Management of Stress in Fish for Sustainable Aquaculture Development

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Abstract :The estimated contribution of aquaculture to global supplies of fish has risen tremendously over the years. This is due to the fact that aquacultural practice, all over the world is becoming more and more intensive; utilizing available resources thereby enhancing maximum productivity. An inevitable part of intensive aquaculture is manipulation of fish, which include handling, stocking, sorting, confinement, transportation and other farm operations right from hatchery to the final commercial stage. However, these procedures produce disturbances which elicit stress responses, leading to decreased performance, altered peripheral leucocytes distribution such as heterophillia and lymphocytopenia, as well as increased susceptibility of fish to diseases, and in extreme cases leads, to mortality. A sound and working knowledge of the importance of stress in aquaculture management and its consequence in causing decline in yield, which will ultimately lead to loss in profit. This aspect of fishery management must be adequately looks into. Hence, this paper review critically the causes, mechanisms of stress, consequences and various ways of effectively alleviating it in aquacultural practice, so as to enhance the sustainability of aquaculture as a major source of fish supply for the growing population in the world. [Ugwemorubong Ujagwung Gabriel, Ojo Andrew Akinrotimi. Management of Stress in Fish for Sustainable

Aquaculture Development. Researcher. 2011;3(4):28-38]. (ISSN: 1553-9865). <u>http://www.sciencepub.net</u>.

Key words: stress, fish, management, aquaculture, sustainable yield

1. Introduction

Over the past decades aquaculture has expanded, intensified, and diversified in response to an increasing demand for fish as a source of protein globally. This is because production from capture fisheries has reached their maximum potential possible, as the catch is dwindling with each passing day (Akinrotimi et al 2007a). According to FAO (2006), fish supplies from capture fisheries will, therefore, not be able to meet the growing global demand for aquatic food and protein. Hence there is the need for viable alternative fish production systems that can sufficiently meet this demand and aquaculture fits exactly into this role (FAO, 2005; Akinrotimi et al., 2007a).

The estimated contribution of aquaculture to global supplies of fish, crustaceans and mollusks increased from 3.9% in 1970 to 27.3% in 2000 (FAO 2002). From the year 2000 and beyond, aquaculture has grown in leaps and bounds (Table 1) making it the fastest growing food producing industry in the world (NACA/FAO, 2000). Although rapid growth of aquaculture in the past two decades has enabled the world fisheries supply to keep pace with population growth (Boyd et al. 1998; Savas, 1998), the future of aquaculture still lies in increasing production efficiencies and intensities, so as to produce more fish using less land water and financial resources (Jamu and

Ayinla, 2003).

As aquaculture world over is becoming more and more intensive, this involves manipulation of fish and other farm management procedures which include confinement, handling. liming fertilization, transportation and other operations from the hatchery to the final commercial stage (Angelids et al., 1987; Gabriel et al., 2007a). According to Pickering (1981) these management procedures as crucial as they are, produce some level of disturbances, which can elicit a stress response leading to decreased fish performance (Maule and Shreck, 1990), alterations of the peripheral leucocyte distribution, such as heterophilia and lymphocytopaenia (Ellsaesser and Clem 1986, Ainsworth et al., 1991; Gabriel et al. 2007a) increased susceptibility to diseases (Pickering and Pottinger 1985; Maule et al. 1989) and in extreme, cases leads to mortality (Akinrotimi et al. 2007b). It should be noted that a very devastating effect of stress or the stock may occur during application of these management procedures with and without apparent warning. This then raises the question of proper monitoring of stress in fish, in order to reduce to the barest minimum the negative effects of such management procedures. This paper critically reviews the causes, mechanisms and consequences of stress in cultured fish and suggests various ways such stresses can be effectively managed.

2. Description of stress in fish

Stress is any condition that causes physical or psychological discomfort which results in the release of stress-related hormones or specific physiological responses (Foster and Smith, 2007). Stoskopf (1993) observes that stress is present virtually in the lives of all living organisms and can be described as the latent force that brings about physical, psychological and physiological change and adjustment. Unfortunately the term "stress" is used inconsistently. It is sometimes taken to mean the environmental alteration (stressor) itself and at other times the response of the fish, population and ecosystem (Pickering, 1981). Selve (1950) defined stress as "the sum of all the physiological responses by which an animal tries to maintain or re-establish a normal metabolism in the face of physical and chemical force". But this definition according to Wedemeyer et al. (1990) does not consider the fact that the outcome of stress may be negative for an individual but positive for the population. For example, mortality of individual fish due to exhaustion from over crowding may actually enhance survival of the population when space or food supplies are limiting.

