

THE EFFECTIVENESS OF COMBINATION OF SNAKEHEAD FISH, TEMULAWAK, AND MENIRAN EXTRACTS ON POST OVARIOHYSTERECTOMY WOUND HEALING IN CATS

Fadeli Bermani¹, R Harry Soehartono², Guntur Berlian³, and Riki Siswandi^{2*}

¹Postgraduate Program in Animal Biomedical Sciences, SKHB, IPB University, Bogor, Indonesia

²Division of Surgery and Radiology, SKHB, IPB University, Bogor, Indonesia

³Mega Medica Pharmaceuticals, Jakarta, Indonesia

*Corresponding author: rikisis@apps.ipb.ac.id

ABSTRACT

This study aims to determine the effectiveness of combination of snakehead fish, ginger and meniran extract (GTM) on healing wounds after ovariohysterectomy (OH) in cats. Twenty healthy female cats aged 1-4 years, weighed 2-4 kg were divided into three groups with different doses of GTM and compared with the group without treatment ($n=5$ cats/group). The GTM dose was extrapolated from the human dose using allometric scaling, with the basal dose for G:T:M were 40:3.2:4.8 mg/kg BW (group 1, GTM1x). Treatment groups 2 (GTM2x) and 3 (GTM4x) were given a dose of GTM that was multiplied by 2 and 4 of the basal doses, respectively. OH was performed via a flank approach with the same operator and anesthesia. Discharge, swelling and erythema scores were significantly lower on days 3, 4 and 5 post-operative among all GTM groups compared with control. Pain evaluation using Baseline Tactile Semmes Monofilaments showed significantly lower in GTM2x and GTM4x compared to GTM1x and control. The 4A-vet pain scale showed lower pain in GTM2x from 1 to 5 days postoperative. The number of leukocytes, erythrocytes and platelets was in the normal range indicates the absence of postoperative infection. Postoperative albumin levels showed a significantly higher difference in GTM2x compared to control. The treatment given GTM2x showed the fastest healing interval (3.2 ± 0.44 days). The combination of GTM can reduce pain, reduce wound scores and accelerate post-OH wound healing with the most effective dose was GTM2x.

Key words: cat, meniran, ovariohysterectomy, snakehead fish, temulawak

ABSTRAK

Penelitian ini bertujuan mengetahui efektivitas kombinasi ekstrak ikan gabus, temulawak, dan meniran (GTM) terhadap penyembuhan luka pasca ovariohisterektomi (OH) pada kucing. Dua puluh kucing betina sehat berusia 1-4 tahun, berat 2-4 kg dibagi menjadi tiga kelompok dengan dosis GTM yang berbeda dan dibandingkan dengan kelompok tanpa perlakuan ($n=5$ kucing/kelompok). Dosis GTM diekstrapolasi dari dosis manusia menggunakan allometric scaling, yaitu G:T:M 40:3.2:4.8 mg/kg bobot badan untuk kelompok 1 (GTM1x). Kelompok perlakuan 2 (GTM2x) dan 3 (GTM4x) diberikan dosis GTM yang masing-masing dikalikan dengan 2 dan 4 dari dosis awal. OH dilakukan melalui pendekatan flank dengan operator dan anestesi yang sama. Skor discharge, pembengkakan dan eritema lebih rendah secara signifikan pada hari 3, 4, dan 5 pasca operasi antara semua kelompok GTM dibandingkan dengan kontrol. Evaluasi nyeri dengan Baseline Tactile Semmes Monofilaments menunjukkan skor yang lebih rendah pada GTM2x dan GTM4x dibandingkan GTM1x dan kontrol. Skala nyeri 4Aet menunjukkan nyeri lebih rendah pada GTM2x dari 1 hingga 5 hari pasca operasi. Jumlah leukosit, eritrosit dan platelet pada kisaran normal menunjukkan tidak adanya infeksi pasca operasi. Kadar albumin pasca operasi menunjukkan perbedaan signifikan lebih tinggi pada GTM2x dibandingkan kontrol. Perlakuan yang diberikan pada GTM2x menunjukkan interval penyembuhan tercepat ($3,2 \pm 0,44$ hari). Kombinasi GTM dapat mengurangi nyeri, menurunkan skor luka dan mempercepat penyembuhan luka pasca OH dengan dosis paling efektif pada GTM2x.

Kata kunci: kucing, meniran, ovariohysterectomy, ikan gabus, temulawak

INTRODUCTION

The most common surgical procedure performed in veterinary practice is the removal of ovaries and uterus (ovariohysterectomy/OH) in female cats and dogs. This procedure is carried out because it has benefits, such as preventing reproductive tract diseases, attenuating unwanted behavior related to hormonal activity and reducing the population of stray dogs and cats (Detora and McCarthy 2011). The most frequent side effects after OH are intra-abdominal bleeding during and after surgery, erythema, hemorrhage, swelling, vaginal discharge, urinary incontinence, loose stitches and infection. Surgery can cause bleeding which results in the loss of albumin as a blood component. Albumin has benefits in post-operative healing such as reducing inflammatory reactions, stimulating the formation of new cells and restoring damaged body tissue (Sugiantanti *et al.* 2018).

One source of albumin that can be consumed by humans comes from snakehead fish (*Channa striata*).

Several studies have shown the potential of snakehead fish extract in the wound healing process. For example, research conducted by Suryanti *et al.* (2020) explained that the use of snakehead fish extract can produce a positive effect in speeding up the wound healing process after cesarean surgery. Snakehead fish extract contains high levels of albumin and has an important role in the wound healing process (Yuliana *et al.* 2022). Apart from that, snakehead fish extract is also known to increase fibroblast proliferation which contributes to the wound healing process, as well as reducing swelling in wounds (Siswanto *et al.* 2016).

