

## Monitoring of Behavioral Responses in Great Sturgeon, *Huso huso* After Sub-Lethal Exposure to Water soluble fraction of Diesel Oil

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**Abstract:** Great sturgeon (*Huso huso*) is native to the Caspian Sea and represents a valuable species for caviar production. Thus, considering the growing cases of environmental pollution involving spills of oil distillate products into continental waters in the recent years in Caspian sea, the aims of the present study were to investigate the efficacy of Water Soluble fraction of Diesel oil to induce toxicity on Great sturgeon. We also investigate if diesel oil suppressed the swimming behavior in this species or not. Five different concentrations of WSD (0, 10, 100, 500 and 1000 ppm) were assessed. Narcosis, inaction and death occurred with different concentrations of diesel oil (100, 500 and 1000 ppm) rapidly occurred within 48 hrs after exposure to the WSD. After 38 hrs all infected fish in treatments were dead. With regard to toxicity effects of diesel oil, dosages upper than 500 ppm will be dangerous and it is not possible to come back to normal situation after 72hrs exposure.

**Key words:** Caspian sea • Great sturgeon • Oil pollution • Toxicity

### INTRODUCTION

In recent decades, oil pollution has become a universe environmental issue in that marine and inland water ecosystems are threatened greatly. So the evaluation and prediction of the effects of oil pollution on marine environment and aquatic animals have become a very urgent and important issue [1].

Acute and chronic exposure to WSD oil and derivatives can induce a variety of toxic symptoms in aquatic animals. Petroleum hydrocarbons can produce free radical as a mediator in aquatic animals [2] and this modulation can lead to the biochemical changes.

Sturgeons are valuable species, which are currently highly endangered. The rearing of these species has seen considerable progress in past years. Beluga (*Huso huso*) is one of the sturgeon species with high growth rates and appears to be very suitable for aquaculture [3]. It is spawned artificially in aquaculture facilities with the aim of restocking to improve its population. So some studies were done on effects of toxicity agents on brood stocks and fries [4]. Raising sturgeons for producing

broodstocks in order to reduce dependency on natural populations is very promising [5]. During culturing and breeding practices, stressful functions such as handling and transportation might affect its survival and growth, so using toxicity agents could be helpful [6].

In command to examine the efficacy of diesel oil as toxicity on Great sturgeon, *Huso huso*, we conducted an experiment. We also investigate to determine whether diesel oil anesthesia suppressed the normal behavior and swimming in this species or not.

### MATERIALS AND METHODS

**Fish and Rearing Condition:** The experiments conducted on juvenile Great sturgeon (average weight: 200 g) produced at the Institute of Aquaculture of the Marjani for Sturgeon, Golestan, Iran. Prior to the study, fish were maintained in groups in 400-L aquariums in an indoor facility. For the purpose of the study, fish were housed separately in experimental aquaria and acclimated to it for a minimum of one week. The aquaria shared a common source of water with a steady temperature of 25°C.

Throughout the acclimatization period and during the experiment; environmental conditions were monitored and maintained within a narrow range of variable. Fish were kept under natural photoperiodic conditions, fed on hand with handmade pellet and fasted for 24 hrs prior to each experiment.

#### Toxicity Preparation of WSD and Experiment:

A part of commercial crude oil (Purchase from Gas station, Gorgan-Iran) was added to four parts of water in a glass container. The mixture was then exposed to intense sunlight for 6 hrs, simulating a diesel spill in tropical conditions. After that the upper insoluble phase was discharged and the remaining water phase was collected [4].

Since many Scientifics add toxicity agents directly to water baths to achieve the desired dose [7], we applied prepared diesel oil solution into water. As far as diseased or weakened animals are much more susceptible to toxicity treatment, seven healthy Fish were anesthetized by immersing them in a bath containing toxicity agent so that it is absorbed through the gills and rapidly enters the blood stream. Aeration provided extra oxygen required during induction which causes increased respiration. To prevent abrading the skin of the fish, the handler wore wet latex gloves and gently transferred the fish into the container.

For the toxicity effect, a video cassette recorder (DSC-W80, Japan) was used to record fish behavior for subsequent analyses [8]. Two observers made decision using the table 1, according to the Iwana *et al.* [9].

The recovery tank used the same water as toxicity bath (at a similar temperature and chemistry) supplied with flow-through water at a high exchange rate to ensure that fish were always in contact with clean water. Water quality was carefully controlled during the experiment. Toxicity and Recovery times before deaths were recorded from the time place the fish in toxicity and recovery tank to the nearest second using an electronic stop-watch [10].

## RESULTS AND DISCUSSION

Cumulative mortality of Great sturgeon exposed to different dose of sub-lethal doses of diesel oil is in table 1. A summary of the average time to toxicity stages at each of the tested dosages is presented in table 2. Response time at tested dosages was rapidly occurred in 72hrs after exposure to the diesel oil. All experimental fish were successfully revived and no mortalities observed before 48hrs post-treatment (Table 3).

It was shown that the Great sturgeon exposed to the WSD tested concentrations in our experiment; get in toxicity phase in 72 hrs. death time for all treatments was less than 48 hrs. Toxicity induction as well as recovery phase was significantly affected by concentration, toxicity and death times were increased by elevating toxicity agent dosage.

