# Effect of Clove Oil as an Anaesthetic on The Fry of Fishes, Silver Dollar (Metynnis schreitmuelleri Ahl), Sword Tail (Xiphophorus helleri Heckel) and Pearl Gourami (Trichogaster leeri Bleeker)

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### **Abstract**

Efficacy of Clove oil as an anesthetic was examined on the fry of ornamental fishes Silver dollar (Metynnis schreitmuelleri Ahl), Swordtail (Xiphophorus helleri Heckel) and Pearl gourami (Trichogaster leeri Bleeker) in order to slow down the movement for short duratio while handling for scientific studies. The two sized groups (A and B) of each fish with eight concentrations (10, 20,30,40,50,60,70 an  $80 \text{mg } \Gamma^1$ ) of clove oil were tested. The fry of Silver dollar in the group 'A' having average size of  $3.50 \pm 0.01$  cm indicated  $38.66 \pm 0.33$  an  $268.33 \pm 4.40$  seconds as induction and recovery times, while in the group 'B', fry with average size of  $6.00 \pm 0.02$  cm, the induction an recovery times were  $50.00 \pm 0.57$  and  $371.00 \pm 6.00$  seconds respectively when treated with 80 mgl<sup>-1</sup> concentration of clove oil. This induction and recovery times in respect of both the groups of Sword tail fry were  $285.33 \pm 0.33$ ,  $379.00 \pm 0.57$ ,  $187.00 \pm 0.57$  and  $98.33 \pm 0.3$  seconds respectively when exposed to  $80 \text{ mgl}^{-1}$  concentration. A positive dependence was observed between fish length, induction an recovery time. However, there was no effect of any clove oil concentration as an anesthetic on the fry of both size groups of Pearl gourami. The concentration of  $80 \text{ mgl}^{-1}$  of the clove oil observed to have quick induction of sedation on Silver dollar and Swordtail in shortest the scheme of the second of the

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### Introduction

Clove oil has been used for a number of years to anaesthetize fishes in seawater (Patrick and Adeline, 2000) and best-known local anesthetic in dental sugary. Clove oil is distilled from *Eugenia caryophyllata* stems, buds and leaves. The active ingredients are phenol derivatives essentially the C<sub>10</sub>H<sub>12</sub>O<sub>2</sub> euganol compound (Taylor and Roberts, 1999). The effects of clove oil as an anesthetic were studied by researchers on marine and fresh water fishes (Keene *et al.*, 1998; Patrick and Adeline, 2000 and Woody *et al.*, 2002).

## Materials and Methods

The commercially available clove oil mixed with 3-4 volumes of ethanol to make it water-soluble. Eight concentrations (10, 20, 30, 40, 50,60,70 and 80 mg [-1] of this mixture were tested on two different sized (Group-A and B) fry of Silver dollar, Metynnis schreitmuelleri Ahl  $(3.50 \pm 0.01 \text{cm}, 6.00 \pm 0.02 \text{ cm});$ Sword tail, Xiphophorus helleri Heckel {2.82 ±1.48 cm, 6.15 ± 1.13cm) and Pearl gourami, Trichogaster leeri Bleeker  $(3.90 \pm 0.45 \text{ cm}, 6.50 \pm 0.25)$ cm). The range of concentrations was selected on the basis of the results reported by Woody et al. procedure guidelines this (2002). During recommended by Hicks (1989) were followed. The experiments were conducted in triplicate in glass aquaria 15"x12"x12" containing 3 l water. Induction

time and recovery time were recorded. The water temperature was  $28.5\pm1.0$  °C throughout the experiment.

# Results and Discussion

The results of induction time and survival percentage of both groups of Silver dollar and swordtail fishes exposed to eight clove oil concentrations are presented in Table 1. There was significant difference (P< 0.05) among the treatments for induction and recovery times in both groups. The Silver dollar fry of group-A treated with 80 mgl<sup>-1</sup> showed quick induction i.e  $38.66 \pm 0.33$ seconds with recovery time of  $268.33 \pm 0.40$  seconds (Table 1). There was no effect of 10 mgl-1 concentrations on the induction of both sized Silver dollar fishes. The induction time in respect of group-A Silver dollar fishes was not significantly different (P>0.05) between 70 and 80 mgl 1. However recovery time differed significantly (P<0.05) for both treatments. The similar trend was observed with group-B Silver dollar fry when subjected to these concentrations. The recovery times for both groups of fishes observed to have increased with increase in concentrations of the clove oil (Table1)

