



***PATHOLOGICAL CHANGES IN GILLS, LIVER AND KIDNEY OF GOLDFISH
(*Carassius auratus*) WHEN EXPOSED TO CLOVE OIL USING THE FISH
ANAESTHESIA DELIVERY SYSTEM***

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FPV 2018 30

**PATHOLOGICAL CHANGES IN GILLS, LIVER AND KIDNEY OF
GOLDFISH (*Carassius auratus*) WHEN EXPOSED TO CLOVE OIL USING
THE FISH ANAESTHESIA DELIVERY SYSTEM**

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A project paper submitted to the

Faculty of Veterinary Medicine, Universiti Putra Malaysia

In partial fulfillment of the requirement for the

DEGREE OF DOCTOR OF VETERINARY MEDICINE

Universiti Putra Malaysia

Serdang, Selangor DarulEhsan

MARCH 2018

CERTIFICATION

It is hereby certified that we have read this project entitled “Pathological Changes in Gills, Liver and Kidney of Goldfish(*Carassius auratus*)When Exposed to Clove Oil using the Fish AnaesthesiaDelivery System”, by Hanisah Nordin and in our opinion it is satisfactory in terms of scope, quality, and presentation as partial fulfillment of the requirement for the course VPD 4999-Project.

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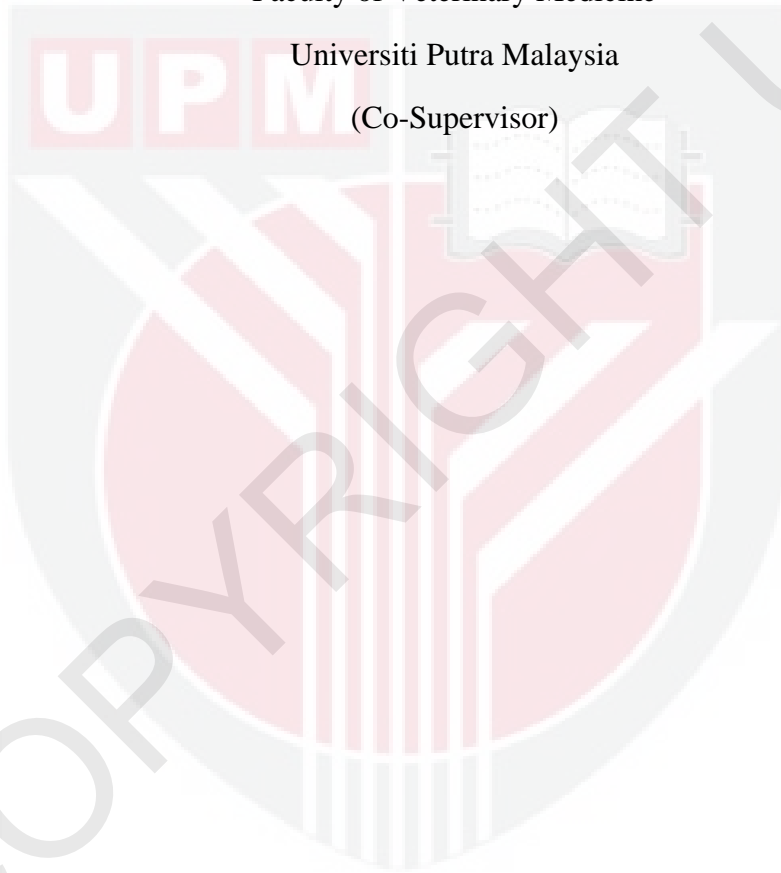
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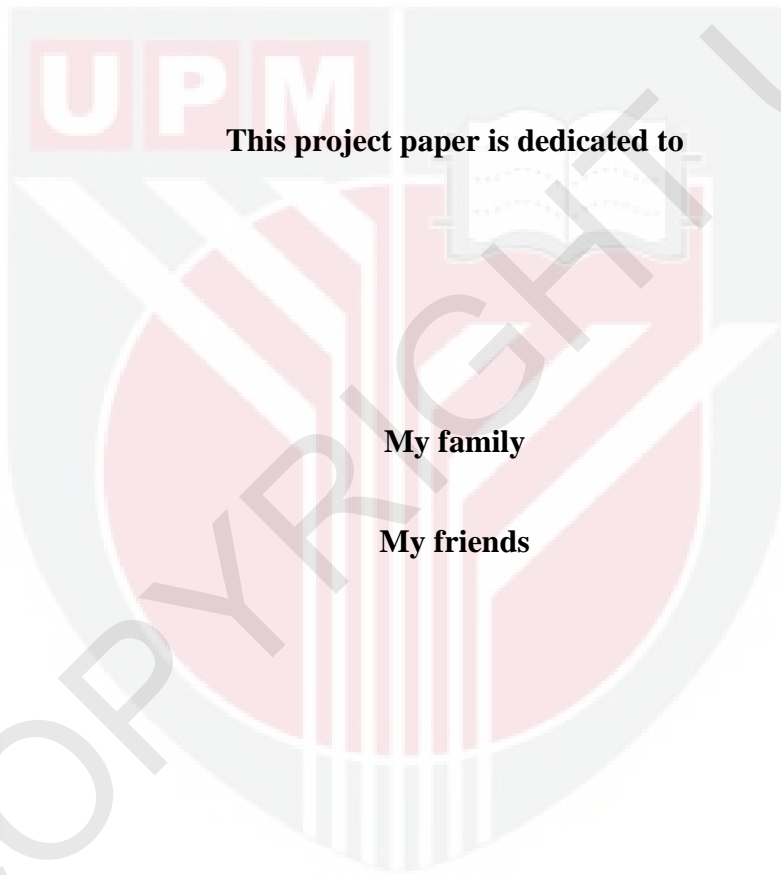