Longer recovery times before death which was observed in fish with diesel oil could be an additional advantage in activities such as morphological evaluations, biopsy and stripping which are required long handling periods outside the water [4, 11].

Table 1: Cumulative mortality of Great sturgeon (n=21, each concentration)

Concentration (ppm)	No. of mortality			
	24hrs	48 hrs	72 hrs	96 hrs
Control	0	0	0	0
10	0	0	0	0
100	0	0	0	0
500	0	5	6	12
1000	11	19	19	21

Table 2: Stages of Toxicity and Recovery

Stages of Toxicity	Description
I	Loss of equilibrium
II	Loss of gross body movements but with continued opercular movements
III	As in stageII with cessation of opercular movements
Stages of Recovery	Description
I	Body immobilized but opercular movements just starting
II	Regular opercular movements and gross body movements beginning
III	Equilibrium regained and pretotoxicity appearance

From Iwana *et al.* [40], modified by Ackerman *et al.* [12].

Table 3: Effects of diesel oil on toxicity and recovery of Great sturgeon

Dose (mgL <sup>-1</sup> )	Death time(s)	Recovery times before death
10	-	-
100	-	-
500	48 hrs	38 hrs
500	72 hrs	36 hrs
500	96 hrs	17 hrs
1000	24 hrs	11 hrs
1000	48 hrs	7 hrs 30 min
1000	72 hrs	2 hrs 11 min
1000	96 hrs	2 hrs 20 min

Since environmental factors affect the efficacy of toxicities in fish, it is not surprising that the relationship between diesel oil dosage and water temperature was also significant ( $p < 0.05$ ) regarding toxicity and recovery times before death. As an ectoderm animal; body temperature of fish closely follows their environments which result in temperature-related physicochemical passage of the drug into the fish.

Therefore, at lower water temperatures, higher doses or longer exposure times to toxicity agents required due to the decrease in absorption rate [4]. This suggests that the levels of diesel oil used in our trial may have been very high with rapid effect on fish treated with this toxicity.

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#### REFERENCES

1. Stubblefield, W.A., G.A. Hancock, W.H. Ford and R.K. Ringer, 1995. Acute and subchronic toxicity of naturally weathered Exxon Valdez crude oil in mallards and ferrets. Environ. Toxicol. Chem., 14: 1941-1950.
2. Achuba, F.I. and S.A. Osakwe, 2003. Petroleum-induced free radical toxicity in African catfish (*Clarias gariepinus*). Fish Physiol. Biochem., 29: 97-103.
3. Moreau, R., K. Dabrowski, S. Czesny and F. Chila, 1999. Vitamin C vitamin E interaction in juvenile lake sturgeon (*Acipenser fulvescens*), a fish able to synthesize ascorbic acid. J. Appl. Ichthyol., 15: 250-7.
4. Molinero, A. and J. Gonzalez, 1995. Comparative effects of MS 222 and 2-phenoxyethanol on gilthead sea bream (*Sparus aurata* L.) during confinement. Comp. Biochem. Physiol. A., 111: 405-414.
5. Hedayati, A., V. Yavari, M. Bahmani, M. Alizadeh and T. Bagheri, 2008. Study of Some Gonadic Growth Index of Great Sturgeon (*Huso huso*) cultured in Brackish Water Conditio. Bulg. J. Agric. Sci., 14: 93-99.
6. Cataldi, E., P. Di Marco, A. Mandich and S. Cataudella, 1998. Serum Parameters of Adriatic sturgeon *Acipenser naccarii* (Pisces: Acipensiformes): effects of temperature and stress. Comp. Biochem. Physiol. A., 121: 351-354.
7. Seol, D.W., J. Lee, S.Y. Im and I.S. Park, 2007. Clove oil as an anaesthetic for common octopus (*Octopus minor*, Sasaki). Aquac. Res., 38: 45-49.
8. Cooke, S.J., C.D. Suski, K.G. Ostrand, B.L. Tufts and D.H. Wahl, 2004. Behavioral and physiological assessment of low concentrations of clove oil anaesthetic for handling and transporting largemouth bass (*Micropterus salmoides*). Aquaculture, 239: 509-529.
9. Iwama, G.K., J.C. McGeer and M.P. Pawluk, 1989. The effects of five fish anaesthetics on acid-base balance, hematocrit, cortisol and adrenaline in rainbow trout. Can. J. Zool., 67: 2065-2073.
10. Mylonas, C.C., G. Cardinaletti, I. Sigelaki and A. Polzonetti-Magni, 2005. Comparative efficacy of clove oil and 2-phenoxyethanol as toxicities in the aquaculture of European sea bass (*Dicentrarchus labrax*) and gilthead sea bream (*Sparus aurata*) at different temperatures. Aquaculture, 246: 467-481.
11. Munday, P.L. and S.K. Wilson, 1997. Comparative efficacy of clove oil and other chemicals in anaesthetization of *Pomacentrus amboinensis*, a coral reef fish. J. Fish Biol., 51: 931-938.