There was no effect of 10 and 20 mg  $\Gamma^1$  concentrations on the induction of both sized Swordtail fishes. However, these fishes remained stationary in the media indicating stage I- light

sedation as described by Durve and Raja (1965). The induction time was higher in 30 mgl<sup>-1</sup> concentration of clove oil in group- A Sword tail fishes and it was reduced as concentration increased from 30 mgl<sup>-1</sup> to

have been discussed by Pickford and Atz (1957). The use of anesthetics for fisheries experiments like transport of live fishes or handling fishes for tagging, fin clipping, weighing and stripping have been well

Table 1. Induction time and survival rate of two groups of Silver dollar fry (Metynnis schreitmuelleri Ahl) and Swordtail fry (Xiphophorus helleri Heckel) treated with varying concentrations of clove oil.

Size group	Concentration (mg l <sup>-1</sup> .)	Exposure Time (Sec.)	Induction Time (Sec.)	Recovery time (sec)
Group A*	10	600	0	0
	20	600	538.66±1.43	90
	30	600	357.53±0.53	165
	40	600	98.66±0.24	220
	50	600	59.46±0.35	225
	60	600	43.33±0.88	227
	70	600	39.66±0.33	230
	80	600	38.66±0.33	260
Group B**	10	600	0	0
	20	600	602.13±1.619	90
	30	600	401.86±2.4369	175
	40	600	176.53±0.3716	300
	50	600	63.33±0.133	300
	60	600	55.33±0.33	300
	70	600	52.00±1.52	310
	80	600	50.00±0.57	360
Group A*	10	600	0	0
	20	600	0	0
	30	600	413.66±0.33	150
	40	600	359.66±0.33	155
	50	600	339.00±0.57	158
	60	600	309.00±0.57	160
	70	600	289.00±0.33	162
	80	600	285.33±0.33	162
Group B**	10	600	0	0
	20	600	0	0
	30	600	599.00±0.57	95
	40	600	479.66±0.57	95
	50	600	419.66±0.33	95
	60	600	399.33±0.66	95
	70	600	385.33±0.333	97
	80	600	379.00±0.57	97

\*P>0.5 (CD = 2.14); \*\* P>0.05 (CD = 3.59), Survival (%) is 100 %

80 mgl<sup>-1</sup>. The recovery times of the group-A fishes treated with 50 and 60 mgl<sup>-1</sup> clove oil concentration was almost same (179.66  $\pm$  0.33 and 180.00  $\pm$  0.57 seconds). The survival rate was 100 % among all the treatments. In group-B Swordtail fishes induction time was 379.00  $\pm$  0.57 seconds when exposed to 80 mgl<sup>-1</sup>clove oil concentrations while recovery time was 98.33±0.33 seconds (Table 1) and differed by 88.67 seconds from group-A fishes. There was no effect of clove oil on Pearl gourami probably because of their habit of gulping the atmospheric air.

Several methods have been used to induce anesthesia among the various species of fishes. (Abramonitz, 1937; Parker, 1937, 1939; Abramonitz et al., 1940) like immersing fishes in crushed ice or ice water (Osborn, 1938, 1941) and electric shock to reduce the movement of fishes lasting about two minutes (Haskell, 1940). The disadvantages of such methods

recognized (Durve and Raja, 1965; Thienpont and Niemegeers, 1965; Saha et al., 1965; Natarajan and Renganathan, 1960; Sreenivasan, 1962; Keene et al., 1998; Munday and Wilson, 1997; Taylor and Roberts, 1999). Woody et al. (2002) recommended 50 mgl<sup>-1</sup> of clove oil for anaesthetizing Salmon ranging in length from 400 to 550 mm with induction time of about 3 minutes and recovery time of 10 minutes. In the present study, the induction and recovery times with 50 mgl<sup>-1</sup> of concentration was observed to be  $59.46 \pm 0.35$ ,  $63.33 \pm 0.13$ ,  $239.40 \pm$ 0.23 and 306.26  $\pm$  1 for Silver dollar and 339.00  $\pm$ 0.57,  $419.66 \pm 0.33$ ,  $179.66 \pm 0.33$  and  $94.66 \pm 0.66$ seconds for Sword tail fishes respectively. Munday and Wilson (1997) evaluated the efficacy of clove oil with other anesthetics like quinaldine, benzocaine, MS 222, 2- Phenoxyethanol and noted that clove oil was effective at much lower concentration than the other chemicals. They also stated that clove oil was