Meniran (*Phyllanthus niruri*) and ginger (*Curcuma xanthorrhiza*) extracts also have potential in the wound healing process. Meniran extract contains flavonoids, tannins and saponins which can help the wound healing process through several mechanisms, including fibroblast proliferation, increased formation of capillary blood vessels and suppressing inflammatory reactions (Mufidah *et al.* 2023). Temulawak extract contains curcumin, which has anti-inflammatory activity and

plays a role in the inflammation, proliferation and remodeling phases of the wound healing process, so it can reduce wound healing time (Wathoni 2016).

Previous research has shown that snakehead fish extract has the ability to reduce inflammatory reactions in wounds in experimental animals, such as mice (Ramadhanti *et al.* 2021). However, the use of combination of snakehead fish, ginger, and meniran (GTM) extract in the wound healing process in small animals such as cats has never been carried out. This study aims to evaluate the effectiveness of using a combination of GTM extracts on wound healing after ovariectomy in cats.

MATERIALS AND METHODS

This research was carried out from January-March 2023 at Hikari Vet Clinic, Sentul. All procedures in this research were approved by the IPB University Animal Ethics Commission (Number: 11/KEH/SKE/II/2023). This study used 20 female domestic cats with body weight of 2-5 kg and aged between 1-4 years who were divided into 4 groups. Each group consists of 5 cats. Cat was declared healthy with a physical examination and blood examination 2 days before the operation is carried out. Acclimatization before surgery was carried out for a week by administering the antibiotic amoxicillin syrup (dose 10 mg/kg BW, PO) for a week and the antiparasitic ivermectin (dose 0.02 mL/kg BW, SC). Feed was given twice a day and water *ad libitum*.

The extracts of snakehead fish, ginger and meniran in this study used ChannaFit[®] produced by PT MMP in factory facilities that have been certified by BPOM-RI with drug status as herbal medicine (POM TR 203641891). The main raw materials were snakehead fish extract, ginger extract, meniran extract. Each 1 mL of syrup contains 50 mg of snakehead fish extract, 4 mg of ginger extract, 6 mg of meniran extract. Snakehead fish extract produced by PT MMP had albumin content of 21.09% in every 100 g, so in 50 mg of snakehead fish extract contained 10.54 mg albumin (Berlian *et al.* 2023). The production stages follow good traditional medicine manufacturing procedures. The production process includes raw material preparation, weighing, mixing, filling, testing and quality control.

The dose calculation for cats was carried out using the *allometric scaling* (dose conversion) method against the recommended dose for humans so that the dose obtained for cats was snakehead fish extract (G) 40 mg/kg BW, ginger (T) 3.2 mg/kg BW and meniran (M) 4.8 mg/kg BW. This dose was then given to treatment group 1 (GTM1x). In the second treatment group, the dose of GTM given was twice than that of GTM1x, namely snakehead fish extract at a dose of 80 mg/kg BW

(albumin 16.8 mg), ginger 6.4 mg/kg BW and meniran 9.6 mg/kgBW, hereinafter referred to as the GTM2x group. Third group (GTM 4x) used 4 times dose of GTM1x namely snakehead fish extract at a dose of 160 mg/kg BB (albumin 33.6 mg), ginger 12.8 mg/kg BB and meniran 19.2 mg/kg BB. The control group (KK) in this treatment did not using albumin extract from snakehead fish , ginger and meniran in post-operative treatment.

Surgical Procedures and Postoperative Care

OH surgery via flank followed the procedure carried out by Vera *et al.* (2023). Operations were performed by the same surgical operator so that there were no variations between operations. Postoperative treatment was carried out by administering the antibiotic amoxicillin syrup at a dose of 10 mg/kg BW 2 times a day for 5 days to avoid secondary infections and meloxicam injection at a dose of 4 mg/kg BW once a day for 5 days as an analgesic. The GTM combination in liquid form was given orally 2 times a day for 5 days after operations with different doses according to the treatment group.

Wound Healing Parameters

The clinical appearance of the wound was observed every day and a wound score was assessed including four parameters, namely discharge, swelling, erythema, and dehiscence (Sylvestre *et al.* 2002). The wound score assessment was summed up from these four parameters to get the final total wound score (Table 1).

The duration of wound healing was observed by visual observation and recorded every day until the wound healed. The wound was considered healed if the surgical wound was dry, the incision marks have covered and the wound score only showed erythema or swelling (minimum final wound healing score = 1).

Evaluation of Pain/Pain Expression

4A-vet scoring system

The 4A-vet method was method for measure pain scale that evaluate several parameters including general behavior, interactive behavior, heart beat frequency, reaction palpation of the surgical site and intensity response (Grandemange *et al.* 2013). Cats were clinically evaluated daily until the fifth postoperative day. Each parameter was calculated on a scale of 0 to 4 and the final value was obtained by adding up the values of all parameters so that generate value end on the composite pain scale (SN) between 0 until 18. Three levels of pain are defined based on the final value, namely: weak (level 1; SN: 1-5), moderate (level 2; SN: 6-10), and severe (level 3; SN: 11-18).

Assesment of Pain Expression using Baseline Tactile Semmes-Weinstein Monofilaments

Table 1. Criteria or evaluate the wound appearance

Results	Score			
	0	1	2	3
Go out liquid (Discharge)	-	Serosanguinous	Purulent	
Swollen	-	0-2 mm	2-5 mm	>5 mm
Erythema	-	0-2 mm	2-5 mm	>5 mm
Dehiscence	-	0-20%	20-50%	>50%

Expression of pain in surgical wounds was assessed using Baseline Tactile Semmes-Weinstein Monofilaments. Pain assessment with this tool follows the procedure carried out by Vera *et al.* (2023). The pressure value on the device was measured every day at the same time until the 5th post-operative day. Larger values indicated less pain expression.

Hematology and Blood Biochemistry

Cat blood samples were taken via saphenous vein and then analyzed using hematology analyzer (RT-7600 for vet, China) and biochemical analyzer (Pointcare cV3, China). Hematology and blood biochemistry examinations were carried out on the 2nd day before surgery and on the 4th day after surgery. The blood biochemical parameters observed were albumin and glucose.