According to Esch and Herzer (1998) stress, then can better be defined as "the effect of any

Table 1. World fisheries and aquaculture production

environmental alterations or force that extends homeostatic or stabilizing processes beyond their normal limits, at any level of biological organization". While Barton (1997) defined stress as "the response of the cell, or organism, to any demand placed on it such that it causes an extension of a physiological state beyond the normal resting state". According to Chrousos (1997), stress in fish can be considered as a state of threatened homeostasis that is re-established by a complex of adaptive responses. If the intensity of the stressor is overly severe or long lasting, however, mechanisms physiological response may be compromised and it can become detrimental to the fish's health and well being, or maladaptive a state associated with the term "distress" (Barton and Iwama 1991: Barton 2002).

Stress can be physical, psychological or environmental. It can either be short and sudden or long and chronic. While mild, short-term stress has few serious health effects, compared to long-term stress, it should be noted that short-term stress contribute to many of the illnesses and deaths in aquarium fish (Tullock, 2001). This description is based on the type of stressors acting on the fish specifically over a period of time.

Production (million tones)	2000	2001	2002	2003	2004	2005
Inland						
Capture	8.8	8.9	8.8	9.0	9.2	9.6
Aquaculture	21.2	22.5	23.9	25.4	27.2	28.9
Total Inland	30.0	31.4	32.7	34.4	36.4	38.5
Marine						
Capture	86.8	84.2	8.4.5	81.5	85.8	84.2
Aquaculture	14.3	15.4	16.5	17.3	18.3	18.9
Total Marine	101.1	99.6	101.0	98.8	104.1	103.1
Total Capture	95.6	93.1	93.3	90.5	95.0	93.8
Total Aquaculture	35.5	37.9	40.4	42.7	45.5	47.8
Total World Fisheries	131.1	131.0	133.7	133.2	140.5	141.6

3. Stressors common in fish cultivation

Stressors are real or perceived challenges to an organism's ability to meet its real or perceived needs (Greenbery et al., 2002). Fishes are exposed to stressors in nature as well as in artificial conditions such as in aquaculture or in the laboratory (Iwama et al., 2006). Stressors that challenge homeostasis, often regarded as the most urgent of needs are the best known (Table 2). When an organism's capability to maintain homeostasis within a specific range is exceeded, responses are evoked that enabled the organisms to cope by either removing the stressor or facilitating co-existence with it (Antelman and Caggiula, 1990).

Fish under intensive culture conditions are

exposed to a regime of acute and chronic stressors, which have adverse effects on growth, reproduction immunocompetence, and flesh quality (Barton et al., 1987; Maule et al., 1989; Shreck et al., 2001). These stressors, which are peculiar to fish in aquaculture range from chemical, biological, physical and procedural. The most prevalent which induce stress in fish is the procedural stressors, from daily management practices in fish farming (Table 2).

3. Indicators of stress in fish

There are various direct and indirect quantitative parameters used as indicators of stress in fish (Barton and Iwama, 1991). This ranges from physiological status to physical observations on the fish (Table 3.). These include changes in the level of cortisol in the plasma, alterations in immunoglobulin levels, and changes in the haematological parameters. As in other vertebrates, the blood concentration of costicosteroid hormones is a major index of stress in fish and elevated levels of these hormones arise from activation of the hypothalamus-pituitary-internal (HPI) axis (Wendelaar-Bonaga, 1997). Physical signs of stress from observation differs with stressors and varies from one species of fish to another.

4. Fish responses to stress in aquaculture

Fish responses to stressors in aquaculture vary according to the source, effect, environment and characteristics of stressor (Akinrotimi et al., 2009). However, the features of stress reactions are generally common to most forms of stressors. Stress response of fish is an integrated reaction with behavioural, neural, hormonal and physiological elements all combined together to provide fish with the best possible chance of survival (Pickering, 1993). Also, Akirontimi et al. (2007c) observed that male and female fish differs in the way they response to stress. Therefore sex is a crucial factor in the way fish respond to stress. Physiological responses of fish to environmental stressors have been grouped broadly as primary, secondary and tertiary (Barton, 2002).