superior except the quinaldine for anaesthetizing Pomacentrus amboinensis, a coral reef fish as the recovery time with clove oil was 2-3 times longer than recovery from other chemicals. Woody et al. (2002) observed positive dependence between fish length and induction time but not with recovery time in respect of Sockeye Salmon (Oncorhynchus nerka). In the present study both induction and recovery times indicated positive dependence with fish length for both the fish species. The results revealed group-A fishes has less induction time compared to group-B fishes. Recovery times in group-B Silver dollar fishes was more than group-A fishes but exactly opposite results were observed in case of sword tail fishes. This indicated that the response to the anesthesia in terms of induction and recovery times might vary among the fish species. The concentration of 80 mgl<sup>-1</sup> was highly effective. The 10 and 20 mgl<sup>-1</sup> dose of clove oil could be used during transportation of Silver dollar and Sword tail fishes respectively as this dose was observed to have reduced the movement of fishes and brought them to light sedation stage. Clove oil, a natural cost effective anesthetic can become an alternative to the standard MS-222, Phenoxyethanol, Quindaldine or Benzocaine, which are hazardous, expensive and sometimes less effective (Munday and Wilson, 1997).

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### References

- Abramonitz, A.A. Hisaw, F.L., Boettiger, F. and Papandrea, D.N. 1940. The origin of the diabetogenic harmone in the dogfish. Biol. Bull. Woods Hole, 78: 189-201.
- Abramonitz, A.A. 1937. The opercular approache to pituitary. Science, 85: 609.
- Durve, V.S. and Dharma Raja, S.K. 1965. Effects of anesthetics on the behaviour of mullet fingerlings and the scope of using these in different fishery procedures-I. J. Mar. Biol. Ass. India, 1965, 8 (1): 28-56.
- Haskell, D.G.1940. Stunning fish by electricity. Progr. Fish. Cult., 49: 33-34.
- Hicks, B. 1989. Anesthetics: sweet dream s for fragile fish. Canadian Aquaculture, March –April 89: 29-31.
- Keene, J.L., Noakes, R D.G. Moccia, D. and Soto, C.G. 1998. The efficacy of clove oil as an anesthetic for rainbow trout, Onchorynchus

- Applied Fisheries & Aquaculture 2002 Vol. II (2): 7-9
  - mykiss (Walbaum). Aquaculture Research. 29: 89-101.
- Munday, P.L. and Wilson, S.K. 1997. Comparative efficacy of clove oil and other chemicals in anaesthetization of Pomacentrus amboinensis, a coral reef fish. J. Fish Biol., 51: 931-938.
- Natarajan, M.V. and Renganathan, V. 1960. A note on the possibilities of utilizing quinaldine in transporting live fish. Curr. Sci., 29: 393.
- Osborn, C.M. 1941. Studies on the growth of integumentary pigments in the lower vertebrates II. The role of the hypophysis in the melanogenesis in the common carfish (Ameiurus meals). Biol.Bull.Woods Hole. 81:352-363.
- Osborn, C.M.1938. The role of melanophoredespersing principle of the pituitary in the colour changes of the catfish. J. Exp. Zool., 79(2): 309-330.
- Parker, G.H. 1937. Integumentary color changes of elasmobranch fishes especially of *Mustelus*. Proc. Am. Phil. Soc., 77(3): 223-247.
- Parker, G.H. 1939. General anesthesia by cooling. Proc. Soc. Exp. Biol., 42:186-187.
- Patrick D. and Adeline, C. 2000. Clove oil used as an anesthetic with juvenile tropical marine fish. SPC Live Reef Fish Information Bulletin, 9: 1-7.
- Pickford, G.E. and Atz, J.S. 1957. The physiology of the pituitary gland of fishes. New York Zoological Soc., 613 pp.
- Saha, K.C., B.P. Sen and Banerjee, S.M.1965. Use of Sodium Amytal in transport of fry of Indian major carps. Ann. Biochemi. Exptl. Med., 15(2): 133-134.
- Sreenivasan, A. 1962. Use of tranquilizing drugs and narcotics in reducing oxygen consumption by fish. Indian J. Fish., 9(2): 738-745.
- Taylor, P.W. and Roberts, S.D. 1999. Clove oil: an alternative anesthetic for aquaculture. North American Journal of Aquaculture, 61: 150-155.
- Thienpont, D. and Niemegeers, C.J.E. 1965.
  Propoxate (R 7464): a new potent
  anesthetic agent in cold-blooded
  vertebrates. Nature, 205:1018
- Woody, C.A., Nelson, J. and Ramstad, K. 2002. Clove oil as an anesthetic for adult sockeye salmon: field trails. J. Fish Biol., 60: 340-347.