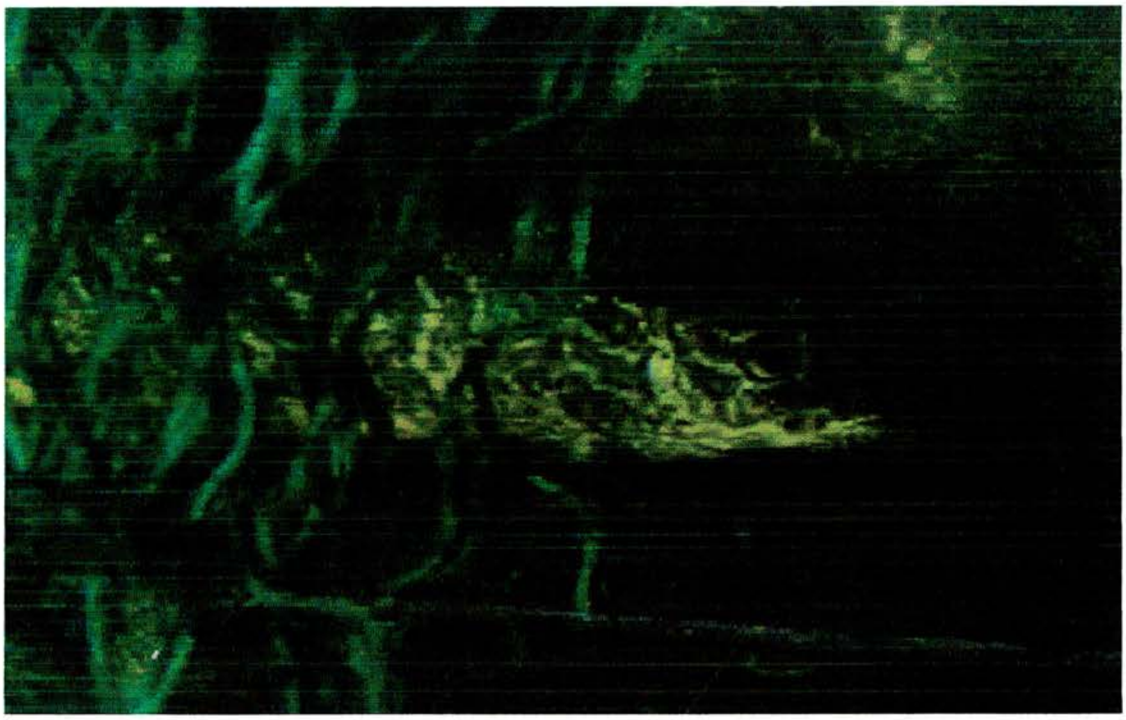
Predator recognition in Atlantic salmon, and interactions with pike

Lorraine A. Hawkins



Thesis submitted for the degree of doctor of philosophy,
University of St Andrews

October 2005

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Abstract

This thesis investigates predator recognition skills of Atlantic salmon, *Salmo salar*. In particular it focuses on their innate ability to recognize predators and their capacity to enhance predator recognition skills through learning. The thesis also addresses the behaviour of a predator of Atlantic salmon, the pike, *Esox lucius*. The spatial distributions and social interactions of juvenile pike are explored, and the potential for interactions between salmon and pike in Scotland is discussed. Olfactory experiments showed that salmon have the innate ability to recognize pike and distinguish them from other species. Newly-hatched salmon have finely-tuned innate responses to predator odours determined by the species of predator and the strength of the odour. The anti-predator responses of wild-progeny salmon that were reared either in wild (predator-present) or in hatchery (predator-absent) environments for ten months were broadly similar. However, a proportion (*c.* 30%) of hatchery-reared fish showed a delayed response to a predator cue, relative to wild-reared fish, demonstrating that some hatchery fish were slower to realize the potential danger. Eight months of hatchery-rearing did not lead to a decline in the innate response to predator odour. However, at age *c.* three months, predator-naïve salmon showed an enhanced response to predator odour compared to younger or older fish. This coincided with an increase in the ability to learn predator odour recognition and supported the hypothesis that a period of heightened receptivity to predator odour enabled fish to build on existing abilities with learnt abilities. A field-based experiment that released two groups of hatchery-reared salmon smolts, one group that was exposed to cues that stimulated learning of predator recognition and a second control group, found no difference in survival rate between groups. The high mortality of both groups (> 95%) was suggested to be due to pike predation. Experimental results suggested that the potential for interactions between salmon and pike could be spatially wide-ranging. The habitat use of juvenile pike was not simply a preference for physical habitat types but was also influenced by social interactions, which led to individuals not selecting for vegetated habitat. There was no evidence of territoriality in juvenile pike; in contrast, pike were found to aggregate under habitat-specific conditions.

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SECTION I

CHAPTER ONE

General introduction

When do baby harp seals shut their eyes and hold their breath (Lydersen & Kovacs 1995)? Why do crucian carp inhabiting some pools develop deep bodies but in other pools remain more streamlined (Brönmark & Miner 1992)? What induces salamanders to hatch early from their eggs (Sih & Moore 1993)? Why does the spider crab decorate itself with algae (Wilson 1987)? Why do minnow schools jump and 'explode' (Magurran & Pitcher 1987)?

These animals are all behaviourally controlling their risk of predation. Predation is a major selective force in the evolution of some morphological and behavioural characteristics of animals (Lima & Dill 1990), from the induced morphological defence of carp, to the evasion tactics of minnows and the 'freezing' behaviour of baby seals. As most animals are always at some predation risk, many aspects of animal decision-making and behaviour are governed by their perception of risk (Lima & Dill 1990, Ydenberg 1998). This thesis explores how fish perceive and respond to predators and how these responses may influence their fitness.

(I) HOW DO ANIMALS RECOGNIZE AND RESPOND TO PREDATORS?

To address the question of how animals recognize and respond to predators, we must understand the process through which recognition occurs, and how responses are produced. The study of predator recognition in animals must incorporate how the animal's sensory systems are designed to collect information regarding predators and how they process the information, how constraints may act on acquiring, processing and retaining this information, and the function of acquiring, processing and retaining such information, ultimately relating to its effect on fitness. To understand the responses to predators we must consider the optimal design of anti-predator responses and the capacity of animals to perform responses, how biases and constraints may influence information processing and behavioural responses and trade-offs between these responses and other factors influencing fitness (Figure 1).

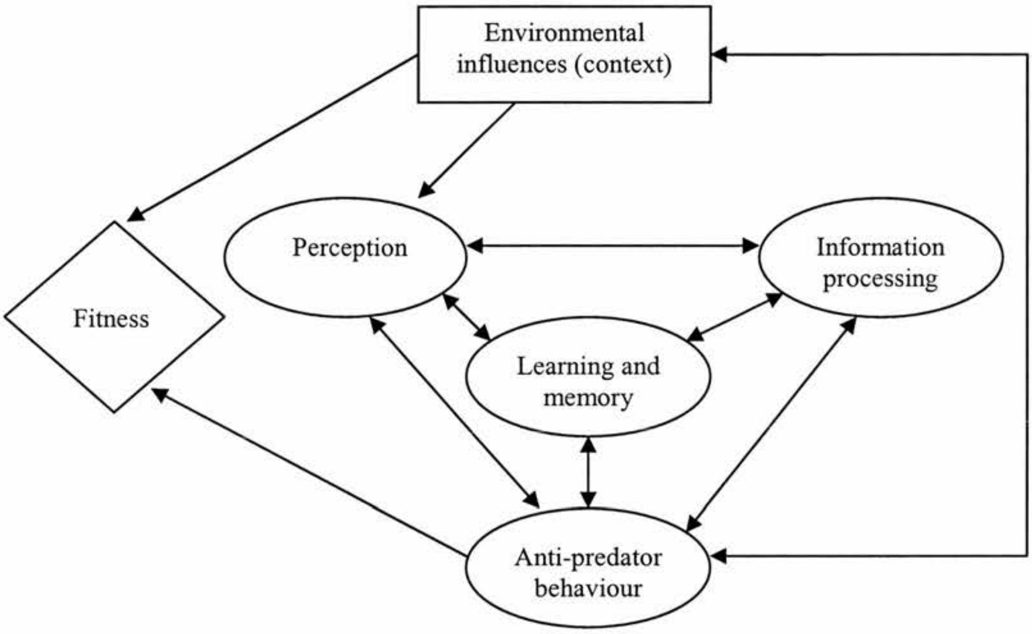

Figure 1. Processes governing anti-predator responses and influencing fitness (adapted from Dukas 1998a).

Cognition is characterized as the neuronal processes concerned with the acquisition and manipulation of information by animals. Cognitive abilities influence all aspects of predator recognition. An animal will perceive huge quantities of information through its sensory systems. How does it filter out the information regarding predation risk that will be relevant to its fitness, for processing? At the next step, most information that is processed and utilized for working memory will subsequently be lost; how does an animal chose which information should be retained in long-term memory?

The capacity of animals to recognize predator cues may have a stronger influence on fitness than capacities to recognize other cues, such as those pertaining to foraging or mate searching. Therefore we may expect memories of predator cues to be formed more readily. The building of information into long-term memory is referred to as learning. Learning is defined as the acquisition of or change in memory that allows an animal to alter its subsequent responses to certain stimuli (Dukas 1998b), and more typically as a change in behaviour as a result of experience (Kieffer & Colgan 1992). Aspects of predator avoidance that may be learned include learning to ignore neutral stimuli, learning not to overreact to predator stimuli (this is likely a trade-off situation), learning how to avoid attack (e.g. by learning location of attack) and learning about novel risks.

Learning may be seen as an adaptation to environmental variation (Dukas 1998b) and therefore environmental conditions can determine whether or not information is retained as a memory. Furthermore, information that is learnt may be lost from memory if it is no longer relevant; for example, information regarding predation may have seasonal or ontogenetic relevance. Furthermore, as maintenance of memories requires resource expenditure, the optimality of long-term memory is determined by a cost-benefit trade off (Dukas 1998c).

However, given the direct link between capacity to recognize predators and survival, a lot of information relating to predator recognition will probably always be essential to animals, over spatial and temporal environmental axes. Thus we should expect animals to retain more innate responses to predators, compared to learning about food, mates and other resources (Dukas 1998b). As natural selection removes individuals that fail to recognize predators, it is not surprising that many aspects of predator responses in fish are innate. Individual fitness may be further influenced through sub-lethal effects of predators, such as by a reduction in an animal's activity, growth and fecundity (Ydenberg 1998). The influences of information processing and decision making on an animal's fitness is encompassed in the study of cognitive ecology.

(II) FISH COGNITION

Historically, fish have been epitomized in the perception of the General Public as forgetful and stupid animals. So research from the last two decades may come as a surprise to some people, but fish have strong cognitive abilities! The cognitive processes of fish required for learning spatial information have similar mechanisms to mammals and birds (Broglio *et al.* 2003). An edition of the journal *Fish and Fisheries* (Issue 4, 2003), reviewing learning abilities of fish, cited over 500 research papers demonstrating that fish do, indeed, learn. In some aspects of learning, fish may even surpass abilities of rats and human infants (Sovrano *et al.* 2002).

Learning plays a critical role in the development of fish behaviours. I will not attempt to review all of this literature (it requires at least one issue of a journal!); instead, I will simply highlight the different fields in which learning capacities of fish are being investigated.

1. Learning and memory are important for foraging. Learning can lead to improvements in prey search, capture and handling efficiency (Reiriz *et al.* 1998, Warburton 2003). For example, repeated experience can improve the efficiency of prey recognition, attack and manipulation by fish (Ware 1971, Colgan *et al.* 1986, Hughes & Croy 1993). Fish build spatial memories of patch profitabilities, which influences patch exploitation rates (DeVries *et al.* 1989, Mackintosh *et al.* 1971), and memories of distributions of competitors (Croy & Hugh 1991). Furthermore, the ability to forget memories may also be adaptive, as this may allow individuals to track temporal variation in food resources (e.g. Mackney & Hughes 1995, Warburton *et al.* 1998).

2. Fish learn their environmental surroundings and landmarks to allow orientation and navigation (e.g. Rodriguez *et al.* 1994, Lopez *et al.* 1999, Sovrano *et al.* 2002). Spatial information used to form map-like representations of the environment is processed in a sub-structure of the brain (Broglio *et al.* 2003). A well-known

example would be the ability of juvenile salmon to 'imprint on' (memorize) the chemical cues specific to their home stream, which are likely to be chemicals derived from the local soil and vegetation (Hasler 1966; Hasler *et al.* 1978; Kieffer & Colgan 1992). This memory enables them to return to the same place as an adult (Stewart *et al.* 2004).

3. Memory capacity enables fish to distinguish between individuals based on previous experience (Griffiths 2003). This may influence who they associate with; for example, recognition of conspecifics that cooperated in predator inspection visits in the past may lead individuals to associate with them in the future (Milinski *et al.* 1990a,b). Individual recognition is important for shoal formations and the transfer of information between individuals in a shoal (Krause *et al.* 2000). For example, it may facilitate the social learning of behaviours (where individuals acquire new behaviours or information regarding their environment via observation of, or interaction with, other individuals).

4. Learning is important for predator recognition and anti-predator responses. Despite strong innate anti-predator skills in many fish species (Kats & Dill 1998), there is evidence that many aspects in the anti-predator repertoire of fish are learned (Kieffer & Colgan 1992, Kelley & Magurran 2003). Learning may allow fish to enhance and adjust their unlearned anti-predator dispositions (Magurran 1990, Kelley & Magurran 2003), for example, through incorporating information concerning local and temporal predation risk.

(III) APPLICATION OF THE STUDY OF FISH COGNITION

(i) Hatchery-reared salmonids

The study of cognitive abilities of fish has application in the preparation of fish reared in hatcheries for release into the wild. Salmonids are reared in culture facilities on a massive scale worldwide. In some cases the life cycle is completed within the hatchery. However, more usually, fish are stocked out into the natural environment to complete some part of their life cycle. The stocking of salmon into the wild is done for two broad purposes: either to facilitate the self-sustaining capacity of the population in the wild or to enhance natural production of salmon above carrying capacity by artificial means (Aprahamian *et al.* 2003). A problem facing both objectives is that in some circumstances hatchery-reared salmonids may have poor survival, relative to wild fish from the same population (Felton *et al.* 1990, Shively *et al.* 1996, Brown & Laland 2001, Jonsson *et al.* 2003), although relative survival may be strongly influenced by age/development stage at release and density of stocking.

There are two underlying causes of this. Firstly, breeding from individuals that have been held in a hatchery environment (hatchery-bred or 'domesticated' fish) can lead to genetic divergence from the founding population (Leider *et al.* 1990, Petersson *et al.* 1996, Einum & Fleming 1997, Fleming & Einum 1997), such that the hatchery genotype may not be suitably adapted for survival in the wild. Secondly, phenotypic divergence can occur between wild and hatchery fish within a generation (Weber & Fausch 2003). This may result from the influence of environmental conditions on ontogeny and the potential to acquire learned traits (Fleming *et al.* 1997, Olla *et al.* 1998, Einum & Fleming 2000). Furthermore, the high mortality suffered by juvenile salmonids in the wild (Elliot 1989a, Brännäs 1995, Einum & Fleming 2000) is largely bypassed in a hatchery environment (Swain *et al.* 1991, Fleming *et al.* 1994, Dellefors & Johnsson 1995), such that hatchery fish have a range of phenotypes reflecting low – high fitness in the wild.

Behavioural traits that are strongly dependent on cognitive processes may diverge between salmon that are reared in a hatchery for a period extending past endogenous feeding and those that are reared in the wild. For example, hatchery-reared salmon fry are often poorer competitors than wild-reared fish, and so are less successful in obtaining territories (Metcalfe *et al.* 2003). This implies reduced access to quality feeding patches and shelter. The reduced foraging skills (Olla *et al.* 1998, Sundström & Johnsson 2001), combined with the higher metabolic rates of hatchery-reared compared to wild fish

(Herbert *et al.* 2001) may make them more vulnerable to starvation (Henderson & Letcher 2003). Hatchery-reared salmon fry also have poorer swimming abilities (McDonald *et al.* 1998), making them more vulnerable to predators. Compounding this is that hatchery-reared fish are less risk-averse (Dellefors & Johnsson 1995, Fernö & Järvi 1998), may have reduced camouflage (Donnelly & Whoriskey 1991) and poorer predator recognition and anti-predator skills than their wild counterparts (Järvi & Uglem 1993, Alvarez & Nicieza 2003, Berejikian *et al.* 2003, Hawkins *et al.* 2004a). For these reasons, salmonids reared in the hatchery for an extended period may not be suitable for stocking if the goal is to enhance existing wild populations (e.g. Aprahamian *et al.* 2003).

(ii) Atlantic salmon, *Salmo salar* L.

The Atlantic salmon, *Salmo salar* L., is an anadromous species which spawns in freshwater, although some members of the population will complete their entire life cycle in freshwater. The developmental stages of the Atlantic salmon are described below and in Fig. 2. The terminologies of the developmental stages are those provided by Allan & Ritter (1977). Because many life history processes are dependent on temperature and/or day length, the timings of ontogenetic shifts are given for Scottish salmon.

Atlantic salmon spawn in the gravel beds of rivers between November and January in Scotland (Mills 1971). Eggs are buried in a series of nests in the gravel, which, when adjacent, form a redd (Webb & Hawkins 1989, Baglinière *et al.* 1990). The eggs hatch from late February to early April, depending on temperature. At hatching, the fish - termed alevins - remain within the gravel bed and feed on their yolk sac for 4 - 8 weeks (M. Miles, pers comm).. The young fish then emerge from the gravel and start feeding exogenously. During the transition period between endogenous and exogenous feeding the fish are called fry. Once yolk sac absorption is complete the fish are called parr.

Parr typically remain in the river for 1 - 3 years. Some parr, particularly males, become mature during this stage and are termed mature, or precocious, parr. The anadaromous individuals undergo a complex of morphological, physiological and behavioural changes

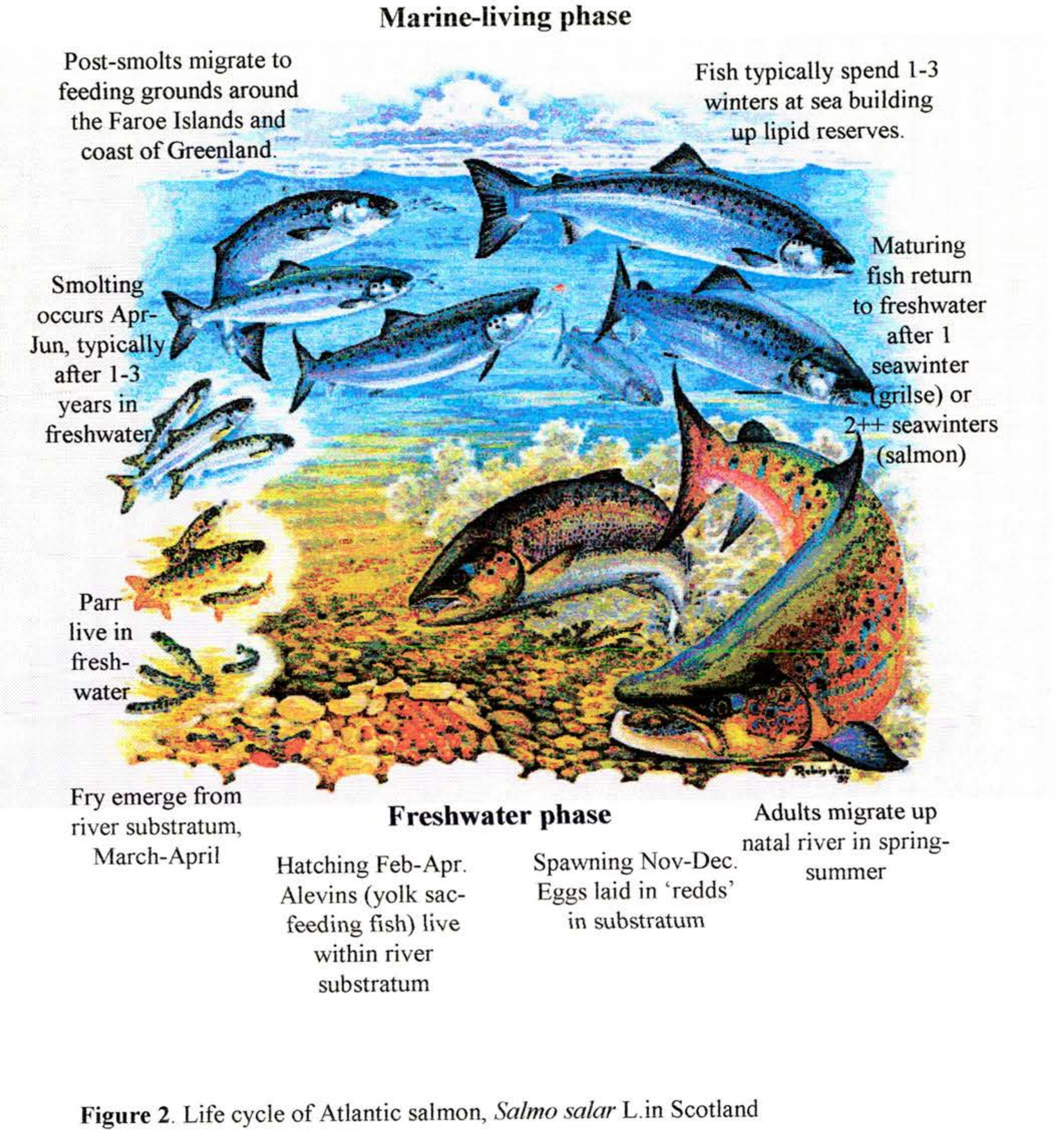

Figure 2. Life cycle of Atlantic salmon, *Salmo salar* L.in Scotland

Picture courtesy of Robin Ade

associated with moving from a freshwater to a marine environment (Hoar 1976, McCormick *et al.* 1998). This is smolt metamorphosis, and fish undergoing this process are termed smolts. Metamorphosis occurs during the spring (April - June) and is triggered by photoperiod and temperature (McCormick *et al.* 1998); however, the determination of whether an individual will smolt or not occurs the previous late-summer to autumn (Thorpe *et al.* 1980).

Smolts that survive the downstream migration and enter the estuary are termed post-smolts. These fish migrate thousands of kilometers in the ocean to feeding grounds off of the Faroe Islands and the coast of Greenland. The marine phase of the life cycle allows rapid growth, and many fish will return to estuaries after having built up sufficient lipid reserves during only one winter at sea. These maturing one-sea winter fish return to their natal river during spring and summer are called grilse. Fish that return after spending more than one winter at sea are called salmon. Mature parr may also smolt and then migrate to the marine environment before spawning again.

The early critical period

After emergence the fry may disperse and are highly territorial (Kalleberg 1958). The transition from endogenous to exogenous feeding has been termed the critical period (Hjort 1914, Elliott 1989a,b) as mortality may be high (Fleming 1996, 1998, Elliott 1994) due to competition for resources (Nislow *et al.* 2004) and acute vulnerability to fish predators (Garcia de Leanniz *et al.* 2000). Mortality during this critical period may be highly density-dependent (Elliott 1989a,b, Einum & Nislow 2005) and may constitute a population bottleneck (Shuter 1990). However, in other situations the critical period may be much more prolonged (e.g. Egglishaw & Shackley 1977, Gardiner & Shackley 1991). Alternatively, population size may be determined by the habitat carrying capacity and regulated by self-thinning (Grant & Kramer 1990) and this may extend over the first summer of life (Steingrímsson & Grant 1999). High over-winter mortality also appears to be common (Armstrong 1997) and would be expected to bring populations below carrying capacity in spring unless there is compensatory immigration. Thus there is evidence that both early critical periods and self-thinning influences survival in different locations.

(iii) Stocking of Atlantic salmon in the UK

In the UK, there are special cases when Atlantic salmon are stocked into rivers as part of mitigation and rehabilitation measures; for example, to compensate for loss of habitat due to hydro-electric schemes. The goal of this stocking is to enhance the locally existing wild salmon population, and typically, salmon are stocked as fry before exogenous feeding begins. Such stocking can be highly effective (e.g. Egglishaw & Shackley 1973, Jones 1994, Crozier *et al.* 1997, Anon. 2003).

The case for stocking of later developmental stages of Atlantic salmon in the UK could be argued, under certain circumstances, to avoid periods of high mortality in the wild. However, it should be considered that by-passing periods of high mortality may reduce selection for traits that will benefit subsequent generations.

The first instance where stocking might be appropriate is at the parr stage, early in the first year of life. Hatchery-rearing has the potential to by-pass egg and early-fry mortality; for example, 85 - 98% mortality occurs in the wild between egg fertilization and the first summer (Jessop 1984, Einum & Fleming 2000). Predation of these young fish in the wild could be responsible for up to 48.6% of total mortality in the first year (Henderson & Letcher 2003). However, as discussed above, the occurrence of a bottleneck at this early life period may not always be the case (Armstrong 1997), and requirement for such stocking may be limited.

The second possibility is stocking at the smolt stage to avoid mortality from piscivorous birds and fish. Such mortality may exceed 50% (Larsson 1985, Kennedy & Greer 1988, Jepsen *et al.* 1998, 2000). There may also be special cases where stocking of smolts is required because freshwater rearing habitat is limiting, such as is common in Baltic countries.

However, the effect of rearing salmon in the hatchery until later life stages can actually be poorer survival, compared to wild fish (see Table 1). Any advantages of bypassing periods of mortality in the wild may be offset by a reduction in fitness of hatchery-reared compared to wild-reared fish. There is some evidence that the later in ontogeny that fish are released into the wild, the more divergent their behaviours are from wild-reared fish, and generally the lower their survival rate (Table 1). However, the low survival rates of the stocked fish do not necessarily reflect poor quality of the fish. This is because their survival in the wild will also depend on the number of fish stocked relative to the carrying capacity of the river, which is further influenced by the numbers of wild fish already present in the river. Furthermore, stocking densities vary between studies, and there may be strong density-dependent mortality after stocking (Crisp 1995). Management that simply entails 'dumping' huge quantities of fish into a convenient access point of the river will have much lower survival rates than when fish are stocked across a larger area at lower densities.

Table 1. Survival rates mean (range) for Atlantic salmon (From Aprahamian *et al.* 2003, with additions).

Life stage	Wild-reared fish		Hatchery-reared fish	
	Mean (range) percentage survival in the wild	Reference	Mean (range) percentage survival in the wild	Reference
Survival from fertilized egg to:				
Eyed egg			89 (85-95)	MacKenzie & Moring (1988)
Hatching			31 (7-61)	Pauwels & Haines (1994)
			74 (45-88)	MacKenzie & Moring (1988)
Summer 0+	2 (1-3)	Jessop (1984)	4	Kennedy & Strange (1981)
	7 (5-8)	Jessop (1984)		
	7 (6-8)	Elson (1957)		
	8	Elson (1957)		
	10 (9-11)	Meister (1962)		
	12 (7-20)	Jessop (1984)		
	15 (11-18)	Jessop (1984)		
Summer 1+	5 (4-6)	Elson (1957)		
2+ smolt	3 (1-6)	Symons (1979)		
Survival from eyed egg to:				
Emergence	6	Gustafson-Marjanen & Moring (1984)	5 (3-7)	Gustafson-Marjanen & Moring (1984)
Summer 0+			19	Kennedy & Strange (1981)
Autumn 0+			13 (12-14)	Egglishaw & Shackley (1973)
			- (17-19)	Kennedy & Strange (1981, 1986)
Survival from alevins to:				
Autumn 0+			8 (1-30)	Mills (1969)
			20 (9-30)	Egglishaw & Shackley (1973)
			26 (18-34)	Orciari *et al.* (1994)
			42 (5-53)	McMenemy (1995)
Survival from summer age 0+ to:				
Autumn 0+			- (6.7-22.7)	Elson (1957)
			- (7.8-41.3)	Farooqi & Aprahamian (1995)

			- (10.0-42.3)	Milner (1993)
			13 (11-15)	McCrimmon (1954)
Summer 1+	17 (1-29)	Côté & Pomerleau (1985)	- (4-33)	Elson (1957)
	- (14-32)	Kennedy & Strange (1981)		
	47	Kennedy & Strange (1981)		
	50 (41-59)	Meister (1962)		
	51 (22-88)	Egglishaw & Shackley (1980)		
	53 (45-68)	Orciari *et al.* (1994)		
2+ smolt	20	Elson (1957)	- (3-12)	Elson (1957)
	38	Elson (1975)	8	Elson (1975)
Survival from summer age 1+ to:				
2+ smolt	19 (14-23)	Orciari *et al.* (1994)	- (2-30)	Elson (1957)
	45 (14-79)	McMenemy (1995)	26.2 (14.7-43)	Jokokokko & Jutila (1998)
Survival from 1+ smolt to:				
Adult (1 seawinter)			1 (0.1-2.9)	Crozier & Kennedy (1993)
Survival from 2+ smolt to:				
Adult (1 seawinter)	0.7 (0.5-2)	Chadwick (1986)	0.05 (0.01-0.1)	Baum (1983)
	6 (1-12)	Chadwick (1986)	1 (0.3-3)	Harvie & Amoro (1996)
	6 (1-17)	Jessop (1984)	1 (0.4-4)	Harvie & Amoro (1996)
	7 (3-12)	Chadwick (1986)	2 (0.4-5)	Harvie & Amoro (1996)
	8.2 (3.9-12.0)	Crozier & Kennedy (1993)	2.3 (0.6-8.2)	Crozier & Kennedy (1993)
			4 (1-6)	Isaksson (1979)
Adult (2 seawinter)	0.6 (0.3-0.9)	Chadwick (1986)	0.1 (0-0.7)	Crozier & Kennedy (1993)
	0.6 (0.4-1)	Chadwick (1986)	0.1 (0.03-0.2)	Harvie & Amoro (1996)
	1.1 (0.4-1.9)	Crozier & Kennedy (1993)	0.2 (0-0.5)	Isaksson (1979)
			0.5 (0.1-1.5)	Harvie & Amoro (1996)
			0.7 (0.2-1.3)	Baum (1983)
			0.7 (0.2-3)	Harvie & Amoro (1996)
Adult (1-3 seawinter)	4	Piggins (1979)	- (0.004-0.36)	Meyers (1994)
	8 (4-13)	Piggins (1979)	0.07 (0.008-0.17)	Stolte (1994)
			0.45	Piggins (1979)
			2.4 (1-4)	Piggins (1979)
			2.7 (0.5-7)	Piggins (1979)
			2.3	Piggins (1979)

Survival from 1+ and 2+ smolts to:				
Adult (2 seawinter)			0.1 (0.0-0.7)	Crozier & Kennedy (1993)
Adult (1-2 seawinter)	- (2-6)	Shearer (1961)	- (0.2-2)	Shearer (1961)
	10 (2-16)	Carlin (1967)	- (0-11)	Carlin (1967)
	23 (20-26)	Österdahl (1969)	10 (6-13)	Österdahl (1969)
	21	Isaksson (1979)	1-2	Isaksson (1979)
			2	Isaksson (1979)
			8	Isaksson (1979)
			0.06 (0.001-0.16)	Rideout & Stolte (1988)
			0.035	Rideout & Stolte (1988)
			0.33	Rideout & Stolte (1988)
Adult (1-3 seawinter)	8.9	Jonsson *et al.* (1991)	4.6	Jonsson *et al.* (1991)
			0.3	Anon. 2003

Given the limitations of the survival data (Table 1) for comparing across studies, it is perhaps instead possible to compare survival rates of hatchery- and wild-reared fish within studies and generalize the overall trend. Table 1 does suggest that hatchery-reared smolts have lower fitness in the wild compared to wild-reared smolts. This may be due to environmental differences between rearing habitats – e.g. in feeding regimes, conspecific density, exposure to predators and interactions with conspecifics – such that phenotypic traits are selected in the hatchery which are not adaptive to living in the wild (Johnsson *et al.* 1996, Fernö & Järvi 1998, Aprahamian *et al.* 2003). Furthermore, a hatchery environment may not provide the opportunity for fish to develop and refine adaptive skills (Sundström & Johnsson 2001, Alvarez & Nicieza 2003). For example, although fish may have the capacity to perform anti-predator behaviour, it may not occur without specific experience (Griffin *et al.* 2000). Given these negative influences of hatchery rearing on survival, is it conceivable that fish could be stocked at later stages of development (as 0+ parr or smolts) to enhance population abundances?

(iv) Life skills training

The cognitive abilities of fish may provide an answer: the behaviours of hatchery fish that influence fitness in the wild environment could be enhanced or restored through learning (Suboski & Templeton 1989, Brown & Laland 2001). This has been termed 'life skills training' (Suboski & Templeton 1989). Such behaviours may pertain to foraging, predator recognition, predator avoidance and social interactions. For example, providing simple experiences through modifying the hatchery environment (e.g. by incorporating structure into rearing tanks, providing overhead cover or underwater feeding systems) may influence the subsequent behaviour of fish when released into the wild. Such modifications can affect the social hierarchy of hatchery salmon, such that in the wild their territory distribution is more similar to wild reared salmon, leading to increased carrying capacities of streams (Berejikian *et al.* 2001). More generally, introducing variability into the hatchery environment (e.g. water flow rate and direction) may increase hatchery fishes' behavioural repertoire to prepare them for living in the natural environment, possibly through the enhancement of brain development which facilitates cognition (Marchetti & Nevitt 2003, S. Winberg, pers comm). The area that has received most attention is the potential to enhance predator recognition abilities and anti-predator responses. In the hatchery environment, avoidance-conditioning of fish using predator cues and alarm cues (discussed later in this chapter) can increase alertness to predator cues and anti-predator behavioural responses and may increase survival in encounters with predators (Berejikian *et al.* 1999, 2003, Mirza & Chivers 2000, Vilhunen & Hirvonen 2003).

In practical terms, the potential to 'train' fish to avoid predators is probably the most simple to apply, out of the techniques outlined above, as it should not affect hatchery maintenance procedures. However, there have been only few tests of how this effects survival in the wild, and methodological problems exists with these studies (Thompson 1966, Kanayama 1968, Berejikian *et al.* 1999). What is still required is the demonstration that the enhancement of anti-predator responses in the hatchery environment can improve fitness in the natural environment.

(v) Under what circumstances would training be useful in the UK?

In Scotland, predation from pike (*Esox lucius* L.) on migrating salmon smolts may be ac~~c~~ute (Mills 1965). This may be especially so in water bodies that form part of hydro-electric schemes, as they provide habitat that is ideal for pike (Raat 1988). Pike predation on salmon is a controversial issue in Scotland, and indiscriminate culling of pike is a management method that may be "approved of by most [salmonid] anglers and riparian owners" (Mann 1980), and has been a key feature in trout fishery management for decades (Mann 1985). However, removing all pike from a system is virtually impossible. Furthermore, the occurrence of pike in Scotland could be beneficial, in terms of the economic impacts of attracting pike anglers to rural areas, and by offering an alternative fishery to the declining salmon populations. The creation of reservoirs for hydro-electric schemes, which is favourable in the current climate of producing green energy, means that a lot of suitable habitat for pike already exists.

Is it possible to reduce the loss of salmon to pike predation, and hence, is there a means to enhance the success of rehabilitation stocking of salmon? Are there alternatives to pike culling? To address such applied questions we must first answer fundamental questions regarding predator cognitive abilities in salmon, behaviour of pike, and the qualitative and quantitative nature of the interactions between these two species. Firstly, do salmon have an innate ability to recognize pike as a predator? This is a particularly interesting question as there is a debate whether pike is a native species to Scotland, and hence whether salmon would have evolved under the risk of pike predation. If no innate abilities exist, can salmon learn to recognize pike, or, if they are predisposed to recognize pike, can they enhance and refine existing abilities through learning processes? If salmon are capable of learning to recognize predators, can we stimulate these learnt abilities in a hatchery environment (i.e. life skills training), such that we can 'improve' the abilities of fish used in rehabilitation programmes? Does this translate into an increase in survival in the wild? Are there ontogenetic limitations on their capacity to learn, i.e. is there a time period when training fish to recognize predators would be most successful? Or if training is not successful, are there other approaches to reducing the impacts of pike predation, such as habitat modifications? What information would we need to evaluate this?

(IV) AIMS OF THIS THESIS

This thesis addresses some of the questions outlined above. Experiments investigate the innate and learnt predator recognition abilities of Atlantic salmon, and how their capacity to acquire predator recognition may influence their survival in a natural environment. Experiments have also established the habitat use of a predator, the pike, and the behavioural interactions between these two species. The objective was to investigate two alternative approaches to culling for limiting pike predation on salmon.

(1) In Sections II and III, the possibility of compensating for predator-recognition deficiencies in hatchery-reared fish by encouraging the development of learnt predator recognition abilities is evaluated (Suboski & Templeton 1989, Brown & Laland 2001). Chapters two and three investigate innate predator recognition abilities and data presented in chapter four compares anti-predator responses of hatchery- and wild-reared fish. Chapter five examines how innate abilities change during the course of hatchery-rearing and whether predator recognition skills can be learnt in a laboratory environment. Chapter six assesses how learnt skills can influence survival of hatchery-reared salmon in the wild.

(2) In Section IV, the habitat use (chapter seven) and distributions (chapter eight) of juvenile pike is assessed, to provide basic information on habitat requirements that is currently lacking. This information is required to evaluate the potential to reduce pike predation on salmon through habitat manipulation, by reducing encounter rates between the two species.

Section V concludes by discussing the factors that influence pike-salmon interactions (chapter nine). The remainder of chapter one discusses predator recognition in fish, capacities of fish to learn predator recognition and the application to enhancing anti-predator skills of hatchery-reared salmonids. The predator-prey interaction is then considered from the aspect of the predator and existing knowledge on pike distributions is described.

(V) PREDATOR RECOGNITION IN FISH

This research on the predator recognition abilities of salmon has primarily focused on recognition of olfactory cues. Olfactory recognition is particularly important in the aquatic environment (Kats & Dill 1998) and has been demonstrated in a diverse range of fish families, including characins, gouramies, gobies, cyprinids, cichlids, poeciliids, damselfishes, rainbowfishes and salmonids (reviewed by Kats & Dill 1998). Olfaction may be especially important for detecting ambush predators such as the pike, due to the lack of visual cues available to the prey.

A predator cue may elicit a variety of behavioural responses in prey fish, which may help them to evade the predator (Lima & Dill 1990, Kats & Dill 1998). Many species of salmonids have been shown to respond to predator odour cues alone (e.g. Rehnberg & Schreck 1987, Hirvonen *et al.* 2000, Vilhunen & Hirvonen 2003, Hawkins *et al.* 2004b). Salmonids may respond to cues with a variety of anti-predator behaviours (Table 2).

Table 2. Anti-predator behaviours of salmonids

Anti-predator behaviour	Predator cue eliciting response	Species	Reference
Area avoidance	Predator odour	Brook trout (*Salvelinus fontinalis*)	Keefe (1992)
	Predator exposure	Pink (*Oncorhynchus gorbuscha*) and chum (*O. keta*) salmon	Magnhagen (1988)
	Predator odour	Coho salmon (*O. kisutch*)	Rehnberg & Schreck (1987)
Swimming haphazardly ('dashing')	Predator odour	Arctic charr (*Salvelinus alpinus*)	Vilhunen & Hirvonen (2003)
Remaining still ('freezing')	Predator odour	Arctic charr	Vilhunen & Hirvonen (2003)
	Predator odour	Rainbow trout (*O. mykiss*)	Brown & Smith (1998)
Sheltering	Visual cue	Brown trout (*Salmo trutta*)	Alvarez & Nicieza (2003)
Reduction of activity	Predator model	Atlantic salmon (*Salmo salar*)	Metcalfe *et al.* (1987)
	Predator exposure (safe)	Coho salmon	Martel (1996)
Reduction of foraging	Predator exposure (safe)	Coho salmon	Martel (1996)
	Predator odour	Atlantic salmon	Gotceitas & Godin (1991)
Increase in school formation	Predator exposure	Atlantic salmon	Handeland *et al.* (1996)
Reduction in conspecifics interactions	Predator exposure (safe)	Coho salmon	Martel (1996)

The most widely studied of the olfactory cues used in predator recognition are kairomones, or predator odours, which are released continuously from a predator. The chemical components of kairomones likely derive from intestinal by-products (Courtenay *et al.* 1997), mucus (Williams & Moore 1985), faeces (Brown *et al.* 1996) and/or urine (Moore *et al.* 1994). Diet may have a strong influence on the composition of kairomones; for example, through the presence of enzymes related to a piscivorous diet (Fish 1960) or through the effect of diet on the predator's mucus (Saglio & Fauconneau 1985). Dietary cues may therefore indicate the risk of predation to the prey (Mirza & Chivers 2001a, 2003a).

The acute olfactory abilities of salmon are reflected in the specificity of their responses to predator odours. Salmonids assess dietary cues, and can distinguish between predators fed a piscivorous diet and those fed invertebrate diets (Keefe 1992), and even between predators feeding on conspecifics and heterospecifics (Mathis & Smith 1993, Brown 2003, Mirza & Chivers 2003a, Vilhunen & Hirvonen 2003). Can salmon distinguish between predatory species, regardless of diet? Are they capable of refined discriminatory powers of predator cues? In chapters two and three of this thesis experiments are described that test the innate abilities of Atlantic salmon to recognize predator odours, and their abilities to distinguish between odours from different species and of different concentrations (the latter potentially reflecting risk of predation).

A second means of olfactory predator recognition utilizes a chemical alarm signalling system. Many species of fish, including salmonids (Chivers & Smith 1998, Mirza & Chivers 2001b), have specialized epidermal cells that contain damage-released chemical alarm cues, or alarm substance (AS; Smith 1992). AS is only released when these cells are damaged, such as when predator's teeth puncture the skin. AS invokes an innate fright responses in conspecifics (and some heterospecifics) who detect it (Von Frish 1941). Responses to AS have been documented in most aquatic taxa (Pfeiffer 1974, Howe & Sheikh 1975, Sleeper *et al.* 1980, Lawrence 1991, Purrington *et al.* 1991).

The detection of AS in conjunction with predator odour is a strong signal to alert conspecifics to the risk of predation (Brown 2003). Furthermore, a novel predator can be identified as dangerous if its odour is associated with AS (Mirza & Chivers 2003a), as a predator's dietary odour cue can be 'labelled' with AS (Wisenden 2001).

The recognition of unknown predator odour through its association with AS exemplifies a means of learning predator identification, through releaser-induced recognition learning (Suboski 1990). More generally, this type of learning is stimulated by exposing a fish to a neutral cue combined with an aversive cue (i.e. one that elicits an innate fright response), resulting in the fish demonstrating a learned aversion to the neutral cue. Depending on the neutral stimuli, this can lead to an acquired aversion to predator odour cues (Brown & Smith 1998, Berejikian *et al.* 1999, Mirza & Chivers 2000), predator visual stimuli (Chivers & Smith 1994a, Brown *et al.* 2001a), habitat patches (Chivers & Smith 1995), and even non-threatening stimuli, such as cues of non-predatory species (Chivers & Smith 1994b, Magurran 1989) or light (Moreira & Volpato 2004). Releaser-induced recognition learning of predators has been demonstrated in salmonids (Brown & Smith 1998, Berejikian *et al.* 1999, 2003, Mirza & Chivers 2000).

It has been suggested that the absence of predators in the hatchery environment may deny hatchery-reared salmonids the opportunity to develop existing, or acquire learnt, anti-predator skills, through mechanisms such as releaser-induced recognition learning (Kieffer & Colgan 1992, Olla *et al.* 1992, Dellefors & Johnsson 1995). Furthermore, in an environment where no anti-predator skills are required, existing responses may be lost over ontogenetic time (Griffin *et al.* 2000, Mirza & Chivers 2000, Brown & Laland 2001). This may explain the reduced predator recognition and anti-predator skills of hatchery-reared compared to wild-reared fish (p. 7). The effect of rearing environment on predator recognition is tested in chapter four by comparing anti-predator responses of wild-progeny salmon that have been reared either in the wild or in the hatchery.

However, it may be possible that hatchery-reared salmonids can learn predator recognition (Wiley *et al.* 1993, Brown 2003). In support of this, there is evidence that experience with predators can improve anti-predator responses in salmonids (e.g. Kieffer & Colgan 1992, Järvi & Uglem 1993, Berejikian 1995, Healy & Reinhardt 1995), suggesting that the phenotypic plasticity to learn is not lost. Furthermore, salmonids can also acquire anti-predator skills through releaser-induced recognition learning (e.g. Brown & Smith 1998, Berejikian *et al.* 1999, 2003, Mirza & Chivers 2000). Generally, however, the success of these learning techniques in a laboratory environment has not been replicated in the field. Few studies have looked at how anti-predator training techniques can influence survival of fish in a wild environment, and those that have (Thompson 1966, Kanayama 1968, Berejikian *et al.* 1999) have found only limited success.

Chapter five of this thesis tests how rearing fish in the hatchery to different developmental stages affects innate predator recognition skills. The capacity for fish to learn is corresponding tested, to determine whether releaser-induced recognition learning can enhance existing skills. Chapter six reports on a field-based experiment that tested how the same methods used to stimulate learning in the hatchery could influence survival of hatchery-reared salmon smolts stocked into the wild.

(VI) PIKE DISTRIBUTIONS

In order to understand the impact on prey, it is necessary to learn about the behaviour of the predator, and the circumstances under which predator and prey interact. The northern pike, *Esox lucius* L., is a top predator of northern temperate and boreal aquatic systems. It is distributed throughout most of Britain but is absent from much of northern Scotland (Davies *et al.* 2004), and may have been introduced to Scotland from S.E. England (Wheeler *et al.* 1977, Davies *et al.* 2004). Pike are solitary, sit-and-wait predators (Diana *et al.* 1977, Eklöv 1992, Eklöv & Diehl 1994), although they are reported to move in search of fish schools, and adults may even follow prey fish (Grimm & Klinge 1996). Their ambush hunting strategy (Savino & Stein 1989a,b) relies primarily on visual cues (Raat 1988, Casselman 1996).