4.1. Primary Response

Stress reaction arises from the activation of the sympathetico-chromaffin system and the hypothalamic pituitary-interrenal axis (Pickering, 1981). Changes of parameters of these systems are named as primary stress responses (Vosyliene and Kazlauskiene, 1999). Primary response involves the initial neuroendocrine responses, and include the release of catecholamines from chromaffin tissues (Randall and Perry, 1992; Reid 1998) al.. and the stimulation of et the hypothalamic-pituitary-interrenal (HPI) axis culminating in the release of stress hormones, cathecolamines and cortisol circulation (Wendelaar Bonga, 1997; Mommsen et al., 1999). Cortisol is released from the interrenal tissue located in the head (anterior) kidney, in response to several pituitary hormones, but most potently to adrenocorticotrophic hormone, ACTH (Balm 1997). The approximate resting and stressed levels in the plasma of salmonids are between 20-70 nmoles/L for adrenaline, and 40-200ng/ml for cortisol (Iwama et al, 2004). These values may serve as general basal data. Individual conditions including species differences, general characteristics, prior rearing history and local

environment will modify the plasma values for control and stressed states (Barton et al., 2002).

4.2. Secondary Response

This response includes changes in plasma and tissue ion and metabolic levels, haematological features, and heat shock or stress proteins (HSPs) all of which related. Plasma cortisol concentrations in selected juvenile freshwater fishes before and one hr after being subjected to an identical 30-sec aerial emersion, handling stressor to physiological adjustments such as metabolism, respiration, acid-base in status, hydromineral balance, immune function and cellular responses were also affected (Pickering, 1981; Iwama et al., 1997; 1998; Mommsen et al., 1999; Barton, et al. 2002). In this type of response the stress hormones activate a number of metabolic pathways (Table 4) that result in alterations in blood chemistry and haematology (Randall and Perry 1992; Vijayan et al.; 1994; Iwama et al., 2004).

Also, worthy of mention is the plasma glucose concentration which often times has been used as an indicator of stressed state in fish, and perhaps may be the most commonly measured secondary response parameters to stressors in fish (Barton et al., 2002).

According to Iwama et al., (2006) the plasma glucose concentration in circulation is dependent upon glucose production and its clearance from the circulation. The glucose produced, when fish is under stress supplies energy to tissues such as the brain, gills and muscles in order to cope with the increased energy demand. Liver is the main source of glucose production by glycogenolysis and is achieved or gluconeogenenesis. Adrenalin and cortisol have been linked to increased glucose production in fish (Vijayan et al., 1994).

4.3. Tertiary Response

Tertiary response refer to aspects of whole animal and population level performance such as changes in growth, condition, overall resistance to disease, metabolic scope for activity, decreased reproductive capacity and ultimately survival (Wedemeyer and McLeay, 1981; Wedemeyer et al. 1990; Iwama et al. 2004). All these alterations according to Barton et al. (2002) may be associated with stress. Mediated energy-repartitioning that diverts energy subtrates from vital life processes, such as reproduction and growth in order to cope with the enhanced energy demand associated with stress, have detrimental consequences on cultured species.

Table 2.	Stressors common in intensive aquaculture	
S/No.	Stressor	Occurrence (%)
А	Chemical stressors	
1.	Poor water quality (Low DO., improper pH)	20
2.	Pollution- intentional pollution: efficient, wastes, sewage; -accidental pollution: spills: insecticide, pesticide	10
3.	Diet composition imbalance diet	10
4.	Nitrogenous and other metabolic wastes i.e accumulation of ammonia or nitrite	35
В	Biological stressors	
1.	Population density over crowding	5
2.	Social dominance	1
3.	Micro organisms-pathogenic and non pathogenic	2
4.	Macro organisms-internal and external parasites	2
С	Physical stressors	
1.	Temperature	1
2.	Light	1
3.	Sounds	0
4.	Dissolved Gases	1
D	Procedural stressors	
1.	Handling	3
2.	Transportation	5
3.	Sorting/Grading	3
4.	Disease Treatments	1
Source:	Field Survey (2007)	