Data Analysis

Data were presented as means with standard error distribution. The research data were analyzed using the analysis of variance (ANOVA) test and continued with the Tukey test with a 95% confidence interval using the

Prism Graph P ad version program. 9 (GraphPad Software Inc., United States). Area under curved (AUC) value was also calculated to conclude the quantitative information obtained from graph for overall wound score parameters and pain score (end score of 4A-vet and Baseline Tactile Semmes-Weinstein Monofilaments).

RESULTS AND DISCUSSION

Wound Healing Process

Observation of post-operative OH wounds in the control group on days 3, 4, and 5 found the production of serosanguinous discharge (Figure 1). There was no visible discharge production observed in GTM1x, GTM2x, GTM4x (Figure 1a). In addition, it was seen that GTM2x had lower swelling scores compared to the control group on days 3, 4, and 5 after surgery (Figure 1b). E rythema in GTM2x showed a significantly ($P<0.05$) lower score on days 4 to 5 after surgery compared with the control group (Figure 1c). All cats did not show any dislodging of sutures. The overall wound score was significantly ($P<0.05$) lower in the GTM2x

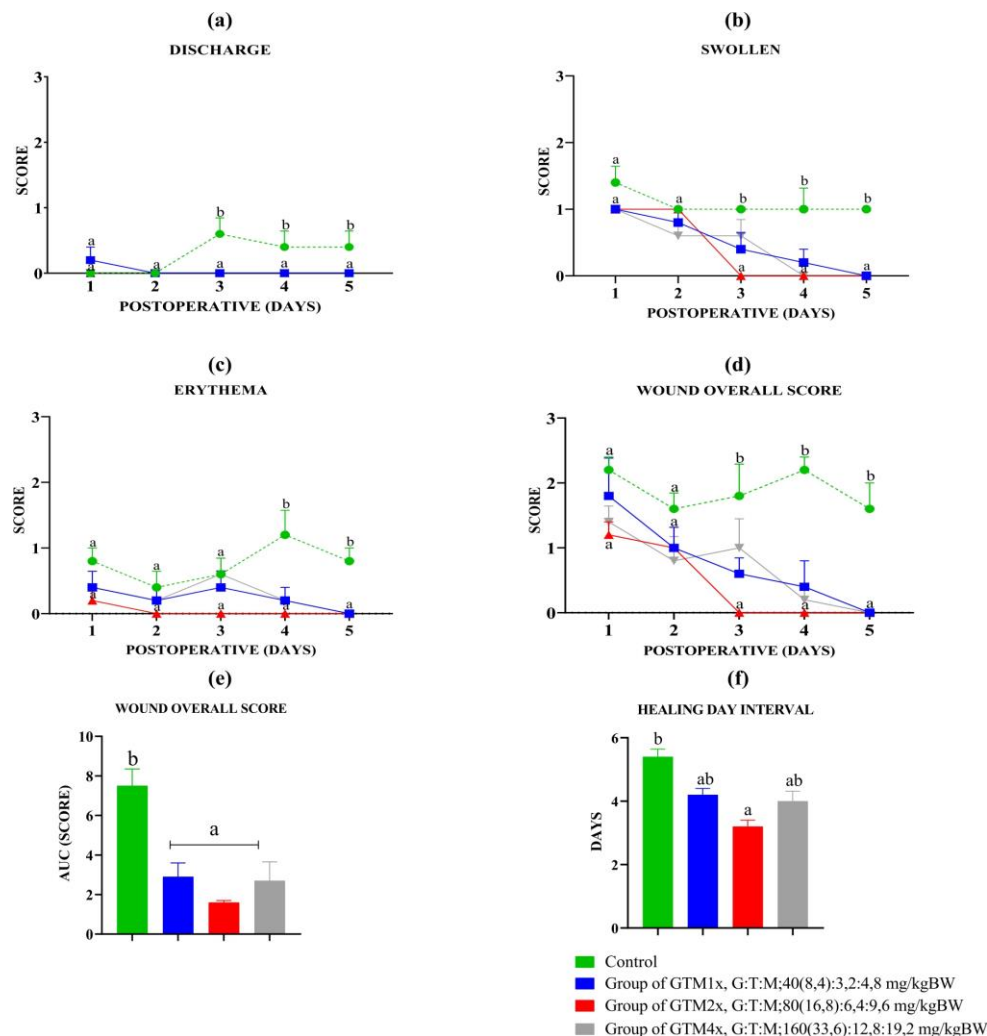


Figure 1. Wound score and healing intervals score. Different superscripts indicated significant difference among group on the same day ($P<0.05$). Data is presented in mean + SEM (Error bar). $n=5$ /group

compared with the control group on days 3, 4, and 5 post-operatively (Figure 1d).

Postoperative discharge OH can occur from days 1 to 14 after surgery (Kiani *et al.* 2014), and significantly more discharge comes out through the flank approach due to the large number of muscle and fat layers that are incised (Coe *et al.* 2006). Discharge on the wound is a protein that contains nutrients, enzyme mediators, leukocytes, electrolytes and growth factors (Harding 2008). The discharge produced in this study had a clear liquid consistency and there was no inflammatory fluid with a smell and a thick consistency, indicating that there was no infection in the wound area and the group given GTM extract showed a little production discharge compared to control and the discharge was no longer visible after the 3rd post-operative day, indicates that giving GTM extract can reduce production discharge in post OH wounds in cat.

Erythema is a natural response of the body as part of wound healing process. When the skin is injured, blood flow increases to the area to carry the necessary substances to repair the damaged tissue. Erythema of the wound could consider as a normal sign of the healing process, however, excessive erythema or in a long time period could be sign of infection (Li *et al.* 2021). In this study, it was observed that the erythema with the combined administration of GTM extract lasted shorter and showed no sign of infection.