The primary habitat of pike is slow-moving or still water bodies with abundant vegetation (Grimm 1981, Raat 1988, Greenberg *et al.* 1995, Eklöv 1997, Jepsen *et al.* 2001), although they also occupy rivers in which vegetation or backwaters can provide areas of low water velocity (Mann 1976, Paragamian 1976, Penczak *et al.* 1976, Raat 1988). Spawning occurs in the UK between February and May in littoral areas (Frost & Kipling 1967, Bregazzi & Kennedy 1980). Vegetation is essential for spawning and nursery areas as it provides structure for eggs and yolk sac-feeding fry to adhere to (Wright 1990).

Adult pike frequently occur in vegetated habitats (Raat 1988, Casselman & Lewis 1996), which may be due to prey distributions (Cook & Bergersen 1988, Chapman & Mackay 1990, Jepsen *et al.* 2001). However, adult pike are not restricted to vegetated habitats outside of spawning time, and also utilize open water (Diana 1980), even extensively so during the winter when less vegetation is available (Diana *et al.* 1977, Cook & Bergersen 1988). Water turbidity may also influence habitat preference. Chapman & Mackay (1984), Vøllestad *et al.* (1986) and Cook & Bergersen (1988) all found that pike used open water more when water turbidity increased. It is likely that turbidity reduces visual contact with prey (Skov *et al.* 2002) and possibly (conspecific) predators.

Tracking studies have provided a lot of information concerning habitat use of adult pike; however, there is only scant information on the habitat use of juveniles (< 2 - 3 years old), probably as a result of the difficulties of sampling juveniles. Juvenile life stages are thought to be strongly associated with vegetated littoral areas (Grimm 1981, Raat 1988, Eklöv & Hamrin 1989, Savino & Stein 1989b, Bry 1996, Casselman & Lewis 1996, Eklöv 1997), and young-of-the-year (YOY) pike may be restricted to dense emergent and submerged vegetation in their first summer (Grimm 1981). This may be due to the distribution of their prey (Eklöv 1997) or the requirement for structural protection from predators (Holland & Huston 1984, Eklöv 1997, Skov & Berg 1999, Skov *et al.* 2002). These influences are discussed further in chapter seven. Similar to adults, water turbidity may influence habitat use (Skov *et al.* 2002), perhaps through its influence on encounters with predators and/or prey.

Whilst aquatic vegetation is important for pike habitat, it is less clear how vegetation density determines habitat suitability. Vegetation density can influence visual contact with predators (including conspecifics) and prey. However, the limited information on the effects of vegetation density is conflicting. Casselman & Lewis (1996) suggest that optimal vegetation cover is 35 - 80%. Yet Savino & Stein (1989b) found that hunting success did not change with vegetation density, and Eklov & Hamrin (1989) report a negative correlation between vegetation density and hunting success in a field enclosure experiment. Clearly, other factors must influence habitat suitability. For example, water cloudiness may affect foraging success. Skov *et al.* (2002) found that structured habitat was preferred by pike only under clear-water conditions. It was suggested that in clear water conditions, pike require habitat structures for crypsis to enable successful ambush of prey, but in turbid water additional cover is not necessary (Skov *et al.* 2002).

On a macro-scale, juvenile pike may select vegetation densities inversely related to body size (Eklöv 1997). However, field studies of juveniles are based on broad-scale sampling techniques such as electrofishing. Such sampling methods cannot deal with patchiness of vegetation and the microhabitat of juveniles (e.g. individuals may locate on vegetation/bare habitat boundaries). Semi-natural and enclosure experiments suggest that juveniles (age 0+) are not restricted to vegetation patches (Eklöv & Diehl 1994, Hawkins *et al.* 2003). Correlations between juvenile pike numbers and vegetation cover may thus not be so straightforward.

The paucity of information concerning microhabitat requirements is addressed experimentally in chapter seven. Conspecific interactions may also play a role in the habitat use of juvenile pike; however, the mechanisms through which they occur are not clear. Although cannibalism is a well-known interaction that could strongly influence individual's habitat use (Eklöv 1997), there is behavioural evidence that interactions between juvenile pike do not routinely end in cannibalism or overt aggression (Eklöv & Diehl 1994, Nilsson *et al.* 2000, Hawkins *et al.* 2003). Therefore the influence of social interactions on distributions was experimentally tested (chapter eight).

Overlap between salmon and pike distributions

The habitat requirements of pike largely restrict interactions between pike and salmon in Scotland to lochs and reservoirs, as well as the rivers and streams up- and down-stream of lochs. Therefore interactions between pike and salmon are mainly limited to the smolt migration period when smolts traverse through lochs. Smolt migration may coincide with the post-spawning feeding period of pike (Jepsen *et al.* 1998, 2000), thus potential predation on smolts could be very high. Atlantic salmon parr may also undertake migrations into lakes in the spring and autumn (Hutchings 1986, Ryan 1986, Matthews *et al.* 1997). Parr may remain in lakes until smoltification or maturation (Hutchings 1986, Matthews *et al.* 1997), ~~such~~ therefore there is the potential for pike-salmon parr interactions as well (Mills 1965).

What are the predation impacts of pike on salmon in Scotland? The information that is available to address this question, and areas where information is still required, is concluded in the general discussion (chapter nine). The potential for reducing predation impacts on salmon through (1) life skills training to enhance predator recognition of salmon and (2) manipulation of habitat to reduce pike-salmon interactions will be discussed.

SECTION II

CHAPTER TWO

Innate predator recognition in newly-hatched Atlantic salmon[*]

ABSTRACT

It is well established that fish can learn to associate odours from potential predators with risk and alter their behaviour accordingly. However, newly-hatched individuals have few opportunities for acquired predator recognition and may depend on unlearnt (innate) responses. This study considered (1) whether newly hatched Atlantic salmon (alevins) exhibit innate predator recognition and (2) whether techniques to stimulate releaser-induced recognition learning could enhance responses to predator cues. This investigation showed that the response to predator (pike) odour was not affected by previous exposure to pike odour and conspecific tissue extract but was consistent with innate recognition of pike as predators. Trials conducted using odour from a non-piscivorous species confirmed that the fish were not simply reacting to a novel stimulus.

INTRODUCTION

The ability to assess local predation risk accurately is crucial if animals are to avoid attack and balance predator evasion with other activities such as foraging (Helfman 1989, Mathis

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& Vincent 2000, Chivers *et al.* 2001, Engström-Öst & Lehtiniemi 2004). Prey species belonging to many taxa, including birds and mammals (Owings & Coss 1977, Curio *et al.* 1983), amphibians (Sih & Kats 1994, Griffiths *et al.* 1998), reptiles (VanDamme & Castilla 1996) and crustaceans (Wahle 1992), show an innate ability to recognise predator odour cues. However, other studies contrastingly find that prey may not respond to predators on their first encounter, and instead experience is required for prey to give appropriate anti-predator responses in future encounters (e.g. Herzog & Hopf 1984, Suboski 1990, Owings *et al.* 2001).

Fish are capable of assessing subtle differences in predation risk (Laland *et al.* 2003). There are two important categories of chemical cues that are implicated in predator detection by fish. First, there are the 'damage-released alarm cues', sometimes referred to as 'alarm substance' or 'Schreckstoff', that are liberated when the skin of a prey fish has been abraded during predator attack (von Frisch 1941). Adaptive responses to conspecific alarm substance have been documented in a wide variety of taxa including ostariophysans, salmonids, gobies, poeciliids, sticklebacks, percids and cichlids (Pfeiffer 1977, Chivers & Smith 1998). These responses are generally regarded as innate (Göz 1941, Pfeiffer 1962). The second type of cues, are 'predator odours'. In some cases (Hirvonen *et al.* 2000, Berejikian *et al.* 2003, Vilhunen & Hirvonen 2003) fish may recognise the scent of a predator the first time they detect it, but in others (Chivers & Smith 1994a, Chivers *et al.* 1995a, Mathis *et al.* 1996, Brown & Smith 1998, Mirza & Chivers 2000) recognition is acquired through learning. Learning typically occurs when a novel predator odour is paired with an aversive stimulus, such as conspecific alarm substance (see Brown (2003) for a review). Learnt responses may build upon and refine existing abilities (Chivers & Smith 1994a, Wisenden 2000, Brown 2003, Kelley & Magurran 2003).

The early-life stages of many animals, including fish, are extremely vulnerable to predation (Fuiman & Magurran 1994, Sih & Kats 1994). In such cases, innate predator recognition can be expected to be selected for. Salamander larvae, for example, have been shown to respond to predators within 18 h of hatching (Sih & Kats 1994). This is adaptive since larvae drift between pools after hatching, and individuals may soon drift into detection range of a predator.

Predator recognition capabilities in extremely young individuals are especially acute in the case of salmon, which face a critical risk period when they emerge from their nests in the protective river substratum to feed, once yolk sac absorption is completed. Emergence is a critical period when predation mortality is great (Elliott 1989a,b), and predation risk is influenced by timing of emergence (Brännäs 1995, Einum & Fleming 2000). Emergence carries a large risk but, equally, delaying emergence reduces an individual's ability to compete for food and feeding territory. In view of this early-life predation risk, it seems reasonable to hypothesize that salmon have an innate response to predators. In support of this hypothesis, Berejikian *et al.* (2003) demonstrated a degree of innate predator recognition in free-swimming Chinook salmon, *Oncorhynchus tshawytscha*, fry two months after emergence. However, they used domesticated hatchery-reared salmon, in which anti-predator responses may have been selected against. Their work raises the intriguing possibility that predator recognition may be even more pronounced in wild fry and poses the question of whether innate predator recognition occurs at earlier stages of development.

Newly-emergent salmon may not rely entirely on innate recognition. Courtenay *et al.* (1997) have shown that coho salmon, *Oncorhynchus kisutch,* can learn conspecific odours between 0 - 14 d after yolk sac absorption, and that this learnt recognition is retained for over two months. Clearly, by this stage the fish have developed an olfactory sensory system sufficient to detect predator chemical cues. In fact, there is evidence that the olfactory apparatus is essentially complete upon hatching in the rainbow trout, *Oncorhynchus mykiss* (Hara & Zielinski 1989), and within 7 d of hatching in cutthroat trout, *Oncorhynchus clarki* (Jahn 1972). These studies suggest that newly-hatched salmonids are capable of detecting predator odour cues.

This study tested whether newly-hatched (yolk sac feeding) Atlantic salmon (27-40 d old) of wild parentage, responded to odour of pike, a specialist piscivore, relative to odour of an omnivorous fish and a control. Fish were pre-exposed to pike odour combined with conspecific salmon tissue, either before or immediately after hatching, to encourage learnt responses to pike odour through releaser-induced recognition learning (Suboski 1990). It is uncertain whether salmon eggs are sufficiently developed to detect odours; however, it is known that sensory neurones develop early in the egg phase, following which neural activity is generated in response to amino acids (Jahn 1972, Hara & Zielinski 1989). Therefore, there may be potential for response to predator odour even at the pre-hatch stage.

A potential tool for evaluating predator recognition abilities in fish is the measurement of ventilation (opercular) rate. It is well known that ventilation rates of fish respond to factors that raise metabolic rate, such as temperature (Burton 1979) and handling (Laitinen & Valtonen 1994). Pfeiffer (1962) reported that ventilation rates increased in response to alarm substance and Metcalfe *et al.* (1987) observed an increase in opercular rate of hatchery-reared Atlantic salmon in response to a predator model. Recently, Barreto *et al.* (2003) demonstrated hyperventilation in tilapia, *Oreochromis niloticus*, exposed to a visual predator cue, and Sneddon *et al.* (2003a) have shown that rainbow trout, *Oncorhynchus mykiss*, increase respiration rate as part of a fear response to a novel object. This study uses the opercular response of Atlantic salmon as a measure of their recognition of predator odours.

METHODS

Atlantic salmon eggs were stripped from females obtained from the River Almond, Perthshire, Scotland (56º43'N - 3º53'W), in November 2002 and reared at the Fisheries Research Services Almondbank field station.

Prior exposure

Salmon were exposed to an aversive cue and predator odour simultaneously prior to testing responses to predator odour alone. This prior exposure method can lead to acquired predator recognition through encouraging a learnt association between the two cues (releaser-induced recognition learning; Suboski 1990). Conspecific tissue extract was used for the aversive stimulus to encourage an association between predator odour and injured conspecifics. Prior exposure was given to either eggs (2-3 months post-fertilization; between 27 and 1 d prior to hatching) or newly-hatched alevins (3-17 d old), and a third group were left unexposed (control exposure). These three groups are here on collectively referred to as exposure groups.

Predator odour

Predator odour was obtained from one stock tank (120 x 38 x 38 cm, 173 l) that contained 22 juvenile pike (*c.* 14 cm L_T). No pike have been recorded in the R. Almond, from which the hatchery water is drawn; the control alevins were thus naïve to pike odour. Pike were fed a diet of dead Atlantic salmon parr, but a week prior to, and during the experiments, diet was limited to earthworms to prevent salmon cues from being incorporated in pike odour. In juvenile pike, gut evacuation after a meal takes 5 d at a temperature of 5 °C (Bevelhimer *et al.* 1985). During our experiment the minimum temperature was 5 °C; because gut evacuation time decreases with increasing temperature, 7 d would be sufficient for residual salmon cues to be eliminated from pike dietary odour. The pike holding tank was supplied with filtered water from the R. Almond. A total water change was carried out three times per week to flush any residual salmon odour cues from the system. A 20 ml sample of water from the pike holding tank provided one dose of predator odour (PO).

Conspecific tissue extract

Salmonid skin contains alarm substance, and when released into the water (when the skin is broken), conspecifics that detect it show an innate fright response (Mirza & Chivers

2001). Therefore an extract of the skin of Atlantic salmon, termed salmon skin extract (SSE), was used as the aversive stimulus. SSE was obtained by removing the skin from 12 humanely-killed Atlantic salmon (*c.* 8 cm L_T), then homogenizing the skin with 310 ml distilled water, followed by filtering through wool and then filter paper. This solution was separated into 5 ml containers (single doses) and frozen at -20°C until required.

A second cue was prepared as an aversive stimulus, termed salmon egg extract (SEE). SEE was obtained by homogenizing 81 Atlantic salmon eggs with 310 ml of distilled water. The solution was then filtered, centrifuged (3000 rpm for 30 min) and frozen in 5 ml containers until required. Exposure was one dose of each odour per day, supplied simultaneously to all individuals in the exposure treatments.

(i) Is salmon skin extract (SSE) an aversive stimulus?

An initial experiment was undertaken to test whether SSE did indeed invoke an innate fright response in yearling Atlantic salmon (parr). Individual hatchery-reared salmon (L_F 79.5 ± 5 mm; mean ± 95% CI) were placed into aquaria (0.9 x 0.4 x 0.35 m) containing a layer of large gravel and a single shelter (a 20 x 15 cm board raised 4 cm above the gravel by a single support). Fish were given 2 d to settle into this environment, during which time they were fed on bloodworm. The opercular response of the fish to either 15 ml of SSE (N = 18) or 15 ml distilled water (DW; N = 10) was then tested. Opercular rate was measured for at least three 30-s periods to obtain a stabilized opercular rate (pre-stimulus rate). The stimulus (SSE or DW) was then injected into the tank, and after allowing 20 s for it to circulate, four 30-s counts of opercular rate were made (post-stimulus rate).

(ii) The effect of prior exposure on the response to predator odour

Salmon eggs were raised in 15 boxes, each containing 100 eggs, under standard hatchery conditions. Boxes were assigned equally to the three exposure groups: EG, which was exposed to PO + SSE + SEE during the egg stage; HA, which was exposed to PO + SSE + SEE at the newly-hatched stage; and CO, a control exposure group that received no exposure at any development stage. PO, SSE and SEE were presented simultaneously. Every day, for 27 d before hatching, and for 14 d after hatching, egg boxes were placed simultaneously, for 1 h, into one of three identical tanks (0.4 x 0.3 x 0.3 m), supplied with a constant flow of river water. One dose each of PO, SEE and SSE was placed in a separate tank filled with 12 l of river water. This solution was administered to the tank containing the treatment group (EG or HA) at the rate of 12 l h^{-1}. Afterwards, the boxes were thoroughly flushed with river water to avoid transmitting odours to the other groups in the hatchery.

Stimulus odours

At 27 - 40 d post-hatching, individuals (feeding on yolk sac; size *c.* 20mm L_T) in each of the three exposure groups were tested with one of four stimulus odours. These comprised (1) the river water supplied to the hatchery (control), (2) water from a different river basin (R. Earn; novel water), and two fish species odours – a sample of water obtained from holding tanks of either (3) roach, *Rutilus rutilus,* or (4) pike.

Pike odour was obtained in the same manner as described for the exposure phase. Roach odour was obtained from two holding tanks (90 x 38 x 45 cm, 154 l) each containing five roach, *c.* 12-14 cm L_T. As with pike, roach were fed an earthworm diet. Holding tanks were supplied with filtered water from the R. Almond and a total water change was carried out three times per week. Roach odour was used because although this species may consume fish alevins should it encounter them, it is largely herbivorous and insectivorous and thus likely to present less of a threat to salmon alevins than pike. The responses to these two different species may therefore shed light on the capacity of alevins to assess level of predation threat. Although the fish density was greater in the pike holding tank than the roach tanks, both fish densities would produce odour concentrations that are far greater than alevins would experience in the wild, thus, odours of both species should stimulate the strongest response. Equalizing the densities in roach and pike holding tanks is unlikely to equalize odour concentration as there is no evidence that different species produce similar concentrations of odours; indeed, it has been shown that individuals of the same species may produce different quantities of odour (Courtenay *et al.* 1997).

Test protocol

Individuals were tested with only one stimulus odour. The odours were presented when alevins were 27- 40 d old, and thus at least 10 d since the HA-group had experienced prior exposure cues. Their response was measured in terms of change in opercular rate.

Alevins from each of the exposure groups (EG, HA, CO) were sampled at random from one of their five assigned egg boxes. Equal numbers of individuals from the three exposure groups were tested each day, so that ontogenetic differences did not influence group responses. One alevin at a time was gently pipetted into a 1cm diameter piece of tubing supplied with a trickle flow of river water. After 2 min acclimatisation, opercular rate was recorded for 20 s (pre-stimulus opercular rate). The water supply was then switched to one of the stimulus odours. After one min elapsed, ensuring that this new water flow reached the alevin, opercular rate was measured again (post-stimulus opercular rate). Comparison of pre- and post-stimulus opercular rates indicated the magnitude of response to the different stimulus odours.

The experiment was a crossed two-factor design to test the effect of three exposure groups (two treatments and one control) and four stimulus odours (two fish odours and one control). For each of the exposure groups, sample sizes for the stimulus odours were: control odour (10); pike odour (15); roach odour (15); novel water odour (5) (Table 1). The first three stimulus odours were tested randomly throughout the period (thus there should be no effect of developmental stage); unfortunately, novel water was tested later in the experiment (fish were $\geq$ 35 d old), thus we cannot rule out an interaction with development for this stimulus. For this reason, sample size for this odour was much lower.

Table 1. Sample sizes for the experiment.

	Exposure group		
Stimulus odour	Eggs (EG)	Newly-hatched (HA)	Control (CO)
Pike	15	15	15
Roach	15	15	15
Novel water	5	5	5
Control	10	10	10

Data analysis

The response to each of the four stimulus odours was normally distributed (Kolmogorov-Smirnov, D > 0.05, *P* > 0.15 in all cases). Parametric statistics were therefore used. A two-factor ANOVA tested the effect of prior exposure and stimulus odour type (having three and four levels, respectively) with N = ((15 + 15 + 10 + 5) x 3) = 135. The proportional increase in opercular rate after exposure to odour was calculated for each individual and these values were arcsine-transformed for statistical analysis.

Pre-stimulus opercular rates showed no significant differences amongst exposure groups (ANOVA with *post-hoc* Bonferroni correction, *P* > 0.52) or in the odour tests (*P* > 0.05), with the exception that alevins exposed to novel water had higher pre-stimulus opercular rates than alevins exposed to other odours (*P* < 0.001). It is possible that this is because novel water was collected the day before testing, and oxygen content may have decreased.

RESULTS

(i) Is salmon skin extract (SSE) an aversive stimulus?

Salmon parr showed a significantly greater opercular response to SSE (opercular rate increased by 15.9 ± 5.8 %; mean ± 95% CI) than to distilled water (opercular rate increased by -0.7 ± 2.5%; ANCOVA; $F_{1,27} = 15.25$, $P = 0.001$; Fig. 1).

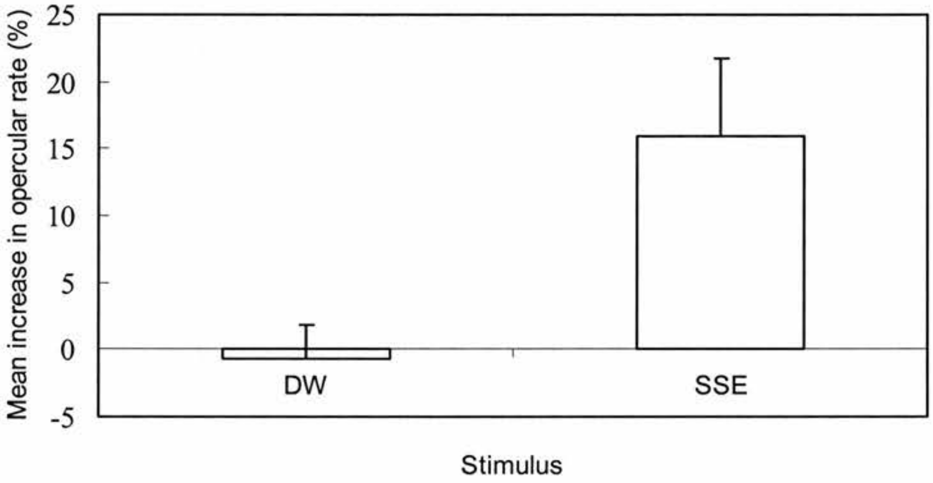

Figure 1. Opercular response of salmon to distilled water (DW; N = 10) or salmon skin extract (SSE; N = 18). Bars indicate 95% CI.

(ii) The effect of prior exposure on the response to predator odour

Overall, exposure group (EG, HA, CO) and stimulus odour (pike, roach, novel water, control) significantly affected opercular beat rate (ANOVA: $F_{11,123} = 10.36$, $P < 0.001$).

The type of stimulus odour presented had a significant effect on opercular rate ($F_{3,131} = 35.28$, $P < 0.001$; Fig. 2). The greatest response was elicited by pike odour (opercular rate increased by 9.29 ± 1.89%; mean ± 95% CI). Presentation of roach odour increased opercular rate by 4.13 ± 1.07%; this response was significantly lower than the response to pike odour (Bonferroni, $P < 0.001$). The response to the control odour was minimal (0.07 ± 0.47%); novel water odour ($P = 0.043$), roach odour ($P = 0.001$) and pike odour ($P <$

0.001) elicited greater responses than the control odour. The proportion of fish showing an increase in opercular rate (post-stimulus increase ≥ 2 beats min^{-1}) also depended on stimulus odour: 99.8% of fish responded to pike odour, compared with 88.9% to roach odour, 50% to novel water and 6.9% to the control odour.

The effect of exposing fish to predator odour combined with conspecific cue either at the egg stage or newly-hatched stage, or not at all (EG, HA, CO), did not affect subsequent response to the stimulus odours ($F_{2,132} = 0.46$, $P = 0.633$). There was no interaction between exposure group and response to the stimulus odours ($F_{6,123} = 1.06$, $P = 0.393$).

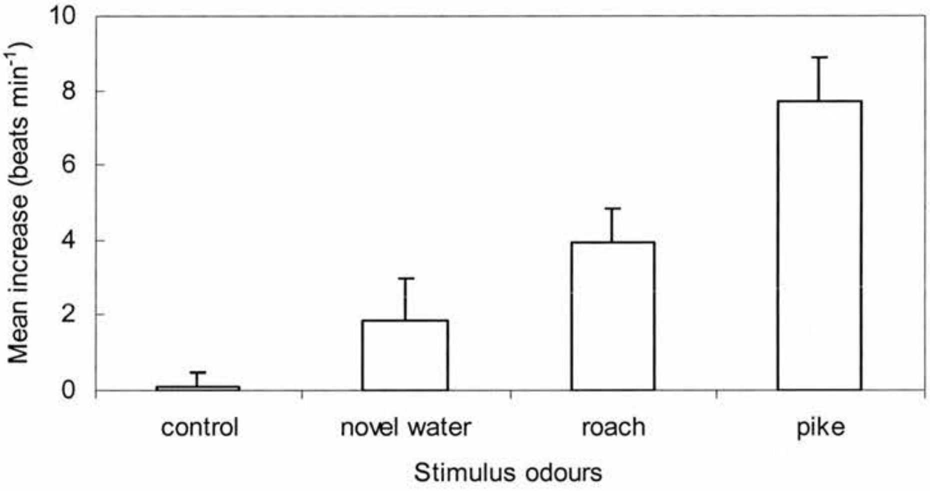

Figure 2. Increase in opercular rate of alevins in response to water from a different river (novel water; N = 15) and water containing either odour of roach; (N = 45) or pike; (N = 45), compared to control (natal river water; N = 29). Data is pooled for all exposure groups. Bars indicate 95% CI.

DISCUSSION

This study shows that newly-hatched salmon recognize the odour of a potentially important predator, the pike. This predator recognition appears to be innate since it was not dependent on prior exposure to salmon alarm substance and predator odour, either at the late-egg or hatching stage. Indeed, individuals that were naïve to pike odour showed similar levels of response to those individuals that had previously been exposed to pike odour combined with alarm substance multiple times.

The non-piscivorous roach odour generated an opercular response that was lower than the response to pike odour, but was nonetheless significant. It is possible that roach, like any large generalist feeder, may predate on salmon hatchlings, if given the opportunity; however, the perceived threat to salmon of roach relative to pike can be expected to be lower. This hints that the alevins discriminate between pike and roach, suggesting that innate predator recognition is highly specific. Vilhunen & Hirvonen (2003) also found that Arctic charr showed highly specific responses to potential predators, depending on the predator's diet. This study found that when the same diet was provided to potential predators, naïve salmon were still able to distinguish between different levels of threat, as defined by piscivorous tendencies. Chivers & Smith (1994b) reported that naïve minnows retain fear of pike longer than they retain fear of goldfish, and Magurran (1989) found that minnows acquired a stronger response to pike (a natural predator) than to a non-native and non-predatory cichlid. An even more specific response was found by Kusch *et al.* (2004); they showed that minnows exhibited a greater antipredator response to small pike than large pike, which, they suggest, was because small pike are more likely to prey on minnows. These studies all suggest an innate template for recognition of pike as predators. Such a predisposition for naïve prey to respond to predatory species over non-predators of the same taxon has also been found in birds (e.g. Curio *et al.* 1983) and mammals (Griffin *et al.* 2001, 2002).

In contrast, other studies have suggested that innate abilities may provoke general responses to predator cues in fishes, but that learning is required to refine the response (Kieffer & Colgan 1992, Olla *et al.* 1994, Fuiman & Magurran 1994, Kelley & Magurran 2003). Predator recognition can occur through releaser-induced recognition learning, by the method of presenting a predator cue and a fright stimulus together, thus encouraging an association between predator and threat (Brown 2003). It appears that learning did not build on the innate response of salmon because exposing newly-hatched fish to conspecifics alarm cues and predator odour did not increase their subsequent response to predator odour alone. The absence of learning in newly-hatched salmon may reflect the absence of opportunities to learn in nature. Atlantic salmon alevins live largely in the river substratum and have limited ability to interact with conspecifics. This limits their opportunity to learn through observing predatory attacks on conspecifics, or through social learning (i.e. copying other fish that show anti-predator responses).

However, learned predator responses may become more important once fry emerge after yolk sac absorption and begin to interact, at which time they may have the opportunity to observe predator attack on conspecifics. Thus, the possibility remains that a learned anti-predator response in salmon is manifested in later life. In fact, this is suggested in the light of studies that have found salmonids to be capable of learning at later developmental stages (e.g. Brown & Smith 1998, Berejikian *et al.* 1999, 2003, Mirza & Chivers 2000) - fish exposed to predator odour and conspecific alarm substance subsequently showed enhanced predator recognition. It is possible that salmonid ontogeny includes a sensitive period when the learning of odour recognition occurs, as has been found in birds and mammals (Gagliardo *et al.* 2001, Moriceau & Sullivan 2004). This study may contrast with these other studies because very young (yolk sac-feeding) fish were tested. In salmon, a sensitive period may occur later when the social environment changes as exogenous feeding begins, and/or further development of the olfactory bulb and neural network, which may improve cognition and learning abilities. Furthermore, because most sympatric fish have the capacity (i.e. gape size) to eat salmon hatchlings, an innate response to general fish cues may be highly adaptive in young salmon. This general response might then be lost as the fish reach a size that prohibits predation, and potentially, the loss of an innate general response might be associated with the development of more specific (learnt) responses. For example, young, predator-naïve European rabbits show a less differentiated antipredator response than do adult predator-naive rabbits; however, over time young rabbits developed the same refined responses as the adults, even without any predator experience (Pongracz & Altbacker 2000).

Repeated prior exposure to a predator odour cue, as applied in this study, may reduce any anti-predator behavioural response through habituation, that is, animals may judge a cue to be a false alarm if it is not followed by any harmful consequence. Habituation has been demonstrated in a range of animals (Hinde 1960, Manning 1979) including in Pacific and Atlantic salmon (Järvi & Uglem 1993, Berejikian *et al.* 2003). However, there was no evidence of habituation to predator odour in this study because individuals that were exposed to pike odour on at least 14 occasions still exhibited a stress response when they subsequently re-experienced the odour.

This is the first study to show that prey fish respond to predator odour cues by increasing their ventilation frequency. Seals have been shown to increase ventilatory frequency in response to a predator (Lydersen & Kovacs 1995), similar to the increased breathing rate that occurs in humans in response to a hazardous situation (Hickam *et al.* 1948). In fish, Von Frisch (1941) documented that presentation of alarm substance (Schreckstoff) to minnows produced an increase in breathing rate. Two other studies have documented this physiological response in fish to a visual predator cue without any associated olfactory cues (Metcalfe *et al.* 1987, Barreto *et al.* 2003). These support the finding that the response of salmon alevins to predator odour does indeed represent predator recognition, rather than an increase in cue sampling rate due to presence of an unknown odour. This does not exclude the possibility that a ventilation response may reflect an increase in vigilance; if this is the case then results presented here show that odour of a piscivorous fish induces a greater state of vigilance than an omnivorous fish odour, again supporting innate recognition of predation risk.

The ventilation response appears to have great value as a tool for investigating predator recognition because it was demonstrated to be very sensitive; 99.8% of fish responded to the predator odour cue. There appear to be no other studies that document such a consistent response to a predator odour cue, and find that such a result supports the hypothesis that Atlantic salmon has an innate ability to recognize a sympatric predator. In comparison, for example, Vilhunen & Hirvonen (2003) detected a behavioural response to a predator odour cue in less than half of the Artic charr exposed to it, based on freezing behaviour. However, Vilhunen & Hirvonen (2003) also used domesticated fish, and they note that this may influence strength of response. Johnsson *et al.* (2001) found that domestication reduced the behavioural and heart rate response of Atlantic salmon to a predator. This study used progeny of wild fish.

The composition of predator diets (Brown 2003) is known to mediate the acquisition of predator recognition in fish. Because pike in this study were fed earthworms rather than fish, dietary odour would not have suggested an association between pike and prey fish, which may otherwise have confounded the experiment. Moreover, the roach were also fed earthworms; thus diet cannot explain the magnitude of the response to pike odour. The possibility of a dose-dependent response to odours cannot be excluded. Different species and even different individuals of the same species may produce different amounts of odour (Courtenay *et al.* 1997). Thus, pike odour may have elicited a greater response than roach odour simply because the pike odour was more intense. However, both odours were presented at concentrations that are likely to be much greater than alevins would experience in the wild, especially in flowing water. Therefore both odours should provide a strong olfactory cue. There is evidence that a threshold concentration must be reached to elicit a response (Kusch *et al.* 2004), and this was clearly attained in this study. Unpublished data (see chapter three) showed that predator odour taken from a tank with 2.6 g of predatory fish l^{-1} invoked a maximum ventilation response in Atlantic salmon; further increases in concentration (g of fish l^{-1}) did not increase response. In this study, roach and pike odour were taken from tanks containing at least 4 g of fish l^{-1}.

The trials conducted using river water from an unfamiliar source elicited a significant opercular response compared to the control, suggestive of a neophobic response. It is possible that pike and roach odours elicit a greater response than novel water because metabolic by-products in fish cues are more recognizable and provoke greater vigilance. This nevertheless suggests the remarkable possibility that the salmon are distinguishing between metabolic differences in pike and roach odours. However, it should be remembered that novel water was tested towards the end of the experiment. As breathing rate generally decreases with increasing size in fish (Schmidt-Nielsen 1996), it is possible that growth of fish during the experiment could interact with breathing response to a threatening situation.

By quantifying a physiological response, namely opercular beat rate, this study has demonstrated that newly-hatched salmon recognize predator odour. Pike are top predators in much of the Holarctic and their range overlaps, to a large degree, that of the Atlantic salmon. It is suggested that there may be a genetic template that allows salmon to identify pike odour cues on first encounter. In contrast to other studies, this study found predator recognition to be consistent across individuals, which may be due to the use of a physiological measure of recognition and/or the use of wild rather than domesticated salmon.

SECTION II

CHAPTER THREE

Innate abilities to distinguish between predator species and cue concentration in Atlantic salmon

ABSTRACT

Atlantic salmon increase their ventilation rate in response to predator odour cues. In this study, the ventilation response of newly-hatched Atlantic salmon to odours of high- and low-risk predators at five concentrations (10-200%), compared to a control odour, was measured. Concentration may be interpreted as predator proximity and/or number of predators present and thus represents a gradation of predation risk. Predator species (pike and minnow) and odour concentration affected the number of individuals responding, and the strength of individual response. Fewer individuals responded to minnow odour than pike odour, and the proportion of individuals responding also declined with concentration. Salmon exhibited a stronger response towards high-risk predator (pike) odour than the low-risk (minnow) odour for a given concentration, and ventilation rate also increased with concentrations of both odours. This study demonstrates that Atlantic salmon have the innate ability to evaluate risk in terms of odour concentration and predator species.

INTRODUCTION

Chemical cues emitted by predators can provide substantial amounts of information to prey regarding predation risk, including the species of predator (Watt & Young 1994), whether the predator is actively foraging (Phillips 1978) and the size of the individual predator (Kusch *et al.* 2004). The strength of the chemical cue may provide information such as distance to the predator or the number of predators close by (Horat & Semlitsch 1994), whilst chemicals present in dietary cues may reveal what the predator has recently been feeding on (Wilson & Lefcort 1993, Mirza & Chivers 2001, 2003a, Vilhunen & Hirvonen 2003).

Signal detection theory (Green & Swets 1966) predicts that a subject (prey) evaluates a signal, such as predator chemical cues, against a predetermined threshold level (e.g. concentration of cue). When the signal reaches the threshold criterion it is positively identified and the subject responds accordingly. A prey may thus show an 'all-or-nothing' response to predator odour (e.g. Brown *et al.* 2001a, Marcus & Brown 2003, Mirza & Chivers 2003b). A lack of response to a cue – even if it has been detected - may be because of the risk that a weak stimulus is actually a 'false alarm' (when a subject falsely identifies a signal that is not present).

To evaluate predation risk, prey may differentiate between odour cue concentrations. A low concentration of predator odour may mean that a predator was in the area but has since left, or is too far away to make a successful strike, and hence poses no real risk. For example, both fathead minnows, *Pimephales promelas*, and rainbow trout, *Oncorhynchus mykiss*, show strong anti-predator behaviour in response to alarm substance (AS), or 'Schreckstoff' (Brown *et al.* 2001b, Mirza & Chivers 2003b), which is released from the skin of conspecifics when damaged, for example when attacked by a predator (Smith 1992, Brown 2003). Both species are able to detect very low levels of the alarm substance (or the active component of it), yet in experiments neither species showed behavioural responses to these low levels (Brown *et al.* 2001b, Mirza & Chivers 2003b). The authors suggest that a weak predator cue signals that the risk is low and therefore it is not beneficial for the prey to engage in a behavioural response. Mirza & Chivers (2003b) describe this as the 'observable response threshold'. However, although a weak stimulus may not produce a behavioural response, other responses such as increased vigilance may be stimulated (Brown *et al.* 2004).

In contrast to the detection/rejection of predator signals, the risk-sensitive predator-avoidance hypothesis (Helfman 1989) predicts that prey animals use predator cues to evaluate risk and produce an anti-predator response appropriate to this level of risk. This strategy is used to minimize the costs of mistakenly performing an anti-predator response when there is actually no risk, such as foregoing foraging opportunities or territorial defence. This hypothesis is supported by evidence that the strength of an anti-predator response performed by the prey depends on the strength of the chemical cue received (e.g. Harvey *et al.* 1987, Horat & Semlitsch 1994, Seely & Lutensky 1998, Forward *et al.* 2003, Dupuch *et al.* 2004, Kusch *et al.* 2004). For example, tadpoles, *Rana* spp., showed a continuous decline in activity when exposed to increasing concentrations of predator odour (Horat & Semlitsch 1994). These tadpoles appear to be balancing the risk of predation (by reducing activity) with opportunities for finding food (by increasing activity).

These two hypotheses need not be mutually exclusive. An individual may first determine whether a cue is in fact, a real signal, and secondly determine the level of response that is appropriate to the predicted risk. Furthermore, the risk posed to different individuals will vary due to differences in their response capacities and the level of risk they are willing to accept before responding.

Atlantic salmon have innate predator recognition and can distinguish between odour cues that represent high and low predation threats (chapter two). Juvenile salmon increase their ventilation (opercular) rates in response to predator odour (chapter two) and visual predator cues (Metcalfe *et al.* 1987; chapter four), suggesting that opercular rate reflects predator recognition and possibly vigilance (chapter two). Because an opercular response is less costly than an overt behavioural response, in terms of energetic requirement and activities foregone, we might expect to be able to detect an opercular response at lower stimulus levels (i.e. below the observable response threshold).

In this study, the opercular response of newly-hatched Atlantic salmon to varying concentrations of odours from two species of fish, presenting high- and low-risks of predation, was measured. The hypotheses tested were that (1) Positive responses will occur more often at higher odour concentrations (signal detection theory), and that the magnitude of the response will be concentration-dependent (risk-sensitive predator avoidance). (2) Response to the high-predation risk cue will elicit more positive responses, and these will always be of greater magnitude than responses to the low-risk cue. (3) Individual response threshold will vary due to individual variation in risk perception and valuation of acceptable risk level.

METHODS

Newly-hatched (yolk-sac feeding; 17-25 d old) Atlantic salmon alevins were exposed to one of three odours. These were pike odour (PO), European minnow (*Phoxinus phoxinus*) odour (MO) and a control (the local river water; CO). Five concentrations of PO and MO were tested. Each fish was exposed to only a single concentration of a single odour (i.e. there was no replication of individuals). In total 335 fish were used. The number of individuals tested with each odour/concentration is presented in Table 1. The odours/ concentrations were tested randomly throughout the nine-day period.

Fish

Salmon alevins were obtained from six families. These were the progeny of wild fish, taken from the River Almond, Perthshire, Scotland, 56°43’N -3°53’W. Eggs were stripped from broodstock in November/December 2003 and held at the Fisheries Research Services Almondbank field station at ambient temperature. Eggs hatched 10 – 12 March 2004. Experiments were conducted between 28 March and 5 April.

Table 1. Numbers of individuals tested with each odour type and concentration and numbers showing a positive response (opercular rate increase ≥ 2 beats min^{-1})

Concentration (%)	Pike odour (PO)		Minnow odour (MO)		Control (CO)	
	N tested	N responding	N tested	N responding	N tested	N responding
0	-	-	-	-	40	5
10	30	14	30	9	-	-
50	30	18	30	9	-	-
75	30	21	30	16	-	-
100	30	27	30	20	-	-
200	25	23	30	25	-	-
Total	145	103	150	79	40	5

Odours

Pike are top predators in temperate and boreal aquatic systems, and so present high predation risk to juvenile salmon. Minnows are generalist feeders. Their gape size is sufficient to make them potential predators of salmon hatchlings. However, Maitland & Campbell (1992) report that they rarely eat other fish. Thus the perceived danger from minnows is likely to be lower than from pike.

The three odours were samples of water taken from tanks containing pike, minnows or no fish. PO was a water sample from one of two 133 l tanks, each containing one pike (340 and 344 g; biomass of 2.6 g l^{-1}). MO was a water sample from a tank filled with 25 l of water and containing seven minnows, totalling 66 g (biomass of 2.6 g l^{-1}). To remove any piscivorous dietary cues in the water samples, pike were fed earthworms for 2 weeks prior to and during odour collection. This time period allowed for any previous dietary cues, still residue in the pike, to be eliminated. Minnows were fed bloodworms. The control tank was set up with pump and filtration, as were the minnow and pike tanks, and filled with river water. All three tanks had partial water changes every 3 d.

Five concentrations of PO and MO were tested: 10%, 50%, 75%, 100% and 200%. 100% equated to a sample of water taken from the respective tanks. Concentrations of less than 100% were achieved by diluting the samples with the required amounts of the CO odour. A concentration of 200% PO was obtained by placing the two pike into the same 133 l tank for 2 d. 200% of MO was obtained by reducing the water in the minnow tank by 50% for 2 d. These different methods of obtaining 200% were chosen because (i) it was deemed unethical to hold the pike in a smaller tank for any length of time, and (ii) we could not have captured the number of minnows required to provide a biomass of 5.2 g l^{-1} in a 133 l tank.

Protocol

Individual alevins were chosen at random and were pipetted into a short section of transparent tubing, which was drip-fed river water from a tank via a peristaltic pump. Alevins were left for 2 min and then a sequence of 30 s counts of opercular rate were made until two subsequent counts differed by no more than 1 beat 30 s^{-1} (pre-stimulus opercular rate). The water supply was then changed to a separate tank containing either PO, MO or CO. 30 s was allowed for this new water supply to reach the alevin, and then opercular rate was measured for two further 30 s periods (post-stimulus opercular rate).

Data Analysis

An individual whose opercular rate increased by < 2 beats min^{-1} after exposure to PO or MO was considered to show no response. The subset of the data that included only individuals showing an opercular rate increase $\geq$ 2 beats min^{-1} (Table 1) was analyzed separately. The distributions of opercular beat rates of these responsive individuals before and after exposure were tested for normality (Anderson-Darling test). Nine out of the 11 predator species/concentration groups showed a normal distribution in opercular rates. As there was a strong linear relationship between pre- and post-stimulus opercular rates in all groups, data were analyzed with the parametric analysis of covariance (ANCOVA; post-stimulus opercular rate as the dependent variable and pre-stimulus opercular rate as the covariate). Predator species (three levels) and odour concentration (six levels) were fixed/predictor factors.

RESULTS

Presence of Response

The ~~number~~ proportion of salmon alevins that did not respond to odour cues (opercular rate increase of < 2 beats min^{-1}) depended on predator species (pike; PO, minnow; MO; control; CO; logistic regression: Wald = 12.402, N = 335, $P < 0.001$) and odour concentration (0, 10, 50, 75, 100, 200%: Wald = 34.717, N = 335, $P < 0.001$; Fig. 1). A greater proportion of individuals responded to PO than MO for any given concentration. At lower odour concentrations fewer individuals responded. For example, at 100% concentration, 90% of fish responded to PO and 66.7% to MO, whereas at 10% concentration 46.7% of fish responded to PO and only 30% responded to MO.

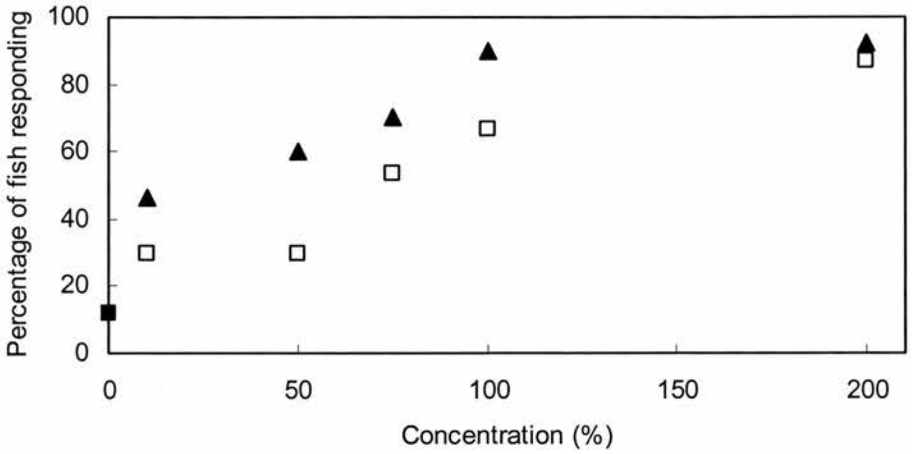

Figure 1. Percentage of fish responding (opercular rate increase ≥2 beats min^{-1}) to pike odour (PO; ▲), minnow odour (MO; □) or control (CO; ■) at varying concentrations.

Strength of Response

Predator species (PO, MO) and odour concentration strongly affected the opercular rate of alevins, even after excluding the 113 individuals (out of 295) that did not respond to PO or MO (ANCOVA: $F_{1,222} = 6.938$, $P = 0.009$ and $F_{4,222} = 3.935$, $P = 0.004$, respectively). There was no interaction between predator species and odour concentration ($F_{4,222} = 0.936$, $P = 0.444$).

The response to PO was always greater than the response to MO, for any given concentration (Fig. 2). The strength of response also increased with increasing concentration of PO (opercular rate increased by 3.3% (2.0 - 4.5) in response to 10% PO and up to 4.8% (3.6 – 9.1) in response to 200% PO; median (IQ range)). This was generally true of MO (1.9% (1.8 – 2.7) increase in response to 10% MO and up to 3.8% (2.1 – 5.5) increase in response to 200% MO), except that the response to 100% MO (4.6% (2.1 – 6.9)) was greater than the response to 200% MO. The increase in response to the control (CO) was 0% (-2.0 – 0).