Table 3. Indicator of stress in some teleost fish

Species	Stressor	Indicator of Stress	References		
Sarotherodon	Confinement	Reduced Haemoglobin increased	Gabriel et al. (2007b)		
melanotheron		differential count	Gabriel et al (2007c)		
C. gariepinus	Acclimation	Distortion in blood parameter	Gabriel et al. (2004)		
S. melanotheron	Salinity changes	Increased WBC	Anyanwu et al. (2007)		
		Reduced RBC			
S. melanotheron	Transportation	Distortion in blood parameters	Akinrotimi et al. (2007c)		
Tilapia guirenesis	Transportation		Akinrontimi et al. (2009)		
Oreochromis niloticus	Transportation	Reduced PCV	Orji (2005)		
Sceianops ocellatus	Transportation	Hormonal dysfunction	Robertson et al. (1987)		
C. gariepinus	Poor water quality	Vertical swimming	Field survey (2007)		
fingerlings		-	• • • •		
C. gariepinus	Fertilizer effluents	Altered tail/opercular beat	Bobmanuel et al. (2006)		
/hybrid/ O. niloticus		frequencies/increased mucus			
2		production			
C. gariepinus	Chemical	Distortion in tissue chemistry	Onusiriuka and Ufodike		
			(2000)		
C. gariepinus	Chemical	Reduced plasma enzyme activities	Gabriel and George (2006)		
Tilapia guineensis	Industrial effluents	Reduced leucocyte levels	Davids et al. (2002)		
C. gariepinus	Chemotheurapetants	Changes in blood component	Musa and Omoregie (1999)		
O. niloticus	Chemotherapeutants	Depression of haemopoesis	Omoregie and Oyebanji		
			(2002)		
Salvinus calpinous	Chemotherapeutants	Spinal deformities	Toften and Jobling (1996)		
C. gariepinus	Crude oil	Distortion of haematocrit and	Gabriel et al. (2001)		
- *		haemoglobin			

5. Efects of stress in cultured fish

The effects of stress in cultured fish ranges from alteration of fish defence mechanisms decreased protective barriers of fish, damaging of scales and skin, inflammation and antibody production. Stress, induced by handling or chemical (disease treatment) often causes some changes in mucus which decreases its effectiveness as a chemical barrier against invading organisms. Stress upsets osmoregulation by disrupting the normal electrolyte (sodium, potassium and chloride) balance which results in excessive uptake of water by fresh water fish and dehydration in salt water fish, thereby disrupting energy balance (Maule, 1994; Norris, 2000). Scales and skin are the most commonly damaged by handling stress. Any injury to the skin or removed scale creates on opening for invasion by pathogenic organisms.

Stress according to Akinrotimi et al. (2007c) often in extreme cases lead to mortality, which is more pronounced in the early stage of the fish life (Table 5) because they are more fragile and vulnerable to external stressors. But as they advance, the effect of stress becomes reduced, but often manifests in retarded growth, impaired reproductive ability in tilapias and some other fish that breed in captivity.

6. Remedy for controlling stress

Considering the devastating effect stress can have cultured species, the challenge before the in aquaculturists is to develop strategies that will maximally reduce stress in cultured species so as to enhance sustainable aquaculture production. Although in aquaculture practices, the strategies for handling acute and chronic stress may differ, as their level of intensity differ (Tullock, 2001). The cultured fish may appear to be doing well, until one day gets sick and dies, and then a few weeks later another one, and so on, until the whole stock is affected (Stoskopf, 1993). If there is any mortality in the fish stock, there is probably a source of stress that needs to be identified and remedied. This can be done in one or more of the following ways or combination of the options:

Parameters	Stimuli			
	Chemical	Starvation	Confinement	
Morphological:				
Weight of fish		*	*	
Weight of tissues		*	*	
Condition factor		*	**	
Tissue-somatic indices	*			
Cardio-respiratory			**	
Ventilation frequency	*		**	
Heart rate		*	**	
Meant RR interval duration		*	**	
Rhythm stability index			**	
Ventilation waves				
Haematological:				
Erythrocytes	*	*		
Leucocyte	*	*	**	
Haemoglobin	*		**	
Haematocrit	*		**	
Glucose		*	**	
Neutrophyls	*		**	
* Significant differences from the contro 0.01)	bl (p \leq 0.05), ** Extrem	ely significant differer	nces from the control	

Source: (Vosyliene and Kazlaoskiene, 1999, Gabriel et al 2007a).

6.1. Maintenance of good water quality in culture medium

Good water quality involves preventing accumulation of organic debris and nitrogenous wastes, preventing ammonia build up, maintaining appropriate pH and temperature depending on the species that is being cultured (Table 6). It also involves most importantly maintaining dissolved oxygen levels of at least 5mg/L.