DEDICATIONS

This project paper is dedicated to

My family

My friends



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ACKNOWLEDGEMENTS

First and foremost, my sincere gratitude and appreciation for my project supervisor, Dr. Mazlina Mazlan for her patience, guidance, knowledge and support throughout this entire project.

In addition, I would also like to thank my co-supervisors, Dr. Mohd Fuad Matori and Prof. Dr. Mohd Hair Bejo for sharing their experiences and thoughts for the improvement of this project.

Also, not forgetting the staff and post-graduate students of the Veterinary Histopathology Laboratory and Animal Health Unit of the Faculty of Veterinary Medicine, including Madam Latifah, Madam Jamilah, Mr. Azmi and Mr. Zainal, whom have guided and assisted me with this project.

My heartfelt gratitude towards Nabila Farahin Ishak, my FYPmate without whom I would have gone through a major breakdown and also to my wonderful friends Adeline, Syahirah, and Crystal for all their support and for their presence in my life.

I would very much like to extend my deepest gratitude and appreciation towards Aquatic Animal Health Unit, Faculty of Veterinary Medicine, Universiti Putra Malaysia and the Dr's that have kick started this project and those who have helped me in making this project a success.

Lastly, I thank my family, for all their love and support throughout my lifetime.

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LIST OF ABBREVIATIONS

G1	Group 1
G2	Group 2
G3	Group 3
G4	Group 4
MS-222	Tricaine methanesulfonate
%	Percent
=	Equal
<	Less than
>	More than
-	Negative
+	Plus
++	Two plus
+++	Three plus
AUP	Animal Utilisation Protocol
cm	Centimetre
FDA	Food and Drug Administration
GRAS	Generally Recognized As Safe
H&E	Haematoxylin & Eosin
IACUC	Institutional Animal Care and Use Committee
L	Liter
mg	Milligram
mg/L	Milligram/Liter
mL	Milliliter
Ppt	Parts-per-trillion

SD	Standard deviation
SPSS	Statistical Package for the Social Sciences
TBL	Total body length



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ABSTRAK

Abstrakdaripadakertasprojek yang dikemukakankepadaFakultiPerubatan Veterinaruntukmemenuhisebahagiandaripadakeperluankursus VPD 4999-Projek