Measurement of AUC values in wound healing showed that AUC values in all treatment groups were significantly lower ($P < 0.05$) compared to the control group (Figure 1e). These findings indicated that the post-OH inflammatory reaction in the treatment group was successfully suppressed by administering GTM which

results in faster healing compared to the control group (Figure 2).

The results showed that GTM2x was significantly different in accelerating the wound healing process compared to the control group. The fastest healing interval occurred in GTM2x with an average of 3.2 ± 0.2 days (Figure 1f). Research conducted by Vera *et al.* (2023) found that OH cat wounds healing through flanks approach takes around 5.14 ± 0.26 days. Munif *et al.* (2022) reported that OH wound healing through flanks cat need time up to 10 days post surgery, so that it reinforces that the administration of a combination of GTM extracts can accelerate wound healing after OH cat surgery.

Snakehead fish extract contains various substances such as albumin, N- arachidonylglycine, fatty acids, vitamins A, B, E, and D, and minerals (Kadir *et al.* 2019). These components have the ability to inhibit the activity of $\text{TNF-}\alpha$, $\text{IFN-}\gamma$, IL-10, and reduce IgE production (Susibalan *et al.* 2018). Curcumin contained in ginger has an important role as an anti-inflammatory agent by suppressing the production of cytokines such as $\text{TNF-}\alpha$, IL-6, and IL-8, thus it can reduce symptoms of post-operative inflammation (Hussain *et al.* 2022). Meniran contains flavonoids such as quercetin, rutin, niruriflavone, and astragalin which work by inhibiting p38-MAPK activity, thereby reducing the production of $\text{TNF-}\alpha$ and IL-6 (Hikmah and Triastuti 2022). These three combinations can synergistically reduce inflammatory symptoms such as swelling, discharge and erythema after OH surgery.

Pain Evaluation



Figure 2. Wound healing on the fifth day after surgery. A= GTM1x, B= GTM2x, C= GTM4x, D= Control

Observations of general behavior at 1 to 5 days postoperatively showed variation in scores between groups. The control group showed general behavioral responses such as vocalization, licking the surgical site, and restlessness with higher scores compared to GTM2x followed by GTM4x and GTM1x. (Figure 3a). Observation of the induced interactive behavior, such as responses to calls and petting, showed that GTM2x had lower scores compared to the control group which had the highest score (Figure 3b). This condition showed that the cats in the control group did not show a

significant response to calls and caresses, presumably because of the pain felt by the cats in this group.

The mean heart rate frequency in GTM1x, GTM2x and GTM4x showed lower values at 1 day postoperative compared to the control group, with similar mean scores at 2 to 5 days postoperative in all groups. (Figure 3c). Palpation reactions in the wound area showed that the control group showed a higher response and experienced pain compared to GTM1x, GTM2x and GTM4x (Figure 3d). Response intensity at 1 to 4 days postoperative in GTM2x displayed a calmer response compared to the