Post-hoc Tukey's HSD (for unequal N) test revealed that the response to 200% PO was significantly greater than responses to 100% PO ($P < 0.001$), 75% PO ($P < 0.001$), 200% MO ($P < 0.001$) and 75% MO ($P = 0.021$). The absence of significant differences between other concentration/odour type groups was attributed to the fact that a greater proportion of individuals showed no response at lower concentrations, meaning that sample sizes were reduced. This is indicated by the inability of the *post-hoc* test to find a difference in response between concentrations of 200 and 50 or 10% (N = 9 - 18), despite finding a difference compared to 75 and 100% (N = 16 - 27).

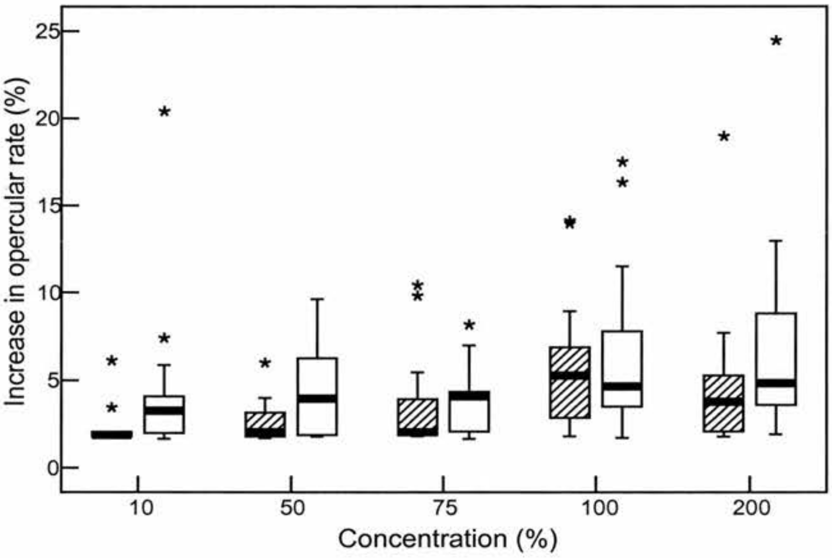

Figure 2. Percentage increase (median with IQ range) in opercular rate after exposure to pike odour (PO; □) and minnow odour (MO; ▨) at varying concentrations. Includes only individuals that showed a response ≥ 2 beats per min.

DISCUSSION

Predator-naïve Atlantic salmon exhibited a ventilation response to predator odour cues. More individuals responded to pike (high-risk predator) odour than minnow (low-risk predator) odour, and the proportion of individuals responding also increased with concentration of both odours. For any given concentration, the strength of the ventilation response was always greatest to pike odour than minnow odour, and the strength of the response was also concentration-dependent.

This study suggests that Atlantic salmon have an innate ability to evaluate predation risk. Predator odour concentration represented predation risk, as it can signify predator proximity and/or number of predators in the vicinity (Horat & Semlitsch 1994). Predator-naïve salmon demonstrated the ability to distinguish between different concentrations of predator odours, and the strength of the response increased with increasing concentration of both pike and minnow odour. This supports Helfman's (1989) hypothesis that the strength of an anti-predator response is appropriate for the level of predation risk. An innate ability to discriminate different concentrations of a predator odour has apparently only previously been documented in tadpoles of *Rana* spp. (Horat & Semlitsch 1994) and in cloned water fleas, *Ceriodaphnia reticulata* (Seely & Lutnesky 1998).

Furthermore, a greater proportion of individuals responded to higher concentrations of odour. As presumably all individuals were capable of detecting chemical cues to within an order of magnitude, it seems that the fish first chose whether or not to give a response (predicted by signal detection theory), and then adjusted the level of response (predicted by the risk-sensitive avoidance hypothesis). This supports the first hypothesis. The absence of a response at low concentrations by some individuals may be due to the risk of the cue being a 'false alarm' (Fig. 3; the noise distribution curve to the right of the criterion threshold). At higher concentrations there is probably no possibility of making a false alarm (i.e. no overlap of the noise and signal distribution curves; Fig. 3).

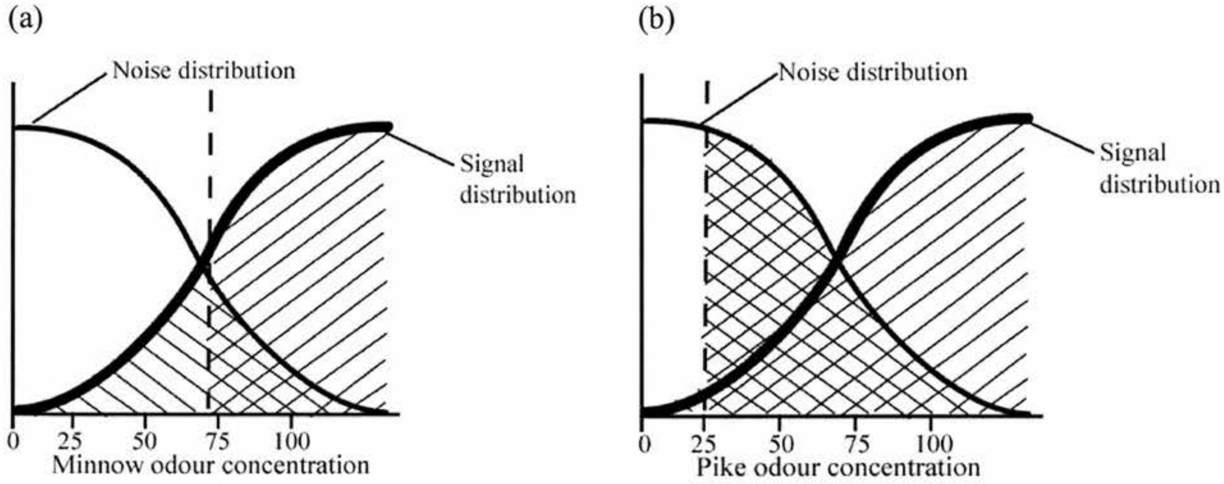

Figure 3. Schematic of signal detection, where the signal is (a) minnow odour and (b) pike odour. Criterion threshold (----) drawn at concentration that elicited a positive response in 50% of individuals. The signal distribution curve to the right of the criterion threshold (▨) represents a correct detection and the signal curve to the left (▧) represents a missed detection. The noise distribution curve to the left of the criterion threshold represents a correct rejection (□) whereas the curve to the right represents a false detection (▩).

Individuals that did respond always showed a greater increase in opercular rate in response to pike odour than minnow odour at a given concentration, supporting hypothesis two. Previous work (chapter two) also showed that newly-hatched Atlantic salmon can distinguish between odours of high- and low-risk predators. Furthermore, a greater proportion of individuals responded to pike odour than minnow odour, for a given concentration, which also supports hypothesis two. This suggests that some individuals have a lower criterion threshold for responding to pike odour than minnow odour (Fig. 3; the areas of the signal distribution curves lying to the right of the criterion threshold).

However, the cost of a lower criterion threshold is that there is a greater probability of making a false alarm. As pike represent greater risk than minnows, ignoring a true signal (rather than acting on a false alarm) would likely be of more consequence with pike odour than minnow odour. Again, it appears that fish are choosing firstly whether a positive response to the signal is warranted, and secondly assessing the level of response that is appropriate.

That some individuals responded to the lowest concentration of odours, whilst others did not respond to the same odour of one magnitude greater strength, demonstrates individual variability in a response threshold (hypothesis three). This finding is consistent with observations of Kusch *et al.* (2004) and Dupuch *et al.* (2004). This variation in response may be because of variation in risk perception. For example, Brown *et al.* (2001a,b) have suggested that response threshold of minnows may be population-specific, with minnows from high predation-risk sites exhibiting more anti-predator behaviour at lower concentrations of predator cue, compared to individuals from low predation-risk sites. In our case, all salmon were from the same river; however, there may have been genetic variation in ability to perceive risk between and/or within family groups. Alternatively, variation may be due to individual variability in the cost of making a response, reflecting the physiological state of the fish. For example, minnows that are hungry do not respond to alarm substance and predator odour, whereas satiated minnows do give an anti-predator response (Brown & Smith 1996). These individuals apparently make the decision to ignore predator warnings when lack of food is a greater threat, although the cues may encourage greater vigilance (Brown & Smith 1996).

Other studies have documented both continuous increases in prey response to increasing concentration of chemical cue (e.g. Harvey *et al.* 1987, Seely & Lutensky 1998, Dupuch *et al.* 2004, Kusch *et al.* 2004) and also an 'all-or-nothing' response above a threshold concentration (e.g. Ward *et al.* 1996, Marcus & Brown 2003, Mirza & Chivers 2003b). Two studies (Brown *et al.* 2001b, Mirza & Chivers 2003b) suggest that although fish may not show a behavioural response to low concentrations of a cue, they are in fact detecting it. Recently, Brown *et al.* (2004) showed that glowlight tetras, *Hemigrammus* *erythrozonous*, did not respond to low concentrations of conspecific Schreckstoff unless it was paired with a secondary (visual) cue. They suggest that low concentrations of the cue increased the fishes' vigilance to other cues, and that they reacted if any further information gathered suggested risk of predation.

Because increased ventilation rate has a lower cost associated with performing it than an overt anti-predator behaviour (e.g. reduction in activity or time spent motionless), we may expect to see a ventilation response in lower-risk situations. This could allow detection of small responses that occur below an 'observable response threshold' (Mirza & Chivers 2003b). Pettersson *et al.* (2000) suggested that the electrophysiological limit of carp to detect predator odour is a magnitude lower than the odour concentration that elicits a detectable anti-predator response. However, this study also showed that the low (mean) responses to low odour concentrations may be partly due to variation in individuals' responses. Fewer individuals responded to low odour concentrations (pike and minnow), whereas high concentrations produced consistent responses across individuals.

Pike odour used in this study was obtained from one of two pike. A total of seven minnows were required for minnow odour. This should allow for any differences in quantity of odour produced by individual fish of the same species (Horat & Semlitsch 1994, Courtenay *et al.* 2001). The assumption was that odour concentration is linearly related to biomass, and that the relationship is the same for pike and minnow odour. As it is unclear what chemical(s) the salmon are detecting in the cue (intestinal by-products (Courtenay *et al.* 1997), faeces (Brown *et al.* 1996), urine (Moore *et al.* 1994) and mucus (Williams & Moore 1985) are all candidates) it is not possible to confirm this assumption. However, it seems reasonable that the amount of urine/mucus/intestinal or other cues produced is related to body size, and Courtenay *et al.* (2001) found that salmon response to conspecific odour was correlated with the biomass of conspecific odour-producers, which they related to the amounts of chemicals produced. If skin mucus was a significant component of the cue (Williams & Moore 1985), then the difference in the responses to minnow and pike odour would be more acute, owing to the greater surface area of minnows than pike.

This study showed that salmon used two pieces of information, namely predator species and odour concentration, to guide the extent of ventilation response. There was a threshold level of cue for response which varied among individuals, and positive responses was also risk-sensitive (*sensu* Helfman 1989), as stronger responses were given to higher concentrations of odour and towards the high-risk predator. This finely-tuned ability to discriminate odour type and strength relies on innate responses.

SECTION II

CHAPTER FOUR

Predator-induced hyperventilation in wild- and hatchery- reared juvenile salmon*

ABSTRACT

Following exposure to a visual predator stimulus (a brown trout), the opercular rate of Atlantic salmon, increased by 35.3 ± 11.0% (mean ± 95% CI). The time taken for opercular rate to decline to baseline levels depended upon the occurrence of any associated locomotory activity. Opercular rates of fish that dashed when exposed remained elevated for 38.2 ± 20.6 min, whereas those of individuals that did not move ('freezers') recovered within 7.2 ± 2.9 min. The duration that opercular rate remained elevated was positively correlated with the magnitude of the elevation, which was higher in 'dashers' than freezers. The maximum opercular rate in freezers was similar between wild fry and hatchery-reared fry (from wild parents). There was a significant delay, however, in hatchery compared with wild fry in the time until peak ventilatory response and onset in the decline phase. This difference in opercular response suggests that hatchery fish were slower to realize fully the potential danger from the predator. Any delay in response can be directly attributed to the effect of hatchery-rearing environment, rather than domestication or hatchery selection effects.

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INTRODUCTION

Hatchery rearing of salmonids can have a negative effect on their performance capacities in the wild, in terms of foraging, movement, migration and predator avoidance (Johnsen & Ugedal 1989, Quinn 1993, Brown & Warburton 1999, Brown & Laland 2001). This occurs in three contexts: through genotypic selection, the expression of phenotypic traits and learning (Weber & Fausch 2003). However, it is often not determined whether observed differences in behaviour can be attributed to an affect acting on the genetic or learnt components of such traits (Keiffer & Colgan 1992, Wisenden 2001).

A reduction in appropriate anti-predator responses has been demonstrated in many studies of hatchery-reared salmonids (e.g. Berejikian 1995, Brown & Smith 1998, Berejikian *et al.* 1999). Nearly all studies have investigated the responses of domesticated salmon, i.e. fish that are the progeny of hatchery-reared broodstock, and so do not separate the effects of genetic selection through generations of breeding with effects arising from rearing wild progeny in an artificial environment. Järvi & Uglem (1993), however, showed that even the progeny of wild fish reared in the hatchery may benefit from training to respond appropriately to predators, possibly because the rearing environment denied them the opportunity to practice and refine their anti-predator responses (Suboski & Templeton 1989, Brown & Laland 2001). There is little information on how hatchery-reared fish fare against predators when released into the wild as fry or parr (Mather 1998), which highlights the need for more detail on how the hatchery-rearing process influences responses of fish to predators.

A suitable starting point is to determine whether the abilities of fish to recognize and respond appropriately to predators are affected by rearing environment. Metcalfe *et al.* (1987) observed an increase in opercular rate of hatchery-reared Atlantic salmon fry in response to a predator model. There are two reasons why opercular rate could have increased in these contexts. Firstly, hyperventilation may have been linked to repayment of an oxygen deficit following 'dashing' or fleeing bursts (Shelton 1970). Secondly, it has been suggested that ventilatory frequency may increase in preparation for possible physical responses to the predator, such as fleeing or fighting (Hickam *et al.* 1948, Espmark & Langvatn 1985, Lydersen & Kovacs 1995) even when there is no prior locomotory activity. It is well established that heart and breathing rates of deer, sheep and birds change in response to stress and in the absence of obvious locomotory and post-prandial influence in (e.g. Moen *et al.* 1978, MacArthur *et al.* 1979, Causby & Smith 1981, MacArthur 1982). In salmonid fish, heart rate increases following predator attack (Hojesjo *et al.* 1999, Johnsson *et al.* 2001) and the level of increase is correlated to the duration of any associated activity.

In Metcalfe *et al.*'s (1987) study, individual fish required up to 3 h to recover from the predator exposure before attaining their resting opercular rate. However, the study used predator-naïve hatchery salmon, and this raises the possibility that the fish had imperfect perception of predation threat due to a lack of predator experience, which led the fish to overreact (Fraser & Gilliam 1987). It is possible that anti-predator responses of wild fry become refined through experience (Kieffer & Colgan 1992, Berekijian 1995) and weaker individual responses may be weeded out through selective predator mortality (Johnsson *et al.* 2001, Kelley & Magurran 2003).

This study investigated a frequent claim that the predator response of hatchery fish would be weaker or inappropriate compared to that of wild fish because the hatchery environment denies fish the experience required to perform well in predator encounters (Johnsson 1993, Dellefors & Johnsson 1995). The hyperventilation response was measured to determine whether a visual predator stimulus produces the same response in wild and hatchery-reared (wild origin) Atlantic salmon. This quantitative measure has the potential to provide insight into predator recognition abilities that may be affected by rearing conditions, and have previously only been hinted at (e.g. Berejikian 1995, Alvarez & Nicieza 2003).

METHODS

Fish

All wild and hatchery fish were progeny of wild brood stock from ten families that were collected from the River Almond, Perthshire, Scotland in September 2002. All eggs were held at the Fisheries Research Services Almondbank Field station, Perthshire. The hatchlings were subsequently split into two groups. One of these was stocked out into the Ballinloan Burn, a tributary of the River Braan, Perthshire (NN969408) in spring 2003 before first-feeding, and the second group was reared on in the hatchery. The hatchery fish used in these experiments were taken from the latter group while wild fish were collected from the river by electrofishing in September 2003. As no natural spawning occurs in this river section, these were the surviving individuals from the spring stocking. Wild and hatchery fish were sized matched (fork length (L_F); mm; mean ± SD: wild fish, 90.3 ± 5.8; hatchery fish, 90.8 ± 4.6) because ventilation frequency is inversely proportional to body size (Schmidt-Nielsen 1996) and because of a potential size effect on risk assessment; for example, larger individuals may be more willing to risk predator-exposure when foraging (Johnsson 1993). Sample sizes were N = 26 wild fish and N = 29 hatchery fish.

Experiments

The trials were conducted within a glass-sided indoor stream separated into 15 identical compartments, housed at Fisheries Research Services Almondbank field station. The compartments were enclosed at each end by mesh grids, through which flowed a continual water supply from the River Almond. Each section was 0.9 x 0.35 x 0.17 m and contained one shelter (17 x 12 cm), placed against the glass. An individual fish was placed in each section and could be observed through the glass whilst the observer remained within a darkened hide. A continuous food supply was maintained by individual feeders in each section, which released bloodworms for 3 s every 30 min. The provision of individual food supplies was important because hungry fish may show less response to a predator cue (Dill & Fraser 1984, Gotceitas & Godin 1991, Hojesjo *et al.* 1999). Experiments were conducted between 30 September 2003 and 3 November 2003, when mean daily average water temperature was 6.6° C (range: 1.9 – 10.8 °C). Light was from overhead 400 Watt, 6000 lux bulbs (Philips SON Agro) set to an ambient photoperiod.

Fish were allowed 4 d to settle into the compartments. On days two and three, 10 min behavioural observations were made per fish to compare settlement of hatchery and wild fish in the test environment. The proportion of time spent within the shelter, resting on the substratum and maintaining a feeding position, and the number of food items taken, number of movements (> 4 cm) and opercular rate was recorded. Responses of individual fish to a predator model were then measured on day five. An observer within the darkened hide was able to view the salmon fish such that opercular movements were clearly visible, minimizing any potential for observation error. Opercular rate immediately prior to predator exposure (resting rate) was calculated by the observer counting opercular movements for 30 s every minute for three consecutive minutes and then taking the mean. The predator model was then suspended in the test compartment by a second person standing on a gangway behind the observation compartments. The model was a brown trout, *Salmo trutta*, specimen which had been killed for unrelated purposes and frozen. The trout was tied to a stick by 0.3 mm diameter fishing wire and held *c.* 20 – 30 cm in front of the subject fish for 30 s. This provided close-range exposure to a realistic predator model which ensured that all salmon had seen the predator (the water was always clear), without putting them at risk of actual predation. Immediately after the trout was removed, opercular rate was measured again by taking four counts each of 30 s at intervals of 60 s.

The placement of fish into the test sections and exposure to the predator model was repeated four times (runs) using different fish and a different predator model per run. The same observer recorded opercular rates throughout the runs. The four trout used were 215, 202, 245 and 355 mm L_F, as brown trout of this size are one of the main fish predators of young Atlantic salmon in Britain (Mills 1971). Wild and hatchery fish were placed in alternate sections and this order was changed over the four runs. Exposure to the trout was initiated at the downstream section, so as to avoid exposing other fish to trout odour before testing. The latter two runs (N = 11 wild fish, N = 14 hatchery fish) also looked at the duration that opercular rate remained elevated after exposure to the model, thus enabling comparison with Metcalfe *et al.*'s (1987) study.

Data analysis

Opercular rate before and after exposure were compared using *t*-tests corrected for multiple pairwise comparisons. To compare the responses of wild and hatchery fish independent two-sample *t*-tests (assuming unequal variance) and ANOVA were used. The latter compared wild and hatchery fish as separate (fixed) factors and also included individual body length and run number as random factors. Where percentage change in opercular rates was analyzed, data were arcsine-transformed. Repeated measures ANOVA was used to analyze the change in individual opercular response in the 4 min post-exposure period. This required calculating the proportional change between opercular rates of subsequent counts and then applying an arcsine transformation (subtraction of 1 from each datum was necessary for this).

A variance-ratio test was performed to determine whether the samples of wild and hatchery fish had showed equal variance in opercular response. Opercular rate before and after exposure of both wild and hatchery fish were normally distributed (Kolmogorov-Smirnov; $F < 1.03$, $P > 0.276$), therefore an *F*-test was used to compare variance in opercular rates of wild and hatchery fish.

RESULTS

(i) Adaptation to the test environment

On days two and three of the trial, wild and hatchery fish spent similar amounts of time sheltering (two-sample *t* test; $t_{53} = 0.9$, $P = 0.366$), resting on substrate outside of shelter ($t_{53} = 1.2$, $P = 0.232$) and holding a feeding position ($t_{53} = 0.2$, $P = 0.855$). Most fish remained in their shelter the majority of the time. The number of food items consumed was very low for both wild and hatchery fish (mean ± 95% CI; 0.01 ± 0.01 and 0.01 ± 0.01 food items min^{-1}). Resting opercular rate was lower in wild fish (35.8 ± 3.5 beats min^{-1}) than hatchery fish (50.4 ± 3.2; $t_{54} = 6.3$, $P < 0.001$). Overall, wild and hatchery fish appeared to have adapted to the testing environment similarly.

(ii) Behavioural response to predator model

Seven (26.9% of) wild fish exhibited 'dashing' (disorientated swimming) after exposure, although two of these dashed but remained under the shelter. Dashing was also exhibited in nine (31.0% of) hatchery fish (all of which left the shelter), including two individuals that darted out of the shelter to the place furthest away from the trout. 'Rocking', whereby a fish resting on the substratum rocks from one pectoral fin to the other (Sneddon *et al.* 2003b) was also observed in two (6.9%) of the hatchery fish.

Activity such as dashing increases oxygen demand and thus opercular rate, which would occur in addition to a fright response. Because dashing may affect subsequent behaviour (predator response), I compared the responses of 'dashers' and 'freezers' (the latter defined as individuals that did not dash). Fish that exhibited dashing were of the same size as individuals that responded by freezing (two-sample *t* test; wild: $t_{10} = 0.8$, $P = 0.441$; hatchery: $t_{13} = 0.6$, $P = 0.576$).

(iii) Physiological response to predator model

Before exposure to the model, wild fish had significantly lower opercular rates than hatchery fish (mean ± 95% CI; 42.0 ± 3.8 and 53.2 ± 3.6 beats min^{-1}, respectively; two-sample *t* test; $t_{53} = 4.2$, $P < 0.001$). Resting opercular rates were not correlated with body size (Pearson; $r = -0.168$, $N = 55$, $P = 0.222$). There was no difference in the variance in opercular rates of wild and hatchery fish (ANOVA; $F_{2,36} = 1.03$, $P = 0.954$). All individuals except a single hatchery fish demonstrated an elevation in opercular rate in response to the model of at least 4 beats min^{-1} during the 4 min post-exposure.

Increase in opercular rate

Opercular rate increased from (mean ± 95% CI) 41.8 ± 4.5 to 50.2 ± 4.5 beats min^{-1} during the first 4 min post-exposure in wild 'freezers' (N = 19; an increase of 22.4 ± 6.5 %; paired *t* test; $t_{18} = 7.6$, $P < 0.001$; Fig. 1) and from 54.2 ± 4.2 to 66.2 ± 5.5 in hatchery 'freezers' (N = 20; 23.6 ± 10.0 % increase; $t_{19} = 5.4$, $P < 0.001$; Fig. 1). The response of wild and hatchery 'freezers' was not significantly different (ANOVA; $F_{1,3} = 0.0$, $P = 1$). Individual body length did not influence response ($F_{1,29} = 0.0$, $P = 0.887$) but run number (a correlate of temperature and predator model) had a nearly significant effect ($F_{3,3} = 8.2$, $P = 0.058$). Linear regression showed that opercular response was related to predator model size ($F_{1,37} = 14.0$, $P = 0.001$), such that response magnitude was greater with larger models. A linear regression with temperature was also significant ($F_{1,37} = 9.03$, $P = 0.005$); the trend was for opercular response to increase with decreasing temperature. There was no difference in the variance of opercular rates of wild and hatchery fish ($F_{2,36} = 0.60$, $P = 0.277$).

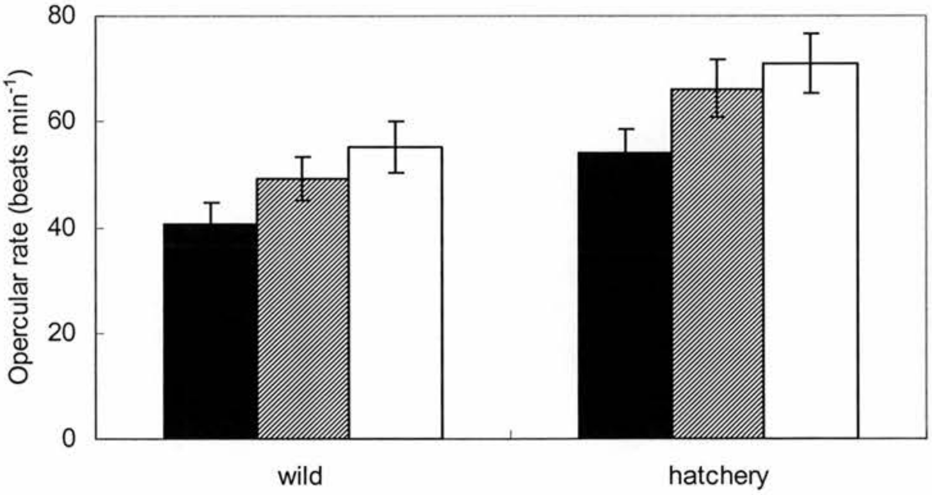


Figure 1. Mean opercular rate of wild and hatchery 'freezers' before (■) compared to mean opercular rate after (▨) and maximum rate after (□) exposure to predator model.

The maximum opercular rate recorded for each fish during the 4 min post-exposure was also analyzed. The maximum response of wild 'freezers' was significantly lower than that of hatchery 'freezers' (t_{36} = 4.2, $P < 0.001$), mirroring differences in resting opercular rate. However, the maximum opercular rate of wild 'freezers' after exposure was, on average, 37.3 ± 10.0 % greater than resting opercular rate, whilst hatchery 'freezers' increased by 33.3 ± 12.0 % (Fig. 1) - this relative difference between wild and hatchery 'freezers' was not significant (ANOVA, $F_{1,3} = 1.6$, $P = 0.302$). Body size did not influence the opercular response ($F_{1,29} = 0.4$, $P = 0.540$) but run number did ($F_{3,3} = 14.4$, $P = 0.026$).

Dashing had a strong effect on opercular rate increase. Wild 'dashers' showed a greater maximum opercular rate than wild 'freezers' (mean ± 95% CI; dashers: 80.8 ± 9.6 beats min^{-1}; freezers: 55.2 ± 2.5 beat min^{-1}; $t_9 = 4.7$, $P = 0.001$). Similarly, hatchery 'dashers' showed a greater maximum rate than hatchery 'freezers' ('dashers': 92.9 ± 11.5 beats min^{-1}; 'freezers': 71.1 ± 2.8 beat min^{-1}; $t_{12} = 3.5$, $P = 0.004$) (Fig. 2). The maximum opercular rate was similar for wild (N = 7) and hatchery (N = 9) 'dashers' (two-sample *t* test; $t_{13} = 0.2$, $P = 0.127$; Fig. 2).

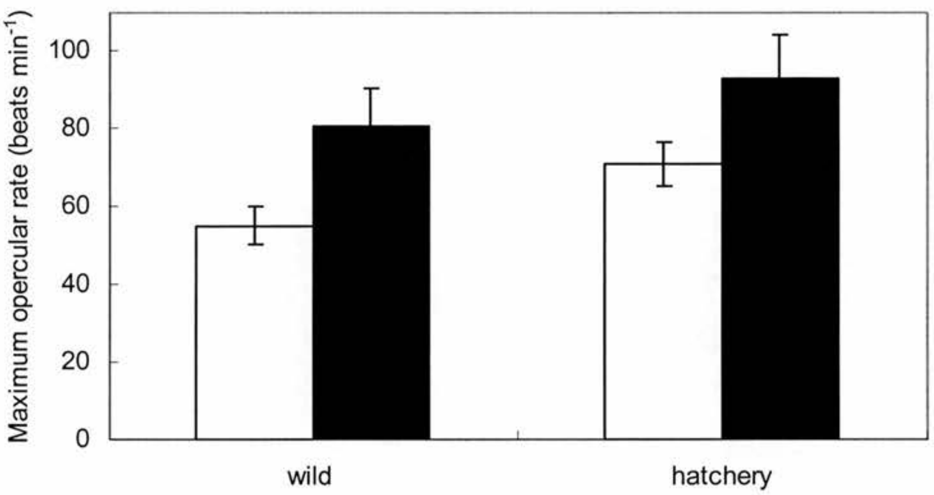


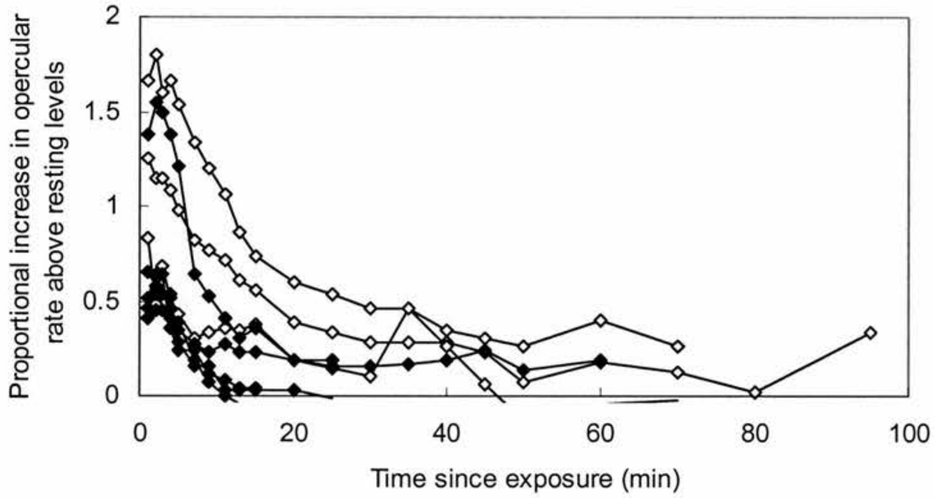
Figure 2. Maximum opercular rate after presentation of a predator model of wild and hatchery fish that exhibited dashing ('dashers'; ■) compared to individuals that remained still ('freezers'; □).

Duration of response

Exposure to the model initiated a rapid and dramatic increase in opercular rate, followed by a progressive decline in rate. The time taken for opercular rate to decline to pre-exposure levels was calculated for runs three and four (N = 11 wild and N = 14 hatchery fish). Pre-exposure rate was defined as being when opercular frequency was within 2 beats min^{-1} of the mean pre-exposure rate and did not decrease in the following count.

'Dashers' (N = 3 wild, N = 5 hatchery) had elevated opercular rates for (mean ± SD) 38.2 ± 29.8 min (Fig. 3(a)), whereas 'freezers' (N = 8 wild, N = 9 hatchery) showed a return to resting opercular rate in 7.2 ± 6.1 min (Fig. 3(b)). The response pattern was qualitatively similar for wild and hatchery 'freezers', half of which recovered within 5 min.

(a)



(b)

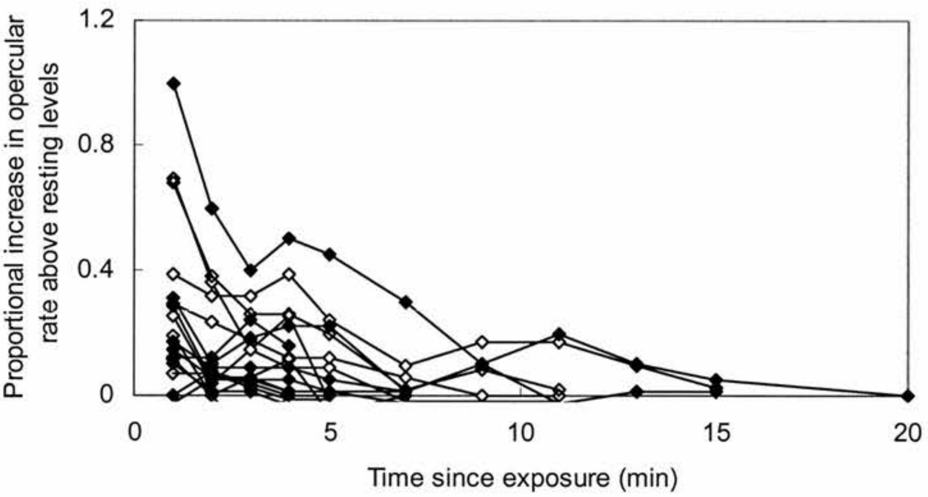


Figure 3. Elevation in opercular rate after exposure to a predator model in (a) fish that exhibited dashing (N = 8) and (b) fish that remained still (N = 17), for wild (◇) and hatchery fish (◆).

There was a strong linear correlation between time taken for opercular rate to return to resting levels and the maximum increase in opercular rate for wild and hatchery 'freezers' (ANCOVA; $F_{14,17} = 4.0$, $P = 0.033$; Regression equation: Duration = 0.103*(maximum increase (%)) + 4.174). There was no difference between wild and hatchery fish in this relationship ($P = 0.705$) and there was no effect of run number ($P = 0.205$).

Initiation of response

Wild fish showed a trend of maximum opercular rate in the first 30 s after exposure followed by a significant progressive reduction in rate over the next 3 min (Repeated measures ANOVA; $F_{2,17} = 7.8$, $P = 0.004$); this trend was not apparent in hatchery fish ($F_{2,18} = 1.2$, $P = 0.334$). This was because six out of 20 hatchery fish showed a delayed response to the model, such that their maximum response occurred either 60-90, 120-150 or 180-210 s following exposure: In comparison a delay was observed in one of 19 wild fish (2-tailed Mann-Whitney; Z = 2.0, $P = 0.043$). Because of this effect the initial opercular response of hatchery fish (first 30 s post-exposure) was significantly lower than their maximum rate during the 4 min post-exposure period (paired t test; $t_{19} = 2.6$, $P = 0.018$) but not for wild fish ($t_{18} = 1.7$, $P = 0.104$). Consequently, the reduction in wild fish opercular rates were significantly greater than seen in hatchery fish throughout the 4 min post-exposure period (two-sample t test; $t_{36} \leq 2.0$, $P \leq 0.050$, for each min post-exposure).

DISCUSSION

Atlantic salmon showed a large (35.3 ± 11.0%) increase in ventilation rate after exposure to a predator model. Significantly, the ventilation response was delayed in 30% of hatchery-reared fish compared to only 5% of wild fish of the same parentage. However, the peak and duration of ventilatory response was remarkably similar between wild and hatchery individuals. Some fish (wild and hatchery) exhibited dashing in response to the predator stimulus whilst others showed no behavioural response. An increase in ventilatory rate occurred both in the presence and absence of dashing behaviour, but the extent and time course of the ventilation response was strongly affected by this behaviour.

An increase in gill ventilation following dashing is readily explained by elevated metabolic rate during repayment of the oxygen debt that follows anaerobic activity (Olson 1998). The rapid increase followed by a more gradual decline in ventilatory rate is a typical response to burst activity (Beamish 1978).

The function of the increase in gill ventilation of those fish that froze following exposure to the predator model is less clear. This response may be in preparation for possible subsequent activity, for example, if the perceived predator attacked. This explanation has been favoured by a number of workers studying responses of cardiovascular and respiratory systems of other taxa to predator exposure (Hickam *et al.* 1948, Causby & Smith 1981, Espmark & Langvatn, 1985, Ydenberg & Dill 1986, Lydersen & Kovacs 1995). However, escape bursts could be undertaken anaerobically and there is clearly not a requirement for the fish to be physiologically geared up, although it is possible that efficiency of escape might be enhanced by preparation. Increased ventilation frequency also has potential costs. First, the metabolic costs of ventilating the gills are high and during ventilation may constitute 25% of the total metabolic rate of a fish (Hughes & Shelton 1962). Secondly, movements of the operculae possibly make the salmon more visible to potential predators and this might be considered a cost. Some fish respond to presence of predators by reducing physiological activity to increase crypsis (Hanlon *et al.* 1999, Stoddard 2002, Losey 2003); notably, fish exhibit bradycardia (a common response of fish to aversive stimuli; Labat 1966) and so may reduce electrical activity, which is used by hunting sharks to locate their prey (Montgomery 1988).

Alternatively, it is also possible that far from being a cost, in terms of attracting predators, heightened gill ventilation is associated with fish actively signalling their presence to the perceived predator, to convey that the predator has been spotted (Hasson 1991, Holley 1993). Signalling awareness may act to deter the predator from pursuing the potential prey (Ap Rhisiart 1989, Godin & Davis 1995, but see Milinski & Boltshauser 1995) because some predators are less successful in attacking prey that are aware of their presence (Webb 1982, FitzGibbon 1989). Furthermore, opercular movements may signal to conspecifics the presence of danger, similar to fin-flicking reported in glowlight tetras, *Hemigrammus erythrozonus* (Brown *et al.* 1999). The sight of fin-flicking results in conspecifics increasing their anti-predator responses. Interestingly, some of the salmon exhibited rocking behaviour, whereby they rocked from side to side on their pectoral fins. This behaviour could also subtly increase visibility of the prey to the predator. Rocking has been observed previously as a response to pain when fish have been injected with bee venom (Sneddon *et al.* 2003b). However, in this study the behaviour was more generally exhibited by fish and may have a function in the context of response to perceived predators.

Hatchery-reared and wild fish showed similar overall magnitudes of increase and time-courses of recovery of ventilatory rate following exposure to the predator model. Therefore, the apparently large response observed by Metcalfe *et al.* (1987) was not simply overreaction by naïve hatchery-reared fish but a true response to predator threat. There was, however, the small, but possibly important difference that it took longer for the hatchery-reared fish to increase ventilation rate to a peak. The heart rate responses of wild- and hatchery-reared brown trout also differ immediately after presented with a predator cue (for the first 20 s), but after this time period, heart rate responses of the two groups parallel each other (Anon 2002).

There is evidence for both innate (e.g. Gerlai 1993, Huntingford & Wright 1993, Brown & Warburton 1997, Jordão & Volpato 2000, Utne-Palm 2001, Barreto *et al.* 2003) and learnt (e.g. Jarvi & Uglem 1993, Chivers & Smith 1994, Brown *et al.* 1997, Brown & Smith 1998, Brown & Warburton 1999, Utne-Palm 2001) visual predator recognition abilities in fishes. Together, these studies suggest the existence of genetic capabilities that may require fine-tuning by experience (e.g. Pfeiffer 1963, Waldman 1982, Magurran

1989, Fuiman & Magurran 1994), hence anti-predator skills may be phenotypically plastic (West-Eberhard 1989, Thompson 1991, Holopainen *et al.* 1997). The finding that some hatchery salmon have a delayed response compared to wild salmon may be taken to suggest that experience of predators (which is denied in hatchery-rearing) enhances existing predator recognition capabilities such that the ventilatory response is more rapid. Similarly, anti-predator responses may be most appropriate in those individuals that have experienced predators (e.g. Brown & Warburton 1999, Kelley & Magurran 2003) or experience of predators might strengthen responses (Magurran 1990). However, an alternative (and not mutually exclusive) explanation for the results observed is that selective predation mortality removed those wild fish with relatively delayed ventilatory responses (Johnsson *et al.* 2001, Kelley & Magurran 2003). It is also possible that hatchery reared fish have been perturbed by general maintenance activities (e.g. cleaning tanks), which reduced their general vigilance (Johnsson *et al.* 2001). Also, to the contrary, it may be argued that these disturbances have allowed fish to practice their fright responses and anti-predator skills (Kelley & Magurran 2003).

Hatchery-reared salmon had a significantly higher resting opercular rate than wild-reared salmon. This may indicate a difference in metabolic rates due to the rearing environment. Such a difference may relate to capacity to process food, which could be higher in the food-rich hatchery environment. Other differences between the habitats may also have an effect by influencing the activity levels of the fish. It is also conceivable that there is a tendency for individuals with relatively high metabolism rates to suffer relatively high mortality in the wild.

At present, there is little evidence to suggest that hatchery-reared progeny of wild parents suffer higher mortality than wild fish of the same parentage specifically due to a lack of appropriate anti-predator skills. However, this study indicates a difference in the ventilatory response between these groups of fish that may have a role to play in predator avoidance in the few days after stocking.

SECTION II

Concluding remarks

(i) Use of multiple sensory cues

Olfactory recognition of pike may be paramount for prey fish, as pike are ambush predators which may provide little in the way of visual cues. Pike spend long periods of time in restricted areas with little water exchange (Raat 1988), thus localized olfactory cues may provide reliable information to prey fish on the potential danger nearby. However, for many other predator species, such as active foragers and species that alter hunting strategies to suit environmental conditions, cues from multiple sensory modalities will potentially be available.

It seems obvious that prey will incorporate information on potential risk gained from different sensory systems because a more detailed picture can be built up. Until recently, however, there have been few investigations into how prey fish integrate information from multiple senses, with the exception of marine fish larvae (e.g. Fuiman & Magurran 1994, Higgs & Fuiman 1996, Poling & Fuiman 1997). Nonetheless, visual and olfactory cues are relatively easy to study in the laboratory, and there is increasing information on how these two sensory modalities complement each other.

Firstly, different sensory modalities may be relied on under different environmental conditions. Hartman & Abrahams (2000) demonstrated that when the primary source of predator information for minnows (visual) is reduced, they are more willing to use alternative (olfactory) cues. This sensory compensation (*sensu* Hartman & Abrahams 2000) may occur when fish change between different habitats; for example, olfaction may be of greatest use in turbid systems (Waldman 1985) or where visibility is poor (e.g. structured habitats; Chivers *et al.* 1997).

Secondly, multiple senses may be required to detect the whole suite of predatory species. For example, olfaction may be essential to detect slow moving, inconspicuous, cryptic or sit-and-wait (e.g. pike) predators (Fuiman & Magurran 1994, Hirvonen *et al.* 2000), but vision may be essential to detect fast-moving predators (Hirvonen *et al*. 2000).

Thirdly, different sensory modalities may be used sequentially when evaluating predation risk. For example, in weak flow regimes, chemical cues may allow prey to recognise potentially high-risk habitats (Chivers & Smith 1995) or predator feeding grounds, due to their considerably longer efficacy than visual cues (Brown *et al*. 1997). However, visual cues may secondarily be required to evaluate actual risk (Chivers *et al*. 2001), such as to assess risk based on predator body size (Chivers *et al*. 2001, Lehtiniemi 2005), to identify the specific location of the predator, or indeed to confirm whether the predator is still present (Liley 1982, Wisenden 2001). Thus cues detected initially may prime the fish for subsequent stimuli (Wisenden *et al*. 2004) and may increase vigilance (Brown *et al.* 2004). Both visual and chemical cues may be required to invoke the full anti-predator response of a prey fish (e.g. Brown *et al*. 2004, Wisenden *et al*. 2004, Lehtiniemi 2005).

Conversely, a visual predator cue alone may not demonstrate sufficient risk to invoke an anti-predator response. For example, the predator may not be hunting or may not have detected the prey. Although vision can provide information such as motivation of the predator to strike, through posture (Licht 1989) or behaviour (Murphy & Pitcher 1997), some behaviours may be unreliable as they could be manipulated by the predator (Brown *et al*. 2000). Predator odour cues combined with a visual sighting may confirm actual risk (e.g. Brown & Godin 1999) through the presence of conspecific AS in its dietary odour cue (Wisenden *et al.* 2004). Furthermore, if a novel species is sighted, dietary cues can confirm whether the species is dangerous (Wisenden 2001) and may thus allow learnt recognition of predator visual cues (Chivers & Smith 1994a, Brown & Godin 1999).

(ii) Conflicting cues and context-dependence

Response to predator cues depends not only on the sensory modality in which it is received (Hartman & Abrahams 2000), but also on the context in which it is perceived (Magurran *et al.* 1996, Irving & Magurran 1997) and the motivation (Murphy & Pitcher 1997, Höjesjö *et al.* 1999) and past experience of the recipient (Kieffer & Colgan 1992, Healy & Reinhardt 1995). Prevailing conditions, such as habitat (and availability of shelter) and presence of conspecifics may influence the actual predation risk for a given predator cue, and this may influence the magnitude of the anti-predator response given (Magurran *et al.* 1996, Irving & Magurran 1997).

This has strong implications regarding inferences made about behaviour in the wild based on behaviour observed in the laboratory. Firstly, anti-predator responses may be greater in the context of the laboratory environment because there is seldom any escape or shelter for prey fish, and therefore risk of predation would be perceived as greater relative to the natural environment. For example, AS invokes a strong fright response by fish in the laboratory, but under wild conditions it invokes weaker (Wisenden *et al.* 2004) or no (Magurran *et al.* 1996) responses.

Secondly, under natural conditions, prey fish may receive information regarding predation risk from multiple senses (vision, olfaction, mechanoreception). However, under laboratory conditions, typically a fish is presented with a cue to one sensory modality. This may not be sufficient to interpret predation risk, and therefore anti-predator responses may not reflect natural behaviour.

Thirdly, the state of the prey fish may also influence its response to predator cues. Brown & Smith (1996) observed no fright response in fasting minnows, which they suggested was due to the trade off between foraging and anti-predator responses. However, in the laboratory, food is generally readily supplied, and therefore fish do not experience the foraging trade-offs of wild fish.

Laboratory investigations into anti-predator behaviours should aim to minimize the problems outlined above. This requires the provision of appropriate environmental conditions, including suitable habitat for the species, the presence of conspecifics, if appropriate, and natural lighting conditions. Food supply is important in two aspects. Firstly, food supply should be appropriate to the foraging behaviour of the species (e.g. substrate-, drift- or surface-feeding), as anti-predator and foraging behaviours are often linked (Ydenberg 1998). Secondly, consideration should be given to the quantity of food supplied before and during experiments, as this may influence the trade-off between foraging and predator avoidance (Lima & Dill 1990). These same considerations should also be given to live predators used in experiments, especially the influence of stress on feeding motivation. Finally, to achieve a realistic situation, more than one predator cue may need to be provided, as anti-predator responses may be influenced by the type of cue, whether other cues can confirm or contradict risk, and the order in which cues are supplied.