PERUBAHAN PATOLOGIK PADA INSANG, HATI DAN GINJAL IKAN EMAS(*Carassius auratus*) BERIKUTAN PENDEDAHAN KEPADAMINYAK CENGGIHING PADA DURASI BERBEZA DENGAN MENGGUNAKAN SISTEM PENGHANTARAN ANESTESIA IKAN

Oleh

Hanisah Nordin

2018

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Prof. Dr. Mohd. Hair Bejo

Perubahan patologi pada insang, hati dan ginjal ikan Emas (*Carassius auratus*)berikutan pendedahan kepada minyak cengkih dengan menggunakan sistem penghantaran anestesia ikan telah dikaji. Dalam kajian ini, tempoh pengendalian semasa pembedahan dalam ikan Emas adalah 5 minit (G1), 15 minit (G2) dan 30

minit (G3) menggunakan minyak cengkih sebagai anestesia dengan kepekatan sebanyak 50 mg/L manakala ikan Emas dalam kumpulan 4 (G4) bertindak sebagai kumpulan kawalan. Sampel insang, ginjal dan hati telah diambil daripada semua kumpulan untuk penilaian secara makroskopik dan mikroskopik. Hasilnya menunjukkan bahawa walaupun tidak ada perubahan kasar yang diperhatikan, terdapat perubahan secara histopatologi pada insang iaitu pembengkakan sel epitelium, pengangkatan sel epitelium, edema dan pendilatan saluran darah pada ikan dalam G1, G2 dan G3 dengan skor lesi tertinggi diperhatikan dalam G3. Di samping itu, keputusan menunjukkan bahawa minyak cengkih tidak mempunyai kesan berbahaya terhadap buah pinggang dan hati secara makroskopik mahupun mikroskopik. Kesimpulannya, kajian ini mengesahkan bahawa tempoh pendedahan kepada minyak cengkih selama 5 minit dan 15 minit adalah lebih selamat dalam ikan Emas kerana kurang kesan lesi yang ditunjukkan oleh ikan khususnya pada insang dan sistem penghantaran anestesia ikan menggunakan minyak cengkih menyokong anestesia optimum untuk menjalankan prosedur pembedahan dalam ikan Emas. Oleh itu, kajian ini mencadangkan bahawa tempoh pendedahan kepada minyak cengkih selama 5 minit dan 15 minit adalah lebih selamat kerana tidak memberi kesan patologikal pada insang. Sebagai kesimpulan, hasil kajian ini menunjukkan minyak cengkih boleh digunakan sebagai anestesia untuk pembedahan yang berkaitan dengan ikan Emas dengan menggunakan sistem penghantaran anestesia ikan.

Kata kunci: minyak cengkih, ikan Emas (*Carassius auratus*), anestesia, perubahan patologi, insang, hati, buah pinggang, sistem penghantaran anestesia ikan

ABSTRACT

Abstract of the project paper presented to the Faculty of Veterinary Medicine in
partial requirement for the course VPD 4999-Project

PATHOLOGICAL CHANGES IN GILLS, LIVER AND KIDNEY OF GOLDFISH (*Carassius auratus*) WHEN EXPOSED TO CLOVE OIL USING THE FISH ANAESTHESIA DELIVERY SYSTEM

By

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2018

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The pathological changes in gills, liver and kidney of Goldfish (*Carassius auratus*) following exposure to clove oil using the fish anaesthesia delivery system were studied. In this study, the duration of maintenance stage during surgery in Goldfish were 5 minutes (G1), 15 minutes (G2) and 30 minutes (G3) using 50 mg/L clove oil while the Goldfish in group 4 (G4) served as the control. The gills, kidney and liver were sampled from all groups for macroscopic and microscopic evaluation. The

results revealed that although there was no gross changes observed, histopathological alterations of gills characterised by epithelial swelling, epithelial lifting, oedema and dilatation of blood vessels were present in fish in G1, G2 and G3 with the highest lesion scoring was observed in G3. In addition, results showed that clove oil has no harmful effect towards kidney and liver both macroscopically and microscopically. In conclusion, this study verified that the duration of exposure to clove oil for 5 minutes and 15 minutes are relatively safer in Goldfish as less injurious effects were observed in gills and the fish anaesthesia delivery system using clove oil can provide optimum anaesthesia to conduct surgical procedures in Goldfish. Thus, this study suggests that the duration of exposure to clove oil for 5 minutes and 15 minutes is relatively safer as there were less pathological effects on the gills. In conclusion, the result from this study revealed that clove oil can be used as anaesthesia for surgery related to Goldfish by using the fish anaesthesia delivery system.