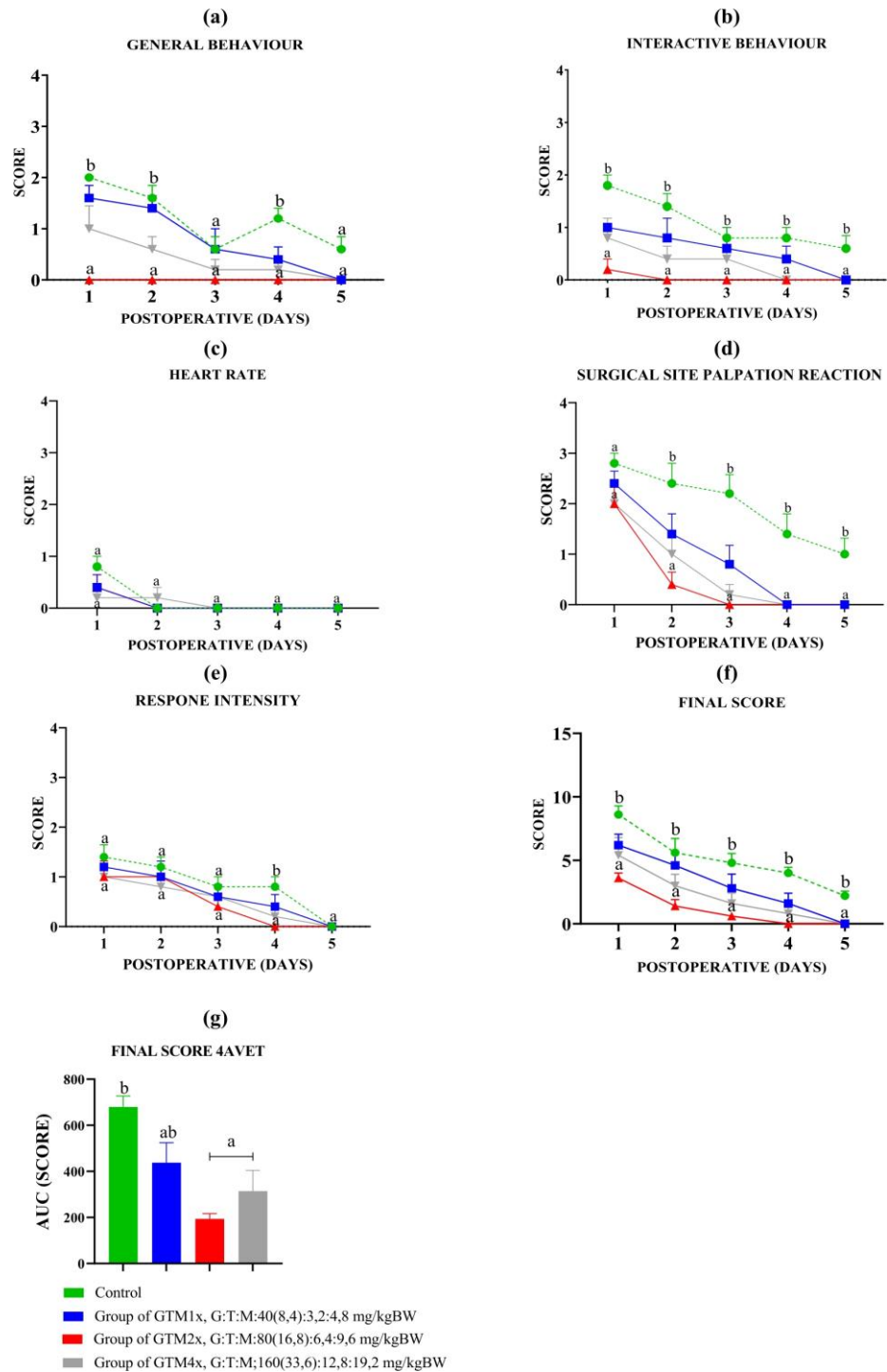


Figure 3. Pain scores evaluation using 4A-vet system. Different superscripts indicated significant difference among group on the same day ($P < 0.05$). Data is presented in mean + SEM (Error bar). $n = 5/\text{group}$

control group who displayed aggressive and defensive behavior when approached (Figure 3e).

Overall, the 4A-vet system score indicated a lower pain score on the GTM2x from 1 to 5 postoperative day (Figure 3f). The total average score within 1 to 5 post-operative days in GTM1x, GTM2x and GTM4x were below 5, so it was categorized as a weak pain level, while the control group had an average value below 10, which indicated a moderate pain level. The AUC value of 4A-vet score showed that GTM2x and GTM4x had significantly lower values compared with the control group, indicating that GTM2x and GTM4x experienced lower pain after OH surgery (Figure 3g).

Pain measurement using the Baseline tactile Semmes-Weinstein monofilament device showed a significant difference between GTM2x and control on days 3, 4, and 5 after surgery. The AUC values showed that GTM2x and GTM4x had higher values and were significantly different from GTM1x and control. This indicated that GTM2x and GTM4x experienced lower pain compared to GTM1x and control. Higher values indicated less expression of pain or tenderness (Figure 4).

Research by Vera *et al.* (2023) reported that the OH flank approach has higher pain because it cuts 3 layers of muscle and the presence of nociceptor nerves in the flank incision area muscles (Grint *et al.* 2006). However, the present study showed that administering GTM can suppress pain as seen in 4A-vet system parameters and Baseline tactile monofilament. Other complications observed is difficulty in removing the uterus intact, ovaries that are dislodged back into the abdomen making them difficult to find, and difficulty in finding ovaries and uterine branches on the opposite side e (Stone 2003). In the study we conducted, no complications were found during flank OH procedures, which indicates the need for experience for surgeons who will perform flank OH procedures.

Pain in post surgery wounds was related to the release of arachidonic acid metabolites through cyclooxygenase activity and prostaglandin biosynthesis (Franzotti *et al.* 2000). Snakehead fish extract has been shown to significantly inhibit the formation of prostaglandins (Saffar *et al.* 2011). In addition, *P. niruri* contains

flavonoids, especially quercetin, which has an analgesic effect (Porto *et al.* 2013). Curcumin contained in ginger also has an analgesic effect because it can inhibit the cyclooxygenase enzyme, thus inhibiting the transformation of arachidonic acid into prostaglandins (Wulandari 2018). Combination GTM has the potential to work synergistically in reducing pain in post OH due to each component extract can hinder formation of prostaglandins and provide analgesic effect. This research provided evidence that administering a combination of GTM extracts can be an alternative for treating higher levels of pain in OH through a flank approach.

Hematology and Blood Chemistry

In this study, the mean values of hematological parameters in all groups showed that total leukocytes (WBC), erythrocytes (RBC), and platelets (PLT) were within the normal range (Table 2). This indicates that no post-operative infections occurred and there were no negative effects from administering the combination GTM extract on hematological profile.

Glucose level values from various treatments are shown in Figure 5. There was a decrease in glucose levels in GTM1x, GTM2x and GTM4x, while the control group experienced showed an increase trend. The largest decrease in glucose levels occurred in GTM4x with a decrease of around 1.07 mmol/L (15%) from baseline, followed by GTM2x with a decrease of 0.61 mmol/L (7.1%), and GTM1x with a decrease of 0.078 mmol/L (1.21%). In the control group, there was an increase of 0.05 mmol/L (0.5%). The results of statistical analysis showed there were no significant differences in blood glucose levels between treatment groups. Administration of snakehead fish extract has a tendency to reduce blood glucose (Mustafa *et al.* 2013). Snakehead fish extract can regenerate 68.78% of the islets of Langerhans in the pancreas and reduce blood glucose levels by up to 34.42% (Aisyatussoffi and Abdulgani 2013). The results of this study indicated that administration of GTM extract has the potential to regulate blood glucose levels without causing hyperglycemia or hypoglycemia in cats after OH surgery.

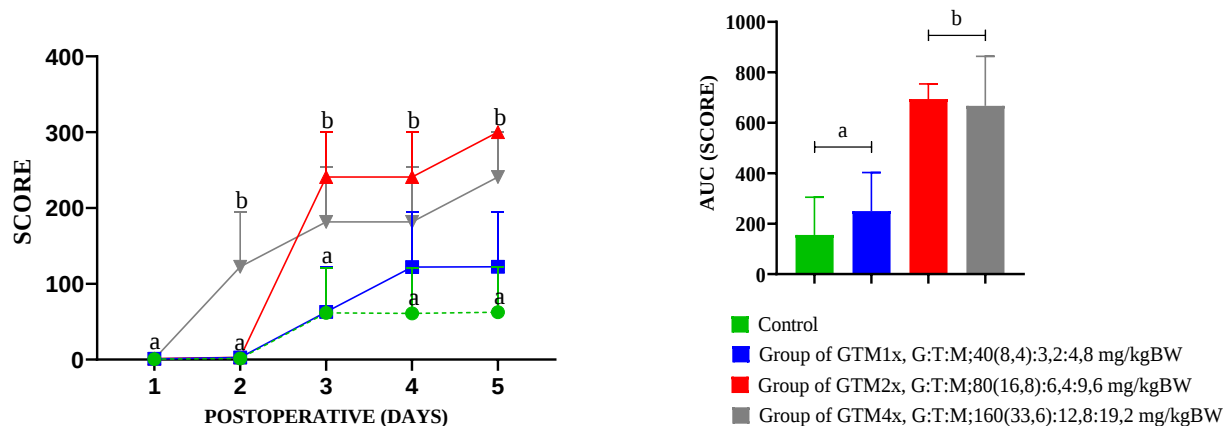


Figure 4. Pain scores evaluation using Baseline tactile semmes-weinstein monofilament tool. Different superscripts indicated significant difference among group on the same day ($P < 0.05$). Data is presented in mean $\pm$ SEM (Error bar). $n = 5/\text{group}$