SECTION III

CHAPTER FIVE

Sensitive periods for acquired predator recognition in hatchery-reared salmon

ABSTRACT

Atlantic salmon demonstrated innate predator recognition skills which were retained for at least eight months in a hatchery environment. Salmon aged *c.* three months old showed a greater response to predator odour than younger or older fish, suggesting a period of heightened receptivity to predator odour cues. It was hypothesized that this age group showed enhanced receptivity to cues because it corresponds to an ontogenetic period when predator encounters in the wild are much greater than would previously have been experienced. This age group also showed learnt predator recognition abilities, in contrast to younger fish that relied on innate responses. It is suggested that this represents a sensitive period, during which fish are predisposed to improve existing predator recognition skills through learning.

INTRODUCTION

Atlantic salmon have innate predator recognition skills (chapter two). The progeny of wild Atlantic salmon have reduced anti-predator skills if reared in a hatchery environment rather than in the wild (Järvi & Uglem 1993, chapter four). It has been hypothesized that the absence of predators in the hatchery environment denies fish the opportunity to develop existing, or acquire learnt, anti-predator skills (Kieffer & Colgan

1992, Olla *et al.* 1992, Dellefors & Johnsson 1995). Furthermore, in an environment where no anti-predator skills are required, existing responses may be lost over ontogenetic time (Griffin *et al.* 2000, Mirza & Chivers 2000, Brown & Laland 2001).

Many species of salmonids, including Atlantic salmon, show improved anti-predator skills after experience with predators (e.g. Kieffer & Colgan 1992, Järvi & Uglem 1993, Berejikian 1995, Healy & Reinhardt 1995). Salmonids can also acquire anti-predator skills through releaser-induced recognition learning (Suboski 1990). In this instance, learning is stimulated by exposing a fish to a predator cue combined with an aversive cue (i.e. one that elicits an innate fright response), resulting in the fish learning that the predator cue is aversive (e.g. Brown & Smith 1998, Berejikian *et al.* 1999, 2003, Mirza & Chivers 2000). For example, alarm substance (AS) is a chemical contained in epidermal cells and is released only if the skin is damaged; for example, when a predator attacks the fish. AS is an aversive cue which triggers an innate fright response in salmonids (Mirza & Chivers 2001), and indeed many other fish families (Chivers & Smith 1998).

It has been suggested that the abilities of fish to learn could be used to compensate for the lack of experience as a result of rearing in a hatchery environment (Suboski & Templeton 1989, Brown & Laland 2001), and potentially to increase survival of hatchery-reared fish released into the wild (Berejikian *et al.* 1999, 2003, Mirza & Chivers 2000). This has been termed 'life skills training' (Suboski & Templeton 1989). One aspect of this would be to improve the response to predators of hatchery-reared fish.

It may be possible to enhance existing (innate) predator recognition abilities of fish through learning (Magurran 1990, Griffin *et al.* 2000, Berejikian *et al.* 2003). Although previous work (chapter two) found no effect of releaser-induced recognition learning on the response to predator odour in newly-hatched Atlantic salmon (< 40 days old), such training techniques require individuals to be predisposed to learn (Griffin *et al.* 2000). I propose that learned enhancement of predator recognition may occur at ecologically appropriate periods during ontogeny of salmon, similar to the capacities of birds for song learning (Marler 1970) and filial imprinting (Gottlieb 1961), which are dependent on the developmental state of the bird (Bateson 1979). There must be a functional requirement for learning to occur in a particular developmental period, as opposed to showing continuous predispositions to learn (Bateson 1979). Therefore I suggest that salmon will show enhanced predator recognition abilities during periods when predation risk is likely to be high.

In this study I first measured how the response to predator odours of hatchery-reared (progeny of wild fish) Atlantic salmon changed during ontogeny. Ventilation (opercular) rate was used to measure the response of juvenile salmon. The first hypothesis was that (innate) response to predator odour would decline during hatchery rearing due to a lack of reinforcement of predation threat. Secondly, I hypothesized that response to the odour of a low-risk predator (a small, generalist fish species) would decline more rapidly during development than the response to a high-risk predator (hypothesis two), because vulnerability to the former would decline with increasing prey body size. In the second part of the study, I looked at whether releaser-induced recognition learning could enhance existing responses to predator odour at different ages. I hypothesized that fish would be primed to learn predator recognition during an ontogenetic stage when predation risk is likely to be high (hypothesis three).

METHODS

Salmon

All salmon used in the experiments were progeny of adults captured in the River Almond, Perthshire, Scotland, 56°43'N -3°53'W. Eggs/fish were hatchery-reared at the Fisheries Research Services Almondbank field station, Perthshire, Scotland. Eggs/fish were kept in a flow-through system with water from their natal river at ambient temperature. Fish used for experiment one (conducted in 2004) were the progeny of six parent crosses spawned in December 2003 and hatched 10 – 12 March 2004. Experiment two (conducted in 2005) used progeny from four parent crosses that were spawned in December 2004 and hatched 28 February 2005.

(i) Innate responses during captive-rearing

I measured the opercular response of Atlantic salmon at three ages of development to three odours. The ages tested were 17 – 25 d old (category I; yolk-sac feeding alevins); 74 – 103 d old (category II); and 194 - 252 d old (category III). The odours tested were pike odour (PO; a high-risk predator), minnow (*Phoxinus phoxinus*) odour (MO; a low-risk predator) or a control odour (CO; the local river water). Each fish was tested at only one age and exposed to only one odour; in total, 255 fish were used (Table 1). The three odours were tested sequentially during the experiments. Experiments were conducted between 28 March - 5 April (category I; Fork length, L_F *c*. 22 mm), 24 May - 24 June (category II; 36.9 ± 1.4 mm (mean ± 95% CI)) and 20 September – 17 November (category III; 77.36 ± 2.35 mm).

Table 1. Number of individuals tested in the nine treatment groups.

Age (days) Odour	17 - 25	74 - 103	194 - 252
Pike; PO	30	22	30
Minnow; MO	30	22	28
Control; CO	41	23	29
Total per age group	101	67	87

Predator odours

The three odour cues were water samples from tanks containing pike, minnows or no fish. No pike have been recorded in the R. Almond, from which the hatchery water is drawn; thus all salmon were naïve to pike odour. In total, seven pike (437.3 ± 189.1 g; mean ± 95% CI) and 47 minnows (3.0 ± 0.4 g) were used to obtain odour. The stocking levels of pike and minnows were chosen to equalize biomass in both species tanks to 2.6 g l^{-1}. To eliminate piscivorous dietary cues, pike were fed earthworms for 2 weeks prior to and during experiments. Minnows were fed bloodworms. All three tanks were filled with hatchery water and had similar oxygen supply and filtration, and partial water changes were carried out every 3 d.

Protocol

To test category I, individual alevins were chosen at random and pipetted into a section of transparent tubing (1 cm diameter), which was drip-fed river water via a peristaltic pump. After 2 min acclimatization, opercular rate was recorded for two 30 s counts (pre-stimulus opercular rate). Subsequent counts were done if the two counts differed by > 1 beat 30 s^{-1}, until opercular rate had stabilized The water supply was then switched to a beaker containing either PO, MO or CO. This new water supply was delivered for 30 s. Opercular rate was then measured for one further 30-s period (post-stimulus opercular rate).

Categories II and III were tested in transparent observation chambers (150 x 100 x 100 mm; Fig. 1). Three sides of the chambers were covered in black plastic and the fourth was used for observations. An individual fish was gently netted from one of the rearing tanks and placed into a chamber in the late afternoon, and left approximately 18 h to settle. The chamber had one inlet (1 cm diameter) and two outlets. Hatchery water held in a separate tank was re-circulated through the observation chambers during this period.

The following morning one 12 l tank was filled with CO and a second 12 l tank was filled with PO, MO or CO. The water supply for the chamber was then switched to the first tank containing CO. This was pumped into the chamber through tubing connecting to one of the chamber's outlets. 30 min was allowed for the fish to adjust to the directional change in water flow. The opercular rate of the fish was then measured for three 30 s counts (pre-stimulus rate), or until there was ≤ 1 beat 30 s^{-1} difference between counts. Then a valve shut off the water supply from the first tank and water was pumped from the second tank (PO, MO or CO) to the chamber via the second outlet. After 45 s I measured the opercular rate of the fish again, for three 30 s counts (post-stimulus rate). Experiments were conducted in a dark room with a single light source (40 watts) focused on the observation chamber. The observer was behind the light source and therefore not visible to the fish.

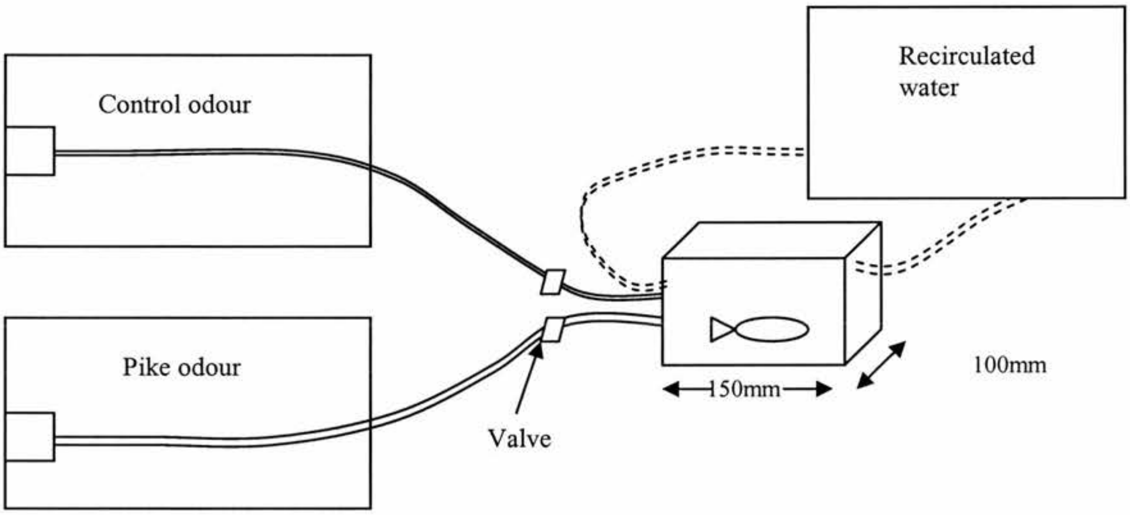


Figure 1. Observation chamber for testing categories II and III.

Data analysis

Pre- and post-stimulus opercular rates for each category were normally distributed (Anderson-Darling; $P > 0.063$); therefore parametric analysis of covariance (ANCOVA) was used. The post-stimulus opercular rate was the dependent variable, the pre-stimulus opercular rate was the covariate, and the type of odour and category were fixed factors. *Post-hoc* comparisons of groups used Tukey's Honestly Significant Difference (HSD) test for unequal N. Figures were produced using data on proportional changes between pre- and post-stimulus opercular rates. These data were not normally-distributed and are therefore presented as median ± IQ range.

The effect of body length was tested in separate ANCOVAs for categories II and III (L_F; included as a random factor). An additional analysis was done for category III because their size-distribution was bimodal, comprising larger (S1) individuals that were preparing to smolt (change to the marine form) at 1+ year, and the remaining smaller (S2) individuals that would smolt at 2+ years or later (Thorpe 1977, Thorpe & Metcalfe 1998). Size-frequency distributions were produced for each fish family using additional fish sampled at random from the stock tanks (N = 60 - 90 per family). The fish used in the experiment were then compared to their family size-frequency distribution, and assigned to either an S1 or S2 group. 12.6% (N = 11) individuals of intermediate size could not be assigned to either groups and were excluded from this analysis. Size (S1 or S2) was included as a random factor in ANCOVA.

(ii) Learnt predator recognition

This experiment was designed to test how the response to PO was affected by prior exposure to cues which may stimulate learning. The cues used for prior exposure were PO and a solution of salmon skin extract (SSE). Previous trials showed that SSE elicits a strong opercular response in salmon on the first encounter (chapter 2). PO + SSE were presented to the fish simultaneously. 7 d later I measured the response to PO presented alone. The effect of prior exposure was tested at two ages: (1) 25 – 26 d old (category I; exposed 18 March), and (2) 109 - 132 d old (category II; exposed 17 June – 8 July), approximately corresponding to categories I and II in the first experiment.

Preparation of chemical cues

PO was obtained from one tank holding two mature female and two mature male pike (age 3+; 698 ± 75 g; mean ± SD). In line with previous experiments, a sample of water taken from the tank was diluted to obtain a concentration of 2.6 g l^{-1}.

SSE was prepared the morning of use, with skin removed from salmon parr (age 1+ fish) from two different families of the same river population as tested fish. For category I, one batch of SSE was prepared (as all fish were exposed on the same day) with skin removed from two salmon parr (L_F 97 and 115 mm). The skin was homogenized with 70 ml of water and the solution was filtered through wool. Two 20 ml samples of solution were pipetted out and used for the prior exposure. For category II, SSE was prepared a total of nine times (four fish were exposed at one time). Each batch of SSE used skin of a single parr (98.8 ± 10.6 mm) which was homogenized with 50 ml of water and then filtered through wool.

Prior exposure to learning cues

Alevins (category I) were placed into egg boxes (10 x 10 x 15 cm) 16 d after hatching. Four egg boxes were each filled with 70 alevins and placed in a single tray in the hatchery. Two days later the egg baskets were placed in separate tanks filled with 9 l of river water and oxygenated by an air stone. After a 2 h acclimatization period the prior exposure cues were added. Two tanks received 200 ml PO + 20 ml SSE (treatment group) and two tanks received 220 ml of distilled water (DW) (control group). After 30 min the egg baskets were returned to the hatchery tray.

Four fish per day from category II were selected at random from a stock tank in the afternoon and placed in a mesh basket (10 x 10 x 18 cm) suspended in a tank filled with 200 l of hatchery water on a through-flow system. The prior exposure cues were added the following day at noon. These were either 400 ml PO + 50 ml SSE (treatment group) or 450 ml DW (control group). After 2 h the basket was placed into an identical tank for a further 6 d. Presentation of the treatment and control cues was alternated daily and control and treatment groups were each held in two identical tanks.

Testing response to PO

The response of category I to PO was tested 7 - 8 d after prior exposure to the learning cues. Tests used the same apparatus as the first experiment. The protocol differed slightly from previously. After opercular rate had stabilized, the water supply was switched to a second beaker also containing CO. After 30 s the opercular rate was measured again (one 30 s count). The water supply was then switched to a beaker containing PO and after 30 s the opercular rate was measured a third time (one 30 s count). The increase in response to PO was compared with the increase in response to the CO for each individual. N = 30 alevins from each prior-exposure group (control or treatment) were tested.

Category II individuals were tested 7 d after prior-exposure to the learning cues. Tests used the same apparatus as the first experiment. Individual fish were placed into observation chambers the afternoon before testing. The following morning opercular rate was measured at least three times to obtain a stabilized opercular rate. The water supply of the chamber was then changed to a tank containing CO. After 30 s, opercular rate was measured in two 30 s counts. The water supply was then changed to a tank containing PO. After a further 30 s, opercular rate was measured again for two 30 s counts. The increase in opercular rate in response to the CO was compared with the increase in rate in response to PO for each individual. N = 27 fish from each prior-exposure group (control or treatment) were tested.

Data analysis

Pre- and post-stimulus opercular rates for each category/prior-exposure group tested were normally distributed (Anderson-Darling; all $P > 0.062$). ANCOVA used the opercular rate after addition of PO as the dependent variable and opercular rate after addition of CO as the covariate. Category (I or II) and prior-exposure group (treatment or control) were fixed factors. The effect of L_F for category II was tested in a separate ANCOVA. Figures were produced using percentage differences between pre- and post-stimulus opercular rates. This data was normally-distributed and therefore presented as mean ± 95% CI.

RESULTS

(i) Innate responses during captive rearing

Opercular response depended on odour type presented (pike, PO; minnow, MO; control, CO; ANCOVA: $F_{2,254} = 57.71$, $P < 0.001$) and the age of the fish (17 - 25, 74 - 103, 194 – 252 d; corresponding to categories I, II and III; $F_{2,254} = 16.84$, $P < 0.001$, Fig. 2). There was a significant interaction between category and odour type ($F_{4,254} = 6.98$, $P < 0.001$).

The response to PO was greater than the response to MO and CO (HSD post-hoc test; $P < 0.003$), with the exception that category I showed a similar response to PO and MO ($P = 1.000$). Responses to MO were greater than to the CO ($P < 0.001$) with the exception that the response of category III to MO and CO were not significantly different ($P = 0.679$).

Fish at all ages responded to MO, with opercular rate increasing between 2.1 (0.0 – 6.1) and 3.5 (1.7 – 7.7) % {median (IQ range)}. The response to PO was similar for categories I and III {increase of 4.0 (2.1 – 7.7) and 3.7 (0.5 – 9.4) %, respectively}. However, category II showed a significantly greater response to PO {10.0 (4.4 – 26.5) %; $P < 0.001$}.

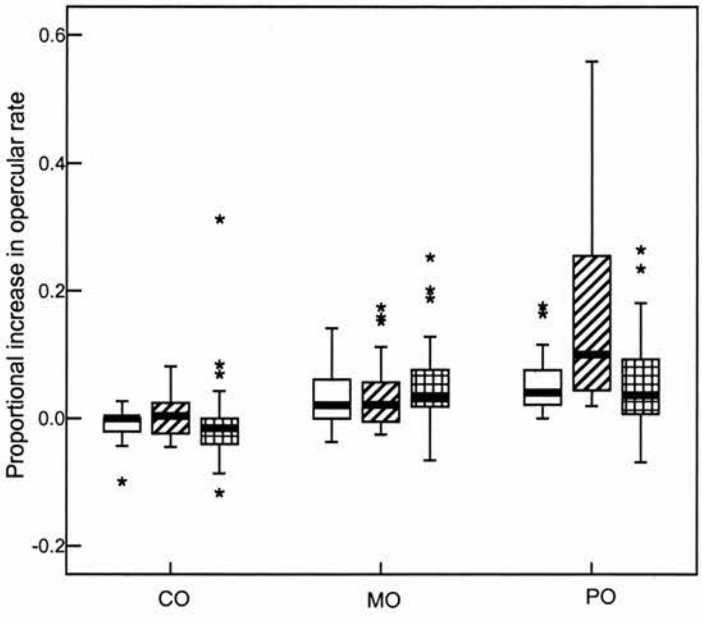


Figure 2a. Increase in opercular rate by categories I (17 – 25 d old; □), II (74 – 103 d old; ▨) and III (194 – 252 d old; ▦) in response to control odour (CO), minnow odour (MO) and pike odour (PO). Boxes represent IQ range, horizontal lines represent median values, whiskers represent < 1.5*IQ range and asterisks (*) represent ≥ 1.5*IQ range.

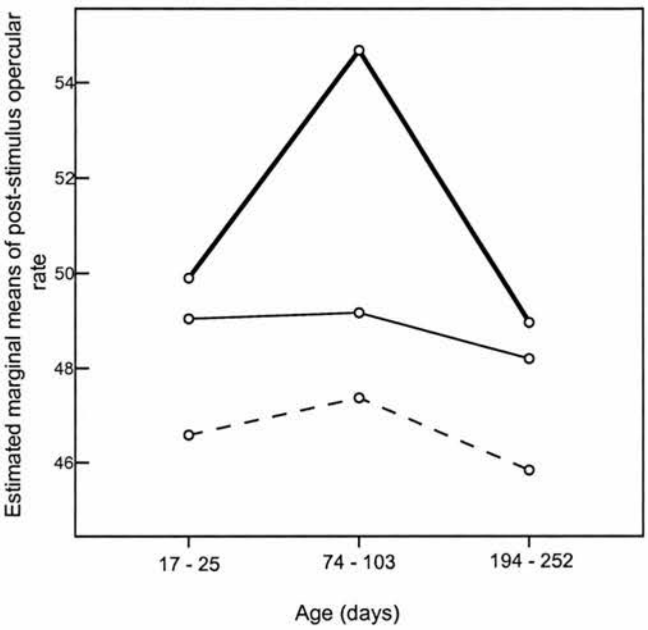


Figure 2b. Profile plot of the estimated marginal means of the post-stimulus opercular rate (adjusted for pre-stimulus opercular rate) to each odour type (control odour, …; minnow odour, —; pike odour, —) by each age group.

There was no effect of body size on the opercular response in categories II and III (ANCOVA; $P > 0.372$) and there was no interaction between size and odour type ($P > 0.264$). There was also no difference in response between large (S1) and small (S2) individuals in category III ($P = 0.320$) and no interaction with odour type ($P = 0.659$).

(ii) Learnt predator recognition

The response to PO differed between categories I (17 – 25 d old) and II (109 – 132 d old) (ANCOVA; $F_{1,113} = 24.24$, $P < 0.001$). There was no significant difference between prior-exposure groups (control and treatment; $F_{1,113} = 1.75$, $P = 0.189$); however there was a significant interaction between category and prior-exposure group ($F_{1,113} = 7.54$, $P = 0.007$).

Analysis of the categories separately showed that this significant interaction occurred because the response of category I individuals was similar in the control and treatment groups (ANCOVA; $F_{1,59} = 1.52$, $P = 0.223$) but the response of category II individuals was significantly greater in the treatment group compared to the control group ($F_{1,53} = 5.93$, $P = 0.018$; Fig. 3). There was no effect of body size on response of category II fish ($P = 0.130$) and no interaction with prior-exposure group ($P = 0.758$).

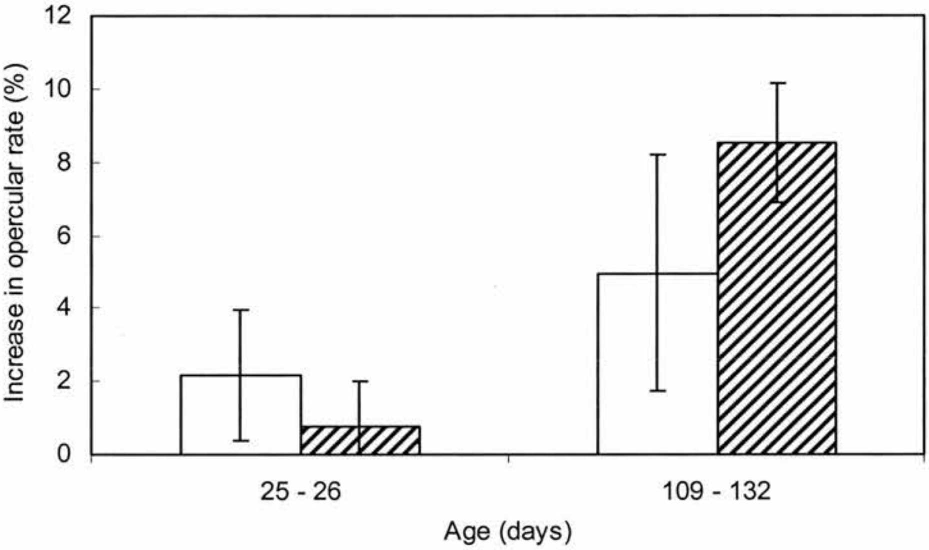


Figure 3. Response to pike odour (PO) by salmon at 25 – 26 d old (category I) and 109 – 132 d old (category II). Treatment groups (▨) were exposed to PO + salmon skin extract (SSE) prior to testing. Control groups (□) were exposed to distilled water prior to testing. Error bars represent 95% confidence limits.

DISCUSSION

Juvenile salmon showed an innate response to predator odour which was retained for eight months without reinforcement. An enhanced innate response to pike odour by fish 10 - 14 weeks old (category II) suggested a period of heightened receptivity to predator odour cues. I hypothesized that this age is a sensitive period during which salmon can develop and refine predator recognition skills, and in support of this contention it was shown that experience with pike odour and alarm substance (AS) led to an enhanced subsequent response to pike odour in category II, but not in category I (*c.* 3 weeks old).

A significant and similar response to pike odour was shown in categories I and III of development, demonstrating that innate predator recognition was not lost during eight months of rearing in a predator-free environment (refuting hypothesis one). There was no difference in the response to minnow and pike odours by alevins (category I), which perhaps reflects the fact that at this stage in their life, salmon may be equally vulnerable to minnows or pike because of their small size. In any case, alevins in natural systems live mainly within gravel and are probably well protected from most fish predators in most circumstances. Categories II and III showed weaker responses to minnow odour than pike odour, suggesting that these fish were able to distinguish between risks of different predator species, which was also shown in chapter two. Category III showed no significantly greater response to minnow odour than control odour, which supports hypothesis two (response to the gape-limited minnow declines quicker than the response to the pike because salmon out-grow vulnerability to the former).

The heightened receptivity to pike odour in category II may facilitate the acquisition of a conditioned response (Morin *et al.* 1989a). The third hypothesis predicted the existence of an ontogenetic sensitive period, during which the capacity to learn predator recognition would be enhanced. These results suggest that this period occurs in category II. It has been suggested that sensitive periods should coincide with a developmental stages at which recognizing and remembering predator cues are particularly important (Bateson 1979).

Why should salmon be particularly responsive to learning predator cues in category II? An important ontogenetic niche shift occurs between categories I and II. Yolk sac-feeding alevins (category I) live within the interstitial spaces in the substratum of river beds, where they are protected from most fish predators. However, once yolk-sac consumption is almost complete (at 4 – 8 weeks old, but highly temperature-dependent (M. Miles, pers comm)), the fish emerge from the substratum (and are termed fry) to begin exogenous feeding. Above-substratum dwelling is likely to render the fish much more vulnerable to fish predators (Symons 1974, Brännäs 1995). Therefore it would be highly beneficial for these fish to be acutely alert to predator cues, and it may be their first opportunity to build on or enhance innate predator recognition skills through experiences with predator cues.

However, the question arises as to why the heightened response to pike odour at category II was lost by category III? In the wild, there is a tendency for salmon to settle into localized home ranges in mid summer after dispersion from the nests in early summer (Crisp 1995, Armstrong *et al.* 2001). Therefore, after category II, these fish would not be entering new areas where they might encounter new predators. There may be a cost to maintaining strong responses unnecessarily. Signal detection theory predicts that a high sensitivity to inappropriate cues carries the risk and cost of a high level of false alarms with no benefits (Green & Swets 1966). Such costs could include spending time in shelter rather than feeding and competing for territories when there is no risk from the perceived predator. In the case of our experimental fish, there would have been no olfactory cues from pike during their time in laboratory rearing tanks between categories II and II, and therefore no stimulus to respond to the predator at category III. An interesting analogous anti-predator response is the ontogenetic development of induced morphological defence that carp, *Carassius carassius*, develop only in the presence of predators (e.g. Brönmark & Miner 1992, Vøllestad *et al.* 2004). Their defence is an increase in body depth, but it does not occur in the absence of predators due to the cost of increased energy requirements for swimming (Brönmark & Miner 1992).

Atlantic salmon have been shown to have sensitive periods for olfactory learning (Morin *et al.* 1989a,b, McCormick *et al.* 1998). There is evidence that the imprinting process, whereby salmon learn and retain the memory of olfactory cues specific to their natal river/conspecifics, which enables them to return to their natal rivers to spawn (Horrall 1981), is strongly associated with a sensitive period occurring when juveniles undergo the smolting (maturation) process. Morin *et al.* (1989a) have shown that a sensitive period may exist for only seven days, and may be linked to high thyroxine levels (Hasler & Scholz 1983). It seems likely that imprinting also occurs at the early stages (Hansen *et al.* 1987, Dittman & Quinn 1996, Nevitt & Dittman 1998) before salmon disperse to rearing areas that may be remote from spawning grounds (Erkinaro & Gibson 1997, Erkinaro *et al.* 1998). If so, then learning of homing cues and predator cues may occur during the same life-stage window. Similarly, a heightened response to new predators would be beneficial when salmon smolt and migrate to new habitats. A general proliferation of neural capacity at these key stages (Jahn 1972, Olsén 1993) may underpin both predator recognition and homing.

The potential for learnt skills to build on existing abilities, as demonstrated in category II, may be bypassed in a predator-free (hatchery) environment, and may be responsible for the poorer anti-predator behaviours of hatchery-reared compared to wild-reared salmon and trout (e.g. Järvi & Uglem 1993, Alvarez & Nicieza 2003). It has been suggested that releaser-induced recognition learning (such as simultaneous exposure to predator cues and AS), may stimulate learnt recognition of predators, and therefore could be used to compensate for lack of experience of hatchery fish (i.e. 'life skills training'; Suboski & Templeton 1989). This work demonstrates the importance of considering ontogeny of the fish species that is to be 'trained', as receptivity to cues may change during development. Even if it is not clear what developmental stage is most adept at learning, an understanding of the life history of the species and the time period when opportunities to learn are present in the wild, may give some guidance.

SECTION III

CHAPTER SIX

A test of how life skills training influences survival of hatchery-reared Atlantic salmon in restocking programmes

ABSTRACT

This study tested how methods used to enhance predator recognition abilities of predator-naïve fish influenced the survival of hatchery-reared Atlantic salmon stocked into the wild. Pre-release exposure to olfactory cues (predator odour and salmon alarm substance) has been shown to stimulate learnt recognition of predators through releaser-induced recognition learning under laboratory conditions. In this field study, pre-release exposure had no effect on survival of salmon smolts migrating through lochs with abundant predator (pike) populations. The hatchery-reared smolts were recaptured 40 days after being released into the river, by which time all of the wild salmon smolts had migrated further downstream. Delayed migration, and/or other behavioural differences of hatchery- to wild-reared fish, may have a greater influence on survival than predator recognition skills.

INTRODUCTION

Hatchery-reared Atlantic salmon may be introduced into rivers in attempts to enhance healthy wild stocks, to compensate for damage caused by anthropogenic disturbance such as hydro-electric structures, and to rehabilitate fragile populations (Aprahamian *et al.*

2003). Furthermore, salmon may be released as smolts, in which case there may be the potential to generate a catch of adult fish that is not dependent on limitations of natural juvenile rearing capacity. Hatchery-rearing to smolt stage is commonplace in Baltic countries and can help support high quality fisheries such as that on the River Delphi in Co. Galway, Ireland. However, survival of hatchery-reared smolts tends to be low relative to wild-reared smolts (Aprahamian *et al.* 2003).

A major factor likely to cause poor survival of released smolts is that they can be particularly vulnerable to predation. For example, Kennedy & Greer (1988) found that most of the salmon smolts consumed by cormorants feeding at the estuary of the River Bush were hatchery-reared fish. Hatchery-reared salmonids may be relatively susceptible to predation (Berejikian 1995, Dellefors & Johnsson 1995, Shively *et al.* 1996) because they exhibit more risk-taking behaviour (Dellefors & Johnsson 1995, Johnsson *et al.* 1996, Fernö & Järvi 1998), have poorer predator recognition skills (Berejikian *et al.* 2003, Hawkins *et al.* 2004a) and show weaker anti-predator responses (Järvi & Uglem 1993, Alvarez & Nicieza 2003) than wild-reared fish. These factors are likely to be especially pronounced in fish reared until smolt stage, as they spend an extensive period under hatchery conditions.

The absence of predators in the hatchery environment means that there are no opportunities for hatchery fish to acquire predator recognition skills and develop anti-predator behaviours (Suboski & Templeton 1989, Kieffer & Colgan 1992, Olla *et al.* 1992, Dellefors & Johnsson 1995). It has been suggested that captive-rearing programmes could rectify this by providing some form of predator experience to the captive animal, to stimulate the learnt acquisition of anti-predator skills (e.g. Ellis *et al.* 1977, Berejikian 1995, Griffin *et al.* 2000). This has been termed ‘life skills training’ (Suboski & Templeton 1989, Suboski 1990).

Predator-naïve animals can acquire recognition of predators through releaser-induced associative learning (Suboski 1989), whereby a novel stimulus (e.g. predator cue) is paired with an aversive cue, leading to learned aversion to the novel stimulus. This technique has been used to improve anti-predator responses of mammals used for reintroduction programmes, including Siberian polecats, *Mustela eversmanni* (Miller *et al.* 1990), and wallabies, *Lagorchestes hirsutus* (McLean *et al.* 1996).

Salmonids also show enhanced responses to predators as a result of releaser-induced recognition learning, under laboratory conditions (Brown & Smith 1998, Berejikian *et al.* 2003) and in manipulated encounters with predators (Mirza & Chivers 2000). Potentially this could improve the success of salmon restocking programmes (Berejikian *et al.* 1999, 2003, Mirza & Chivers 2000, Brown & Laland 2001), as training techniques could be easily and inexpensively applied in a hatchery environment. However, the few studies that have attempted this have met with limited success (Thompson 1966, Kanayama 1968, Berejikian *et al.* 1999).

Pike are strong predators of salmon smolts in many systems (e.g. Mills 1971, Larsson 1985, Carl & Larsen 1994, Jepsen *et al.* 1998, 2000), and may take advantage of the sudden influx of smolts entering water bodies and switch their feeding habitats to this abundant food source. For example, birds may congregate feeding on migrating smolts (Kennedy & Greer 1988), and pike may temporarily 'specialize' in hunting smolts (Jepsen *et al.* 2000). This may present a problem for salmon restocking programmes as the sudden release of fish can attract predators to the area (e.g. Shively *et al.* 1996).

This study tests how pre-release training using the releaser-induced recognition learning methods influence the survival of hatchery-reared Atlantic salmon smolts stocked into the wild. The work was conducted on a river system equipped with a trap and PIT tag detector below a loch system in which predation by pike on salmon smolts is known to occur. Therefore, it was possible to compare survival rates of trained and control fish through in a short section of the natural system in which we would expect losses to be due largely to pike predation (Mills 1965).

The method for training the salmon involved simultaneous exposure to pike odour (neutral stimulus) and a solution composed of salmon skin (aversive stimulus).

Mechanical damage to the skin of salmon releases a chemical alarm substance (AS) from specialized epidermal cells, and this AS triggers an innate fright response in conspecifics that detect it, whereas skin extract from species which do not have AS does not trigger a response (Mirza & Chivers 2001b).

Salmon were trained and released at the onset of smolting, at which time they undergo morphological and physiological changes in preparation for migrating into the marine environment (Hoar 1976, McCormick *et al.* 1998). Atlantic salmon are particularly sensitive to olfactory learning during smolting (Morin *et al.* 1989a,b) and may be predisposed to enhance their olfactory predator recognition skills (Magurran 1990). Furthermore, it is at this stage that the cumulative effects of hatchery rearing throughout life are greatest. Finally, it is much easier to resample smolts than earlier life stages specifically because they migrate and therefore can be trapped with relative ease.

METHODS

Study site

The River Bran, Easter Ross-shire, is in northern Scotland, and is one of the four catchments of the Conon River system. The system is heavily impacted by a hydroelectric scheme which includes eight dams and six power stations. To compensate for the blockage of the migration routes of adult salmon to their spawning grounds, the R. Bran has annual supplemental stocking of wild salmon eggs (eyed ova) and fry, by the Conon and Alness District Fishery Board. Therefore a large proportion of smolts on this system are of hatchery origin.

The release sites for the hatchery-reared smolts were at Caiseachan, 4 km above Loch Achanalt (57°36'N, 4°53'W; Fig. 1). The fish trap (a wolf trap, Fig. 2) used to recapture smolts was at the downstream end of Loch a' Chuilinn. This represents a downstream migration distance of 7.6 km. Lochs Achanalt and a' Chuilinn total 135 ha and contain abundant pike (Mills 1965), estimated to be 3159 (2484 – 8951) aged 1++ individuals {median (IQ range); see Appendix}. The fish trap is adjacent to a barrage, used to supply water to a power station immediately below.

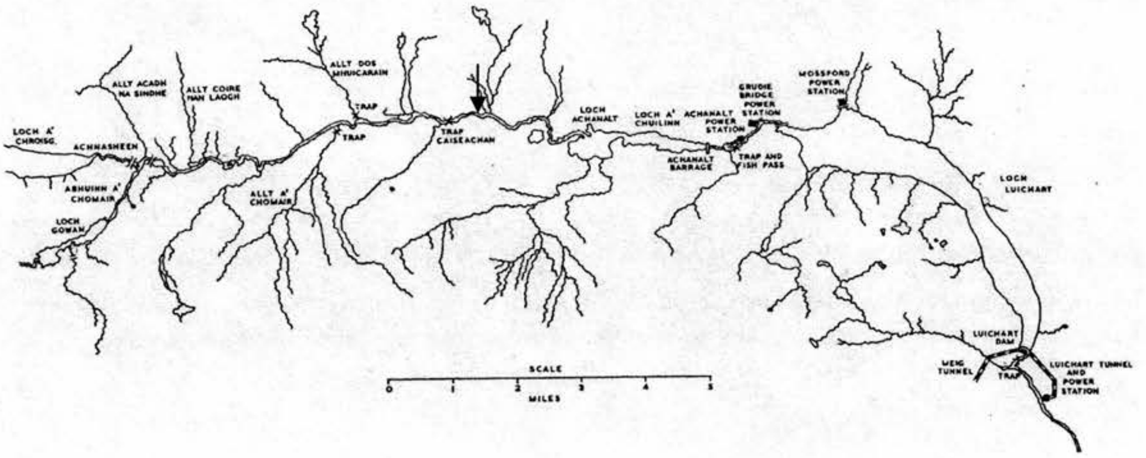


Figure 1. Map of study site. Release site of smolts marked by arrow. Recapture of smolts at fish trap below Achanalt barrage, representing a total migration distance of 7.6 km.

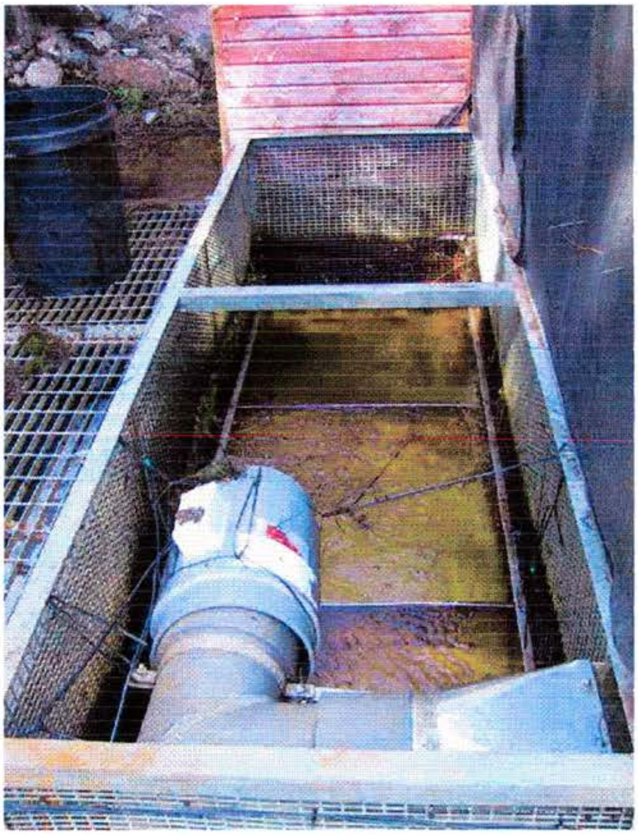


Figure 2. The wolf trap used to recapture smolts below the lochs.

Hatchery-reared smolts

Adult salmon migrating upstream to spawn in 2003 were trapped approximately 15 km below the study section on the R. Blackwater. Fish were stripped of eggs/milt in December 2003. Randomly-fertilized eggs were held in a hatchery, supplied with water from the R. Blackwater, under ambient temperatures. Fish were reared in 1000 l tanks.

N = 1000 age 1+ fish, which were likely to undergo smolting (a size-dependent process), were tagged on 31 March and 1 April 2005 (five weeks prior to release) and held in four 1000 l tanks. The fish were tagged with passive integrative transponder (PIT) tags (12.0 x 2.2 mm diameter, weight 0.1 g; Wyre Micro Design Ltd). A single tag was inserted into the body cavity of each fish via a small incision (*c*. 3 mm) in the abdomen. PIT tags have unique identification numbers which are detected when the fish is in close proximity to a suitable decoder.

Pike

Pike odour used for the pre-release training was a water sample from tanks containing adult and juvenile pike. One adult pike (L_F 84 cm, 4.3 kg), captured (27 April) from the river system, was held in a 1000 l tank. Regurgitation of five salmon smolts confirmed it was feeding post-spawning, and dead hatchery-reared smolts were provided during its captivity. To obtain odour for the training, the pike was placed in a bath (97 x 65 x 68 cm) containing 170 l of water (biomass of 25.4 g l^{-1}) for 40 h. During this time the water was aerated but not filtered and no food was provided.

In addition, six juvenile pike were captured in the fish trap between 13 April and 4 May (L_F 22.5 ± 1.6 cm, 76.9 ± 19.6 g; mean ± SD). These were held in a tank (90 x 37 x 32 cm) and fed earthworms and dead hatchery-salmon parr. 50% water changes were carried out every 2 d. 2 d prior to training the filter was removed from the tank and the water reduced to 65 l (biomass of 7.1 g l^{-1}). Feeding continued during this time.

Pre-release training

Training was applied twice (6 and 10 May), once fish showed strong signs of smolting (silvery colour with black fins; loss of appetite). The procedure was the same for both training sessions. On the morning of each session, I made a solution of salmon skin extract (SSE; the aversive stimulus) using the skin of six salmon smolts (L_F 11.5-13.8 mm). The skin was homogenized with 300 ml of water. Previous work (chapters two and five) has found that exposing salmon to pike odour and SSE of similar quantities enhanced subsequent opercular response to pike odour, compared to the control-exposure group (chapter five). At 12:30 on both days, two of the smolt-rearing tanks received 150 ml of SSE, immediately followed by 4 l of water from the bath containing the adult pike and 4 l from the tank containing juvenile pike (treatment group; N = 490). The other two smolt tanks received 8 l of river water (control group; N = 507).

Release of conditioned smolts

Release of smolts was staggered over a 5-d period, from 11 – 15 May. 100 fish were released each morning and 100 fish every afternoon. Fish from the treatment and control groups were not mixed within release batches. One batch of treatment fish and one batch of control fish were released each day and their timing of release was alternated. Smolts were anaesthetized, measured, weighed and their PIT tag number recorded on the day of their release. Size at the date of release was (mean ± SD) L_F: 129.0 ± 6.3 mm; W: 24.4 ± 3.6 g. Smolts from the treatment and control group were similar size (L_F and W: $t_{994} < 0.39$, $P > 0.700$). After recovery from the anaesthetic the fish were transported (duration 1 h) to their release site.

Two sites were used for releasing fish, 400 m apart. Release sites had strong water flow, thus maximizing chances of smolts navigating downstream. Fish were released from the downstream site in the morning and the upstream site in the afternoon, so as to minimize chances of fish from two groups meeting.

Recovery of smolts

The fish trap was checked every 1 - 4 d. All smolts and juvenile pike in the trap were scanned for PIT tags. All smolts were then transported 16 km downstream by the Conon and Alness District Fishery Board to avoid dams below this site.

RESULTS

4.7% (N = 23) of the treatment group and 4.9% (N = 25) of the control group were recovered ($\chi^2_2 = 0.083$, $P = 0.773$). Three (0.6%) of the recovered smolts from the control group were dead and showed signs of predator damage and one further control smolt was detected inside the stomach of a juvenile pike in the trap.

With the exception of two early-recaptured hatchery smolts, all smolts were recovered between 19 and 29 June, representing a delay between release and recapture of 40.3 ± 7.5 d (mean ± SD; range 4 – 49 d; Fig. 3). The last of the 'wild' smolts (which includes hatchery fish stocked as eyed eggs or newly-hatched fry) entered the trap on 14 June. The mean migration speed of recaptured smolts was 0.23 ± 0.25 km day^{-1}, whereas the two early-recaptured smolts had mean speeds of 1.9 and 0.58 km day^{-1}.

The 48 recaptured smolts (129.5 ± 7.1 mm; 24.8 ± 3.8 g; mean ± SD) were similar in size from the total released group (L_F: $t_{50} = 0.53$, $P = 0.597$; W: $t_{50} = 0.79$, $P = 0.435$).

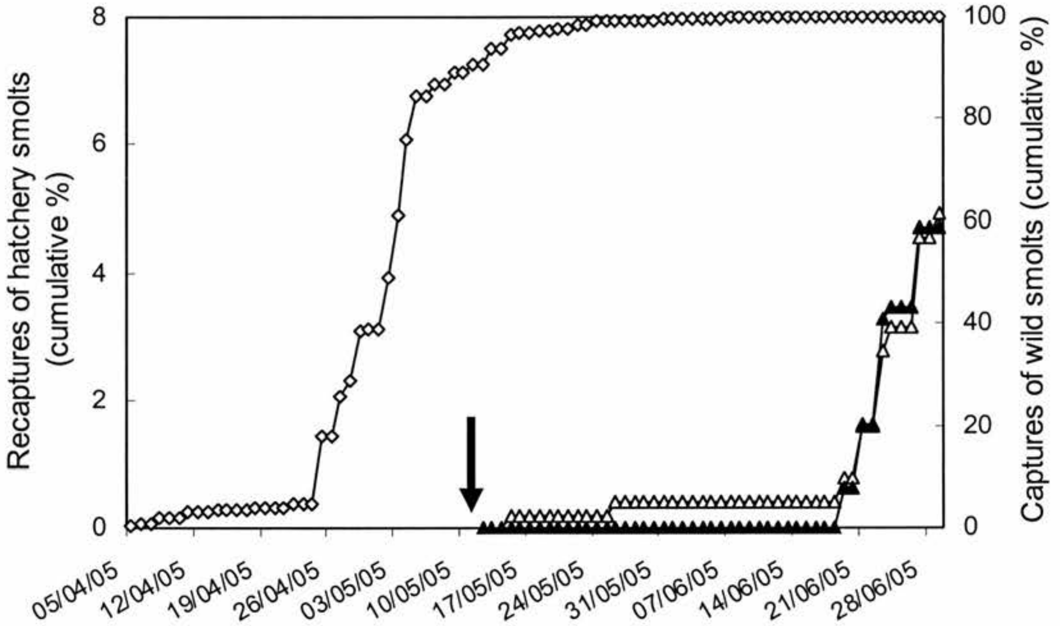


Figure 3. Cumulative percentage of hatchery-reared smolts recovered in fish trap from control group (△) and treatment group (▲), compared to cumulative percentage of wild smolts (◇). Arrow marks release of hatchery-reared smolts.