6.2. Appropriate stocking density

To avoid unnecessary stress, fish should be stocked at appropriate stocking density (Table 7). The stocking rate is the number of fish, which a pond can hold and maintain without exceeding its carrying capacity. The stocking

density therefore depends on the species and the culture systems that are being utilized. Appropriate stocking density prevents the fish from over crowding which often leads to struggling for food, oxygen and survival and ultimately culminates in stress.

Stages of fish	Stress Effect
Fry	Devastating, with high mortality
Fingerlings	Very fatal, with high mortality
Juveniles	Moderate mortality, reduced growth rate
Adult	Less effect compared to juveniles reduced growth rate
Brood Fish	Less gravid
Field survey, 2006	-

Species	pH	DO (mg/l)	Temperature (⁰ C)	Salinity $(^{0}/_{00})$
Clarias gariepinus	6.5-9.5	>0.67	15-35	0-0.5
T. guineensis	5.5-9.6	>0.41	14-33	0-35
Sarotherodon melanotheron	5.4-9.8	>0.41	15-34	0.1-35
Oreochromis niloticus	6.8-100	>0.42	10-35	0.1-1.0
Mugil cephalus	5.5-10.0	>0.46	14-33	5-35
Chrysichthys nigrodigitatus	5.6-9.0	>0.63	18-33	0-35
Ictalurus punctatus	6.5-10.0	>0.66	10-33	0-0.6
Cyprinus carpio	6.7-10.0	>0.65	10-33	0-0.6
Chanos chanos	6.8-10.2	>0.41	10-30	0-0.8
Megalops atlanticus	5.5-9.8	>0.40	10-35	0-36
Source: Boyd (1982)				

6.3. Balanced diet

Fish should be fed with a high quality diet that meets their nutritional requirements depending on the species, age, size and production function (Table 8).

Balanced and complete feed have been observed by Tacon (1994), as an appropriate way of eliminating nutrititional induced stress in fish. The vitamin C included diet, has been observed by Ibiyo *et al.* (2006) to reduce stress to the barest minimum in *Heterobranchus longifilis*.

6.4. Proper sanitation

This implies routine removal of debris from fish tanks, and regular disinfection of containers, nets and other equipments between groups of fish, especially after harvesting and before new stock arrived (Table 9). The culture medium should be washed, cleared and disinfected. It should be noted that organic debris which accumulate at the bottom of tanks is an excellent medium for reproduction of fungal, bacteria, and protozoan agents. Prompt removal of debris will help reduce the number of pathogens and hence pathogen-induced stress.

Table 7. Stocking density of some culturable fishes in earthen and tank culture system.					
Species	Earthen pond (10 ³ fish /ha)	Concrete tank (No of fish/ m ³)			
C. gariepinus	26.6-10	30-40			
T. guineensis	10-20	20-40			
Heterotis niloticus	3.5-8				
O. niloticus	10-20	20-40			
Cyprinus carpio	3-6	20-30			
Sarotherodon melanotheron	15-25	20-30			
Source: Adesulu (2001).					

6.5. Usage of Anesthetics

Anesthetics can be used to minimize stress during transportation, breeding and other farm activities. It is used to sedate bigger fish during breeding, so as to minimize stress. Also, during out break of any disease, anesthetics could first of all be used to stabilize the fish before proper treatment is applied. Selected anesthetics and estimates for optimum doses, as well as induction and recovery times for various fishes are shown in Table 10.

6.6. Proper Management of Fish during Transportation

To alleviate stress due to transportation, fish should be transports very early in the morning or late in the evening. Of recent, air conditioned vehicles, are being used to transport fish over a long distance, to reduce mortality as a result of stress (Table 11).

 Table 8. Dietary protein and energy levels resulting in highest growth rates in various fish species (% of dry diet)

 Fish species
 Crude dietary protein Gross dietary energy

1 isit species	Crude dietary protein	Gross dictary chergy
	(%)	(kj/g)
Chinook salmon (Oncorhynchus tshawytscha)	40-55	19.3-20.4
Rainbow trout (Oncorhynchus mykiss)	40-45	19.1-20.8
Estuary grouper (Epinephelus salmoides)	40-50	14.3-18.1
Gilthead bream (Pagrus auratus)	40-45	22.5-23.2
Red sea bream (Pagrus major)	55-40	21.1-22.2
Smallmouth bass (Micropterus dolomieui)	45-50	18.4
Striped bass (Morone saxatilis)	47-55	24.8
African catfish (Clarias gariepinus)	40-45	18.6
Asian catfish (Clarias batrachus)	30-35	17.2
Channel catfish (Ictalurus punctatus)	35-40	11.5-16.9
Common carp (Cyprinus carpio)	31-40.6	12.8-22.7
Indian major carp (Labeo rohita)	34-36	15.5
Tilapia (Oreochromis aureus) fingerling	34-36	13.4
Tilapia (<i>Tilapia zilli</i>)	30-35	21.8
Tilapia hybrid (O. niloticus x O. aureus)	30-35	17.3
Mullet (Mugil cephalus)	28-30	11.0
Milkfish (Chanos chanos) fry	40-45	15.3
Source: Data adapted from Tacon (1990), De Silva and Anderso	on (1995); Hassan et al. (19	96)