In all treatment groups, there was an increase in albumin levels (Figure 5). The highest levels was occurred in GTM2x with an increase of 8.14 g/L (31.33%) from baseline, followed by GTM4x of 3.82 g/L (13.71%), and GTM1x of 1.96 g/L (7.75%). In control, there was an increase of 1.2 g/L (4.75%). Statistical analysis showed a significant increase in albumin value between GTM2x compared to control. These findings indicated that oral administration of albumin is considered effective in increasing albumin levels in the blood in accordance with the findings. This condition is in accordance with the findings of Fitriansyah *et al.* (2021), that giving supplements with high albumin content can directly increase albumin levels in the blood.

In this study, the high levels of albumin in GTM2x was assumed to have a positive contribution to wound

healing. Albumin has the ability to activate the Epidermal Growth Factor Receptor (EGFR), which stimulates protein kinase for DNA synthesis and cell proliferation in the wound area. Apart from that, albumin also regulates Transforming Growth Factor- β (TGF- β) which plays a role in stimulating angiogenesis, fibroblast proliferation, reducing inflammation, and collagen synthesis in the wound healing process (Utariani *et al.* 2020). Albumin also has the property to accumulate at sites of inflammation and interact directly with various cells via surface receptors (Sleep 2015). This characteristic means that albumin can act as a drug delivery agent to tissues that need it, or in this study, antibiotics and anti-inflammatory drugs given orally are thought to have higher concentrations in surgical tissue. Apart from delivering drugs to targeted tissues, albumin can also

Table 2. Hematology

Parameter	Treatment								Reference Value
	Baselines				Day 4 Post Operation				
	GTM1x	GTM2x	GTM4x	K.K	GTM1x	GTM2x	GTM4x	K.K	
WBC	18.3±2	14.3±3.3	13.5±0.9	18.6±4.4	14.4 ± 1.6	12.2 ± 1.9	15.0±1.7	16.7±2.2	5.5-19.5
LYM#	6.5±2.8	6.3±4.0	5.3±1.4	5.4 ± 0.8	4.8±2.1	4.0±1.2	6.3±1.6	4.5±1.2	0.8-7
MID#	1.4 ± 0.6	1.5±0.7	1.0±0.3	1.3 ± 0.4	1,350.4	0.8±0.2	1.5 ± 0.43	1.2 ± 0.34	0-1.9
GRA#	9.3±3.8	6.5±5.3	8.1±1.0	13.5±6.8	11.2 ± 5.4	6.8±2.7	11.3 ± 10.5	12.4±6.38	2.1-15
LYM%	33.9±10	41.8±23	37.7±8.7	35.1±15.7	27.6±6.1	33.9±4.8	40.0±20.8	24.9±6.9	12-45
MID%	7.4 ± 2.2	11 ± 2.76	7.2±1.2	9.0±3.27	8.0±1.82	6.3 ± 1.04	10.0±4.15	6.7±1.0	2-9
GRA%	41.2±23	42.5±20	54.5±9.5	55.7±18.9	64.3±7.8	52.3±12	49.9±24.9	67.5±6.2	35-85
RBC	6.3 ± 0.2	6.7±0.4	5.9 ± 0.28	6.1 ± 0.47	7.3 ± 0.36	7.4 ± 0.28	6.78 ± 0.34	7.40 ± 0.29	4.6-10
HGB	10.1±1.3	11.6±1.7	10.7±1.4	9.2±2.33	11.5±1.7	10.9±1.6	11.2 ± 1.3	10.5±1.3	9.3-15
MCHC	32.3±1.0	33.2 ± 0.7	33.0±1.8	31.7 ± 0.9	32.8±1.8	33.0±1.8	32.4 ± 1.1	33.5±1.01	30-38
MCH	15.3±1.4	15.5±0.9	16.8±0.6	14.7±1.3	15.5±0.7	16.4 ± 2.3	16.5±0.5	14.5±1.6	13-21
MCV	45.4±3.1	46.7±3.6	51.1±2.7	46.4±3.3	47.4 ± 0.7	47.7±3.6	51.0±2.2	43.4±4.9	39-52
RDW-CV	14.4 ± 1.3	13.5±0.5	13.4 ± 1.2	13.4 ± 0.5	13.2 ± 0.4	15.3±1.5	38.2 ± 56.3	13.4 ± 0.7	14-18
RDW-SD	36.8±5.0	33.7±1.4	38.5±4.6	33.1±2.8	33.5±0.9	37.9±2.9	32.9±2.6	31.1±2.7	35-56
HCT	30.5±2.2	34.9±5.9	31.5±3.4	28.9±6.6	34.8±3.8	31.7±3.7	34.5±3.1	31.3±3.8	28-49
PLT	219±35.9	170.4±34	240.6±34	120.8±20	141±30	295.8±44	244.8±29	115.8±17	100-514
MPV	8.2 ± 0.53	7.9±0.6	8.5±1.1	8.0±1.1	7.8±0.6	8.2±1.2	8.0±0.6	8.0±0.7	5-11.8
PDW	11.5±2.8	12.3±3.3	11.7±4.4	13±5.23	10.0±3.9	13.6±1.7	13.1±2.7	10.9±4.4	10-18
PCT	0.2±0.09	0.2±0.1	0.2±0.1	0.1±0.1	0.1±0.1	0.3±0.06	0.16±0.07	0.13±0.11	0.1-0.5
P-LCR	20.5±7.3	15.4 ± 2.1	22.3±9.1	16.8±6.4	17.3±4.2	15.2±4.3	19.4±9.7	16.6±3.3	13-43

