study of the anti-inflammatory effect of herbal remedy "Quyen ty thang gia giam" in animal model (Đánh giá tác dụng chống viêm cấp của bài thuốc quyên tý thang gia giảm trên mô hình gây phù chân chuột bằng carrageenin). Vietnam Med J. (Tạp chí Y học Việt Nam). 2022; 519(2):30-34.
21. Koster R, Anderson M, De Beer EJ. Acetic acid for analgetic screening. Fed Proc. 1959; 18:412.
22. Woolfe G, Macdonald AD. The evaluation of the analgesic action of pethidine hydrochloride (demerol). J. Pharmacol Exp Ther. 1944; 80:300-307.