DISCUSSION

The recovery rate of hatchery-reared salmon smolts was low (< 5%). Pre-release training with olfactory cues designed to enhance recognition of predators had little effect on their survival in the wild. After release into the river, the hatchery-reared smolts were not recaptured in the trap for 40 days, thus migrating after the wild smolts had moved downstream.

The training technique used in this study was similar to that used by Berejikian *et al.* (1999), who trained and released into the wild hatchery-bred Chinook salmon (*Oncorhynchus tshawytscha*) smolts. They found "preliminary" evidence that training enhanced the survival of fish in the wild, although these results were not replicated in tests of survival conducted in stream channels (Berejikian *et al.* 1999).

The limited ability of this technique to enhance survival in the wild contrasts with laboratory-based studies that have found that releaser-induced recognition learning can improve anti-predator responses and predator recognition of salmonids, using the same techniques of exposing fish to predator odour and salmon skin extract (e.g. Brown & Smith 1998, Mirza & Chivers 2000, Berejikian *et al.* 2003). There are several reasons why this technique would not enhance survival of smolts in the wild. (1) Experiences after release may quickly have 'trained' the control group; only one encounter with a predator would compensate for the training technique applied before release, and odours of pike and dead smolts would likely have been abundant in the lochs. (2) A predation attack may occur before smolts detect the odour of the pike, due to slow transmission times of odour cues. This is particularly plausible if pike take a more active hunting strategy when smolts are abundant (Jepsen *et al.* 1998, 2000). (3) The context in which the learning procedures were applied may affect behavioural responses in the wild. The suites of factors that govern behaviour and influence survival in a wild environment were controlled for in the hatchery environment. For example, Berejikian *et al.* (1999) demonstrated that habitat structure in the rearing environment could influence survival in the wild. (4) It is possible that fish that learn to recognize predator cues in a laboratory/hatchery environment will not recognize the same cues presented in a different environmental context.

The method of training used here did not appear to improve survival of hatchery-reared salmon smolts in the wild. Applying pre-release training in more ecologically-relevant conditions (e.g. in wild-type habitat) may improve the methods. However, other factors (discussed below) may have had greater influence on survival than predator recognition skills.

There was a 40-d period between release and recovery of hatchery smolts, resulting in the hatchery smolt migration occurring after the wild smolts had moved further downstream. This delay may have influenced survival rates to a greater extent than predator recognition abilities. An increased migration time would increase the exposure to pike predation, thus hatchery smolts would have been at risk for a greater period than wild smolts. Mills (1965) has estimated the speed of downstream movement of wild smolts to be 0.67 km day^{-1}, whereas most hatchery-reared smolts in this experiment travelled much more slowly, on average (0.23 km day^{-1}). Survival of wild smolts in these two study lochs is *c.* 50% (Birrell, Armstrong & Brooks, unpublished). Thus, for smolts suffering *c.* 50% mortality during 11 days migrating through this system (Mills 1965), a comparable mortality rate over a migration time of 40 days is in excess of 90%. Hence the greater mortality rate of hatchery smolts (*c.* 95%) compared to wild smolts can largely be explained by time exposed to predation. The time exposed to pike would have been prolonged, as most of the habitat between the release and recapture site is suitable pike habitat, including the two lochs and the river for *c.* 2 km upstream of the lochs. The habitat is reedy and the river stretch is slow-flowing. Both lochs contain abundant pike (Mills 1965) and pike could also move into the lower reaches of the river.

Other factors may have had additional effects on hatchery smolt survival. (1) A delay in migration may have reduced the opportunity for schooling with wild smolts. Hatchery smolts might thus have missed the opportunity to learn predator recognition and anti-predator responses through social learning mechanisms transmitted through the shoal (Krause *et al.* 2000). They could also have gained anti-predator benefits from being part of a school (although this benefit could have been obtained if hatchery smolts had schooled with each other, but recaptures in the trap suggested that this was not the case). (2) Stress as a result of handling and transporting fish prior to release may have reduced their capacity to evade predators (Olla & Davis 1989, Olla *et al.* 1992, Berejikian *et al.* 2003). (3) Poor fin condition (Webb 1977, Mork *et al.* 1989) and muscle condition (Johnston 1982), which is typical of hatchery salmonids, may influence swimming capacity (McDonald *et al.* 1998), and hence predator evasion. It is important to note that these factors are characteristic of all release programmes, and not a peculiarity of the experiment. These results thus have broad relevance to release schemes.

The prolonged period that hatchery smolts spent in the system before recapture could have been due to delaying migration after release. Although I avoided releasing smolts in low current velocity because this can delay smolt migration (Hansen *et al.* 1984), differences between the hatchery and wild environments, such as less daylight in the hatchery, could have delayed the onset of smolting in our fish (McCormick *et al.* 1998, Leonard & McCormick 2001). However, external appearances and behavioural changes of our fish suggested that they would be ready to migrate upon release. Alternatively, stress associated with handling and transporting prior to release may have delayed migration (Mesa 1994, Olla *et al.* 1994), and this may be a general problem of restocking programmes. It is also possible that hatchery smolts did migrate shortly after release, but then spent a long time in the lochs trying to find their way out (Thorpe *et al.* 1981). This may have increased their vulnerability to pike predation (e.g. Jepsen *et al.* 1998, 2000).

It is possible that survival rates of hatchery fish could have been greater than the recapture rates. The loch system in this study was part of a hydro-electric scheme, and the fish trap had to be closed on 1 July because water was then diverted to generate electricity. However, smolts migrating after this time would have little chance of survival, as they would have to pass through river sections used for hydro-power generation. Secondly, during periods of peak water flow, the barrage gates adjacent to the fish trap have to be opened, and then smolts may move downstream through the barrage gates without entering the fish trap. After release of the hatchery smolts the gates were only opened twice (26 May and 4 June). However, from Fig. 3 it is apparent that both groups were migrating downstream at almost identical daily rates, and therefore neither of these factors should affect the comparison of treatment and control groups.

Pike are likely to be the most significant predator of salmon smolts on this river section (Mills 1965; also see Appendix). Smolts would probably have encountered pike frequently as they migrate through lochs following the shoreline (Bourgeois & O'Connell 1988), which is typical habitat of pike (Raat 1988). Predation by brown trout, *Salmo trutta*, may also be significant, but impact of avian predators, including goosanders, *Mergus merganser*, cormorants, *Phalacocorax carbo*, and gulls, *Larus* sp. is likely to be low, although remains unestimated (Mills 1965).

Quantities of alarm substance (AS) used as the aversive cue in this experiment were within or above those used successfully in other studies (e.g. Mathis & Smith 1993a, Chivers *et al.* 1995a, Berjikian *et al.* 1999). The pike odour we used to train fish would likely be greater than would be experienced in the wild, although this depends on water exchange and pike movement. However, a potential concern is that it is not known what strength of predator odour is ecologically relevant. It is unclear whether fish would associate weaker predator odour cues with the same level of predation threat. If they do not, then this training may have little effect on survival due to the high variability in odour cue concentrations in the wild.

Hatchery smolts were released after the two peaks of the wild smolt run (25 - 29 April and 2 - 5 May), to ensure that the smolts were ready to migrate, and thereby reducing their time spent in the river and vulnerable to predation. This would not have affected the comparison between treatment and control groups, but could perhaps have reduced the overall survival rate of hatchery smolts; for example, by limiting the opportunity for hatchery smolts to school with wild smolts, or delaying migration until photoperiod or temperature conditions were less favourable. Alternative methods for release could be considered (e.g. releasing smolts early in the smolting window or releasing fish at night); however, the effects (if any) on survival are unclear.

Presently, there is no evidence that life skills training techniques designed to improve anti-predator skills of hatchery smolts offer a simple and effective management technique for restocking programmes. Alternative methods of releasing fish, in the types of cues used for training, and the development stages when training is given, remain to be tested. The conditions under which odour recognition has survival advantage need further consideration. However, migratory behaviour of hatchery smolts may have overriding effects on survival. The opportunity for hatchery fish to mix with wild fish may facilitate social learning of more appropriate skills, and the earlier on in the life cycle that fish are released, the more relevant their own experiences will be.

SECTION III

Concluding remarks

The laboratory-based learning experiment described in chapter five suggested that anti-predator conditioning using odour cues can stimulate releaser-induced recognition learning, although the enhancement in subsequent response to predator odour was very small. This may have been because salmon had existing (innate) recognition of predator cues. The field release experiment (chapter six) found no detectable increase in survival of hatchery smolts in the wild after anti-predator conditioning, so perhaps the small enhancement in predator recognition skills has little adaptive value. This does not exclude anti-predator conditioning being of benefit for fish lacking anti-predator behaviours, for example 'domesticated' salmonids. However, the field experiment highlighted that non-selective mortality on stocked hatchery fish may be so immense that small effects obtained from anti-predator training may be overwhelmed.

Despite two decades of research into anti-predator conditioning of salmonids, including a few experiments conducted in the field (Thompson 1966, Kanayama 1968, Berejikian *et al.* 1999, chapter six), there is no management advice on how conditioning techniques could be applied to improve survival of fish released into the wild. In Scotland, where hatchery-reared salmon are typically the progeny of wild adults and have innate predator recognition skills, the most appropriate consideration for stocking - if additional stocking of salmon is required at all – is to stock fish at the earliest life stages possible to minimize effects of hatchery-rearing. This can be achieved when natural rearing habitat is not limiting. For example, the River Conon system in northern Scotland (see chapter six) undertakes supplementary stocking of eyed ova and unfed fry to compensate for loss of spawning habitat, and adult return rates of stocked fish are 0.19% (Aprahamian *et al.* 2003). Furthermore, early stocking of salmon allows individuals with low fitness to be selectively removed early-on, thus saving on the cost of rearing in a hatchery. Early stocking will also allow fish to gain valuable experiences in which to learn behaviours conducive to living in the natural (and highly variable) environment. Stocking before a sensitive period for learning is reached (suggested to occur during early-exogenous feeding; chapter five) could allow predatory encounters in the wild to enhance and refine existing anti-predator responses.

In countries where loss of natural rearing habitat for salmon means that stocking of smolts is the only option (e.g. countries surrounding the Baltic Sea) the potential for anti-predator conditioning may offer scope for research. Hopefully, future research will take a more integrated approach and consider the importance of sensory modes and development in learning. For example, conditioning fish with visual and mechanical predator cues (to encompass aspects of predator behaviour and movement) in realistic situations (e.g. taking into account habitat) and during sensitive periods of development (which may vary between salmonid species). More practical aspects requiring research include the timing and methods of releasing smolts. An important aspect will be how quickly smolts will move downstream after release, as this will determine scope for predation.

For Scottish loch systems, there may be alternatives to anti-predator conditioning to consider. As predation on salmon smolts by pike can be extensive in Scotland (Mills 1965), the next section of this thesis (IV) focuses on the behaviour of this predator. Habitat use of pike is investigated to provide insight into the potential for habitat manipulation to reduce predatory encounters between pike and salmon smolts.

SECTION IV

CHAPTER SEVEN

Settlement and habitat use by juvenile pike in early winter[*]

ABSTRACT

The spatial distribution and social behaviour of young-of-the-year pike was investigated in early winter. Reed beds were selected in the early exploration phase but thereafter, pike congregated in the pool habitat. A significant increase in pool occupancy combined with a decrease in reed bed occupancy and movement indicated settlement within three days. There was no evidence of territoriality, which has been reported previously in adult pike: indeed, inter-individual spacing was consistent with social grouping rather than territory defence. The strong preference for reed cover among juveniles reported in other studies that were undertaken during summer was not observed here. Among-individual variation in exploratory behaviour was observed but this was not related to activity levels or habitat use.

INTRODUCTION

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[*] Data from this chapter have been published as Hawkins, L.A., Armstrong, J.D. & Magurran, A.E. (2003). Settlement and habitat use by juvenile pike in early winter. *Journal of Fish Biology* **63** (Suppl. A), 174-186.

Distributions and social organizations of animals often depend fundamentally on individual behaviours (Sutherland 1992). The process of settlement into habitats can be critically important because it can be the time when territories are won or lost (Stamps & Krishnan 1995), familiarity with conspecifics becomes established (Stamps & Krishnan 1994), and hierarchies are formed. Fish have been important models for exploring and understanding these processes. Much of the focus has been on species in which individuals interact strongly during formation of shoals (e.g. Milinski 1988, Pitcher 1992, Day *et al.* 2001) or within hierarchies (Chase *et al.* 2002). The present study examines behaviour of young of the year (YOY) pike, a species for which occurrence of social behaviour and territoriality is uncertain.

Settlement occurs when fish relocate to new home ranges. Many species of fish undergo ontogenetic habitat niche shifts. In some cases these shifts reflect size-dependent variation in preferences for food and shelter. In other cases they may follow changes in the abundance of resources, either through new resources becoming available or previously available resources disappearing. Fishes sheltering among aquatic vegetation in temperate waters experience seasonal changes in habitat availability. The pike is one such fish.

Interactions among size classes of pike, which are cannibalistic (Frost 1954, Craig & Kipling 1983), appear to have a strong influence on the habitat they use (Raat 1988, Bry 1996). During summer, large pike use a range of habitat types, including open water (Diana 1980) and vegetated areas (Grimm 1981), whereas YOY pike are reported to occupy dense emergent and submerged vegetation near the water's edge in early summer (Grimm 1981). This complex habitat provides protection against interspecific- and intraspecific-predation (Holland & Huston 1984, Eklöv 1997, Skov *et al.* 2002). During the summer, vegetation extends progressively out from the shore, and the area used by YOY pike also moves out further within this vegetation, although it is less dense and co-occupied by larger pike (Grimm & Klinge 1996). When the submerged vegetation dies in the autumn, YOY pike lose their shelter.

Displaced animals, such as pike leaving decaying vegetation, are likely to be particularly vulnerable to predation and inefficient at feeding until they have located and learned the structure of their new habitat (Pitcher & Magurran 1983, Warburton 1990). Variations in exploration and settlement processes are therefore likely to be important determinants of individual fitness and ultimately population structure. However, exploration and settlement processes are generally not well understood among animals (Stamps & Krishnan 1995). Some recent studies have revealed between-individual differences among co-occurring conspecific fish in exploration behaviour (Rimmer *et al.* 1983, Wilson *et al.* 1993, Mikheev *et al.* 1994, Armstrong *et al.* 1997, Budaev 1997a, Fraser *et al.* 2001) and settlement in new habitats (Armstrong *et al.* 1997). Such differences may relate to boldness (Wilson *et al.* 1993) and/or dominance (Benus *et al.* 1987, Hessing *et al.* 1994, Verbeek *et al.* 1996, 1999).

There appears to be little or no information on exploration, settlement, space and habitat use of pike displaced during early winter or concerning between-individual variation in traits such as boldness. Dominance relationships may exist because of the large size differences among YOY pike (Wright & Giles 1987), but there are no empirical data on social structure or interference. Nor is there any information about territoriality in juvenile pike; it may occur in adults (Nilsson *et al.* 2000) but has not been reported for fry (Giles *et al.* 1986).

In this study, exploration, settlement and space use of juvenile pike was examined by introducing fish into a replicated and controlled canal environment. The following questions were addressed. (1) Do pike disperse throughout available habitat as they explore and settle? It is expected that habitat use would change as individuals settle into their new habitat; activity levels would correspondingly decline. (2) Is there individual variation in behaviour {e.g. boldness (Wilson *et al.* 1993, Budaev 1997b)} and does this affect patterns of exploration and space use? (3) Are juvenile pike territorial {where territoriality is defined as 'active interference between individuals, such that a more or less exclusive area, the territory, is defended against intruders by a recognizable pattern of behaviour' (Begon *et al.* 1996)}?

METHODS

Fish

Progeny of a single pair of fish from Loch Freuchie (56'3°N 3'5°W), stripped and fertilized in spring 2002, were reared at the Fisheries Research Services Almondbank field station, Perthshire, Scotland. The use of siblings and the common rearing environment ensured that the test for individual differences was conservative. The fish were held in glass aquaria supplied with a through flow of water from the River Almond at ambient temperature and fed a diet of earthworms. A total of 24 pike were used in the experiments; fish were chosen at random by selecting through the glass side of the holding tank and then netting the chosen fish. Fish were size-graded during husbandry, and all individuals were taken from one size-graded holding tank {Total length, T_L: 161 mm (147-174); mean (range)}. Each fish was individually identifiable by its unique natural stripe markings on lateral flanks.

Experiments

The experiments were carried out from 9 October – 2 November 2002 in a section (7.5 m x 1.55 m, 11.4 m^2) of a glass-sided indoor canal isolated by mesh grids at either end. The fish were observed through the glass from within a darkened hide running the length of the canal section. Water was refreshed by a continual trickle feed from the River Almond. The mean daily average water temperature was 7.1°C (range: 2.9 - 10°C). Light was from 400 Watt, 6000 lux bulbs (Philips SON Agro) spaced at 1.8 m intervals along the length of the stream; lights were set to an ambient photoperiod. The pike were not fed during the trials because feeding startled them, and at this time of year they fed infrequently when in the holding tanks.

The experimental section of the canal (Fig. 1) was landscaped with a pool in the middle (1.50 x 1.55 x 0.40 m depth) containing a narrow shelf at the front (height 0.30 m, shelf depth 0.10 m). Either side of the pool was a shallower section (interface; length 3 m, mean depth 0.17 m), and at the far ends of the interface were cane patches of 0.7 m^2 and 0.5 m^2, at a density of 20 canes m^{-2}, representing low-density vegetation stems. This arrangement was designed to include a range of the microhabitat types typically found in near-shore areas of static water bodies inhabited by pike.

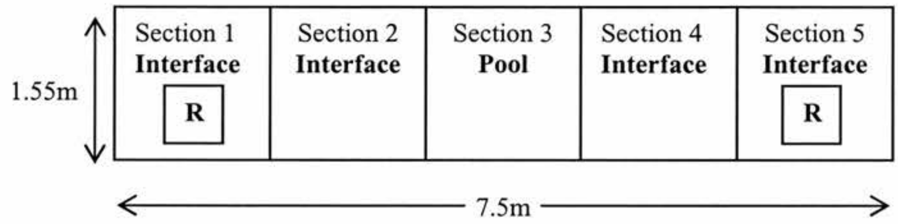

Figure 1. Diagram of glass-sided canal. Groups of 4 pike were introduced into the pool (section 3). R = reed patch.

Six trials (using different pike) were conducted, lasting 5 days each. In the morning of day 1 of each trial, four randomly chosen pike were introduced simultaneously into the pool section (canal density 0.35 fish m^{-2}), and continuous observations were made over the following 3 h. The following information was recorded for each individual: positions; activity (resting on substratum, motionless in water column, slow movement or swimming); time taken to leave the pool; and behaviour when entering a new section. After 3 h, observations were made at least every 15 min of the position and activity of each fish during the periods of daylight over the remainder of the 5 day duration. Continuous 5 min periods of observations were made of all trials to supplement behavioural information. These were done daily (depending on water visibility), morning and afternoon, and within these constraints observation periods were chosen at random.

Observations were disrupted on occasion by heavy rainfall, which increased sediment load in the river water feeding the canal and obscured clear view of the fish. Such weather occurred towards the end of the experiment such that trials 5 and 6 were observed for only 3 and 2 days respectively. The total number of observations ranged from 230–544 per day depending on water clarity.

RESULTS

(i) Initial dispersal and settlement

Fish were observed for the first 3 h after displacement to the canal. Upon introduction to the pool, all fish except one moved towards the substratum in the pool and remained still. The one exception left the pool immediately and returned a few seconds later. The time taken to leave the pool varied greatly between individuals (mean 73.4 min, range 0-181 min, plus one fish that did not leave the pool during the observation period on the first day). During exploration of the interface, movement was characterized by steady swimming interspersed with pauses (of approximately 2-30 s) before the fish moved further through the interface.

Pool, interface and reed habitats accounted for 21%, 68% and 11% of the total area respectively. There was a clear temporal trend in habitat use over the first 3 h (Fig. 2). Pike always selected against the open interface habitat (Fig. 2a). The proportion of observations of pike in the pool decreased from (mean ± SD) 96 ± 6% to 28 ± 27% (repeated measures ANOVA; $F_{4,21} = 7.52$, $P < 0.001$; Fig. 2b) as fish explored first the interface and then the reeds. By the end of the 3 h observation period there was significant positive selection for reed habitat (Fig. 2c), whilst the proportion of time in the pool was not significantly different from that expected with random dispersal throughout the habitat types.

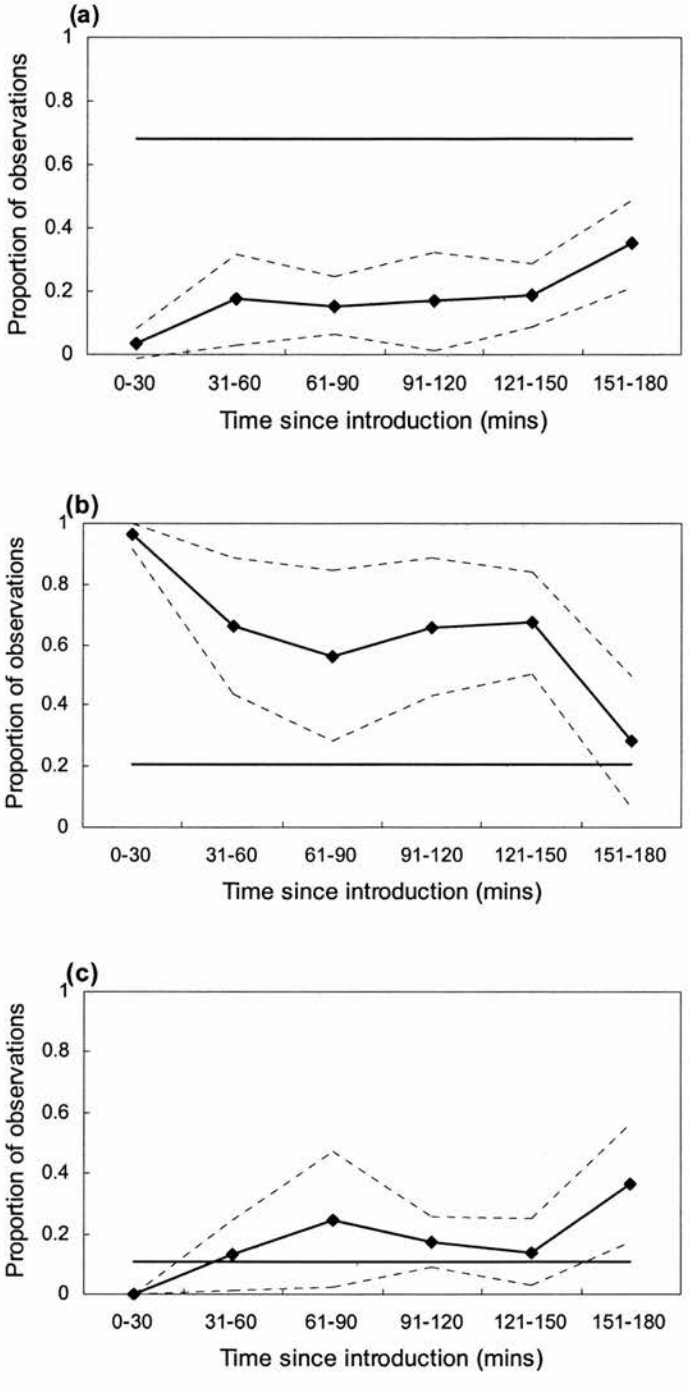

Figure 2. Time spent in (a) the interface, (b) the pool and (c) the reed patches, during the initial introduction (for N = 6 trials). ♦, Proportion of observations in the habitat; ---, 95% CI; —, size of habitat as proportion of total habitat.

Examination of habitat use over the duration of the trial showed a change in pattern following the initial exploration phase. Interface habitat continued to be strongly selected against throughout the trial (Fig. 3a), which corresponded to the pool being selected more than would be expected if habitat was used in proportion to its availability (Fig. 3b). Pool use progressively increased from 44 ± 23% (day 1) to 84 ± 16% (day 3) (repeated measures ANOVA; $F_{2,16} = 4.765$, $P = 0.04$), but did not increase further after day 3 ($F_{1,4} = 0.0$, $P = 1$). There was a decrease in use of reeds between days 1 and 3 (41 ± 17% and 9 ± 12%, respectively; $F_{2,16} = 4.260$, $P = 0.06$), and from day 3 onwards reeds were used no more than expected at random (Fig 3c). No individual spent more than 40% of its time in reed patches after day 2 and most individuals were never observed in reeds. This indicates that the fish settled in the pool by day 3 and some individuals also infrequently visited reed patches.

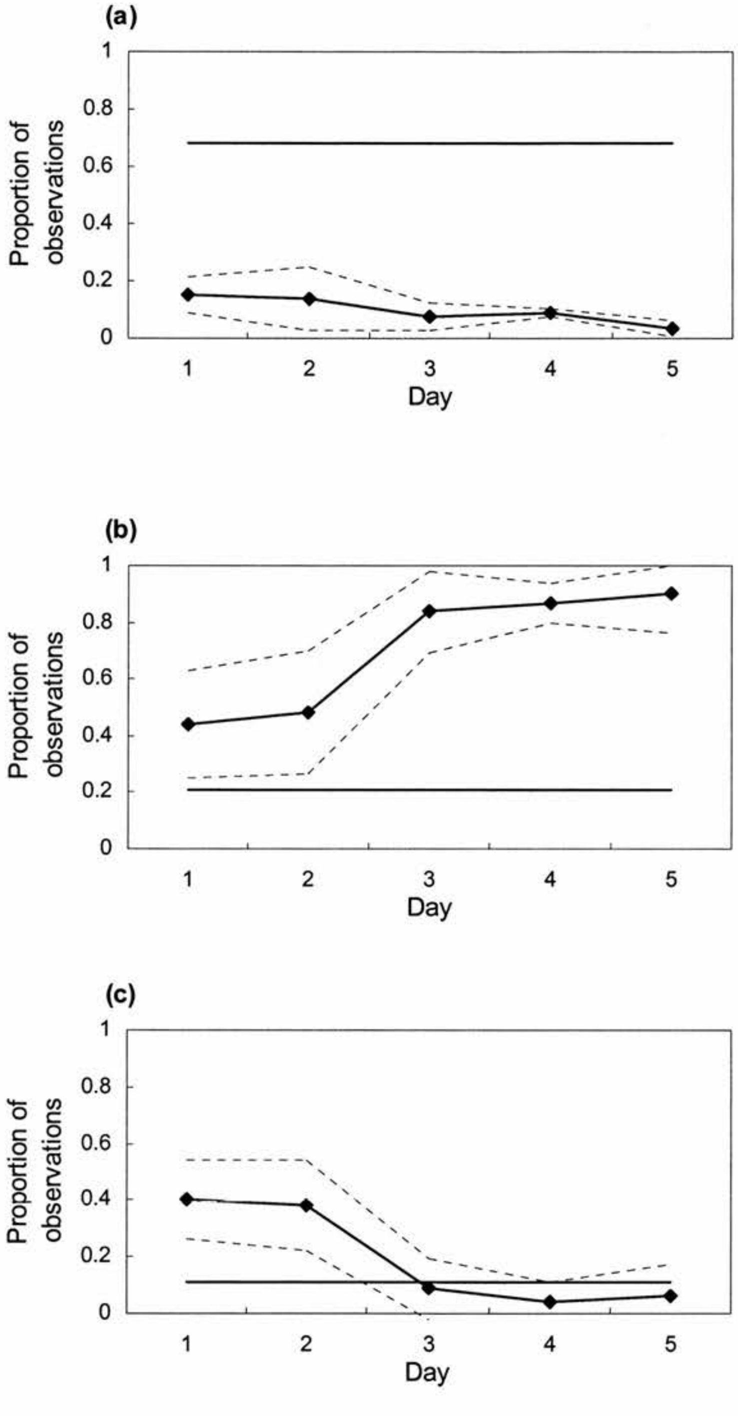

Figure 3. Time spent in (a) the interface, (b) the pool and (c) the reed patches, during the 5 day trial (for N = 6 trials). ◆, Proportion of observations in the habitat; ---, 95% CI; —, size of habitat as proportion of total habitat.

(ii) Movement

Behaviour categories (resting on substrate, still, slow movement [pectoral sculling], swimming) were combined to compare 'still' and 'moving' behaviours, as too few fish were observed either resting on the substrate or swimming after day 1 for statistical analysis. Movement decreased between days 1 and 3 (Repeated measures ANOVA; $F_{2,16} = 9.7$, $P = 0.01$); there was no further decline after day 3 ($F_{1,4} = 1.9$, $P = 0.40$; Fig. 4). This also suggests that fish had stopped exploring and settled by day 3.

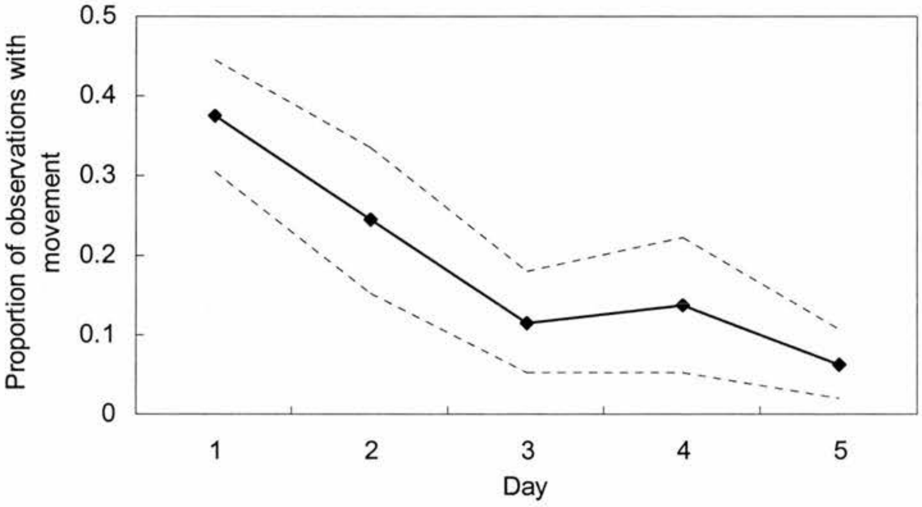

Figure 4. Observations of movement during the 5 day trial (N = 6 trials). ---, 95% CI.

(iii) Individual variation in boldness

Fish were ranked in the order in which they left the pool (the first fish in each trial to leave the pool was rank 1, and so on). There was no difference between ranks in pool occupancy or movement on day 1 (ANOVA; $F_{4,19} = 1.51$, $P = 0.43$; $F_{4,19} = 0.36$, $P = 0.83$, respectively) or following days.

Individuals were also categorised as leaving the pool on their own, in the first instance, or following another fish. These are denoted 'solo explorers' (*S*; N = 16) and 'followers' (*F;* N = 5) (three individuals were not observed when leaving the pool). Followers were characterised by moving out of the pool within 2 s of a lead fish that was < 0.4 m ahead and pausing at the same time as the leading fish. There was significantly higher movement among *S* than *F* for the first 30 min after displacement (Mann-Whitney; Z = 2.43, *P* = 0.03, N = 5) but none thereafter. There was no difference in either habitat occupancy or movement on day 1 (Mann-Whitney; Z = 1.34, *P* = 0.10; Z = 0.62, *P* = 0.56, respectively; N = 5) or subsequent days.

(iv) Spacing and territoriality

All fish occupied the pool after settlement. No aggressive or intimidating behaviour was observed during or after settling, suggesting no overt competition for space. Interspatial distances between individuals in the pool were examined for other evidence of territoriality. Potentially four equal territories could be maintained in the pool of 0.58 m^2, allowing an average spacing of 0.75 m between individuals. However, 73% of all nearest neighbour distances in the pool after settlement were less than 0.5 m (Fig. 5), moreover, 9 ± 14% of distances were less than 0.1 m. As this is less than one body length between conspecifics, this strongly negates the possibility of territories.

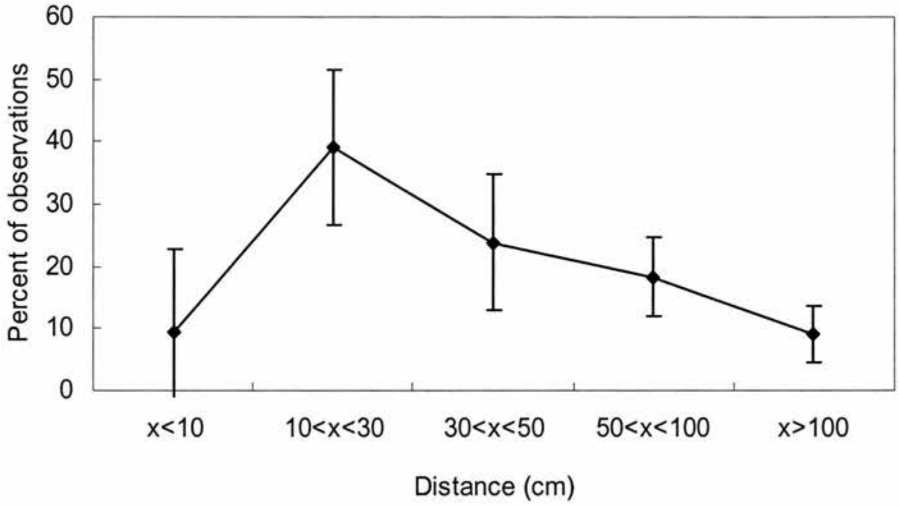

Figure 5. Nearest neighbour distances ± S.D. in pool after settlement (N = 5 trials).

DISCUSSION

Juvenile pike introduced into the pool section of the canal explored the available habitat before settling within about three days as suggested by decrease in activity and stabilization of habitat use. On the first day after displacement, there was selection for reedy areas but after settlement there was strong selection for pool habitat. There was no evidence of territoriality but there was individual variation in boldness.

Dispersion increased as exploration proceeded. Exploration appeared invariably cautious, possibly because movement may trigger attack (Eklöv & Hamrin 1989). Dispersion was moderated by "following" behaviour, which constitutes a form of social interaction and cohesion hitherto apparently not recorded in pike, which is a species generally considered to be solitary after the fry stage. However, as the fish settled, they re-occupied the pool; correspondingly, movement declined and then stabilized once settlement had occurred. The reed beds were strongly selected during the exploration phase before settlement and presumably constituted islands of cover in the interface habitat. It is interesting that many individuals used the reeds for only proportions of their time after settling, as all observations of young pike in the wild find them to exclusively occupy reedy areas (Grimm 1981, Eklöv 1992), although there was no overall selection for vegetation in Eklöv & Diehl's (1994) pond experiment.

It is possible that an absence of predators and/or prey reduced preference for the reeds. Predators were not included in the design because it would likely make observations difficult to interpret. However, the lack of fish predators does not remove anticipated risk of predation. Indeed, many potential predators, such as herons and osprey, would seldom be apparent to fish, even in wild settings. Because avoidance of potentially dangerous habitats even in the absence of predators is an inherent behaviour (Metcalfe *et al.* 1998) and as laboratory-reared fish from wild parents retain this anti-predator behaviour (Metcalfe *et al.* 1987, Magurran 1990), it seems unlikely that pike avoided reeds because they no longer perceived the direct risk of predation.

Food intake by pike at winter temperatures is low (Diana 1983) and digestion times far exceed the overall duration of the experiments (Diana 1983, Armstrong 1986). Therefore, it was anticipated that the intrinsic hunger levels of the subjects were low; hence it is unlikely that occupancy of open water was specifically associated with hunger-induced active hunting (Skov *et al.* 2002). Furthermore, there was no evidence of high hunger levels or active hunting in the studied pike because movement after day 2 was low, and the fish remained grouped, which would increase intraspecific competition for food (Pulliam & Caraco 1984); hungry conspecifics would be less likely to aggregate (Barber *et al.* 1995, Hensor *et al.* 2003). The approach adopted in this study of examining intrinsic expectations of prey availability and predator risk in the available habitats has advantages of standardization over time, among pike and between replicates. However, further study of the effects of different community structures on exploration and habitat choice would also be of value for expanding to specific cases.

There was between-individual variation in propensity to explore, as some individuals were less hesitant than others to leave the pool and enter unfamiliar habitat. It was possible to categorize individuals according to this behaviour. The propensity to explore unfamiliar space has been defined as the shy-bold behaviour continuum (Wilson *et al.* 1993). This continuum has been identified in several taxa, including primates (Stevenson-Hinde *et al.* 1980), canids (MacDonald 1983), rats (Blanchard *et al.* 1986) and several species of fish (e.g. Tulley & Huntingford 1987, Wilson *et al.* 1993, Budaev 1997*b*, Fraser *et al.* 2001). Shy individuals typically retreat or become vigilant in a novel situation, whereas bold individuals act normally or actively explore the novel environment (Wilson *et al.* 1993).

Previous studies have concluded that boldness may affect movement, habitat choice and dispersal distance (Wilson *et al.* 1993, Budaev 1997b, Fraser *et al.* 2001). Contrary to these reported findings, no significant relationship was detected in pike between boldness and exploration or habitat use except in propensity to lead or follow during exploration. This result suggests that a bold strategy such as solo exploration of unknown habitat will result in bold individuals being the first to encounter risk, and in return may gain first access to food and shelter. By following behind bold explorers, shy individuals can reduce risk-taking.

There was no evidence for territorial behaviour or other interference mechanisms. Pike occupying the pool tended to be clumped together and were frequently recorded within one body length of each other (and rarely within 2 cm). As shoaling fish are defined to be within three body lengths of each other (Barber *et al.* 1995), it seems unlikely that strong territorial behaviour was being exhibited. Furthermore, there was never any aggressive, intimidation or orientation and fixation behaviour observed, as would be seen if territorial boundaries were being maintained (Nilsson *et al.* 2000). Individuals did not orientate towards each other even when very close. Therefore it seems that solitary behaviour observed in adult fish (Savino & Stein 1989b, Eklöv 1992, Nilsson *et al.* 2000) is absent under winter conditions or does not develop until after the juvenile stages tested in the present study. A fixed territory- based model of space use would clearly be inappropriate. However, because the fish were siblings it cannot be ruled out that tolerance of conspecifics at close quarters was mediated by kinship. Although experimental work has shown that pike fry do not cannibalize non-siblings more than siblings, after accounting for growth variations between families (Bry & Gillett 1980), it is not possible to eliminate kinship or familiarity as explanations for the absence of territoriality. There is clearly still much scope to investigate the effect of relatedness on social interactions and spacing.

The small size differences among fish used in the trials could potentially influence social status, and consequently affect habitat choice (e.g. Hughes 1992), movement (e.g. Armstrong *et al.* 1997) and space use (e.g. Stamps & Krishnan 1995). However, there was no evidence of dominance among pike or competitive exclusion from a habitat. The positive social interactions observed during following behaviour indicate that the species may use social cohesion strategies to accrue benefits (Magurran & Higham 1988, Elgan 1989). It is possible that grouping together in pools is a response to predation risk at times when vegetative cover is lacking. Such habitat use contrasts with observations of juvenile pike in the wild during summer, when reed-beds are used extensively (Grimm 1981, Eklöv 1992). Adult pike, however, show seasonal habitat variation, tending to occupy vegetation in the summer but rarely in the winter (Diana *et al.* 1977). Occupying deeper water has the potential benefit of reducing the occurrence of an ambush attack from a conspecific concealed amongst reeds, and deeper water may also allow greater manoeuvrability in the event of an attack.

SECTION IV

CHAPTER EIGHT

Aggregation in juvenile pike: interactions between habitat and density in early winter*

ABSTRACT

Juvenile pike, a cannibalistic fish species, aggregates within habitat patches. The advantages to cannibals of aggregating in the absence of other predators and food constraints are not immediately obvious. In this study the basis for this grouping is explored by observing how spatial distributions of juvenile pike are mediated by the presence of conspecifics. Solitary pike preferred shallow-water (0.17 m depth) habitats. When fish density was increased, the average time spent in alternative deep-water habitat (0.33 m) increased, consistent with a despotic type of distribution and suggesting that interference was occurring. In pairs of fish, one pike, nominally the dominant individual, showed a habitat use similar to that of single fish. The second individual mostly occupied deep water, again consistent with a despotic distribution and apparently mediated by intimidation interference. However, dominant pike did on occasion enter deep water, at which times the subordinate pike remained with the dominant fish, appearing to aggregate in the pool. We propose that habitat-specific risk could explain aggregations of pike in deep water. Although remaining in close proximity to dominant individuals in deep water would seemingly put subordinate fish at great risk, the alternative of moving

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to shallow water may increase risk still further by reducing the capacity to perceive and/or evade attacks.

INTRODUCTION

Aggregating can provide benefits to individual animals in terms of predator defence, foraging, mate searching and energy efficiency (Pitcher & Parrish 1993, Lee 1994, Krause & Ruxton 2002). However, grouping may also incur costs, for example, from sharing resources (exploitation competition) (Krause & Ruxton 2002). In addition, high-density living often results in interference, whereby the presence of conspecifics impairs individuals from exploiting a resource effectively (Begon *et al.* 1996). Interference has been documented in many taxa, including insects (e.g. Whitehouse & Lubin 1999), birds (e.g. Goss-Custard *et al.* 2001), reptiles (e.g. James & M'Closkey 2004) and fish (e.g. Holbrook & Schmitt 2002).

Spatial distributions of animal populations that do not suffer interference can be modelled by the Ideal Free Distribution (IFD; Fretwell & Lucas 1970), in which the proportion of a population in a given habitat patch relates to quality of the patch irrespective of total population density. This provides a good approximation of the distributions of some animals (e.g. Harper 1982, Kacelnik *et al.* 1992, Ohashi & Yahara 2002, Lin & Batzli 2004). However, most populations do not fit the IFD because interference commonly occurs (e.g. Yasukawa & Bick 1983, Nakano 1995, Whiteman & Côté 2004) and instead may approach an Ideal Despotic Distribution (IDD; Fretwell 1972). Under the IDD, some individuals sequester and defend good quality patches of habitat, leaving poorer competitors to occupy remaining patches. The IDD may be modelled as a dynamic distribution in which individuals respond to temporal variations in the spatial distribution of resources and occupy the best quality of patch that is locally available (Ruxton *et al.* 1999). This dynamic IDD was developed on the basis that intrinsic quality of each patch was independent of total density of foragers and was diminished only by the presence of consuming animals within the patch.

Intensity of interference varies among species and in the extreme can take the form of cannibalism or fatal wounding from a single encounter. Pike can be considered to be a species in which interference intensity is very high. Juveniles can cannibalize conspecifics *c.* 50-75% of their own body length (Hunt & Carbine 1951, Craig & Kipling 1983, Giles *et al.* 1986) and are particularly vulnerable to attack from conspecifics of similar size when handling prey (Nilsson & Brönmark 1999), as usual defence behaviours are presumably compromised.

In view of this high intensity of interference and the tendency for interference to promote dispersion, the earlier finding (chapter seven) that juvenile (age 0) pike form groups in deep water rather than dispersing to use shallow areas, seems surprising. There are some recorded examples of cannibalistic species grouping. Antlions, *Myrmeleon* spp., for example, aggregate when suitable microhabitats are rare (Gotelli 1993). Aggregations of pondskaters, e.g. *Gerris najas*, probably reflect a temporary high population density relative to carrying capacity prior to cannibalistic cropping (Brinkhurst 1966). The spider *Tegenaria atrica*, tolerates conspecifics under high-food conditions (Pourie & Trabalon 2001) when the presence of alternative food reduces risk of cannibalism and the abundant diet inhibits development of cuticular lipids used in pheromonal communication during aggressive interactions. These mechanisms did not appear to explain the grouping among pike (chapter seven) but it was suggested that anti-predator benefits of shoaling might exceed risks of cannibalism from group living. Alternatively, the distribution may result from despotic occupation of shallow water with subordinate fishes clustering in more marginal deep-water habitat.

The study presented here was designed to assess the behavioural mechanisms that underpin grouping behaviour among young-of-the-year pike. First, I established the preferred habitat of single fish. Then, using pairs and groups of four fish, I observed how density affected habitat use. Aggregation in preferred habitat would support the hypothesis that shoaling occurred. On the other hand, aggregation in more marginal habitat would be consistent with the hypothesis that interference excluded subordinate fish from favoured habitat, consistent with a despotic distribution. Finally, I compared time-space budgets of pairs of fish to determine whether subordinate fish used high quality patches vacated by dominant fish, as predicted by Ruxton *et al.*'s (1999) dynamic IDD.

METHODS

Fish

Progeny of a single female and two male pike from Loch Freuchie (56'3°N 3'5°W), stripped and fertilised in spring 2002, were reared at the Fisheries Research Services Almondbank field station, Perthshire, Scotland. Cannibalism occurred during fry stages (starting at a body length of *c.* 30 mm). As soon as this was noticed fish were size-graded in separate tanks. Some individuals quickly outgrew the other fry in the tanks and again attempted cannibalism, making size-segregation a continuous process during the first year. The fish used in these experiments were taken from three size-graded stock tanks supplied with water from the River Almond at ambient temperature. They were fed a diet of earthworms and dead Atlantic salmon parr. Experiments were conducted in the first winter (18 November – 18 December 2002) when the length of the young-of the-year fish used in the experiments was 145.4 ± 8.7 mm, 134 – 160 mm (L_T; mean ± SD, range). Individuals were chosen for experiments by first taking a stock tank randomly and then selecting an individual at random and ensuring this chosen fish was netted. In total, 45 pike were used and each individual was used once only. Test aquaria thus provided novel environments for all experiments and replicates. Each individual was identifiable by unique natural stripe markings on its lateral flanks. It was not possible to determine the sex of individuals.

For ethical reasons, groups of pike in each replicate had length differences of only 13-18% to prevent cannibalism. Furthermore, the fish were not fed, because it is known that this would make them particularly vulnerable to attack (Nilsson & Brönmark 1999). Therefore, the fish were provided with conditions in which they could exhibit the behaviours that reduced their vulnerability to attack.