Data is presented in mean $\pm$ SEM (Error bar). N=5/ group

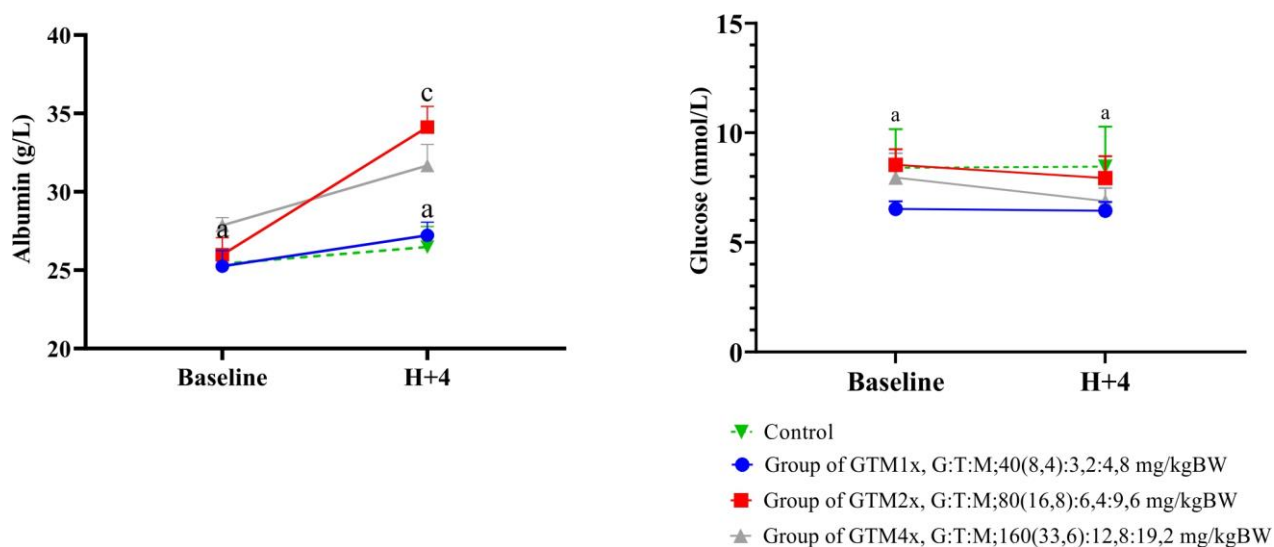


Figure 5. Levels of albumin and glucose in cats treated with GTM. Different superscripts indicated significant difference among group on the same day ($P<0.05$). Data is presented in mean $\pm$ SEM (Error bar). n= 5/group

help remove dangerous biological materials as a byproduct of tissue damage (Bihari *et al.* 2020). This character was seen in wound healing parameters, especially the erythema and discharge scores which were much lower in GTM2x. Although in this study albumin was given in the form of snakehead fish extract, it is considered to provide direct benefits of albumin in wound healing. The combination of snakehead fish with ginger and meniran appears to have effective synergistic potential in wound healing.

CONCLUSION

The use of snakehead fish albumin extract, ginger and meniran can reduce pain, reducing wound scores and speeding up post-healing operation (ovariohysterectomy) in cats with the most effective effect was a dose of snakehead fish extract of 80 mg/kg BW (albumin 16.8 mg), ginger 6.4 mg/kg BW and meniran 9.6 mg/kg BW.

ACKNOWLEDGMENT

Thanks are expressed to Hikari Vet Clinic for the facilities provided for this research.