Keywords: Clove oil, Goldfish (*Carassius auratus*), anaesthesia, pathological changes, gills, liver, kidney, fish anaesthesia delivery system

1.0 INTRODUCTION

General anaesthesia techniques are widely used in aquatic animals for many purposes including for a variety of major and minor surgery, in fisheries research and aquaculture. Clove oil is one of the anaesthetic agent for general anaesthesia in fish.

Clove oil is a dark-brown liquid, a distillate of flowers, stalks and leaves of the clove tree *Eugenia aromatica* (Soto & Burhanuddin, 1995). The major constituent (70 to 90 percent by weight) is the oil eugenol, but clove oil contains a wide range of other compounds that impart its characteristic odour and flavour (Coyle *et al.*, 2004). Similarly, according to Isaacs (1983), clove oil is distilled from stems, leaves and flower buds of *Eugenia caryophyllata* and its active ingredient, eugenol (4-allyl-2-methoxyphenol) makes up 70 to 90% by weight of clove oil. Clove oil also contains eugenol acetate and kariofilen.

The main advantages of clove oil lies in its low cost and its relative safety to both fish and humans (Keene *et al.*, 1998). It is necessary to provide both anaesthesia and analgesia in fish as the interest in ornamental fish medicine is increasing. Goldfish (*Carassius auratus*) is a common and popular pet nowadays. As Goldfish has a long life span, the pet owner will develop sentimental feeling towards their fish. They will be keen enough to proceed with any procedures if the fish develop diseases. Therefore, surgical procedures in Goldfish is not rare anymore. This study can be one of the references to establish the use of clove oil as anaesthesia via the fish anaesthesia delivery system for Goldfish surgery. Even though, a lot of literature exist about the anaesthetic usage on food fish species, not much information seems to be available on ornamental fish aquaculture (Bystriansky *et al.*, 2006).

Other than that, anaesthesia is important for surgery out of water for extended periods of time. Brown (1987) reported that currently, two methods are available for maintaining fish under general anaesthesia. Lewbart & Harm (1999) also stated that for short procedures, anaesthesia is generally achieved by immersing the fish in MS-222 bath. Immersion anaesthesia with alfaxalone can be used to produce an adequate plane of surgical anaesthesia in a Goldfish (Fernandez-Parra *et al.*, 2017).

For longer procedures, continuous anaesthesia delivery can be maintained by pumping anaesthetic-containing water over the gills. Complex recirculation systems which pass an anaesthetic solution over the gills have been constructed and reported by Ross & Ross (1983). An attempt to study the recirculation systems was done by Brown (1987) by using MS-222 on Catfish (*Ictalurus punctatus*). Clove oil recirculation systems has never been attempted and thus, in this study, this method will be attempted. There has been one attempt to study the effects of tricaine as an anaesthetic on Goldfish (*Carassius auratus*) at the different salinities and concentrations, performed by Küçük *et al.* (2016). From their result, MS-222 + salt combination is carried out quickly for Goldfish anaesthesia. It is recommended that 200 mg/L of MS-222 at 12 ppt can be used in Goldfish aquaculture practice. Nevertheless, there is still lack of study in *Carassius auratus* when exposed to clove oil. Thus, this study was conducted to investigate the effect of clove oil at different duration of maintenance stage during surgery in *Carassius auratus*.

This study was undertaken to fulfil the following objectives:

- 1) To investigate the effect of anaesthetic clove oil in *Carassius auratus* when exposed at different duration by using fish anaesthesia delivering system.

- 2) To investigate the gross morphological lesion and histological changes on gills, livers and kidneys on *Carassius auratus* upon the exposure of clove oil at different duration by using fish anaesthesia delivery system.



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