Experiments

The habitat used by individual fish was measured at three population densities – one, two and four fish per tank. 10 tanks (0.9 x 0.4 x 0.35 m) were arranged along three sides of an observation arena, with water supplied as for the stock tanks. Three sides of each tank were covered with black polythene and the fourth side allowed the fish to be viewed by the observer in the centre of the arena. Each tank contained two habitat types – relatively shallow (0.17 m depth) and deep (0.33 m depth) water. The deep-water habitat (pool; 0.3 x 0.4 m) and the shallow habitat (0.6 x 0.4 m) accounted for 0.12 m^2 (35.6%) and 0.22 m^2 (64.4%) of the total surface area, respectively. The substrata of both habitats consisted of a layer of large gravel and were otherwise bare, which is a natural habitat type limited to the winter period. The pool occupied the left side of five of the aquaria and the right side of five of the aquaria. Replicates of each density treatment were distributed evenly throughout the time period and observation tanks. The mean daily average water temperature during the experiments was 5.0 °C (range: 0.3 - 7.5 °C). Fluorescent lighting provided illumination to the aquaria of 220 lux for 18 h per day (00:00 to 18:00).

For all trials, fish were placed into the aquaria in the morning (10:00), and were distributed between the two habitats (shallow- and deep-water) at random. Three days were allowed for fish to settle (see chapter seven). Habitat use was recorded on day four as the proportion of observations that a fish was in each habitat. Observations were made every 30 min between 09:00 and 16:00. N = 7 trials were conducted with single fish (L_T: 140.7 ± 8.4 mm, weight: 20.5 ± 3.0 g, mean ± SD), N = 7 trials of density two (147.0 ± 7.6 mm, 23.2 ± 3.5 g) and N = 6 trials of density four (145.9 ± 9.3 mm, 23.1 ± 5.3 g). Size differences (mean ± SD, range) between individuals sharing an aquarium at density two were: L_T, 6.0 ± 5.1 mm, 1 – 16 mm; weight, 3.0 ± 2.1 g, 0.6 - 6.5 g. Size differences between the largest and smallest fish in density four replicates were 21.3 ± 2.7 mm, 18 – 24 mm, 10.7 ± 4.2 g, 7.7 – 18.8 g.

Data analysis

Habitat preference and effects of density on habitat use

Habitat use was calculated as the proportion of observations in which a fish occupied each habitat. Data were normalized by square root arcsine transformation prior to analysis. Habitat preference of single fish was calculated as the proportion of the observations that an individual was in each habitat, and a mean value was calculated for the seven fish. We refer to these mean values as the expected time a fish would spend in a habitat with no interference (pool use = p and shallow habitat use = 1 – p). Habitat preference was established by comparison of the habitat use of single fish with habitat availability. Comparisons of habitat use across densities used ANOVA followed by *post-hoc* Bonferroni testing.

Aggregation

A null estimate of proportions of time that fish in pairs would co-occur in each habitat if their individual distributions were independent of one another (i.e. no attraction or interference) was calculated from the data for habitat use by individual fish. The expected time for fish occurring in separate habitat is $2(p(1 - p))$, for fish occurring together in pools is p^2 and fish occurring together in shallow habitat is $(1 - p)^2$. Actual frequencies of occurrence were compared with these null values to assess evidence for aggregation and segregation.

RESULTS

(i) Habitat preference and effects of density on habitat use

There was a difference in mean habitat choice among fish densities ($F_{2,17} = 9.890$, $P = 0.001$; Fig. 1). Single fish occurred in the shallow habitat significantly more (shallow habitat occupied 80.3 ± 11.6% (mean ± 95% CI) of the time) than the average time that pairs (33.6 ± 16.2% $P = 0.013$) or groups of four fish (18.2 ± 30.7%, $P = 0.02$) occupied shallow water. Pairs and groups of fish did not differ significantly in habitat use ($P = 0.926$). Single fish showed a significant tendency to use shallow water more than would be expected than if they were distributed at random (Fig. 1).

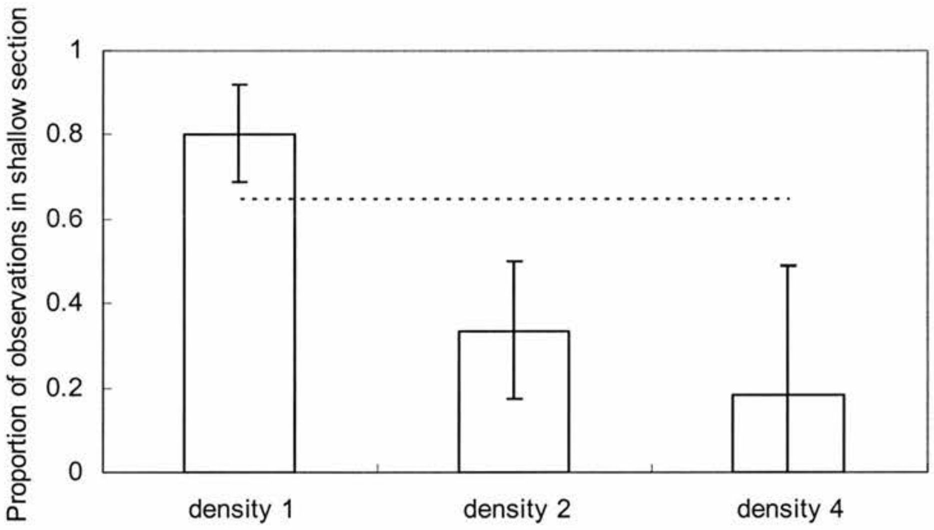

Figure 1. Mean occupancy of the shallow section by fish at varying densities. Error bars indicate 95% C.I. Dashed line represents shallow area as proportion of total area.

(ii) Dominance and avoidance in pairs

Time spent in the preferred shallow section was asymmetric between individuals within pairs of fish (mean ± 95% CI; 62.8 ± 15.8% for one fish compared to 23.8 ± 12.9% for the other; Fig. 2). This suggested a dominance relationship, where the dominant individual is defined as the fish that occupied the preferred shallow section most of the time. Time spent in the shallow section by the dominant fish in the pair was not significantly different from single fish (*t*-test, $t_{12} = 0.669$, $P = 0.516$). By contrast the subordinate fish showed a significantly reduced use of the shallow habitat compared to single fish ($t_{12} = 6.023$, $P < 0.001$). Individuals within pairs occupied different habitats more often than would be expected if there was no interference (proportion of observations with fish in separate sections; mean ± 95% CI: 56.9 ± 9.6%; expected: 31.6 ± 11.9%; Fig. 3).

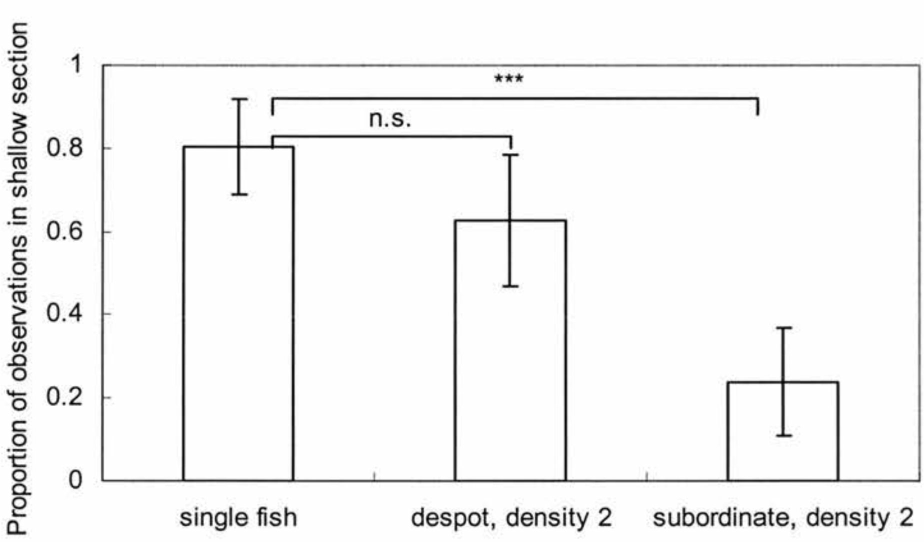

Figure 2. Occupancy of shallow section by pairs of fish compared to single fish. Error bars indicate 95% CI. *** = $P < 0.001$; n.s. = $P > 0.5$.

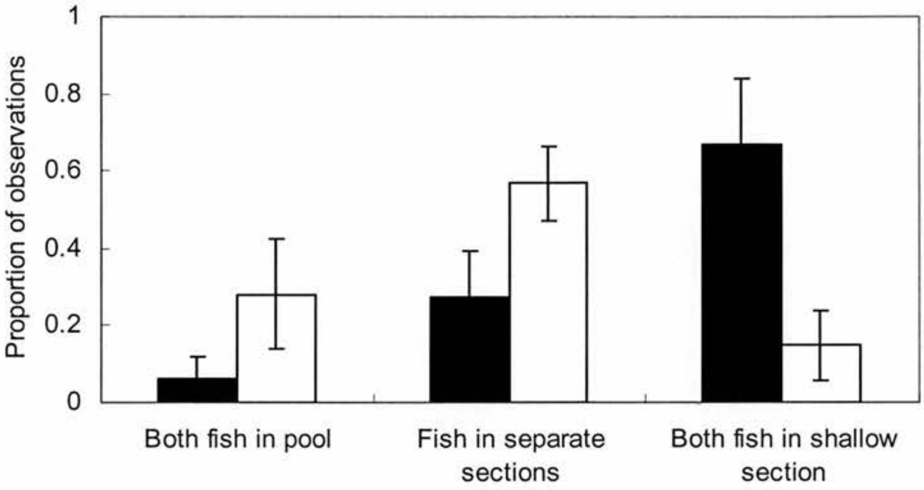

Figure 3. Distribution of fish at density 2 per arena. Predicted values (■) and observed values (□), with 95% confidence intervals.

(iii) Habitat-dependent aggregations in pairs

Occasions when both fish in a pair occupied the same habitat were mostly limited to the pool. Both fish occupied the pool concurrently more than was expected (observed: 28.2 ± 14.4%; expected: 3.9 ± 5.8%; Fig. 3); in contrast, the two fish avoided simultaneously occupying the shallow section (observed: 14.9 ± 9.2%; expected: 64.5 ± 17.6%; Fig. 3).

(iv) Size-dependent effects

Body size (weight and L_T) did not affect habitat use of fish in any of the three density trials (Pearson, $r < 0.401$, $P > 0.170$). Neither was body size related to differences in habitat use between pairs of fish (paired *t*-test, with larger individual compared to smaller individual for each pair; L_T: $t_7 = 1.16$, $P = 0.291$; weight: $t_6 = 0.04$, $P = 0.973$) or within groups of four fish (individual size compared to mean size of the four fish; L_T: $r = 0.363$, df = 24, $P = 0.081$; weight: $r = 0.292$, df = 24, $P = 0.166$).

DISCUSSION

The first aim of this study was to distinguish between the possibilities that aggregations of juvenile pike in the deepest water available (0.33 m) reflect shoaling in favoured habitat or grouping of those fish excluded from favoured habitat. The results showed that solitary pike had a preference for the shallow-water (0.17 m) habitat and that the overall time spent in the least preferred (deeper water) habitat increased with fish density. This indicates a despotic-type distribution with aggregation being a consequence of competitive exclusion. Similarly, a comparison of habitat use by single and pairs of pike showed an overall tendency to disperse.

The second aim was to determine whether subordinate pike used those habitat patches that were preferred by dominant fish when they were vacant. On average, the fish in each pair that used shallow water the most exhibited habitat use indistinguishable from that of single fish, whereas the others tended to use the deeper water (which was deemed by single fish to be of relatively poor quality). This distribution is consistent with the pairs comprising a dominant despot whose habitat use is unaffected by density, and a subordinate fish that tends to be constrained to poor quality habitat. However, it was not simply the case that the subordinate fish avoided the dominant, or used the best patch when it was available (Ruxton *et al.* 1999) because when dominant fish occupied deep water, subordinates tended to remain in the deep-water habitat, thus aggregating with the dominant fish. Although the present study was conducted in small arenas to facilitate the teasing out of mechanisms of interaction among fish, habitat-specific grouping has also been observed in juvenile pike in a large-scale laboratory canal (chapter seven) and among radio-tagged adult pike in the wild (Masters 2004).

There are several plausible explanations for this behaviour. Pike may be more secure when they can observe conspecifics and monitor their behaviour closely to acquire information on their readiness to attack, akin to predator inspection in minnows (Pitcher *et al.* 1986, Pitcher 1992). Pike are generally sit-and-wait predators (Raat 1988) that ambush unsuspecting prey (Casselman & Lewis 1996). Vigilance on the part of subordinate pike may discourage a prospective cannibal from attacking because a successful ambush is less likely (Webb 1982, FitzGibbon 1989).

Subordinate fish may also have a preference for deeper water because it would allow greater manoeuvrability (in three dimensions) to escape from a predatory attack, compared to shallow water. Furthermore, the structural complexity typical of many shallow-water habitats (although not in this experiment) may offer more opportunities for pike to set up ambushes. Shallow water may therefore constitute a favoured habitat for a predator (single or dominant pike) and more dangerous habitat for a potential prey (subordinate pike).

In the present study, there was no observation of overt aggression, which occurs in many species of fish (e.g. Mikheev *et al.* 1994, Adams & Huntingford 1996, Whiteman & Côté 2004), and no individuals had any signs of injury. Instead, interference likely operated through intimidation, in which the mere presence (or possibility of imminent presence) of a dominant individual impairs behaviour of a subordinate pike. Subordinates would presumably have used visual cues to avoid dominants. However, odour cues may have played a role. Areas of the tank consistently inhabited by dominant fish may have become labelled with scent, similar to substrate marking in salmon (Stabell 1987). Subordinates could then use these odour cues to avoid areas preferred by the dominant fish (Griffiths & Armstrong 2002).

Other studies of pike have recorded threat posturing (Raat 1988), movements of individuals towards one another without aggressive encounter (Eklöv & Diehl 1994) and behavioural displays of mouth opening and gill flaring (Nilsson *et al.* 2000), which may all be associated with intimidation. None of these behaviours were observed in the present study, which suggests that more subtle forms of intimidation occurred. It is possible that aggressive interactions occurred but were sufficiently infrequent that our observations did not detect them. However, as even single acts of overt aggression in pike are likely to cause serious damage, they likely pose high risks to both dominant and subordinate fish, such that all individuals may attempt to avoid them. It is also possible that overt aggression observed in previous studies was part of a process of establishing dominance relationships, which had probably already been established in our individuals. Nevertheless, as these pike showed highly skewed spatial distributions, it appeared that dominant individuals used intimidation tactics to control access to resources (space), and the risk of a confrontation escalating was probably very low.

Despite ~~of~~ a difference of up to 64% in body weight within groups of four pike, neither body weight nor length was related to behavioural interactions. By contrast, Eklöv & Diehl (1994) found evidence of size-dependent interference in groups of three wild pike (age 1; 194 – 315 g), despite similar size variation within groups (weight difference up to 62%). However, the occurrence of interference may have been influenced by hunger levels, as the pike in Eklöv & Diehl's (1994) study lost weight during the experiment. It is also possible that initial dominance relationships in pike used in my experiment may have been size-dependent and remained unchanged even if lower-ranked individuals subsequently grew faster than dominant fish (e.g. Abbott *et al.* 1985, Huntingford *et al.* 1990, Stamps & Krishnan 1994). In some instances, kinship or familiarity may affect the use of overt aggression (Brown & Brown 1992, 1993, Chivers *et al.* 1995b). However, the only study of kinship effects in pike found that fry did not cannibalize non-siblings more than siblings, once growth variations between families were accounted for (Bry & Gillett 1980).

Avoidance by subordinate animals of vacant patches preferred by dominant individuals (Griffiths & Armstrong 2002, this study) can be expected to increase the influence of despots across geographic space, particularly when dominant individuals use a network of patches (Armstrong *et al.* 1997, Maclean *et al.* 2005). Risk of damage and mortality may override considerations of energy gain (Ruxton *et al.* 1999) in defining patch quality, although these two parameters should be integrated (Gilliam & Fraser 1987). From the perspective of defining habitat quality, it is interesting that habitat preference of fish in isolation may be quite different from those in groups. For young-of-the-year pike, the availability of shallow habitat that is preferred in isolation and by dominant individuals may be less important than that of deep water that is preferred by subordinates in determining carrying capacity, provided that there is sufficient recruitment of juveniles to use the space fully.

SECTION IV

Concluding remarks

(i) Habitat sharing between pike and salmon

Spatial-temporal overlap in distributions of pike and salmon may occur in (a) riverine habitats and (b) lacustrine habitats (lochs and reservoirs). These are described below.

(a) Riverine habitat

Pike populations occur in rivers containing salmon and trout populations (e.g. Mann 1980). Nevertheless, the diet of these riverine pike does not tend to include salmon fry or parr (Mann 1982), suggesting limited opportunities for predation. There may be spatial segregation of these species within the river, as juvenile salmon preferentially inhabit riffle and run sections of rivers (Symons & Heland 1978, Rimmer *et al.* 1983), whereas very sluggish areas, such as backwaters, tend to be avoided. In contrast, pike are not designed to inhabit fast-flowing water (Raat 1988), although where vegetation and backwaters provide shelter from flow, pike can establish. During the smoltification stage, salmon may be consumed by pike in the river (Mann 1982, Carl & Larsson 1994). In addition, lake-dwelling pike may move into the lower reaches of rivers to hunt smolts (Jepsen *et al.* 1998).

In the autumn, salmon parr, particularly males, may move to deeper and slower-flowing parts of the streams (Allen 1940, 1941, Meister 1962, Saunders & Gee 1964) or descend to larger streams (Riddell & Leggett 1981, Youngson *et al.* 1983). There is opportunity for pike to feed on the autumn-migrating parr (Mills 1965).

(b) Lacustrine habitat

Salmon enter lacustrine habitat during smolt migration. Smolts have been observed to migrate following the shoreline of lakes (Bourgeois & O'Connell 1988), which would suggest high potential for encounters between pike and smolts. A major factor influencing interactions is the time that smolts spend navigating through lacustrine habitat. Smolts can have difficulty finding their way out of lochs (Mills 1965), which would increase their exposure to pike. This is particularly true of hydro-electric reservoirs (Vigg *et al.* 1991, Mesa 1994) where flow is regulated. For example, Jepsen *et al.* (2000) found that salmon and trout smolts traversed the entire reservoir several times but still could not find the way out.

Most smolt migration occurs at night (Thorpe *et al.* 1981, Bourgeois & O'Connell 1988, Olsen *et al.* 2004) and pike are crespuscular predators (Raat 1988), thus it would appear that predator and prey are temporally segregated. However, there is evidence of strong pike predation occurring on smolts (Menzies & Pentelow 1965, Mills 1965, Larsson 1985, Jepsen *et al.* 1998, 2000). It is unclear whether this is due to pike altering their foraging strategy or whether smolts are vulnerable during the daytime in lakes and reservoirs (e.g. because of limited availability of shelters). However, smolts sometimes migrate during the day (Solomon 1978, Olsen *et al.* 2004).

Autumn-migrating salmon parr may make extensive use of lacustrine habitat for overwintering (Hutchings 1986, Ryan 1986, Matthews *et al.* 1997). Migrants, including pre-smolts and precociously-mature males, may stay in lakes until smoltification or maturation. Thus, there is the potential for pike-parr interactions in lacustrine habitat. This autumn migration may be predominantly diurnal (Hutchings 1986), which may further increase the likelihood of interactions. In addition, lakeward migration of parr may also occur in the spring (Hutchings 1986).

Lake populations of pike occur in water bodies with extensive aquatic vegetation (Grimm 1981, Raat 1988, Greenberg *et al.* 1995, Eklöv 1997, Jepsen *et al.* 2001). Vegetation is essential for spawning (Wright 1990); however, it is not clear how important vegetation is for the distribution of pike. Firstly, although juvenile pike are strongly associated with vegetated habitats at the macro-scale (Grimm 1981, Holland & Huston 1985, Raat 1988, Eklöv & Hamrin 1989, Chapman & Mackay 1990, Bry 1996, Eklöv 1997), recent studies (Eklöv & Diehl 1994, chapter seven) suggest that juveniles are not confined to structured habitat. The preference for vegetation density is also unclear and contradict~~ing~~ory (e.g. Eklöv & Hamrin 1989, Savino & Stein 1989b). Distribution may be more dependent on the social structure of juveniles in the locality (chapter eight). Secondly, adult pike are not tied to vegetated littoral areas (Diana *et al.* 1977, Cook & Bergersen 1988) and may also occupy deeper offshore waters (Diana 1980). Thus, the potential for pike-salmon interactions in lakes is wide-ranging.

(ii) Habitat manipulation

The manipulation of habitat has the potential to reduce impact of pike predation on salmon, either through minimising habitat sharing of the two species or by reducing the predation success of pike on salmon. Most interactions are likely to occur between pike and migrating parr and smolts; however, the habitat requirements of migrating parr and smolts are poorly understood, as is the foraging behaviour of pike. Thus, for habitat management, key information is still required. For example, consider the possibility of manipulating vegetation abundance.

Vegetation abundance

Alteration of physical habitat could include removing vegetation from lacustrine sites. This has the potential to reduce habitat for pike to set up ambushes, and hence reduce predation on salmon. Vegetation management could focus on the entrance of rivers into lochs/reservoirs, as this may be an important ambush point for pike, as smolts are funnelled through a small area.

However, the impact of vegetation removal will depend on the importance of vegetation to pike distributions, which - as outlined above - is uncertain. Furthermore, vegetation structure may actually be beneficial for salmon, as habitat structure may promote coexistence of prey and predators (Fraser & Cerri 1982). For example, Fraser & Sise (1980) found that minnows (*Semotilus atromaculatus* and *Rhinichthys atratulus*) utilized habitats with high predator density when the habitats were structurally complex. Vegetation may offer refuge areas to prey (Charnov *et al.* 1976, Stein & Magnuson 1976) and limit predator mobility (Glass 1971). From the aspect of the pike, the influence of complex structure is undetermined – pike may (Cook & Bergersen 1988, Savino & Stein 1989b, Chapman & Mackay 1990, Casselman & Lewis 1996, Jepsen *et al.* 2001, Skov *et al.* 2002, Eklöv 1997), or may not (Eklöv &

Diehl 1994, Greenberg *et al.* 1995, Eklöv & Persson 1996), utilize high-density vegetation to increase hunting success. The requirement for vegetation may be particularly weak if pike adopt an active hunting strategy (e.g. when they are foraging on smolts; Jepsen *et al.* 1998, 2000).

Does vegetation provide cover for pike to ambush smolts, or do smolts use vegetation for cover during periods of low activity? Clearly, habitat manipulation cannot be advocated without more understanding of the effects. Consideration should also be given to how the alteration of pike habitat may effect intraspecific interactions (e.g. cannibalism), which may have indirect effects on interspecific predation.

SECTION V

CHAPTER NINE

General discussion

This thesis has highlighted mechanisms through which fish recognize and respond to predators. My research investigated the role of learning and memory in predator recognition, and asked how predator recognition skills can influence fitness. These fundamental questions were applied to investigate how anti-predator responses of salmon are influenced by hatchery rearing, and the potential for management strategies to enhance fitness of salmon used for restocking. Chapter one posed questions that this thesis aimed to address. This final chapter will review the answers to these questions and suggest where future research could be directed.

Overview

(a) Do salmon have an innate ability to recognize pike as a predator?

Newly-hatched (predator-naïve) Atlantic salmon showed an innate response to the odour of a potentially important predator, the pike (chapter two). The species-specific response to pike suggests the existence of a genetic template which allows salmon to recognize pike on their first encounter. This is an interesting finding as although the distributions of Atlantic salmon and pike overlap in temperate regions, pike would have been absent from Scotland during the ice age and may have been re-introduced in the last few centuries (Davies *et al.* 2004). Innate skills may be regarded as part of an 'arms race' between coexisting predator and prey species (Endler 1991). However, if these species have not co-evolved recently, when was the genetic template formed? Alternatively, perhaps there is some part of the odour cue of pike (not related to diet) that can label the species as a threat to salmon.

In chapter three it was shown that newly-hatched Atlantic salmon have the finely-tuned ability to evaluate predation risk in terms of odour concentration (which may reflect distance to the predator or the number of predators close by) and predator species, relying on innate responses. These newly-hatched fish demonstrated how animals evaluate information and give responses based on the likelihood of erroneously detecting or interpreting a signal. Furthermore, the level of the response was dependent on the strength of the signal, showing how animals may strike an optimal balance between the costs and benefits of reacting to a signal.

The influence of environmental conditions on acquiring and developing adaptive skills was investigated in chapters four and five. The predator recognition skills of salmon reared in a predator-free environment were broadly similar to individuals reared in an environment containing predators (chapter four), demonstrating the importance of innate predator recognition skills. This was confirmed in chapter five, as the innate predator recognition abilities of salmon were not weakened during eight months of captivity (chapter five). However, the rearing environment (which could be considered a learning environment) had quite subtle effects on the responses of salmon to predators. After ten months of rearing in the hatchery, a proportion (*c.* 30%) of fish showed a significant delay in responding to a predator cue, relative to the response of wild-reared fish. This difference suggests that some hatchery fish were slower to realize fully the potential danger from the predator, which may have implications for fitness.

(b) Can salmon enhance and refine existing predator recognition abilities through learning processes? Are there ontogenetic limitations on their capacity to learn?

As innate predator recognition skills are so important in salmon, would we still expect them to have the ability to learn predator recognition skills (bearing in mind the cost of forming and retaining memories)? Newly-hatched salmon did not enhance their anti-predator responses after opportunity to learn predator odour cues (chapter two), suggesting that the cost of developing memories outweighed the benefits, or possibly that the neural system was not far enough advanced to acquire such information. In the wild, alevins would have few experiences with predatory fish, and therefore they may not acquire sufficient benefit from retaining memories to outweigh potential costs. However, given that these benefits would increase substantially with the ontogenetic niche shift from below-to above-substratum/dwelling, due to an increase in the potential for predator encounters, learnt predator recognition skills may then become cost-effective. This change may manifest itself as a sensitive period for learning. In support of this salmon showed a greater (innate) response to predator odour at age *c.* three months, compared to younger and older fish, and this coincided with an increase in the ability to learn predator recognition (chapter five). This supported the hypothesis that a period of heightened receptivity to predator odour cues (at three months old) enabled fish to build on existing abilities with learnt abilities. The function of this sensitive period could be explained by the ontogenetic niche shift from a low predation-risk to a high predation-risk environment.

(c) Can learning of predator recognition be stimulated in a hatchery environment to improve survival of hatchery fish in the wild?

Earlier chapters of this thesis demonstrated (1) the importance of innate predator recognition skills for salmon and (2) that learning could enhance their innate skills, but only to a small extent. It seemed appropriate to question the relevance of learned skills for an individual's fitness. Hatchery-reared salmon smolts were therefore exposed to anti-predator conditioning designed to stimulate learnt recognition of predators, subsequent to their release into the natural environment (which would entail their first encounter with predators). However, this conditioning had little effect on the survival of the salmon smolts, relative to a control group (chapter six). The overall recovery rate of hatchery-reared salmon smolts was very low (< 5%), and one order of magnitude lower than the recovery rate of wild-reared smolts. I suggested that the low survival of hatchery smolts (relative to wild smolts) could be accounted for by an increased time exposed to predation due to delayed migration, and need not be related to anti-predator skills. It was concluded that migratory behaviour and/or other behavioural differences of hatchery- and wild-reared fish may have a greater influence on survival than predator recognition skills. This highlights how the study of a behavioural trait may be confounded by correlation with other behaviours. Laboratory experiments that are designed to test a single behaviour may therefore find a different response to when experiments are conducted under natural conditions.

(d) What information is required to assess alternative approaches to reducing the impacts of pike predation, such as habitat modifications?

The first part of the thesis concentrated on the ability of salmon to avoid predation. Clearly though, the occurrence of a predatory encounter is also related to the behaviour of the predator. At the most fundamental level, encounters will only occur if predators and prey are spatially and temporally integrated. Predatory encounters clearly do occur (e.g. chapter six), so how do these arise? Do the species' habitats overlap at the micro-scale? Or do pike actively pursue migrating salmon, perhaps leaving their preferred habitat ~~niche~~ on a seasonal basis?

Information regarding the micro-habitat use of pike, such as the requirement for vegetation/structure, the influence of conspecifics on spatial distribution, and the use of habitat outside of the littoral zone, is required before we can evaluate the mechanisms driving pike-salmon interactions. From an applied perspective, this information is required before we can contemplate reducing predation on salmon through habitat manipulation. Work by Diana (Diana *et al.* 1977, Diana 1979, 1980) and others (e.g. Cook & Bergersen 1988, Casselman & Lewis 1996) has investigated habitat requirements and behaviour of adult pike. However, there is a paucity of information regarding juvenile pike, and this thesis addressed this in Section IV.

Analysis of spatial distributions found that juvenile pike in the winter congregated in pool habitat and did not select for vegetated habitat at the micro-habitat scale (chapter seven), contrary to general descriptions that vegetation is an essential habitat requirement (Eklöv & Hamrin 1989, Savino & Stein 1989b, Eklöv 1997). Habitat use was not simply a preference for conditions relating to physical habitat but was also influenced by social distributions (chapter eight). Spatial distributions of solitary and groups of pike were consistent with a despotic type of distribution, suggesting that interference was occurring. However, there was no evidence of territoriality (chapters seven and eight). Although it is commonly asserted that pike are highly territorial (e.g. Eklöv 1997, Skov *et al.* 2002), these experiments found that juvenile pike may aggregate under habitat-specific conditions. For YOY pike, the availability of deep water that is preferred by subordinates may determine carrying capacity, provided that there is sufficient recruitment of juveniles to use the space fully. The density of pike in a habitat will determine the predation impact on prey in the habitat, directly, through consumption requirements, and indirectly, through intraspecific interactions such as cannibalism and kleptoparasitism (e.g. Nilsson *et al.* 2000).

How can pike predation on salmon be limited?

This thesis outlined two approaches to reducing pike predation on salmon. Firstly, I investigated the potential to use anti-predator conditioning to enhance predator recognition skills of hatchery-reared salmon. Secondly, I provided information on the habitat requirements of pike, which is fundamental for evaluating the effects of habitat manipulation.

(1) Application of anti-predator conditioning for stocked salmon

An important finding was that salmon have innate recognition of pike, which may have a large impact on the value of anti-predator conditioning. The results presented in chapter five suggested that conditioning may produce only a small enhancement of existing skills. Furthermore, other behavioural divergences of hatchery compared to wild fish may have a much greater impact on the predation mortality of the former. For example, in chapter six it was shown that the migratory behaviour of hatchery smolts may render them more vulnerable to predation. The high mortality on stocked salmon may therefore overwhelm any benefits of anti-predator conditioning.

I have highlighted the influence of ontogeny on the success of anti-predator conditioning, as receptivity to cues and/or capacity to learn may change during development. Further research is required into the factors affecting the capacity of fish to learn. For example, the type of cue used for anti-predator conditioning may influence learning. Are some sensory systems more important than others for acquiring predator recognition skills? Should more than one cue be used, and in what order should they be presented? A next step would be to move away from laboratory experiments that test anti-predator skills under very different conditions to what would occur in the wild. How can we replicate the likely context under which predator cues will be detected in a natural environment? Is it necessary to present cues in appropriate contexts (e.g. specific habitats, time of day)? Or do fish have the ability to learn cues in one context and recognize them in a different context?

At present, there is no evidence that anti-predator conditioning designed to improve anti-predator skills of hatchery-reared salmon smolts offer a simple and effective management technique for restocking programmes. Considering the capacity of fish to learn, it seems likely that experiences gained from living in the natural environment will provide the most opportunities for fish to refine their innate skills. Therefore, for salmon populations that require supplemental stocking, the importance of stocking early in the life cycle is an imperative consideration (Anon 2002).

(2) Habitat manipulation

My research highlights the complexity of spatial distributions of pike, and thereby raises awareness that habitat manipulation may not be a simple solution, but perhaps one best considered on a case-by-case basis. For example, this research has demonstrated the importance of pike population structure on physical habitat requirements.

A potential habitat manipulation considered for lacustrine habitat was the (selective) removal of aquatic vegetation. However, what is most apparent is the need for more specific information. For example, there are conflicting data on how structural complexity of habitat influences foraging of pike (Eklöv & Hamrin 1989, Savino & Stein 1989b). Will the removal of vegetation reduce the hunting success of pike, or will it reduce the protective cover afforded to smolts? And how will it influence pike interactions and cannibalism? Given the ethical implications of pike culling, this area of research should be addressed.

Future directions

There are many facets that govern the outcome of predator-prey interactions. To take a specific example, this thesis posed the question "What are the predation impacts of pike on salmon in Scotland?" Table 1 highlights the information that is available to address this question and the information which is still required.

Table 1. Factors that influence pike predation impacts on Atlantic salmon.

Question	Answered	Research required	References
Anti-predator skills of salmon			
Do salmon have innate recognition of predators?	✓		Brown & Smith (1998), Mirza & Chivers (2000), Berejikian *et al.* (2003), Vilhunen & Hirvonen (2003), Chapter 2
Can salmon learn predator recognition?	✓		Brown & Smith (1998), Berejikian *et al.* (1999, 2003), Mirza & Chivers (2000), Chapter 4
Can salmon learn anti-predator behaviours?	✓		Järvi & Uglem (1993), Healy & Reinhardt (1995), Brown & Smith (1998), Berejikian *et al.* (1999), Mirza & Chivers (2000), Vilhunen *et al.* (2005)
What affects ability to learn?		✓	Berejikian *et al.* (2003), Vilhunen *et al.* (2005), Chapter 5
How does smolt metamorphosis affect sensory systems and predator detection?		✓	Morin *et al.* (1989b)
Can learnt responses increase survival in encounters with predators?	✓		Berejikian (1995), Healy & Reinhardt (1995), Berejikian *et al.* (1999), Mirza & Chivers (2000)
Do learnt responses increase survival in predatory encounters in the natural environment?		✓	Thompson (1966), Kanayama (1968), Berejikian *et al.* (1999), Chapter 6
Frequency of interactions between pike and salmon			
Behaviour of pike			
Do pike show species selection for prey?	✓		Frost (1954), Mills (1965), Mann (1982)
Do pike have a smolt size-preference?		✓	Nilsson *et al.* (2000), Appendix B
Do pike hunt diurnally?		✓	Cook & Bergersen (1988)

Do pike actively hunt smolts?		✓	Jepsen *et al.* (2000)
When do pike spawn, and when is the post-spawning feeding?		✓	
What habitat do pike occupy during smolt migration period?		✓	
What age groups of pike feed on salmon smolts?		✓	Mills 1965, Mann (1982), Appendix B
Behaviour of salmon			
What is the spatial distribution of smolts migrating through lochs?		✓	Bourgeois & O'Connell (1988)
What is the activity pattern of migrating smolts?		✓	Thorpe *et al.* (1981), Bourgeois & O'Connell (1988), Olsèn *et al.*(2004)
What is the habitat selection of autumn-migrating parr?		✓	
Abundances			
Are population sizes of pike known?		✓	Munro (1957), Shafi & Maitland (1971), Treasurer *et al.* (1992), Appendix A
Are pike population size structures known?		✓	
Can numbers of migrating smolts be estimated?	✓		
Are there daily numbers of smolts migrating?	✓		
Are numbers of migrating parr in autumn estimated?		✓	
Is there cyclicity between pike population size and number of smolts surviving?		✓	
Quantitative estimation of pike predation			
How many salmon can a pike eat?	✓		Mann (1982), Diana (1983)
Can number of smolts swamp predation capacity of pike?		✓	
How many smolts will a pike eat during the smolt migration period?		✓	Diana (1983)
Do pike eat autumn-migrating parr?		✓	Mills (1965)
What is the availability of alternative prey species?		✓	

Anti-predator skills of prey

The ability of salmon to recognize pike suggests the intriguing possibility that prey may recognize predators with which they have not co-evolved with recently. We might expect such rapid evolution of anti-predator responses, given the strong selection against individuals that don't respond to predators (Dukas 1998b). More consideration might be given to how animals build templates to recognize predators, such as whether salmon have generalized templates of predators (e.g. Sherman 1985, Evans *et al.* 1993) which were used to build a specific recognition of pike. How can learned information 'update' genetic templates? And can newly-acquired information override genetic presumptions? To address such questions we could exploit situations where predatory species have been introduced, and thus no genetic template exists. The capacity of prey to respond to exotic predators could also be used to test restraints on learning. For example, adaptive responses to exotic predators may depend on similarity with native predator species. This may highlight general cues common to predator species.

My research also suggested the existence of a sensitive period for learning predator cues. Salmon showed greater receptivity to odour cues and a greater capacity to remember these cues during a specific ontogenetic period. Can such a sensitive period also relate to information received through other sensory systems? How common are such sensitive periods for learning predator recognition in animals generally? Are these sensitive periods specific to predator cues, or can other general cues be learned; for example, spatial cues pertaining to foraging information, or in salmon, is sensitivity to predator odour related to sensitivity to odour cues used for homing? More research is required into how development and ontogeny influence learning. To what extent are sensitive periods the result of sensory system development, and under what ecological conditions may sensitive periods arise?

Predator populations

The smolt migration represents a seasonal influx of food to predatory species further down the river system. Can predator populations take advantage of such a food supply, and what is the response of the population once this food supply has disappeared? Can this result in population cyclicity? Can pike take advantage of the seasonally abundance of smolts because they are cannibalistic, and hence at other times of the year may be self-sustaining? Is it therefore an adaptive way of making a temporally-abundant food source last for a whole year? Do other cannibalistic species utilize resources similarly?

There is the potential to use bioenergetics modelling to investigate responses to fluctuations in food supply; for example, to determine how growth rate is influenced. Complex interactions, such as daily variation in food supply, could be included and could test under what situations prey abundance can swamp the predator population. Models could also incorporate population structure, as this may influence population consumption. The model could then be used to demonstrate the outcome of harvesting predator populations.

Cannibalism could also be incorporated into the model. Cannibalism may reduce the impact on prey directly by providing an alternative food source, or indirectly through its effect on population structure. For example, adult pike may regulate numbers of juvenile pike, which may reduce the overall consumption impact of the population (e.g. Mann 1982; see Fig. 1). However, cannibalism may also reduce the likelihood of population crashes due to lack of food, and hence may have an overall negative impact on the prey.

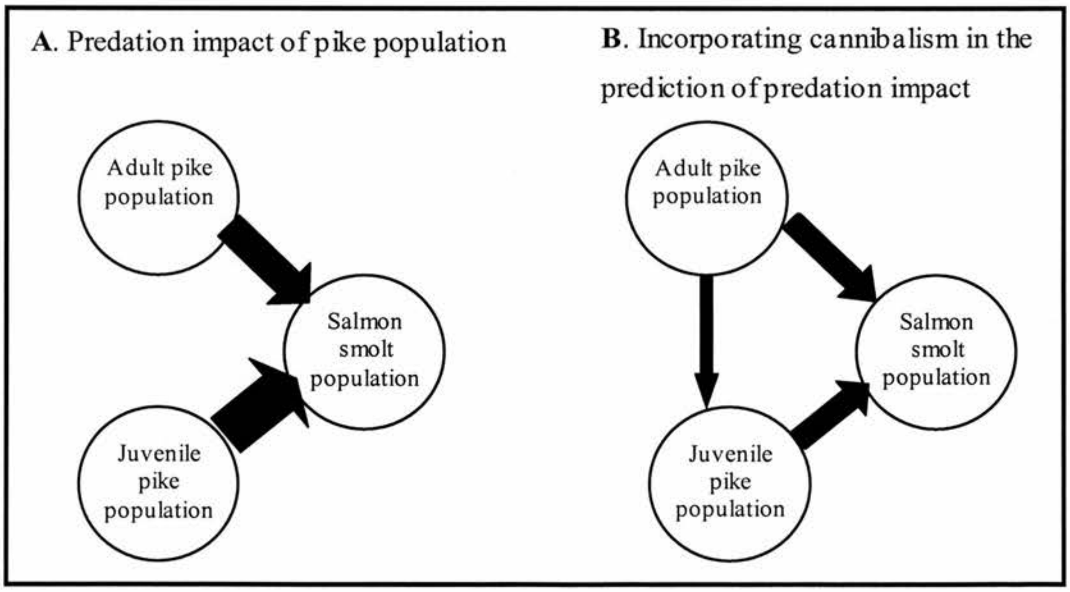

Figure 1. Schematic demonstrating how pike cannibalism may alter predictions on predation impact of salmon smolts.

Concluding remarks

The study of recognition systems in fish is important not only to understand recognition of predators, but also recognition of mates (Kelley *et al.* 1999), kin (Ward & Hart 2003), habitats (Chivers & Smith 1995), interspecific symbionts (Stummer *et al.* 2004), reciprocators (Milinski *et al.* 1990a,b) and individuals (Griffiths 2003). The central design problem in recognition systems is how to optimize the balance between acceptance errors (e.g. failing to recognize a predator) and rejection errors (e.g. falsely identifying a predator) (Sherman *et al.* 1997). Further research into fish cognition could undertake to find how systems are designed to optimize the balance between these errors. Experimental studies could investigate how multiple cues and different sensory inputs are used to minimize errors in detecting predators. A second area requiring further study is how the cognitive capacities of fish change through development; for example, in building recognition templates (Sherman *et al.* 1997). Research into such areas will require neurophysiological studies into the fundamental processes of learning and memory.

Appendix

A. Population size estimates of pike

Data

Data was collated from 19 studies (Table 1) ranging worldwide to evaluate pike population densities and biomasses, with an error margin. These studies provided estimates of population size (either numbers of individuals, density or biomass) for a total of 54 lacustrine sites. Five of the studies were not accessible and are quoted from Raat (1988). Of the remaining 49 estimates, 41 were mark-recapture studies, six were population counts (after poisoning the water body, draining the lake or using visual strip census) and two estimates were produced using virtual population analysis (VPA). 36 (88%) of the mark-recapture studies captured pike using only nets (gill, fyke, or seine nets), which restricted catch to larger individuals (generally adult fish, aged 2++). Other studies used electrofishing or a combination of electrofishing and netting, which provided some estimates of aged 1 (1+) and aged 0 (0+) fish numbers. Six of the populations had additional stocking with 0+ pike ('fingerlings') (Table 2).

Analysis

Median and IQ ranges were estimated for population density (fish ha^{-1}) and biomass (kg ha^{-1}). Estimates were produced for aged 1 and greater (1++) and aged 2 and greater (2++) pike. A limited number of studies also provided estimates of the young-of-the-year (0+) fish (N = 9). The median values were used to estimate the population size of pike in the study lochs. 1+ age pike are capable of eating smolts (pers obs); therefore the 1++ population size of these lochs was calculated for the evaluation of predation impact.

Table 1. Studies estimating pike population sizes.

Location and Reference	Size (ha)	Depth (m) mean (max)	Density (fish ha^{-1}) of age groups estimated (with 95% CI)	Biomass (kg ha^{-1}) of age groups estimated (with 95% CI)	Abundance of prey fish*	Methods
Loch Davan, Scotland Treasurer *et al.* (1992)	41.9	1.2	2++: 5.4 (4.0-8.5)	2++: 5.9	-	Mark-recapture study for five months. Sampling with gill nets and perch traps. Population estimated using Schumacher-Eschmeyer method. Pike > 27 cm caught.
Lake Windermere, England Kipling & Frost (1970)	550	21 (64)	2++: 4.2	2++: 8.4	+	Annual culling using gill netting. Numbers estimated using virtual population analysis (VPA; Paloheimo 1958). Lake is 1476 ha, but netting restricted to < 10 m depth. Population estimates relate to 550 ha. Only first year's estimate used due to culling. Pike >55 cm caught.
Four Netherlands lakes Grimm (1981; 1983)	3.5	1.2 (1.5)	0+: 19.1 (9.7-28.6) 1++: 20.3 (16.9-24.0)	0+: 1.2 (0.4–2.1) 1++: 16.9 (12.9–20.9)	+	Mark-recapture study for five consecutive winters. All lakes had naturally-spawning populations and were also stocked with fingerlings (4-6 cm; 100-300 ha^{-1}). Sampling primarily by electrofishing; also seining and bottom-trawling. Populations estimated with Petersen equation (unmodified). Estimates in this table are mean values of the annual mean estimates. 95% CI are based on N = 5. 0+ pike ≤ 35 cm. Notes that angling may have reduced numbers.
	4.5	1.7 (4)	0+: 48.4 (0-108.4) 1++: 65.3 (30.7-100.0)	0+: 2.8 (1.7-4.0)	+	
	40	1.0 (4.5)	0+: 9.5 (5.1-13.9) 1++: 14.0 (10.7-17.3)	0+: 0.5 (0.3-0.8) 1++: 10.2 (8.1-12.4)	+	
	0.3	1.5	0+: 383.3 (183.3-580.0) 1++: 180.0 (106.7-253.3)	0+: 13.6 (2.1-25.1) 1++: 63.1 (45.3-80.9)	+	
Loch Choin, Scotland Munro (1957)	26.3	1.1 (3)	0+: 60.7 1++: 18.3 2++: 8.3	0++: 5.36	-	Loch poisoned and dead pike counted; therefore a minimum estimate. Pike was the only species present.
Main Lake, England Wright (1990)	17	1.5	1++: 18.5 (15.8-22.4)		+	Mark-recapture study. Sampling with angling, fyke nets, electrofishing and seining. Population estimated with Schumacher – Eschmeyer equation (1987 estimate in study excluded due to bias).