REFERENCES

- Aisyatussoffii N, Abdulgani N. 2013. Pengaruh pemberian ekstrak ikan gabus pada struktur histologi pankreas dan kadar glukosa darah mencit hiperglikemik. *Jurnal Sains dan Seni Pomits*, 2(1):2337-3520.
- Berlian G, Riani C, Kumiat NF, Rachmawati H. 2023. Peptide derived *C. striata* albumin as a natural angiotensin-converting enzyme inhibitor. *Heliyon*, 9(5):1-10.
- Bihari S, Bannard-Smith J, Bellomo R. 2020. Albumin as a drug: its biological effects beyond volume expansion. *Critical Care and Resuscitation*, 22(3):257-265.
- Coe RJ, Grint NJ, Tivers MS, Hotston Moore A, Holt PE. 2006. Comparison of flank and midline approaches to the ovariohysterectomy of cats. *Veterinary Record*, 159(3):309-313.
- Detora M, McCarthy RJ. 2011. Ovariohysterectomy versus ovariectomy for elective. *Journal of the American Veterinary Medical Association*, 239(11):1409-1412.
- Fitriansyah I, Djanas D, Bachtar H. 2021. The effect of oral albumin intake on serum albumin levels in preeclampsia patients with hypoalbuminemia in RSUP Dr. M. Djamil Padang. *Andalas Obstetrics and Gynecology Journal*, 5(1):69-81.
- Franzotti EM, Santos CVF, Rodrigues HMSL, Mourao RHV, Andrade MR, Antonioli AR. 2000. Anti inflammatory, analgesic activity and acute toxicity of *Sida cardifolia* L. (Malva-branca). *Journal of Ethnopharmacology*, 72:237-278.
- Grandemange E, Fournel S, Woehrlé F. 2013. Efficacy and safety of cimicoxib in the control of perioperative pain in dogs. *Journal Small Animal Practice*, 54(6):304-312.
- Grint NJ, Murison PJ, Coe RJ, Pearson AEW. 2006. Assessment of the influence of surgical technique on postoperative pain and wound tenderness in cats following ovariohysterectomy. *Journal of Feline Medicine and Surgery*, 8(1):15-21.
- Harding KG. 2008. Wound exudate and the role of dressings. *International Wound Journal*, 5(1):1-12.
- Hikmah U, Triastuti A. 2022. Mechanism and immunomodulator bioactive compounds of *Phyllanthus niruri* (meniran). *Scientific Journal of Pharmacy*, 18 (2):205-218.
- Hussain Y, Abdullah, Khan F, Alsharif KF, Alzahrani KJ, Saso L, Khan H. 2022. Regulatory effects of curcumin on platelets: An update and future. *Biomedicines*, 10(12):1-13.
- Kadir AA, Kadir AA, Hamid RA, Jais AMM, Omar J, Sadagatullah AN, Badrin S, Win TT, Sirajudeen KNS, Salleh A. 2019. Evaluation of chondroprotective activity of *Channa striatus* in rabbit osteoarthritis model. *BioMed Research International*, doi: 10.1155/2019/6979585.
- Kiani FA, Kachiwal AB, Shah MG, Nizamani ZA, Khand FM, Lochi GM, Haseeb A, Khokbar AM, Oad A, Ansari MI. 2014. Comparative study on midline and flank approaches for ovariohysterectomy in cats. *Journal of Agriculture and Food Technology*, 4(2):21-31.
- Li S, Renick P, Senkowsky J, Nair A, Tang L. 2021. Diagnostics for wound infections. *Advances in Wound Care*, 10(6):317-327.
- Mufidah N, Sunarsih EN, Dini IRE. 2023. Uji aktivitas penyembuhan luka ekstrak etanol meniran (*P. niruri* L.) pada kelinci (*Oryctolagus cuniculus*). *Journal of Research in Pharmacy*, 3(1):65-73.
- Munif MR, Safawat SM, Hannan A. 2022. Left lateral flank approach for spaying in cats. *Open Veterinary Journal*, 12(4):540-550.
- Mustafa A, Riyadi BD, Kristianto Y, Pudjirahaju A. 2013. Pengaruh pemberian sari ikan gabus (*Channa striata*) terhadap perbaikan kondisi diabetes pada tikus wistar (*Rattus norvegicus*) diabetes mellitus. *The Indonesian Journal of Public Health*, 10(1):56-69.
- Porto CRC, Soares LAL, Souza TP, Petrovick PR, Lyra IL, Junior RFA, Langassner SMZ, Ferreira AAA, Guerra GCB. 2013. Anti-inflammatory and antinociceptive activities of *P. niruri* spray-dried extract. *Brazilian Journal of Pharmacognosy*, 23(1):138-144.
- Ramadhanti NA, Sandhika W, Widodo ADW. 2021. The effect of snakehead fish (*Channa striata*) extract on inflammation reaction of skin wound tissue in *Rattus norvegicus* Wistar strain. *Berkala Ilmu Kesehatan Kulit dan Kelamin*, 33(1):48-54.
- Saffar FJ, Ganabadi S, Fakuraz S. 2011. Response of *Channa striatus* extract against monosodium iodoacetate induced osteoarthritis in rats. *Journal of Animal and Veterinary Advances*, 10(4):460-469.
- Siswanto A, Dewi N, Hayatie L. 2016. Effect of haruan (*Channa striata*) extract on fibroblast cells count in wound healing. *Journal of Dentomaxillofacial Science*, 1(2):82-87.
- Sleep D. 2015. Albumin and its application in drug delivery. *Expert Opinion on Drug Delivery*, 12(5):793-812.
- Stone EA. 2003. Ovary and Uterus. In *Small Animal Surgery*. Slatter D. (Ed.). 3rd ed. WB Saunders, Philadelphia.
- Sugiantanti MF, Oesman D, Elfiah U. 2018. Pengaruh kadar albumin serum terhadap penyembuhan luka pada pasien pascaoperasi laparotomi dan lumbotomi di RSD dr. Soebandi Jember. *e-Jurnal Pustaka Kesehatan*, 6(3):383-386.
- Suryanti Y, Hadisaputro S, Nugraheni SA. 2020. The effect of snakehead fish (*Channa striata*) extract on blood leukocyte number and cesarean section wound healing. *Advances in Social Science, Education and Humanities Research*, 443:596-598.
- Susibalan BD, Abdullah B, Lazim NM, Kadir AA. 2018. Efficacy of snakehead fish (*Channa striatus*) in subjects with allergic rhinitis: A randomized controlled trial. *Oriental Pharmacy and Experimental Medicine*, 18(1):209-215.
- Sylvestre A, Wilson J, Hare J. 2002. A comparison of 2 different suture patterns for skin closure of canine ovariohysterectomy. *The Canadian Veterinary Journal*, 43(9):699-702.
- Utariani A, Rahardjo E, Perdanakusuma DS. 2020. Effects of albumin infusion on serum levels of albumin, proinflammatory cytokines (TNF- α , IL-1, and IL-6), CRP, and MMP-8; wound healing in *Sprague dawley* rats. *International Journal of Inflammation*, 20:1-13.
- Vera B, Siswandi R, Soehartono RH. 2023. Ventral median and lateral flank approach for ovariohysterectomy in cats. *Jurnal Kedokteran Hewan*, 17(1):22-29.
- Wathoni N. 2016. Alasan kurkumin efektif mempercepat penyembuhan luka di kulit. *Farmasetika*, 1(3):1-3.
- Wulandari T. 2018. Uji efek analgetik temulawak instan (*Curcuma xanthorrhiza* Roxb) pada mencit jantan dengan metode geliat. *Jurnal Kimia Dasar*, 7(1):1-7.
- Yuliana B, Sartini, Djide N, Djabir YY. 2022. Wound healing effect of snakehead fish (*Channa striata*) mucus containing transdermal patch. *Journal of Applied Pharmaceutical Science*, 12(7):171-183.