CONCLUSION

With the recent upsurge in the world population, especially in the developing countries and the corresponding increase in the need for cheap source of food to satisfy the protein need of the people, coupled with the declining yield in capture fisheries, there has been a massive intensity on aquaculture, as a veritable option in realizing this goal. This level of intensity which involves various ways of handling fish, translates to more stress for the fish. To sustain and increase the yield from aquaculture practice there is the need to create the awareness and understanding of what constitutes stress in fish, notably in the area of physiological mechanism and responses that lead to changes in metabolism, growth, immune functions, reproductive capacity and normal behaviour.

S/no.	Cleaning routines in fish far Equipment/material	Frequency/time of cleaning		
1	Earthen Pond bottom	Desilt pond bottom once a year after harvesting		
2	Tank bottom	Regular removal of debris depending on the age of the fish		
3	Nets	Regular washing and drying of nets		
4	Farm tools	Regular washing and cleaning of tools used in the farm		
5	Tanks	Tanks should be washed at least once in every three months		
6	Farm surrounding	Farm environment should be cleared and cleaned regularly.		
	Field survey (2007)	rann environment should be cleared and cleaned reg		

The indices of fish responses to stimuli varied in their responsiveness to various types of stressors in aquaculture. This is because the effect of different stimuli can occur in an organism through different physiological systems. Different stressor when affecting fish separately cause responses, which can be reversible when appropriate

steps are taken to ameliorate the trend. However, it must be well understood that the response of fish to a stressor is a dynamic process that need to be seriously looked into by fish biologists and aquaculturist. If this is done properly, it will undoubtedly increased production and leads to sustainability of aquaculture industries in the world.

Anaesthetic	Dose	Induction time	Recovery time	Test fish	References
MS 222	25-100	<3 min	<10 min	Salmonids, Carp, Minnows	(Yesaki, 1988)
	250-480 mg/l	<5min	<10min	Atlantic salmon	(Malmstroem et al. 1993)
	75 mg/l	Rapid	3.7-7.1 min	Cod	(Mattson and Ripple, 1989)
	80-100 mg/l	2.6-6.8 min	2.5-1.2min	Tilapia	(Ferriera et al. 1979; Ross and Ross, 1984)
Benzocaine hydrochloride	40mg/1			Cod	(Ross and Ross, 1984)
,	25-50 mg/l	3min	4.3-6.32 min	Salmonids	(Yesaki, 1988)
	55-85 mg/l	3 min	<10 min	Bass	
	50-100 mg/l	1.2-3.9	3.1-2.2	Carp	(Ferriera et al. 1979)
	C C	1.6-6.5 min	2.9-2.2 min	Tilapia	
Lidocaine plus	350 mg/l	53 see	13min	Carp	(Carrasco et al. 1984)
Sodium bicarbonate	pH 6.5 + 642mg/l NaHCo ₃	5min	10min	Carp	(Booke, 1978)
	900 mg/l	5min	12.1min	Adult salmon	(Gilderhus and Marking, 1987)
Metomidate	5-20mg/l	<3min	Rapid	Cod	0, ,
Propoxate	1-4mg/l	<10min	•		(Ross and Ross, 1984)
Quinaldine sulfate	15-40 mg/l	2-4min	1-20min	Salmonids	Gilderhus and
	150 1	1.4.3	4.10	0.1 1	Marking, 1987.
Propanidid	1.5-3mg/l	1-4min	4-10min	Salmonids	(Siwicki, 1984)
Clove oil and AQUI-S	40mg/l	2.5-4min	3min	Rainbow trout (FW, 11 ⁰ C)	(Anderson et al., 1997)
	40-60mg/l	3-4min	12-14min	Raibow trout (FW, 9 ⁰ C)	(Keene et al., 1998)

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2/18/2011