St Peters lake, England Wright (1990)	2	1.0	0+: 22.5 1++: 71.0	0+: 3.0 1++: 53.0	+	Population count after lake was partially drained (therefore minimum estimate). Lake has extensive vegetation.
Dubh Lochan, Scotland Shafi & Maitland (1971)	10	5 (10)	2++: 8.9 (5.1-17.3)	2++: 7.8	-	Mark-recapture study. Sampling primarily with perch traps; some gill netting (<10% of captures). Only pike removed from gill nets in good condition were tagged. Population estimated using Schnabel's method (only 1967 data used). Notes that invertebrates were main food item.
13 lakes, Minnesota Pierce *et al.* (2003)	180	(13)	2++: 36.5			Mark-recapture studies. All lakes sampled once in the spring spawning period with trap nets (modified fyke nets). Re-sampling using gill nets for 2 weeks. Populations estimated with Chapman-modified Petersen equation, and included a 2.4% handling mortality. Pike > 35 cm caught.
	127	(20)	2++: 13.8			
	148	(11)	2++: 54.4			
	151	(12)	2++: 11.7			
	254	(11)	2++: 32.4			
	25	(15)	2++: 35.7			
	96	(14)	2++: 3.3			
	15	(9)	2++: 9.4			
	521	(21)	2++: 8.6			
	48	(11)	2++: 32.1			
	765	(41)	2++: 4.9			
Camerton lake, Minnesota. Pierce & Tomcko (2003a)	28.3	(3)	2++: 59.0	2++: 8.3	+	Methods similar to Pierce *et al.* (2003). Most fish caught were ≤ 3 years. Less than 3% of the population aged 6++. Pike >24 cm caught.
Snaptail lake, Minnesota. Pierce & Tomcko (2003b)	59	(21)	2++: 19.9			Methods similar to Pierce *et al.* (2003).
19 lakes, Wisconsin Margenau *et al.* (1998)	39	3 (10)	2++: 7.8 (6.0-9.6)	2++: 12.4 (11.6-13.2)		Mark-recapture study using similar methods to Pierce *et al.* (2003). Recaptures using trap netting, electrofishing and gill netting. Population estimated with Bailey modification of the Petersen equation. 13 of the 19 lakes had truncated size structures (<25% of the population >53 cm) – suggested that growth rate may slow due to insufficient food resources.
	90†	3 (6)	2++: 38.0 (15.7-60.3)	2++: 29.4 (9.1-49.7)	+	
	62	5 (10)	2++: 7.9 (5.6-10.2)	2++: 5.9 (1.4-10.4)		
	76†	3 (9)	2++: 16.3 (11.4-21.2)	2++: 10.2 (2.7-17.7)	-	
	76	2 (7)	2++: 25.1 (16.2-34.0)	2++: 15.5 (7.7-23.3)		† Lakes with negative growth indexes.

	70†	1 (2)	2++: 23.7 (18.9-28.5)	2++: 14.8 (7.3-22.3)	-	
	98	11 (19)	2++: 9.4 (4.1-14.7)	2++: 7.6 (0-18.2)		‡Lake is stocked because of insufficient natural recruitment.
	110‡	3 (5)	2++: 12.6 (5.7-19.5)	2++: 16.7 (0-39.3)		
	40	3 (6)	2++: 12.0 (5.1-18.9)	2++: 14.3 (0.6-28.0)		
	66†	4 (6)	2++: 15.6 (6.9-24.3)	2++: 10.1 (0-23.8)	-	
	112	4 (7)	2++: 17.8 (9.1-26.5)	2++: 13.3 (0-29.4)		
	112†	4 (13)	2++: 29.0 (19.4-38.6)	2++: 10.8 (2.1-19.5)	-	
	106†	2 (7)	2++: 10.0 (6.2-13.8)	2++: 5.8 (0.3-11.3)	-	
	93†	6 (14)	2++: 15.8 (5.7-25.9)	2++: 13.9 (0-35.9)	-	
	102†	5 (12)	2++: 15.1 (7.4-22.8)	2++: 6.5 (0-14.9)	-	
	104†	3 (11)	2++: 15.3 (9.5-21.1)	2++: 10.3 (0-21.8)	-	
	84	7 (17)	2++: 12.2 (8.4-16.0)	2++: 8.7 (2.1-15.3)		
	57	2 (5)	2++: 2.8 (2.6-3.0)	2++: 3.0 (2.4-3.6)		
	113†	5 (12)	2++: 20.1 (8.6-31.6)	2++: 11.5 (10.9-12.1)	-	
Roi Lake, Alberta Turner & Mackay (1985)	8	(21)	1++: 22.5 (17.6-27.4)			Population count based on observations during strip-census snorkeling. Pike > 17 cm included. Noted to be an unproductive lake. Most fish found in waters <3 m deep.
Dagget Lake, Michigan Beyerle (1971)	5.0	2.1 (4.6)	0+: 2.0 1++: 65.8 2++: 62.2	0+: 0.7 1++: 48.6 2++: 46.1	+	Lakes poisoned and then stocked with fingerlings for three years. Lakes poisoned again and population estimate based on counts.

Emerald Lake, Michigan Beyerle (1971)	2.3	1.8 (2.7)	0+: 10.0 1++: 66.5 2++: 65.7	0+: 1.0 1++: 32.2 2++: 31.9	+	
Maple Lake, Minnesota Seaburg & Moyle (1964)	354	(22)	1++: 29.4		+	Mark – recapture study. Sampling using seine nets. Population estimated with Petersen equation (no Chapman modification). Pike > 35 cm caught.
Grove Lake, Minnesota Seaburg & Moyle (1964)	151	(10.7)	1++: 46.5		+	Mark – recapture study using seining for first capture and angling for recapture. Population estimated with Petersen equation. Pike > 25 cm caught.
Lake Warniak, Poland Ciepielewski (1981). *In* Raat (1988)	38.4		2++: Midpoint (range); 50.2 (20.8-79.6)	2++: Midpoint (range); 25.3 (10.0-40.6)		Estimated by VPA based on 7 years of data.
Murphy flowage, Wisconsin. Snow (1974). *In* Raat (1988)	73		1++: Midpoint (range); 19.9 (5.9 – 33.9)	1++: Midpoint (range); 12.4 (5.0 – 19.8)		Based on 8 years of data (methods not known). Pike > 25 cm caught.
Escabana Lake, Wisconsin Kempinger & Carline (1978a,b). *In* Raat (1988)	119		1++: 6.9 (range; 2.1 – 10.1)	1++: 5.5 (range; 1.9 – 9.9)		Based on 18 years of data (methods not known). Pike > 25 cm caught.
Bucks Lake, Wisconsin Snow & Beard (1972). *In* Raat (1988)	34		1++: Spring: 67.5 (range; 30.6 – 121.8) 1++: Autumn: 89.2 (66.5 – 125.0)	1++: 27.3 (range; 14.9 – 41.2) 1++: 37.0 (32.5 – 47.6)		Methods not known. Both seasonal estimates are included in the analysis.
Lake Demenets, USSR Rudenko (1971)	6.6	3.3 (5)	0+: 24.1 1++: 21.5 2++: 16.7	0+: 0.3 1++: 13.5 2++:13.2	+	Lake poisoned and population estimated by count (therefore a minimum estimate). Biomass calculated from mean weight of age groups and abundances. Pike aged 0-6. Size groups: 0+ = 7-14 cm, 1+ = 17.6-23.5 cm, 2++ > 26 cm.

Alinen Mustajärvi, Finland. Rask & Arvola (1985)	0.7	(6.5)	1++: 24.3	1++: 6.9	+	Population estimate based on catch rates. Sampling with trap nets in the summer. Biomass is a minimum estimate based on fishing yields. Pike aged 1-6. Pike had slow growth rate.
Klicava reservoir, Czechoslovakia Holcík (1968). *In* Raat (1988)	67		1++: 7.9 1++: 2.6	1++: 6.9 1++: 2.3		Methods not known. Two annual estimates provided.

* Based on quantitative or qualitative reports from the authors.

RESULTS

Population density of 2++ pike was 15.5 (9.0 – 28.3) fish ha^{-1} {median (IQ range); N = 40; Fig. 1}. 2++ pike biomass was 10.8 (7.8 – 14.8) kg ha^{-1} (N = 27; Fig. 2). Population estimates of 1++ pike had wider margins of error. 1++ pike density was 23.4 (18.4 – 66.3) fish ha^{-1} (N = 20; Fig. 1). 1++ biomass was 15.2 (6.9 - 39.9) kg ha^{-1} (N = 14; Fig. 2).

Stocked populations accounted for most of the 'outliers' (i.e. estimates greater than 1.5*IQ range) of 1++ and 2++ density and biomass (Figs. 1 and 2). An exceptionally high stocked population density of 1++ pike (Fig. 1; Grimm 1981) may be due to the very small area of the water (0.3 ha); hence a greater proportion of habitat (vegetated littoral zones) would be suitable for the stocked juveniles.

Estimates for age 0+ pike were 22.5 (9.8 – 54.6) fish ha^{-1} (N = 9) and 1.1 (0.6 – 3.0) kg ha^{-1} (N = 8). These estimates were largely based on stocked populations (Table 2) and thus may not reflect natural population sizes.

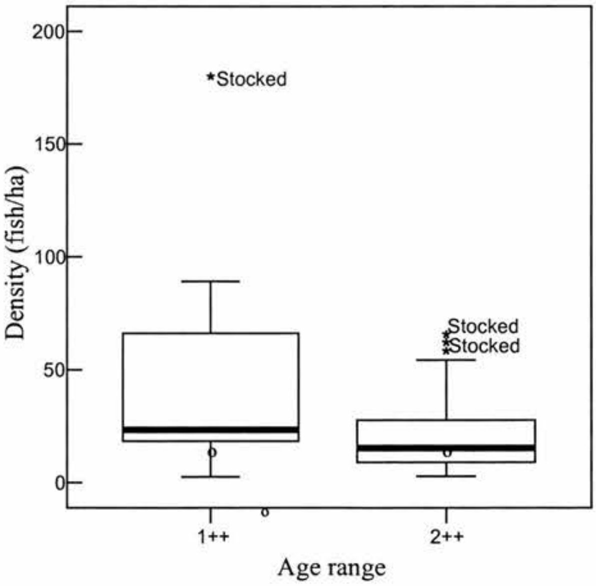

Figure 1. Population densities of 1++ and 2++ pike. Boxes represent IQ ranges and lines mark median densities. Whiskers represent 1.5*IQ range. Outliers (*) represent data

points > 1.5*IQ range; outliers from stocked populations are labelled. Scottish populations; °

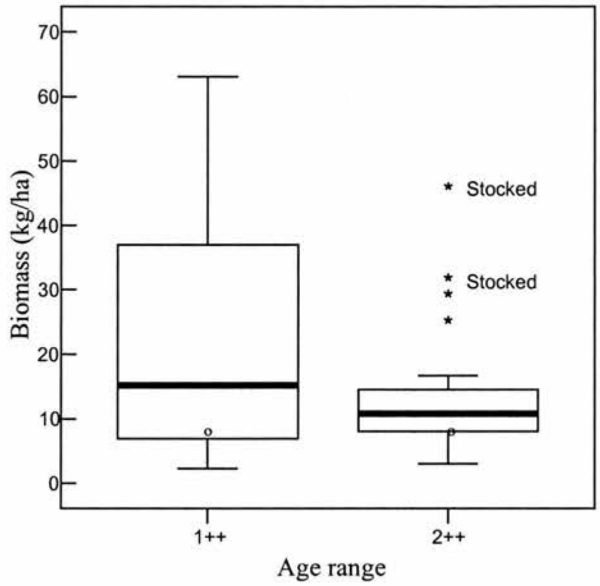

Figure 2. Population biomasses of 1++ and 2++ pike. Boxes represent IQ ranges and lines mark median densities. Whiskers represent 1.5*IQ range. Outliers (*) represent data points > 1.5*IQ range; outliers from stocked populations are labelled. Scottish populations; °

Table 2. Population estimates of lacustrine sites.

	Density (fish ha^{-1})			Biomass (kg ha^{-1})		
	0+	1++	2++	0+	1++	2++
Total N	9	20	40	8	14	27
Stocked N	6	6	3	6	5	3

B. Stomach content analyses of pike

A sample of pike (N = 10; 55.6 ± 16.5 cm; mean ± SD) were taken from the R. Blackwater in northern Scotland between 30 April and 12 May 2004, coinciding with smolt migration. N = 5 individuals were taken from lacustrine habitat and N = 5 individuals from riverine habitat. Capture methods were angling (N = 6), gill netting (N = 2) and shooting (N = 2). Pike caught by angling were humanely killed by a blow to the head and then severance of the spinal cord at the base of the skull. The stomach contents of all individuals were removed and prey items identified. Length (L_F) of prey items was attained where possible, depending on state of digestion.

RESULTS

Pike from this river system were feeding almost exclusively on smolts. All pike sampled had consumed a minimum of one smolt and a maximum of 12 smolts (Fig. 1). Alternative prey species were found in only two pike, both of which had additionally consumed a single minnow, *Phoxinus phoxinus*.

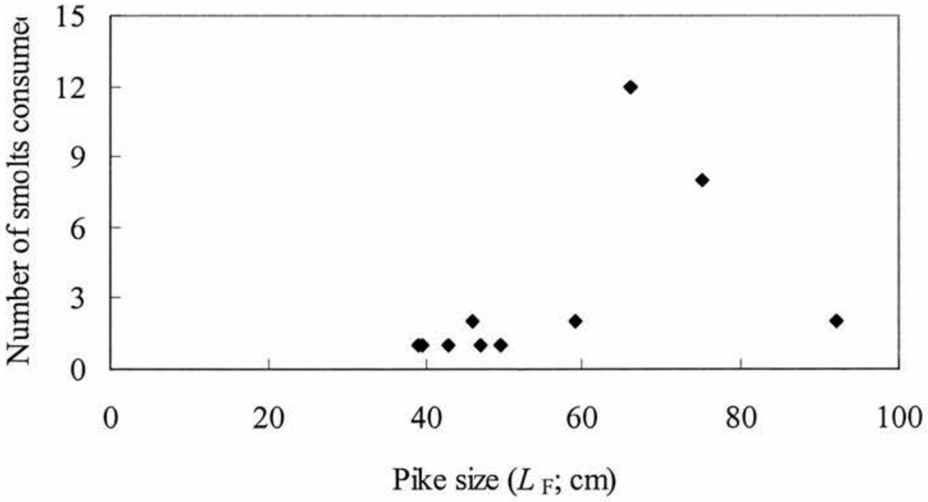

Figure 1. Number of salmon smolts consumed by pike (N = 10) in the study system.

The size of the smolts consumed was 114 ± 8.0 mm (mean ± SD; N = 20). The mean size of smolts captured below the study lochs in 2005 was significantly larger (120.6 ± 9.6 mm; N = 10338; paired t-test (unequal variances); $t_{19} = 3.35$, $P = 0.03$).

References

Abbott, J.C., Dunbrack, R.L. & Orr, C.D. 1985. The interaction of size and experience in dominance relationships of juvenile steelhead trout (*Salmo gairdneri*). *Behaviour* **92**, 241-253.

Adams, C.E. & Huntingford, F.A. 1996. What is a successful fish? Determinants of competitive success in Arctic char (*Salvelinus alpinus*) in different social contexts.
Canadian Journal of Fisheries & Aquatic Sciences **53**, 2446-2450.

Allan, I.R.H. & Ritter, J.A. 1977. Salmonid terminology. *Journal du Conceil, Conseil permanent international pou l'exploration de la mer* **37**, 293-299.

Allen, K.R. 1940. Studies on the biology of the early stages of the salmon (*Salmo salar*). 1. Growth in the River Eden. *Journal of Animal Ecology* **9**, 1-23.

Allen, K.R. 1941. Studies on the biology of the early stages of the salmon (*Salmo salar*). 3. Growth in the Thurso River system, Caithness. *Journal of Animal Ecology* **10**, 273-295.

Ap Rhisiart, A. 1989. *Communication and anti-predator behaviour*. D. Phil. Thesis, University of Oxford, Oxford.

Alvarez, D. & Nicieza, A.G. 2003. Predator avoidance behaviour in wild and hatchery-reared brown trout: the role of experience and domestication. *Journal of Fish Biology* **63**, 1565-1577.

Anon 2002. *Performance and Ecological Impacts of Introduced and Escaped Fish: Physiological and Behavioural Mechanisms*. EC Contract No. FAIR CT 97-1957.

Anon. 2003. Salmon & Sea trout: to stock or not? *FRS Information Leaflet* **22**.

Aprahamian, M.W., Martin Smith, K., McGinnity, P., McKelvey, S. & Taylor, J. 2003. Restocking of salmonids – opportunities and limitations. *Fisheries Research* **62**, 211-227.

Armstrong, J.D. 1986. Heart rate as an indicator of activity, metabolic rate, food intake and digestion in pike, *Esox lucius*. *Journal of Fish Biology* **29** (Suppl. A), 207-221.

Armstrong, J.D. 1997. Self-thinning in juvenile sea trout and other salmonid fishes revisited. *Journal of Animal Ecology* **66**, 519-526.

Armstrong, J.D., Braithwaite, V.A. & Huntingford, F.A. 1997. Spatial strategies of wild Atlantic salmon parr: exploration and settlement in unfamiliar areas. *Journal of Animal Ecology* **66**, 203-211.

Armstrong, J.D., Einum, S., Fleming, I.A. & Rycroft, P. 2001. A method for tracking the behaviour of mature and immature salmon parr around nests during spawning. *Journal of Fish Biology* **59**, 1023-1032.

Backiel, T. 1971. Production and consumption of predatory fish in the Vistula River. *Journal of Fish Biology* **3**, 369-405.

Baglinière, Maisse, G. & Nihouarn, A. 1990. Migratory and reproductive behaviour of female adult Atlantic salmon, *Salmo salar* L., in a spawning stream. *Journal of Fish Biology* **36**, 511-520.

Barber, I., Huntingford, F.A. & Crompton, D.W.T. 1995). The effect of hunger and cestode parasitism on the shoaling decisions of small freshwater fish. *Journal of Fish Biology* **47**, 534-536.

Barreto, R.E., Luchiari, A.C. & Marcondes, A.L. 2003. Ventilatory frequency indicates visual recognition of an allopatric predator in naïve Nile tilapia. *Behavioural Processes* **60**, 235-239.

Bateson, P. 1979. How do sensitive periods arise and what are they for? *Animal Behaviour* **27**, 470-486.

Baum, E.T. 1983. The Penobscot river: an Atlantic salmon river management report. Atlantic Sea Run Salmon Commission, Bangor, ME.

Beamish, F. W. H. 1978. Swimming capacity. In *Fish physiology volume 7* (Hoar, W. S. & Randall, D. J. eds.). pp. 101-187. New York: Academic Press.

Begon, M., Harper, J.L. & Townsend, C.R. 1996. *Ecology: Individuals, populations and communities.* pp. 253. Oxford: Blackwell Science.

Benus, R.F., Koolhaas, J.M. & van Oortmerssen, G.A. 1987. Individual differences in behavioural reaction to a changing environment in mice and rats. *Behaviour* **100**, 105-121.

Berejikian, B.A. 1995. The effects of hatchery and wild ancestry and experience on the relative ability of steelhead trout fry (*Oncorhynchus mykiss*) to avoid a benthic predator. *Canadian Journal of Fisheries and Aquatic Sciences* **52**, 2476-2482.

Berejikian, B.A., Smith, R.J.F., Tezak, E.P., Schroder, S.L. & Knudsen, C.M. 1999. Chemical alarm signals and complex hatchery rearing habitats affect antipredator behaviour and survival of chinook salmon (*Oncorhynchus tshawytscha*) juveniles. *Canadian Journal of Fisheries and Aquatic Sciences* **56**, 830-838.

Berejikian, B.A., Tezak, E.P., Riley, S.C. & LaRae, A.L. 2001. Competitive ability and social behaviour of juvenile steelhead reared in enriched and conventional hatchery tanks and a stream environment. *Journal of Fish Biology* **59**, 1600-1613.

Berejikian, B. A., Tezak, E. P. & LaRae, A. L. 2003. Innate and enhanced predator recognition in hatchery-reared chinook salmon. *Environmental Biology of Fishes* **67**, 241-251.

Bevelhimer, M.S., Stein, R.A. & Carline, R.F. 1985. Assessing significance of physiological differences among three esocids with a bioenergetics model. *Canadian Journal of Fisheries and Aquatic Sciences* **42**, 57-69.

Beyerle, G.B. 1971. A study of two northern pike-bluegill populations. *Transactions of the American Fisheries Society* **100**, 69-73.

Blanchard, R.J., Flannelly, K.J. & Blanchard, D.C. 1986. Defensive behaviors of laboratory and wild *Rattus norvegicus*. *Journal of Comparative Psychology* **100**, 101-107.

Bourgeois, C.E. & O'Connell, M.F. 1988. Observations on the seaward migration of Atlantic salmon (*Salmo salar* L.) smolts through a large lake as determined by radiotelemetry and Carlin tagging studies. *Canadian Journal of Zoology* **66**, 685-691.

Brännäs, E. 1995. First access to territorial space and exposure to strong predation pressure: a conflict in early emerging Atlantic salmon (*Salmo salar* L.) fry. *Evolutionary Ecology* **9**, 411-420.

Bregazzi, P.R. & Kennedy, C.R. 1980. The biology of pike, *Esox lucius* L., in a southern eutrophic lake. *Journal of Fish Biology* **17**, 91-112.

Brinkhurst, R.O. 1966. Population dynamics of the large pondskater *Gerris najas* Degeer (Hemiptera-Heteroptera). *Journal of Animal Ecology* **35**, 13-25.

Broglio, C., Rodríguez, F. & Salas, C. 2003. Spatial cognition and its neural basis in teleost fishes. *Fish and Fisheries* **4**, 247-255.

Brönmark, C. & Miner, J.G. 1992. Predator-induced phenotypical change in body morphology in crucian carp. *Science* **258**, 1348-1350.

Brown, C. & Warburton, K. 1997. Predator recognition and anti-predator responses in the rainbowfish *Melanotaenia eachamensis. Behavioural Ecology and Sociobiology* **41**, 61-68.

Brown, C. & Warburton, K. 1999. Differences in the timidity and escape responses between predator-naïve and predator-sympatric rainbowfish populations. *Ethology* **105**, 491-502.

Brown, C. & Laland, K. 2001. Social learning and life skills training for hatchery reared fish. *Journal of Fish Biology* **59**, 471-493.

Brown, G. & Brown, J. 1992. Do rainbow trout and Atlantic salmon discriminate kin? *Canadian Journal of Zoology* **70**, 1636-1640.

Brown, G. & Brown, J. 1993. Social dynamics in salmonid fishes: do kin make better neighbours? *Animal Behaviour* **45**, 863-871.

Brown, G.E. 2003. Learning about danger: chemical alarm cues and local risk assessment in prey fishes. *Fish and Fisheries* **4**, 227-234.

Brown, G.E. & Smith, R.J.F. 1996. Foraging trade-offs in fathead minnows (*Pimephales promelas*, Osteichthyes, Cyprinidae): Acquired predator recognition in the absence of an alarm response. *Ethology* **102**, 776-785.

Brown, G.E. & Smith, R.J.F. 1998. Acquired predator recognition in juvenile rainbow trout (*Onchorhynchus mykiss*): conditioning hatchery reared fish to recognise chemical cues of a predator. *Canadian Journal of Fisheries and Aquatic Sciences* **55**, 611-617.

Brown, G.E. & Godin, J-G. J. 1999. Who dares, learns: chemical inspection behaviour and acquired predator recognition in a characin fish. *Animal Behaviour* **57**, 475-481.

Brown, G. E., Chivers, D.P. & Smith, R.J.F. 1996. Effects of diet on localized defecation by northern pike, *Esox lucius. Journal of Chemical Ecology* **22**, 467-475.

Brown, G.E., Chivers, D.P. & Smith, R.J.F. 1997. Differential learning rates in chemical versus visual cues of a northern pike by fathead minnows in a natural habitat. *Environmental Biology of Fishes* **49**, 89-96.

Brown, G.E., Paige, J.A. & Godin, J-G. J. 2000. Chemically mediated predator inspection behaviour in the absence of predator visual cues by a characin fish. *Animal Behaviour* **60**, 315-321.

Brown, G.E., Poirier, J.F. & Adrian, J.C. 2004. Assessment of local predation risk: the role of subthreshold concentrations of chemical alarm cues. *Behavioral Ecology* **15**, 810-815.

Brown, G. E., Adrian Jr., J. C. & Shih, M.L. 2001a. Behavioural responses of fathead minnows to hypoxanthine-s-N-oxide at varying concentrations. *Journal of Fish Biology* **58**, 1465-1470.

Brown, G. E., Adrian Jr., J. C., Patton, T. & Chivers, D. P. 2001b. Fathead minnows learn to recognize predator odour when exposed to concentrations of artificial alarm pheromone below their behavioural-response threshold. *Canadian Journal of Zoology* **79**, 2239-2245.

Bry, C. 1996. Role of vegetation in the life cycle of pike. In *Pike: Biology and Exploitation* (Craig, J.F., ed.), pp. 45-67. London: Chapman and Hall.

Bry, C. & Gillett, C. 1980. Reduction of cannibalism in pike (*Esox lucius*) fry by isolation of full-sib families. *Reproduction, Nutrition and Development* **20**, 173-182.

Budaev, S.V. 1997a. "Personality" in the guppy (*Poecilia reticulata*): A correlational study of exploratory behavior and social tendency. *Journal of Comparative Psychology* **111**, 399-411.

Budaev, S.V. 1997b. Alternative styles in the European wrasse (*Symphodus ocellatus*): Boldness-related schooling tendency. *Environmental Biology of Fishes* **49**, 71-78.

Burton, D.T. 1979. Ventilation frequency compensation responses of three thermal estuarine fish exposed to moderate temperature increases. *Journal of Fish Biology* **15**, 590-600.

Carl, J. & Larsen, M. 1994. Predation from pike (*Esox lucius* L.) and zander (*Stizostedion lucioperca* L.) on down-migrating trout smolts in the Bygholm catchment area.

M.Sc. Thesis, Department of Inland Fisheries and University of Aarhus. *In* Jepsen, N., Pedersen, S. & Thorstad, E. 2000. Short communication. Behavioural interactions between prey (trout smolts) and predators (pike and pikeperch) in an impounded river. *Regulated Rivers: Resource Management* **16**, 189-198.

Carlin, B. 1967. Salmon tagging experiments. Translation Series No. 951. *Fisheries Research Board of Canada* [Translated from Swedish, Report LFI Medd., Swedish Salmon Research Institute, January 1965].

Casselman, J.M. 1996. Age, growth and environmental requirements of pike. In *Pike, Biology and Exploitation* (Craig, J.F., ed.). pp 69-102. London: Chapman and Hall.

Casselman, J.M. & Lewis, C.A. 1996. Habitat requirements of northern pike (*Esox lucius*). *Canadian Journal of Fisheries and Aquatic Sciences* **53** (Suppl. A), 161-174.

Causby, L.A. & Smith, E. N. 1981. Control of fear bradycardia in the swamp rabbit, *Sylvilagus aquaticus*. *Comparative Biochemistry and Physiology* **69C**, 367-370.

Chadwick, E.M.P. 1986. Relationship between Atlantic salmon smolts and adults in Canadian rivers. In *Proceedings of the Third International Atlantic Salmon Symposium on Atlantic salmon: Planning for the future* (Mills, D. & Piggins, D. eds.). pp. 301-324. Portland, Oregon: Timber Press.

Chapman, L.J. & Mackay, W.C. 1990. Ecological correlates of feeding flexibility in northern pike (*Esox lucius*). *Journal of Freshwater Ecology* **5**, 313-322.

Charnov, E.L., Orians, E.H. & Hyatt, K. 1977. Ecological implications of resource depression. *American Naturalist* **110**, 247-259.

Chase, I.D., Tovey, C., Spangler-Martin, D. & Manfredonia, M. 2002. Individual differences versus social dynamics in the formation of animal dominance hierarchies. *Proceedings of the National Academy of Sciences of the USA* **99**, 5744-5749.

Chivers, D.P. & Smith, R.J.F. 1994a. The role of experience and chemical alarm signalling in predator recognition by fathead minnows, *Pimephales promelas*. *Journal of Fish Biology* **44**, 273-285.

Chivers, D.P. & Smith, R.J.F. 1994b. Fathead minnows, *Pimephales promelas*, acquire predator recognition when alarm substance is associated with the sight of an unfamiliar fish. *Animal Behaviour* **48**, 597-605.

Chivers, D.P. & Smith, R.J.F. 1995. Fathead minnows (*Pimephales promelas*) learn to recognize chemical stimuli from high risk habitats by the presence of alarm substance. *Behavioural Ecology* **6**, 155-158.

Chivers, D.P. & Smith, R.J.F. 1998. Chemical alarm signals in aquatic predator-prey systems: a review and prospectus. *Écoscience* **5**, 338-352.

Chivers, D.P., Brown, G.E. & Smith, R.J.F. 1995a. Acquired recognition of chemical stimuli from pike, *Esox lucius*, by brook sticklebacks, *Culaea inconstans* (Osteichthyes, Gasterosteidae). *Ethology* **99**, 234-242.

Chivers, D., Brown, G. & Smith, R. 1995b. Familiarity and shoal cohesion in fathead minnows (*Pimephales promelas*): implications for antipredator behavior. *Canadian Journal of Zoology* **73**, 955-960.

Chivers, D.P., Wildy, E.L. & Blaustein, A.R. 1997. Eastern long-toed salamander (*Ambystoma macrodactylum columbianum*) larvae recognize cannibalistic conspecifics. *Ethology* **103**, 187-197.

Chivers, D.P., Mirza, R.S., Bryer, P.J. & Kiesecker, J.M. 2001. Threat-sensitive predator avoidance by slimy sculpins: understanding the importance of visual versus chemical information. *Canadian Journal of Zoology* **79**, 867-873.

Ciepielewski, W. 1981. Population of pike (*Esox lucius* L.) in pond type Lake Warniak in 1969-1978. *Ekol. Pol.* **29**, 35-51. *In* Raat, A.J.P. 1988. Synopsis of biological data on the northern pike, *Esox lucius* Linnaeus, 1758. *FAO Fish. Synopsis 30 Rev. 2.*

Colgan, P.W., Brown, J.A. & Orsatti, S.D. 1986. Role of diet and experience in the development of feeding behaviour in large-mouth bass (*Micropterus salmoides*). *Journal of Fish Biology* **28**, 161-170.

Cook, M.F. & Bergersen, E.P. 1988. Movements, habitat selection, and activity periods of northern pike in Eleven Mile reservoir, Colorado. *Transactions of the American Fisheries Society* **117**, 495-502.

Côté, Y. & Pomerleau, C. 1985. Survie et dispersion d'alevins de saumon atlantique (*Salmo salar*) ensemencés en milieu naturel. *Nat. Can.* 112, 549-558.

Courtenay, S. C., Quinn, T. P., Dupuis, H. M. C., Groot, C. & Larkin, P. A. 1997. Factors affecting the recognition of population-specific odours by juvenile coho salmon. *Journal of Fish Biology* **50**, 1042-1060.

Courtenay, S.C., Quinn, T.P., Dupuis, H.M.C., Groot, C. & Larkin, P.A. 2001. Discrimination of family-specific odours by juvenile coho salmon: roles of learning and odour concentration. *Journal of Fish Biology* **58**, 107-125.

Craig, J.F. & Kipling, C. 1983. Reproduction effort versus the environment, case histories of Windermere perch, *Perca fluviatilis* L., and pike, *Esox lucius* L. *Journal of Fish Biology* **22**, 713-727.

Crisp, D.T. 1995. Dispersal and growth rate of 0-group salmon (*Salmo salar* L.) from point-stocking together with some information from scatter stocking. *Ecology of Freshwater Fish* **4**, 1-8.

Croy, M.I. & Hughes, R.N. 1991. The influence of hunger on feeding behaviour and the acquisition of learned foraging skills by the fifteen-spined stickleback, *Spinachia spinachia* L. *Animal Behaviour* **41**, 161-170.

Crozier, W.W. & Kennedy, G.J.A. 1993. Marine survival of wild and hatchery-reared Atlantic salmon (*Salmo salar* L.) from the River Bush, Northern Ireland. In *Salmon in the Sea, and New Enhancement Strategies* (Mills, D. ed.). pp. 139-162. Oxford: Fishing New Books.

Crozier, W.W., Moffett, I.J.J. & Kennedy, G.J.A. 1997. Comparative performance of native and non-native strains of Atlantic salmon (*Salmo salar* L.) ranched from the River Bush, Northern Ireland. *Fisheries Research* **32**, 81-88.

Curio, E., Klump, G. & Regelmann, K. 1983. An anti-predator response in the great tit (*Parus major*): is it tuned to predator risk? *Oecologia* **60**, 83-88.

Davies, C., Shelley, J., Harding, P., McLean, I., Gardiner, R. & Peirson, G. 2004. *Freshwater Fishes in Britain*. pp. 98-100. England: Harley Books.

Day, R.L., MacDonald, T., Brown, C., Laland, K.N. & Reader, S.M. 2001. Interactions between shoal size and conformity in guppy social foraging. *Animal Behaviour* **62**, 917-925.

Dellefors, C. & Johnsson, J.I. 1995. Foraging under risk of predation in wild and hatchery-reared juvenile sea trout (*Salmo trutta*). *Nordic Journal of Freshwater Resources* **70**, 31-37.

DeVries, D.R., Stein, R.A. & Chesson, P.L. 1989. Sunfish foraging among patches: the patch-departure decision. *Animal Behaviour* **37**, 455-464.

Diana, J.S. 1979. The feeding pattern and daily ration of a top carnivore, the northern pike (*Esox lucius*). *Canadian Journal of Zoology* **57**, 2121-2127.

Diana, J.S. 1980. Diel activity pattern and swimming speeds of northern pike (*Esox lucius*) in Lac Ste. Anne, Alberta. *Canadian Journal of Fisheries and Aquatic Sciences* **37**, 1454-1458.

Diana, J.S. 1983. An energy budget for Northern pike (*Esox lucius*). *Canadian Journal of Zoology* **61**, 1968-1975.

Diana, J.S., Mackay, W.C. & Ehrman, M. 1977. Movements and habitat preference of northern pike (*Esox lucius*) in Lac Ste. Anne, Alberta. *Transactions of the American Fisheries Society* **106**, 560-565.

Dill, L.M. & Fraser, A.H.G. 1984. Risk of predation and the feeding behaviour of juvenile coho salmon (*Oncorhynchus kisutch*). *Behavioural Ecology and Sociobiology* **16**, 65-71.

Dittman, A.H. & Quinn, T.P. 1996. Homing in Pacific salmon: mechanisms and ecological basis. *The Journal of Experimental Biology* **199**, 83-91.

Donnelly, W.A. & Whoriskey, F.G. 1991. Background-color acclimation of brook trout for crypsis reduces risk of predation by hooded mergansers *Lophodytes cucullatus. North American Journal of Fisheries Management* **11**, 206-211.

Dukas, R. 1998a. *Cognitive Ecology.* Chicago: University of Chicago Press.

Dukas, R. 1998b. Evolutionary ecology of learning. In *Cognitive Ecology* (Dukas, R. ed.). pp. 129-174. Chicago: University of Chicago Press.

Dukas, R. 1998c. Constraints on information processing and their effects on behavior. In *Cognitive Ecology* (Dukas, R. ed.). pp. 89-128. Chicago: University of Chicago Press.

Dupuch, A., Magnan, P. & Dill, L. M. 2004. Sensitivity of northern redbelly dace, *Phoxinus eos*, to chemical alarm cues. *Canadian Journal of Zoology* **82**, 407-415.

Egglishaw, H.J. & Shackley, P.E. 1973. An experiment on faster growth of salmon *Salmo salar* (L.) in a Scottish stream. *Journal of Fish Biology* **5**, 197-204.

Egglishaw, H.J. & Shackley, P.E. 1977. Growth, survival and production of juvenile salmon and trout in a Scottish stream 1966-1975. *Journal of fish Biology* **11**, 647-672.

Egglishaw, H.J. & Shackley, P.E. 1980. Survival and growth of salmon, *Salmo salar* (L.) planted in a Scottish stream. *Journal of Fish Biology* **16**, 565-584.

Einum, S. & Fleming, I.A. 1997. Genetic divergence and interactions in the wild among native, farmed and hybrid Atlantic salmon. *Journal of Fish Biology* **50**, 634-651.

Einum, S. & Fleming, I.A. 2000. Selection against late emergence and small offspring in Atlantic salmon (*Salmo salar*). *Evolution* **54**, 628-639.

Einum, S. and Nislow, K.H. 2005. Local density dependence in contiguous habitats: an experimental test using Atlantic salmon. *Oecologia* **143**, 203-210

Eklöv, P. 1992. Group foraging versus solitary foraging efficiency in piscivorous predators: the perch, *Perca fluviatilis*, and pike, *Esox lucius*, patterns. *Animal Behaviour* **44**, 313-326.

Eklöv, P. 1997. Effects of habitat complexity and prey abundance on the spatial and temporal distribution of perch (*Perca fluviatilis*) and pike (*Esox lucius*). *Canadian Journal of Fisheries and Aquatic Sciences* **54**, 1520-1531.

Eklöv, P. & Diehl, S. 1994. Piscivore efficiency and refuging prey: the importance of predator search mode. *Oecologia* **98**, 345-353.

Eklöv, P. & Hamrin, S.F. 1989. Predator efficiency and prey selection: interactions between pike *Esox lucius*, perch *Perca fluviatilis* and rudd *Scardinus erythrophthalmus*. *Oikos* **56**, 149-156.

Eklöv, P. & Persson, L. 1996. The response of prey to the risk of predation: proximate cues for refuging juvenile fish. *Animal Behaviour* **51**, 105-115.

Elgan, M.A. 1989. Predator vigilance and group size in mammals and birds – A critical review of the empirical evidence. *Biological Reviews of the Cambridge Philosophical Society* **64**, 13-33.

Elliott, J.M. 1989a. The critical period concept for juvenile survival and its relevance for population regulation in young sea trout, *Salmo trutta. Journal of Fish Biology* **35** (Suppl. A), 91-98.

Elliott, J.M. 1989b. Mechanisms responsible for population regulation in young brown, *Salmo trutta* I. The critical time for survival. *Journal of Animal Ecology* **58**, 987-1001.

Elliott, J.M. 1994. *Quantitative ecology and the brown trout.* Oxford: Oxford University Press.

Ellis, D.H., Dobrott, S.J. & Goodwin, J.G. 1977. Reintroduction techniques for Masked Bobwhites. In *Endangered birds: management techniques for preserving threatened species* (Temple, S.A., ed.). pp. 345-354. Madison: University of Wisconsin Press.

Elson, P.F. 1957. Using hatchery-reared Atlantic salmon to best advantage. *Canadian Fish Culturist* **21**, 7-17.

Elson, P.F. 1975. Atlantic salmon rivers, smolt production and optimal spawning: an overview of natural production. *International Atlantic Salmon Foundation Special Publication Series* **6**, 96-119.

Endler, J.A. 1991. Interactions between predators and prey. In *Behavioural Ecology. An Evolutionary Approach* (Krebs, J.R. & Davies, N.B. eds.). Oxford: Blackwell Scientific Publications.

Engström-Öst, J. & Lehtiniemi, M. 2004. Threat-sensitive predator avoidance by pike larvae. *Journal of Fish Biology* **65**, 251-261.

Erkinaro, J. & Gibson, R.J. 1997. Interhabitat migration of juvenile Atlantic salmon in a Newfoundland river system, Canada. *Journal of Fish Biology* **51**, 373-388.

Erkinaro, J., Julkunen, M. & Niemela, E. 1998. Migration of juvenile Atlantic salmon *Salmo salar* in small tributaries of the subarctic River Teno, northern Finland. *Aquaculture* **168**, 105-119.

Espmark, Y. & Langvatn, R. 1985. Development and habituation of cardiac and behavioural responses in young red deer calves (*Cervus elaphus*) exposed to alarm stimuli. *Journal of Mammalogy* **66**, 702-711.

Evans, C.S., Macedonia, J.M. & Marler, P. 1993. Effects of apparent size and speed on the response of chickens, *Gallus gallus*, to computer-generated simulations of aerial predators. *Animal Behaviour* **46**, 1-11.

Farooqi, M. & Aprahamian, M.W. 1995. *An evaluation of the survival of Atlantic salmon (Salmo salar L.) fry stocked in eight streams in the Northwest of England,* NRA/NW/FTR/95/8.

Felton, S.P., Wenjuan, J. & Matthews, S.B. 1990. Selenium concentrations in coho salmon outmigrants smolts and returning adults: a comparison of wild versus hatchery reared fish. *Dis. Aquat. Org*. **9**, 157-161.

Fernö, A. & Järvi, T. 1998. Domestication genetically alters the anti-predator behaviour of anadromous brown trout (*Salmo trutta*) – a dummy predator experiment. *Nordic Journal of Freshwater Research* **74**, 95-100.

Fish, G.R. 1960. The comparative activity of some digestive enzymes in the alimentary canal of tilapia and perch. *Hydrobiologia* **15**, 161-178.

FitzGibbon, C.D. 1989. A cost to individuals with reduced vigilance in groups of Thomson's gazelles hunted by cheetahs. *Animal Behaviour* **37**, 508-510.

Fleming, I.A. 1996. Reproductive strategies of Atlantic salmon: ecology and evolution. *Reviews of Fish Biology and Fisheries* **6**, 376-416.

Fleming, I.A. 1998. Pattern and variability in the breeding system of Atlantic salmon (*Salmo salar*), with comparisons to other salmonids. *Canadian Journal of Fisheries and Aquatic Sciences* **55** (Suppl. 1), 59-76.

Fleming, I.A. & Einum, S. 1997. Experimental tests of genetic divergence of farmed from wild Atlantic salmon due to domestication. *ICES Journal of Marine Science* **54**, 1051-1063.

Fleming, I.A., Jonsson, B. & Gross, M.R. 1994. Phenotypic divergence of sea-ranched, farmed, and wild salmon. *Canadian Journal of Fisheries and Aquatic Sciences* 51, 2808-2824.

Fleming, I.A., Lamberg, A. & Jonsson, B. 1997. Effects of early experience on the reproductive performance of Atlantic salmon. *Behavioral Ecology* **8**, 470-480.

Forward Jr, R.B., Tankersley, R.A., Smith, K.A. & Welch, J.M. 2003. Effects of chemical cues on orientation of blue crab, *Callinectes sapidus*, megalopae in flow: implications for location of nursery areas. *Marine Biology* **142**, 747-756.

Fraser, D.F. & Sise, T.E. 1980. Observations on stream minnows in a patchy environment: a test of a theory of habitat distribution. *Ecology* **61**, 790-797.

Fraser, D.F. & Cerri, R.D. 1982. Experimental evaluation of predator-prey relationships in a patchy environment: consequences for habitat use patterns in minnows. *Ecology* **63**, 307-313.

Fraser, D.F. & Gilliam, J.F. 1987. Feeding under predation hazard: response of the guppy and Hart's rivulus from sites with contrasting predation hazard. *Behavioral Ecology and Sociobiology* **21**, 203-209.

Fraser, D.F., Gilliam, J.F., Daley, M.J., Le, A.N. & Skalski, G.T. 2001. Explaining leptokurtic movement distributions: Intrapopulation variation in boldness and exploration. *The American Naturalist* **158**, 124-135.

Fretwell, S.D. 1972. *Populations in a Seasonal Environment*. Princeton: Princeton University Press.

Fretwell, S.D. & Lucas Jr, H.L. 1970. On territorial behaviour and other factors influencing habitat distribution in birds. I. Theoretical development. *Acta Biotheoretica* **19**, 16-36.

Frost, W.E. 1954. The food of pike (*Esox lucius* L.) in Windermere. *Journal of Animal Ecology* **23**, 339-360.

Frost, W.E. & Kipling, C. 1967. A study of reproduction, early life, weight-length relationship and growth of pike (*Esox lucius* L.) in Windermere. *Journal of Animal Ecology* **36**, 651-693.

Fuiman, L.A. & Magurran, A.E. 1994. Development of predator defences in fishes. *Reviews in Fish Biology and Fisheries* **4**, 145-183.

Fuiman, L.A., Smith, M.E. & Malley, V.N. 1999. Ontogeny of routine swimming speed and startle responses in red drum, with a comparison of responses to acoustic and visual stimuli. *Journal of Fish Biology* **55** (Suppl. A), 215-226.

Gagliardo, A., Ioale, P., Odetti, F. & Bingman, V.P. 2001. The ontogeny of the homing pigeon navigational map: evidence for a sensitive learning period. *Proceedings of the Royal Society of London B* **268**, 197-202.

Garcia de Leanniz, C. Fraser, N. & Huntingford, F.A. 2000. Variability in performance in wild Atlantic salmon, *Salmo salar* L., fry from a single redd. *Fisheries Management and Ecology* **7**, 489-502.

Gardiner, W.R. & Shackley, P.E. 1991. Stock and recruitments and density-independent growth of salmon, *Salmo salar* L., in a Scottish stream. *Journal of Fish Biology* **38**, 691-696.

Gerlai, R. 1993. Can paradise fish (*Macropodus opercularis*, Anabantidae) recognize a natural predator? An ethological analysis. *Ethology* **94**, 127-136.

Giles, N., Wright, R.M. & Nord, M.E. 1986. Cannibalism in pike fry, *Esox lucius* L.: some experiments with fry densities. *Journal of Fish Biology* **29**, 107-113.

Gilliam, J.F. & Fraser, D.F. 1987. Habitat selection under predation hazard-test of a model with foraging minnows. *Ecology* **68**, 1856-1862.

Glass, N.R. 1971. Computer analysis of predation energetics in the largemouth bass. In *Systems Analysis and Simulation in Ecology. Volume 1* (Pattern, B.C. ed.). pp. 325-363. New York: Academic Press.

Godin, J.-G. J. & Davis, S.A. 1995. Who dares, benefits: predator approach behaviour in the guppy (*Poecilia reticulata*) deters predator pursuit. *Proceedings of the Royal Society London Series B* **259**, 193-200.

Goss-Custard, J.D., West, A.D., Stillman, R.A., Le, S.E.A., Dit Durell, V., Caldow, R.W.G., McGrorty, S. & Nagarajan, R. 2001. Density-dependent starvation in a vertebrate without significant depletion. *Journal of Animal Ecology* **70**, 955-965.

Gotceitas, G. & Godin, J.-G.J. 1991. Foraging under the risk of predation in juvenile Atlantic salmon (*Salmo salar* L.): effects of social status and hunger. *Behavioral Ecology and Sociobiology* **29**, 255-261.

Gotelli, N.J. 1993. Ant lion zones: causes of high-density predator aggregations. *Ecology* **74**, 226-237.

Gottlieb, G. 1961. Developmental age as a baseline for determination of the critical period in imprinting. *Journal of Comparative Physiology and Psychology* **54**, 422-427.

Göz, H. 1941. Über den art- und individualgeruch bei Fischen. *Zeitschrift für vergleichende Physiologie* **29**, 1-45.

Grant, J.W.A. & Kramer, D.L. 1990. Territory size as a predictor of the upper limit to population density of juvenile salmonids in streams. *Canadian Journal of Fisheries and Aquatic Sciences* **47**, 1724-1737.

Green, D.M. & Swets, J.A. 1966. *Signal Detection Theory and Psychophysics*. New York: Wiley.

Greenberg, L.A., Paszkowski, C.A. & Tonn, W.M. 1995. Effects of prey species composition and habitat structure on foraging by two functionally distinct piscivores. *Oikos* **74**, 522-532.

Griffin, A.S., Evans, C.S. & Blumstein, D.T. 2002. Selective learning in a marsupial. *Ethology* **108**, 1103-1114.

Griffin, A.S., Evans, C.S. & Blumstein, D.T. 2001. Learning specificity in acquired predator recognition. *Animal Behaviour* **62**, 577-589.

Griffin, A.S., Blumstein, D.T. & Evans, C.S. 2000. Training captive-bred or translocated animals to avoid predators. *Conservation Biology* **14**, 1317-1326.

Griffiths, R.A., Schley, A., Sharp, P.E., Dennis, J.L. & Roman, A. 1998. Behavioural responses of Mallorcan midwife toad tadpoles to natural and unnatural snake predators. *Animal Behaviour* **55**, 207-214.

Griffiths, S.W. 2003. Learned recognition of conspecifics by fishes. *Fish and Fisheries* **4**, 256-268.

Griffiths, S.W. & Armstrong, J.D. 2002. Kin-biased territory overlap and food sharing among Atlantic salmon juveniles. *Journal of Animal Ecology* **71**, 480-486.

Grimm, M.P. 1981. Intraspecific predation as a principal factor controlling the biomass of northern pike (*Esox lucius* L.). *Fisheries Management* **12**, 77-79.

Grimm, M.P. 1983. Regulation of biomasses of small (<41cm) northern pike (*Esox lucius* L.), with special reference to the contribution of individuals stocked as fingerlings (4-6cm). *Fisheries Management* **14**, 115-134.

Grimm, M.P. & Klinge, M. 1996. Pike and some aspects of its dependence on vegetation. In *Pike: Biology and Exploitation* (Craig, J.F. ed.). pp. 125-156. London: Chapman and Hall.

Gulin, V.V. & Rudenko, G.P. 1973. Procedure for assessment of fish production in lakes. *Journal of Ichthyology* **13**, 813-823.

Gustafson-Marjanen, K.I. & Moring, J.R. 1984. Construction of artificial redds for evaluating survival of Atlantic salmon eggs and alevins. *North American Journal of Fisheries Management* **4**, 455-461.

Handeland, S.O., Järvi, T., Fernö, A. & Stefansson, S.O. 1996. Osmotic stress, antipredator behaviour, and mortality of Atlantic salmon (*Salmo salar*) smolts. *Canadian Journal of Fisheries and Aquatic Sciences* **53**, 2673-2680.

Hanlon, R.T., Forsythe, J.W. & Joneschild, D.E. 1999. Crypsis, conspicuousness, mimicry and polyphenism as antipredator defences of foraging on Indo-Pacific coral reefs, with a method of quantifying crypsis from video tapes. *Biological Journal of the Linnean Society* **66**, 1-22.

Hansen, L.P., Jonsson, B. & Døving, K.B. 1984. Migration of wild and hatchery reared smolts of Atlantic salmon, *Salmo salar* L., through lakes. *Journal of Fish Biology* **25**, 617-623.

Hansen, L.P., Døving, K.B. & Jonsson, B. 1987. Migration of farmed adult Atlantic salmon with and without olfactory sense, released on the Norwegian coast. *Journal of Fish Biology* **30**, 713-721.

Hara, T.J. & Zielinski, B. 1989. Structural and functional development of the olfactory organ in teleosts. *Transactions of the American Fisheries Society* **118**, 183-194.

Harper, D.G.C. 1982. Competitive foraging in mallards: 'ideal free' ducks. *Animal Behaviour* **30**, 575-584.

Hart, P.J.B. & Pitcher, T.J. 1973. Population densities and growth of five species of fish in the River Nene, Northamptonshire. *Journal of the Institute of Fisheries Management* **4**, 69-86.

Hartman, E.J. & Abrahams, M.V. 2000. Sensory compensation and the detection of predators: the interaction between chemical and visual information. *Proceedings of the Royal Society of London B* **267**, 571-575.

Harvey, C., Garneau, F-X. & Himmelman, J.H. 1987. Chemodetection of the predatory seastar *Leptasterias polaris* by the whelk *Buccinum undatum*. *Marine Ecology Progress Series* **40**, 79-86.

Harvie, C.J. & Amoro, P.G. 1996. Indicies of marine habitat for Atlantic salmon (Salmo salar) and trends in survival of hatchery-origin smolts. *Atlantic Fisheries Research Document* No. **96/140**. Department of Fisheries and Oceans, Dartmouth, NS.

Hasler, A.D. 1966. *Underwater Guideposts*. Madison, Wisconsin: University of Wisconsin Press.

Hasler, A.D. & Scholtz, A.T. 1983. *Olfactory Imprinting and Homing in Salmon*. Berlin: Springer Zoophysiology.

Hasler, A.D., Scholz, A.T. & Horrall, R.M. 1978. Olfactory imprinting and homing in salmon. *American Scientist* **66**, 347-355.

Hasson, O. 1991. Pursuit-deterrent signals: communication between predator and prey. *Trends in Ecology and Evolution* **6**, 325-329.

Hawkins, L.A., Armstrong, J.D. & Magurran, A.E. 2003. Settlement and habitat use by juvenile pike in early winter. *Journal of Fish Biology* **63** (Suppl. A), 174-186.

Hawkins, L.A., Armstrong, J.D. & Magurran, A.E. 2004a. Predator-induced hyperventilation in wild and hatchery Atlantic salmon fry. *Journal of Fish Biology* **65** (Suppl. A), 88-100.

Hawkins, L.A., Magurran, A.E. & Armstrong, J.D. 2004b. Innate predator recognition in newly-hatched Atlantic salmon. *Behaviour* **141**, 1249-1262.

Healy, M.C. & Reinhardt, U. 1995. Predator avoidance in naïve and experienced juvenile chinook and coho salmon. *Canadian Journal of Fisheries and Aquatic Sciences* **52**, 614-622.

Helfman, G.S. 1989. Threat-sensitive predator avoidance in damselfish-trumpetfish interactions. *Behavioral Ecology and Sociobiology* **24**, 47-58.

Henderson, J.N. & Letcher, B.H. 2003. Predation on stocked Atlantic salmon (*Salmo salar*) fry. *Canadian Journal of Fisheries and Aquatic Sciences* **60**, 32-42.

Hensor, E.M.A., Godin, J.-G.J., Hoare, D.J. & Krause, J. 2003. Effects of nutritional state on the shoaling tendency of banded killifish, *Fundulus diaphanous*, in the field. *Animal Behaviour* **65**, 663-669.

Herbert, N.A., Armstrong, J.D. & Björnsson, B.T. 2001. Evidence that growth hormone-induced elevation in routine metabolism of juvenile Atlantic salmon is a result of increased spontaneous activity. *Journal of Fish Biology* **59**, 654-657.

Herzog, M. & Hopf, S. 1984. Behavioural responses to species specific warning calls in infant squirrel monkeys reared in social isolation. *American Journal of Primatology* **7**, 99-106.

Hessing, H.A.M., van Beek, J.A.M., Wiepkema, P.R. & Schouten, W.P.G. 1994. Individual behavioral and physiological strategies in pigs. *Physiological Behaviour* **55**, 39-46.

Hickam, J.B., Cargill, W.H. & Golden, A. 1948. Cardiovascular reactions to emotional stimuli. Effect on the cardiac output, arteriovenous oxygen difference, arterial pressure, and peripheral resistance. *Journal of Clinical Investigations* **27**, 290-298.

Higgs, D.M. & Fuiman, L.A. 1996. Ontogeny of visual and mechanosensory structure and function in Atlantic menhaden *Brevoortia tyrannus. The Journal of Experimental Biology* **199**, 2619-2629.

Hinde, R.A. 1960. Factors governing the changes in strength of a partially inborn response, as shown by the mobbing behaviour of the chaffinch (*Fringilla coelebs*). III. The interaction of short-term and long-term incremental and decremental effects. *Proceedings of the Royal Society of London B* **153**, 398-420.

Hirvonen, H., Ranta, E., Piironen, J., Laurila, A. & Peuhkuri, N. 2000. Behavioural responses of naïve Arctic charr to chemical cues from salmonid and non-salmonid fish. *Oikos* **88**, 191-199.

Hjort, J. 1914. Fluctuations in the great fisheries of northern Europe viewed in the light of biological research. *Rapports et Procés-Verbaux desRréunions. ConseilInternational pour l'Exploration de la Mer* **20**, 1-228.

Hoar, W.S. 1976. Smolt transformation: Evolution, behavior and physiology. *Journal of the Fisheries Research Board of Canada* **33**, 1233-1252.

Höjesjö, J., Johnsson, J. I. & Axelsson, M. 1999. Behavioural and heart rate responses to food limitation and predation risk: an experimental study on rainbow trout. *Journal of Fish Biology* **55**, 1009-1019.

Holbrook, S.J. & Schmitt, R.J. 2002. Competition for shelter space causes density-dependent predation mortality in damselfishes. *Ecology* **83**, 2855-2868.

Holland, L.E. & Huston, M.L. 1984. Relationship of young-of-the-year northern pike to aquatic vegetation types in backwaters of the upper Mississippi River. *North American Journal of Fisheries Management* **4**, 514-522.

Holley, A.J.F. 1993. Do brown hares signal to foxes? *Ethology* **94**, 21-30.

Holopainen, I.J., Aho, J., Vornanen, M. & Huuskonen, H. 1997. Phenotypic plasticity and predator effects on morphology and physiology of crucian carp in nature and in the laboratory. *Journal of Fish Biology* **50**, 781-798.

Horat, P. & Semlitsch, R.D. 1994. Effects of predation risk and hunger on the behaviour of two species of tadpoles. *Behavioral Ecology and Sociobiology* **34**, 393-401.

Horrall, R.M. 1981. Behavioral stock-isolating mechanisms in Great Lakes fishes with special reference to homing and site imprinting. *Canadian Journal of Fisheries and Aquatic Sciences* **38**, 1481 -1496.

Howe, N.R. & Sheikh, Y.M. 1975. Anthopleurine: a sea anemone alarm pheromone. *Science* **189**, 386-388.

Hughes, G.M. & Shelton, G. 1962. Respiratory mechanisms and their nervous control in fish. In *Advances in Comparative Physiology and Biochemistry, Volume 1* (Lowenstein, O. E. ed.). New York: Academic Press.

Hughes, N.F. 1992. Ranking of feeding positions by drift-feeding arctic grayling (*Thymallus arcticus*) in dominance hierarchies. *Canadian Journal of Fisheries and Aquatic Sciences* **49**, 1994-1998.

Hughes, R.N. & Croy, M.I. 1993. An experimental analysis of frequency-dependent predation (switching) in the 15-spined stickleback, *Spinachia spinachia. Journal of Animal Ecology* **62**, 341-352.

Hunt, B.P. & Carbine, W.F. 1951. Food of young pike, *Esox lucius* L., and associated fishes in Peterson's ditches, Houghton Lake, Michigan. *Transactions of the American Fisheries Society* **80**, 67-83.

Huntingford, F.A. & Wright, P.J. 1993. The development of adaptive variation in predator avoidance in freshwater fishes. *Marine Behaviour and Physiology* **23**, 45-61.

Huntingford, F.A., Metcalfe, N.B., Thorpe, J.E., Graham, W.D. & Adams, C.E. 1990. Social dominance and body size in Atlantic salmon parr, *Salmo salar* L. *Journal of Fish Biology* **36**, 877-881.

Hutchings, J.A. 1986. Lakeward migrations by juvenile Atlantic salmon, *Salmo salar*. *Canadian Journal of Fisheries and Aquatic Sciences* **43**, 732-741.

Irving, P.W. & Magurran, A.E. 1997. Context-dependent fright reactions in captive European minnows: the importance of naturalness in laboratory experiments. *Animal Behaviour* **53**, 1193-1201.

Isaksson, A. 1979. Salmon ranching in Iceland. In *Salmon Ranching* (Thorpe, J.E. ed.). pp. 131-156. London: Academic Press.

Jahn, L.A. 1972. Development of the olfactory apparatus of the cutthroat trout. *Transactions of the American Fisheries Society* **101**, 284-289.

James, S.E. & M'Closkey, R.T. 2004. Patterns of body size and habitat use in a lizard assemblage. *Écoscience* **11**, 160-167.

Järvi, T. & Uglem, I. 1993. Predator training improves the anti-predator behaviour of hatchery reared Atlantic salmon (*Salmo salar*) smolt. *Nordic Journal of Freshwater Resources* **68**, 63-71.

Jepsen, N., Aarestrup, K., Okland, F. & Rasmussen, G. 1998. Survival of radio-tagged Atlantic salmon (*Salmo salar* L.) and trout (*Salmo trutta* L.) smolts passing a reservoir during seaward migration. *Hydrobiologia* **372**, 347-353.

Jepsen, N., Pedersen, S. & Thorstad, E. 2000. Short communication. Behavioural interactions between prey (trout smolts) and predators (pike and pikeperch) in an impounded river. *Regulated Rivers: Resource Management* **16**, 189-198.

Jepsen, N., Beck, S., Skov, C. & Koed, A. 2001. Behaviour of pike (*Esox lucius* L.) >50cm in a turbid reservoir and in a clearwater lake. *Ecology of freshwater fish* **10**, 26-34.

Jessop, B.M. 1984. *Relationships between egg deposition, survival and the production of 0+ salmon parr in Maritime rivers.* Research Document No. 84/86. Canadian Atlantic Fisheries Scientific Advisory Committee, Dartmouth, NS.

Johnsen, B.O. & Ugedal, O. 1989. Feeding by hatchery-reared brown trout, *Salmo trutta* L. released in lakes. *Aquaculture and Fisheries Management* **20**, 97-104.

Johnsson, J.I. 1993. Big and brave: size selection affects foraging under risk of predation in juvenile rainbow trout, *Oncorhynchus mykiss. Animal Behaviour* **45**, 1219-1225.

Johnsson, J.I., Petersson, E., Jonsson, E., Björnsson, B.T. & Järvi, T. 1996. Domestication and growth hormone alter antipredator behaviour and growth patterns in juvenile brown trout, *Salmo trutta*. *Canadian Journal of Fisheries and Aquatic Sciences* **53**, 1546-1554.

Johnsson, J.I., Höjesjö, J. & Fleming, I.A. 2001. Behavioural and heart rate responses to predation risk in wild and domesticated Atlantic salmon. *Canadian Journal of Fisheries and Aquatic Sciences* **58**, 788-794.

Johnston, I.A. 1982. Physiology of muscle in hatchery raised fish. *Comparative Biochemistry and Physiology B* **73**, 105-124.

Jokokokko, E. & Jutila, E. 1998. The effects of stocking with salmon parr, *Salmo salar*, on the smolt production in the River Simojoki, Northern Finland. In *Stocking and Introduction of Fish* (Cowx, I.G. ed.). pp. 35-45. Oxford: Fishing New Books.

Jones, G. 1994. *A review of the Welsh region microtagging programme 1984-1993.* Internal Report. National Rivers Authority, Welsh Region.

Jonsson, N., Jonsson, B. & Hansen, L.P. 2003. The marine survival and growth of wild and hatchery-reared Atlantic salmon. *Journal of Applied Ecology* **40**, 900-911.

Jonsson, B., Jonsson, N. & Hansen, L.P. 1991. Differences in life history and migratory behaviour between wild and hatchery-reared Atlantic salmon in nature. *Aquaculture* **98**, 69-78.

Jordão, L.C. & Volpato, G.L. 2000. Chemical transfer of warning information in non-injured fish. *Behaviour* **137**, 681-690.

Kacelnik, A., Krebs, J.R. & Bernstein, C. 1992. The ideal free distribution and predator-prey populations. *Trends in Ecology and Evolution* **7**, 50-55.

Kalleberg, H. 1958. Observations in a stream tank of territoriality and competition in juvenile salmon and trout (*Salmo salar* L. and *S. trutta* L.). *Report of the Institute of Freshwater Research Drottningholm* **39**, 55-98.

Kanayama, Y. 1968. Studies of the conditioned reflex in lower vertebrates X. Defensive conditioned reflex of chum salmon fry in group. *Marine Biology* **2**, 77-87.

Kats, L.B. & Dill, L.M. 1998. The scent of death: chemosensory assessment of predation risk by prey animals. *Écoscience* **5**, 361-394.

Keefe, M.L. 1992. Chemically mediated avoidance behaviour in wild brook trout, *Salvelinus alpinus*: The response to familiar and unfamiliar predaceous fishes and the influence of fish diet. *Canadian Journal of Zoology* **70**, 288-292.

Kelley, J.L. & Magurran, A.E. 2003. Learned predator recognition and antipredator responses in fish. *Fish and Fisheries* **4**, 216-226.

Kelley, J.L., Graves, J.A. & Magurran, A.E. 1999. Familiarity breeds contempt in guppies. *Nature* **401**, 661-662.

Kempinger, J.J. & Carline, R.F. 1978a. Changes in population density, growth, and harvest of northern pike in Escabana Lake after implementation of a 22-inch size limit. *Technical Bulletin of Wisconsin Department of Natural Resources* **104**, p. 16. *In* Raat, A.J.P. 1988. Synopsis of biological data on the northern pike, *Esox lucius* Linnaeus, 1758. *FAO Fish. Synopsis 30 Rev. 2.*

Kempinger, J.J. & Carline, R.F. 1978b. Dynamics of the northern pike population and changes that occurred with a minimum size limit in Escabana Lake, Wisconsin. *Special Publication of the American Fisheries Society* **11**, 382-389. *In* Raat, A.J.P. 1988. Synopsis of biological data on the northern pike, *Esox lucius* Linnaeus, 1758. *FAO Fish. Synopsis 30 Rev. 2.*

Kennedy, G.J.A. & Strange, C.D. 1981. Comparative survival from salmon (*Salmo salar* L.) stocking with eyed and green ova in an upland stream. *Fisheries Management* **12**, 43-48.

Kennedy, G.J.A. & Strange, C.D. 1986. The effects of intra- and inter-specific competition on the survival and growth of stocked juvenile Atlantic salmon, *Salmo salar* L., and resident brown trout, *Salmo trutta* L., in an upland stream. *Journal of Fish Biology* **28**, 479-489.

Kennedy, G.J.A. & Greer, J.E. 1988. Predation by cormorants, *Phalacrocorax carbo* (L.), on the salmonid populations of an Irish river. *Aquaculture and Fisheries Management* **19**, 159-170.

Kieffer, J.D. & Colgan, P.W. 1992. The role of learning in fish behaviour. *Reviews in Fish Biology and Fisheries* **2**, 125-143.

Kipling, C. & Frost, W.E. 1970. A study of the mortality, population numbers, year class strengths, production and food consumption of pike, *Esox lucius* L., in Windermere from 1944-1962. *Journal of Animal Ecology* **39**, 115-157.

Krause, J. & Ruxton, G.D. 2002. *Living in Groups*. Oxford: Oxford University Press.

Krause, J., Butlin, R.K., Peuhkuri, N. & Pritchard, V.L. 2000. The social organization of fish shoals: a test of the predictive power of laboratory experiments for the field. *Biological Reviews* **75**, 477-501.

Kusch, R.C., Mirza, R.S. & Chivers, D.P. 2004. Making sense of predator scents: investigating the sophistication of predator assessment abilities of fathead minnows. *Behavioral Ecology and Sociobiology* **55**, 551-555.

Labat, R. 1966. Electrocardiologie chez les poissons teleosteens: influence de quelques facteurs ecologique. *Annals of Limnology* **2**, 1-175.

Laitinen, M. & Valtonen, T. 1994. Cardiovascular, ventilatory and total activity responses of brown trout to handling stress. *Journal of Fish Biology* **45**, 933-942.

Laland, K.N., Brown, C. & Krause, J. 2003. Learning in fishes: from three-second memory to culture. *Fish and Fisheries*, **4**, 199-202.

Larsson, P.O. 1985. Predation on migrating smolt as a regulating factor in Baltic salmon, *Salmo salar* L., populations. *Journal of Fish Biology* **26**, 391-397.

Lawrence, J.M. 1991. A chemical alarm response in *Pycnopodia helianthoides* (Echinodermata: asteroidea). *Marine Behaviour and Physiology* **19**, 39-44.

Lee, P.C. 1994. Social structure and evolution. In *Behaviour and Evolution* (Slater, P.J.B. Halliday, T.R. eds.). pp. 266-303. Cambridge: Cambridge University Press.

Lehtiniemi, M. 2005. Swim or hid: predator cues cause species-specific reactions in young fish larvae. *Journal of Fish Biology* **66**, 1285-1299.

Leider, S.A., Hulett, P.L., Lock, J.J. & Chilote, M.W. 1990. Electrophoretic comparison of the reproductive success of naturally spawning transplanted and wild steelhead trout through the returning adult stage. *Aquaculture* **88**, 239-252.

Leonard, J.B.K. & McCormick, S.D. 2001. Metabolic enzyme activity during smolting in stream- and hatchery-reared Atlantic salmon (*Salmo salar*). *Canadian Journal of Fisheries and Aquatic Sciences* **58**, 1585-1593.

Licht, T. 1989. Discriminating between hungry and satiated predators: the response of guppies (*Poecilia reticulata*) from high and low predation sites. *Ethology* **82**, 238-243.

Liley, N.R. 1982. Chemical communication in fish. *Canadian Journal of Fisheries and Aquatic Sciences* **39**, 22-35.

Lima, S.L. & Dill, L.M. 1990. Behavioral decisions made under the risk of predation: a review and prospectus. *Canadian Journal of Zoology* **68**, 619-640.

Lin, Y.K. & Batzli, G.O. 2004. Movement of voles across habitat boundaries: Effects of food and cover. *Journal of Mammalogy* **85**, 216-224.

López, J.C., Broglio, C., Rodríguez, F., Thinus-Blanc, C. & Salas, C. 1999. Multiple spatial learning strategies in goldfish (*Carassius auratus*). *Animal Cognition* **2**, 109-120.

Losey, G. S. 2003. Crypsis and communication functions of UV-visible coloration in two coral reef damselfish, *Dascyllus aruanus* and *D-reticulatas*. *Animal Behaviour* **66**, 299-307.

Lydersen, C. & Kovacs, K.M. 1995. Paralysis as a defence response to threatening stimuli in harp seals (*Phoca groenlandica*). *Canadian Journal of Zoology* **73**, 486-492.

MacArthur, R.A. 1982. Cardiac and behavioural responses of mountain sheep to human disturbance. *Journal of Wildlife Management* **46**, 351-358.

MacArthur, R.A., Johnston, R.H. & Geist, V. 1979. Factors influencing heart rate in free-ranging bighorn sheep: a physiological approach to the study of wildlife harassment. *Canadian Journal of Zoology* **57**, 2010-20221.

MacDonald, K. 1983. Stability of individual differences in behavior in a litter of wolf cubs (*Canis lupus*). *Journal of Comparative Psychology* **97**, 99-106.

MacKenzie, C. & Moring, J.R. 1988. Estimating survival of Atlantic salmon during the intergravel period. *North American Journal of Fisheries Management* **8**, 45-49.

MacKenzie, I.J. 1975. The development of a lowland loch as a trout fishery with the aid of rotenone. *Journal of the Institute of Fisheries Management* **6**, 8-12.

Mackintosh, N.J., Lord, J. & Little, L. 1971. Visual and spatial probability learning in pigeons and goldfish. *Psychonomic Science* **24**, 221-223.

Mackney, P.A. & Hughes, R.N. 1995. Foraging behaviour and memory window in sticklebacks. *Behaviour* **132**, 1241-1253.

Maclean. A., Huntingford, F.A., Ruxton, G.D., Morgan, I.J., Hamilton, J. & Armstrong, J.D. 2005. Testing the assumptions of the ideal despotic distribution with an unpredictable food supply: experiments in juvenile salmon. *Journal of Animal Ecology* **74**, 214-225.

Magnhagen, C. 1988. Predation risk and foraging in juvenile Pink (*Oncorhynchus gorbuscha*) and Chum salmon (*O. keta*). *Canadian Journal of Fisheries and Aquatic Sciences* **45**, 592-596.

Magurran, A.E. 1989. Acquired recognition of predator odour in the European Minnow (*Phoxinus phoxinus*). *Ethology* **82**, 216-223.

Magurran, A.E. 1990. The inheritance and development of minnow anti-predator behaviour. *Animal Behaviour* **39**, 834-842.

Magurran, A.E. & Pitcher, T.J. 1987. Provenance, shoal size and socio-biology of predator-evasion behaviour in minnow shoals. *Proceedings of the Royal society of London B* **229**, 439-465.

Magurran, A.E. & Higham, A. 1988. Information-transfer across fish shoals under predator threat. *Ethology* **78**, 153-158.

Magurran, A.E., Irving, P.W. & Henderson, P.A. 1996. Is there a fish alarm pheromone? A wild study and critique. *Proceedings of the Royal Society of London B* **263**, 1551-1556.

Maitland, P.S. & Campbell, R.N. 1992. *Freshwater Fishes of the British Isles.* London: Harper Collins.

Mann, R.H.K. 1976. Observations on the age, growth, reproduction and food of pike *Esox lucius* L. in two rivers in southern England. *Journal of Fish Biology* **8**, 179-197.

Mann, R.H.K. 1980. The numbers and production of pike (*Esox lucius*) in two Dorset rivers. *Journal of Animal Ecology* **49**, 899-915.

Mann, R.H.K. 1982. The annual food consumption and prey preferences of pike (*Esox lucius*) in the river Frome, Dorset. *Journal of Animal Ecology*, **51**, 81-95.

Mann, R.H.K. 1985. A pike management strategy for a trout fishery. *Journal of Fish Biology* **27** (Suppl. A), 227-234.

Mann, R.H.K. & Penczak, T. 1984. The efficiency of a new electrofishing technique in determining fish numbers in a large river in central Poland. *Journal of Fish Biology* **24**, 173-185.

Manning, A. 1979. *An Introduction to Animal Behaviour. 3rd edition*. London: Edward & Arnold.

Marchetti, M.P. & Nevitt, G.A. 2003. Effects of hatchery rearing on brain structures of rainbow trout, *Oncorhynchus mykiss*. *Environmental Biology of Fishes* **66**, 9-14.

Marcus, J.P. & Brown, G.E. 2003. Response of pumpkinseed sunfish to conspecific chemical alarm cues: an interaction between ontogeny and stimulus concentration. *Canadian Journal of Zoology* **81**, 1671-1677.

Margenau, T.L., Rasmussen, P.W. & Kampa, J.M. 1998. Factors affecting growth of northern pike in small northern Wisconsin lakes. *North American Journal of Fisheries Management* **18**, 625-639.

Marler, P. 1970. A comparative approach to vocal learning: Song development in White-crowned sparrows. *Journal of Comparative Physiology and Psychology Monographs* **71**, 1-25.

Masters, J.E.G. 2004. *Temporal variation in the behaviour of the pike* Esox lucius *L.* PhD thesis, University of St Andrews, Scotland.

Mather, M.E. 1998. The role of context-specific predation in understanding patterns exhibited by anadromous salmon. *Canadian Journal of Fisheries and Aquatic Sciences* **55** (Suppl. 1), 232-246.

Mathis, A. & Smith, R.J.F. 1993. Fathead minnows, *Pimephales promelas*, learn to recognise northern pike, *Esox lucius*, as predators on the basis of chemical stimuli from minnows in the pike's diet. *Animal behaviour* **46**, 645-656.

Mathis, A. & Vincent, F. 2000. Differential use of visual and chemical cues in predator recognition and threat-sensitive predator avoidance responses by larval newts (*Notophthalamus viridescens*). *Canadian Journal of Zoology* **78**, 1646-1652.

Mathis, A., Chivers, D.P. & Smith, R.F.J. 1996. Cultural transmission of predator recognition in fishes: intraspecific and interspecific learning. *Animal Behaviour* **51**, 185-201.

Matthews, M.A., Poole, W.R., Dillane, M.G. & Whelan, K.F. 1997. Juvenile recruitment and smolt output of brown trout (*Salmo trutta* L) and Atlantic salmon (*Salmo salar* L) from a lacustrine system in western Ireland. *Fisheries Research* **31**, 19-37.

Martel, G. 1996. Growth rate and influence of predation risk on territoriality in juvenile coho salmon (*Oncorhynchus kisutch*). *Canadian Journal of Fisheries and Aquatic Sciences* **53**, 660-669.

McCormick, S.D., Hansen, L.P., Quinn, T.P. & Saunders, R.L. 1998. Movement, migration, and smelting of Atlantic salmon (*Salmo salar*). *Canadian Journal of Fisheries and Aquatic Sciences* **55** (Suppl. 1), 77-92.

McCrimmon, H.R. 1954. Stream studies on planted Atlantic salmon. *Journal of the Fisheries Research Board of Canada* **11**, 362-403.

McDonald, D.G., Milligan, C.L., McFarlane, W.J., Croke, S., Currie, S., Hooke, B., Angus, R.B., Tufts, B.L. & Davidson K. 1998. Condition and performance of juvenile Atlantic salmon (*Salmo salar*): effects of rearing practices on hatchery fish and comparison with wild fish *Canadian Journal of Fisheries and Aquatic Sciences* **55**, 1208-1219.

McIntosh, A.R., Peckarsky, B.L. & Taylor, B.W. 1999. Rapid size-specific changes in the drift of *Baetis bicaudatus* (Ephemeroptera) caused by alterations in fish odour concentration. *Oecologia* **118**, 256-264.

McLean, I.G., Lundie-Jenkins, G. & Jarman, P.J. 1996. Teaching an endangered mammal to recognise predators. *Biological Conservation* **75**, 51-62.

McMenemy, J.R. 1995. Survival of Atlantic salmon fry stocked at low density in the West River, Vermont. *North American Journal of Fisheries Management* **15**, 366-374.

Meister, A.L. 1962. Atlantic salmon production in Cove Brook, Maine. *Transactions of the American Fisheries Society* **91**, 208-212.

Menzies, W.J.M. & Pentelow, F.T.K. 1965. Fisheries and the development of hydro-electric power in Scotland. *ICES Salmon and Trout Commission C.M.* **2**, pp. 8.

Mesa, M.G. 1994. Effects of multiple acute stressors on the predator avoidance ability and physiology of juvenile Chinook salmon. *Transactions for the American Fisheries Society* **123**, 786-793.

Metcalfe, N.B., Huntingford, F.A. & Thorpe, J.E. 1987. The influence of predation risk on the feeding motivation and foraging strategy of juvenile Atlantic salmon. *Animal Behaviour* **35**, 901-911.

Metcalfe, N.B., Fraser, N.H.C. & Burns, M.D. 1998. State-dependent shifts between nocturnal and diurnal activity in salmon. *Proceedings of the Royal Society of London Series B* **265**, 1503-1507.

Metcalfe, N.B., Valdimarsson, S.K. & Morgan, I.J. 2003. The relative roles of domestication, rearing environment, prior residence and body size in deciding territorial contests between hatchery and wild juvenile salmon. *Journal of Applied Ecology* **40**, 535-544.

Meyers, T.F. 1994. The program to restore Atlantic salmon in the Connecticut River. In *Proceedings of the New England Atlantic Salmon Management Conference on a Hard Look at some Tough Issues* (Calabi, S. & Stout, A. eds.). pp. 11-21. Silver Quill Brook, Camden, Maine: New England Salmon Association.

Mikheev, V.N., Metcalfe, N.B., Huntingford, F.A. & Thorpe, J.E. 1994. Size-related differences in behaviour and spatial distribution of juvenile Atlantic salmon in a novel environment. *Journal of Fish Biology* **45**, 379-386.

Milinski, M. 1988. Games fish play – making decisions as a social forager. *Trends in Ecology & Evolution* **3**, 325-330.

Milinski, M. & Boltshauser, P. 1995. Boldness and predator deterrence: a critique of Godin & Davis. *Proceedings of the Royal Society of London* B **262**, 103-105.

Milinski, M., Pfluger, D., Kulling, D. & Kettler, R. 1990a. Do sticklebacks cooperate repeatedly in reciprocal pairs? *Behavioral Ecology and Sociobiology* **27**, 17-21.

Milinski, M., Kulling, D. & Kettler, R. 1990b. Tit for tat: sticklebacks 'trusting' a cooperating partner. *Behavioral Ecology* **1**, 7-12.

Miller, B., Biggins, D., Wemmer, C., Powell, R., Calvo, L., Hanebury, L. & Wharton, T. 1990. Development of survival skills in captive raised Siberian polecats (*Mustela eversmanni*) II. Predator avoidance. *Journal of Ethology* **8**, 95-104.

Mills, D.H. 1965. The ecology of the young stages of the Atlantic salmon in the River Bran, Ross-shire. *Freshwater and Salmon Fisheries Research* **32**.

Mills, D.H. 1969. The survival of juvenile Atlantic salmon and brown trout in some Scottish streams. In *Symposium on Salmon and Trout in Streams* (Northcote, T.H. ed.). pp. 217-228. H.R. MacMillan Lectures in Fisheries. Vancouver: University of British Colombia.

Mills, D.H. 1971. *Salmon and trout: a resource, its ecology, conservation and management*. pp. 76-84. Edinburgh: Oliver & Boyd.

Milner, N.J. 1993. Assessing the effectiveness of stocking. In *Proceedings of the joint NRA Welsh Region/WSTAA Seminar on Welsh Rivers – Stocking and Taking Stock*. NRA Welsh Region Technical Fisheries Report No. **4**, pp. 49-69.

Mirza, R.S. & Chivers, D.P. 2000. Predator-recognition training enhances survival of brook trout: evidence from laboratory and field-enclosure studies. *Canadian Journal of Zoology* **78**, 2198-2208.

Mirza, R.S. & Chivers, D.P. 2001a. Do juvenile yellow perch use diet cues to assess the level of threat posed by intraspecific predators? *Behaviour* **138**, 1249-1258.

Mirza, R.S. & Chivers, D.P. 2001b. Are chemical alarm cues conserved within salmonids fishes? *Journal of Chemical Ecology* **27**, 1641-1655.

Mirza, R.S. & Chivers, D.P. 2003a. Predator diet cues and the assessment of predation risk by juvenile brook charr: do diet cues enhance survival? *Canadian Journal of Zoology* **81**, 126-132.

Mirza, R.S. & Chivers, D.P. 2003b. Response of juvenile rainbow trout to varying concentrations of chemical alarm cue: response thresholds and survival during encounters with predators. *Canadian Journal of Zoology* **81**, 88-95.

Mirza, R.S., Chivers, D.P. & Godin, J.J. 2001. Brook charr alevins alter timing of nest emergence in response to chemical cues from fish predators. *Journal of Chemical Ecology* **27**, 1775-1785.

Moen, A.N., Dellafera, M.A., Hiller, A.L. & Buxton, B.A. 1978. Heart rates of white-tailed deer fawns in response to recorded wolf howls. *Canadian Journal of Zoology* **56**, 1207-1210.

Montgomery, J.C. 1988. Sensory physiology. In *Physiology of elasmobranch fishes* (Shuttleworth, T.J. ed.). pp. 79-98. Berlin Heidelberg: Springer-Verlag.

Moore, A., Ives, M.J. & Kell, L.T. 1994. The role of urine in sibling recognition in Atlantic salmon *Salmo salar* (L.) parr. *Proceedings of the Royal Society of London B* **255**, 173-180.

Moreira, P.S.A. & Volpato, G.L. 2004. Conditioning of stress in Nile tilapia. *Journal of Fish Biology* **64**, 961-969.

Moriceau, S. & Sullivan, R.A. 2004. Corticosterone influences on mammalian neonatal sensitive-period learning. *Behavioural Neuroscience* **118**, 274-281.

Morin, P-P, Dodson, J.J. & Doré, F.Y. 1989a. Cardiac responses to a natural odorant as evidence of a sensitive period for olfactory imprinting in young Atlantic salmon, *Salmo salar*. *Canadian Journal of Fisheries and Aquatic Sciences* **46**, 122-130.

Morin, P-P, Dodson, J.J. & Doré, F.Y. 1989b. Thyroid activity concomitant with olfactory learning and heart rate changes in Atlantic salmon, *Salmo salar*, during smoltification. *Canadian Journal of Fisheries and Aquatic Sciences* **46**, 131-136.

Mork, J., Jarvi, T. & Hansen, L.P. 1989. Lower prevalence of fin erosion in mature than in immature Atlantic salmon (*Salmo salar*) parr. *Aquaculture* **80**, 223-229.

Moyle, J.B., Kuehn, J.H. & Burrows, C.R. 1948. Fish-population and catch data from Minnesota lakes. *Transactions of the American Fisheries Society* **78**, 163-175.

Munro, W.R. 1957. The pike of Loch Choin. *Freshwater and Salmon Fisheries Research* **16**.

Murphy, L.E. & Pitcher, T.J. 1997. Predator attack motivation influences the inspection behaviour of European minnows. *Journal of Fish Biology* **50**, 407-417.

Nakano, S. 1995. Individual differences in resource use, growth and emigration under the influence of a dominance hierarchy in fluvial red-spotted masu salmon in a natural habitat. *Journal of Animal Ecology* **64**, 75-84.

Nevitt, G.A. & Dittman, A.H. 1998. A new model for olfactory imprinting in salmon. *Integrative Biology* **1**, 215-223.

Nilsson, P.A. & Brönmark, C. 1999. Foraging among cannibals and kleptoparasites: effects of prey size on pike behaviour. *Behavioral Ecology* **10**, 557-566.

Nilsson, P.A., Nilsson, K. & Nyström, P. 2000. Does risk of intraspecific interactions induce shifts in prey-size preference in aquatic predators? *Behavioral Ecology and Sociobiology* **48**, 268-275.

Nislow, K.H., Einum, S. & Folt, C.L. 2004. Testing predictions of the critical period for survival concept using experiments with stocked Atlantic salmon. *Journal of Fish Biology* **65** (Suppl. A), 188-200.

Ohashi, K. & Yahara, T. 2002. Visit larger displays but probe proportionally fewer flowers: counterintuitive behaviour of nectar-collecting bumble bees achieves an ideal free distribution. *Functional Ecology* **16**, 492-503.

Olla, B.L., Davis, M.W. 1989. The role of learning and stress in predator avoidance of hatchery-reared coho salmon (*Oncorhynchus kisutch*) juveniles. *Aquaculture* **76**, 209-214.

Olla, B.L., Davis, M.W. & Ryer, C.H. 1992. Foraging and predator avoidance in hatchery-reared Pacific salmon: achievement of behavioral potential. In *World Aquaculture workshops Number 2; The Importance of Feeding Behavior for the Efficient Culture of Salmonid Fishes.* (Thorpe, J.E. & Huntingford, F.A. eds.). pp. 5-12. Baton Rouge, Louisiana: World Aquaculture Society.

Olla, B.L., Davis, M.W. & Ryer, C.H. 1994. Behavioural deficits in hatchery-reared fish: Potential effects on survival following release. *Aquaculture and Fisheries Management* **25** (Suppl. 1), 19-34.

Olla, B.L., Davis, M.W. & Ryer, C.H. 1998. Understanding how the hatchery environment represses or promotes the development of behavioral survival skills. *Bulletin of Marine Science* **62**, 531-550.

Olsén, K.H. 1993. Development of the olfactory organ of the Arctic charr, *Salvelinus alpinus* (L.) (Teleostei, Salmonidae). *Canadian Journal of Zoology* **71**, 1973-1984.

Olsèn, K.H., Petersson, E., Ragnarsson, B., Lundqvist, H. & Järvi, T. 2004. Downstream migration in Atlantic salmon (*Salmo salar*) smolt sibling groups. *Canadian Journal of Fisheries and Aquatic Sciences* **61**, 328-331.

Olson, K.R. 1998. The Cardiovascular system In *The Physiology of Fishes 2nd Edition* (Evans, D.H. ed.). pp. 129-159. New York: CRC Press.

Orciari, R.D., Leonard, G.H., Mysling, D.J., Schluntz, E.C. 1994. Survival, growth, and smolt production of Atlantic salmon stocked as fry in a southern New England stream. *North American Journal of Fisheries Management* **14**, 588-606.

Österdahl, L. 1969. The smolt run of a small Swedish river. In *Proceedings of the Symposium on Salmon and Trout in Streams* (Northcote, T.G. ed.). pp. 205-216. H.R. MacMillan Lectures in Fisheries. Vancouver: University of British Colombia.

Owings, D.H. & Coss, R.G. 1979. Snake mobbing by California ground squirrels: Adaptive variation and ontogeny. *Behaviour* **62**, 50-69.

Owings, D.H., Coss, R.G., McKernon, D., Rowe, M.P. and Arrowood, P.C. 2001. Snake-directed antipredator behavior of rock squirrels (*Spermophilus variegatus*): Population differences and snake-species discrimination. *Behaviour* **138**, 575-595.

Paloheimo, J.E. 1958. A method of estimating natural and fishing mortalities. *Journal of the Fisheries Research Board of Canada* **15**, 749-758.

Paragamian, V.L. 1976. Population characteristics of northern pike in the Plover river, Wisconsin. *The Progressive Fish Culturist* **38**, 160-163.

Pauwels, S.J. & Haines, T.A. 1994. Survival, hatching, and emergence success of Atlantic salmon eggs planted in three Maine streams. *North American Journal of Fisheries Management* **14**, 125-130.

Penczak, T., Zalewski, M. & Moliński, M. 1976. Production of pike, roach and chub in a selected fragment of Pilica river (Barbel region). *Polskie Archiwum Hydrobiologii* **23**, 139-153.

Petersson, E., Järvi, T., Steffner, N.G. & Ragnarsson, B. 1996. The effect of domestication on some life history traits of sea trout and Atlantic salmon. *Journal of Fish Biology* **48**, 776-791.

Pettersson, L.B., Nilsson, P.A. & Brönmark, C. 2000. Predator recognition and defence strategies in crucian carp, *Carassius carassius. Oikos* **88**, 200-212.

Pfeiffer, W. 1962. The fright reaction of fish. *Biological Reviews* **37**, 495-511.

Pfeiffer, W. 1963. The fright reaction in North American fish. *Canadian Journal of Zoology* **41**, 69-77.

Pfeiffer, W. 1974. Pheromones in fish and amphibia. In *Pheromones: frontiers of biology. Vol. 32* (Birch, M.C. ed.). pp. 269-296. Amsterdam: North-Holland.

Pfeiffer, W. 1977. The distribution of fright reaction and alarm substance cells in fishes. *Copeia* **1977**, 653-665.

Phillips, D.W. 1978. Chemical mediation of invertebrate defensive behaviors and the ability to distinguish between foraging and inactive predators. *Marine Biology* **49**, 237-243.

Pierce, R.B. & Tomcko, C.M. 2003a. Variation in gill-net and angling catchability with changing density of northern pike in a small Minnesota lake. *Transactions of the American Fisheries Society* **132**, 771-779.

Pierce, R.B. & Tomcko, C.M. 2003b. Interrelationships among production, density, growth, and mortalit of northern pike in seven north-central Minnesota lakes. *Transactions of the American Fisheries Society* **132**, 143-153.

Pierce, R.B., Tomcko, C.M. & Margenau, T.L. 2003. Density dependence in growth and size structure of northern pike populations. *North American Journal of Fisheries Management* **23**, 331-339.

Piggins, D.J. 1979. Salmon ranching in Ireland. In *Salmon Ranching*. (Thorpe, J.E. ed.). pp. 187-198. London: Academic Press.

Pitcher, T.J. 1992. Who dares wins – the function and evolution of predator inspection behaviour in shoaling fish. *Netherlands Journal of Zoology* **42**, 371-391.

Pitcher, T.J. & Magurran, A.E. 1983. Shoal size, patch profitability and information exchange in foraging goldfish. *Animal Behaviour* **31**, 546-555.

Pitcher, T.J. & Parrish, J.K. 1993. Functions of shoaling behaviour in teleosts. In *Behaviour of Teleost Fishes* (Pitcher, T.J. ed.). pp. 363-439. London: Chapman & Hall.

Pitcher, T.J., Green, D.A. & Magurran, A.E. 1986 Dicing with death: predator inspection behaviour in minnow shoals. *Journal of Fish Biology* **28**, 438-448.

Poling, K.R. & Fuiman, L.A. 1997. Sensory development and concurrent behavioural changes in Atlantic croaker larvae. *Journal of Fish Biology* **51**, 402-421.

Pongracz, P. & Altbacker, V. 2000. Ontogeny of the responses of European rabbits (*Oryctolagus cuniculus*) to aerial and ground predators. *Canadian Journal of Zoology* **78**, 655-665.

Pourie, G. & Trabalon, M. 2001. Plasticity of agonistic behaviour in relation to diet and contact signals in experimentally group-living of *Tegenaria atrica. Chemoecology* **11**, 175-181.

Pulliam, H.R. & Caraco, T. 1984. Living in groups: is there an optimal group size? In *Behavioural Ecology: An evolutionary Approach* (Krebs, J.R. & Davies, N.B., ed.). pp. 122-147. Oxford: Blackwell Scientific.

Purrington, F.F., Kendall, P.A., Bater, J.E. & Stinner, B.R. 1991. Alarm pheromone in a gregarious poduromorph collembolan (Collembola: Hypogasturidae). *Great Lakes Entomologist* **24**, 75-78.

Quinn, T.P. 1993. A review of homing and straying of wild and hatchery-produced salmon. *Fisheries Research* **18**, 29-44.

Raat, A.J.P. 1988. Synopsis of biological data on the northern pike, *Esox lucius* Linnaeus, 1758. *FAO Fish. Synopsis 30 Rev. 2.*

Rask, M. & Arvola, L. 1985. The biomass and production of pike, perch and whitefish in two small lakes in southern Finland. *Annales Zoologici Fennici* **22**, 129-136.

Rehnberg, B.G. & Schreck, C.B. 1987. Chemosensory detection of predators by coho salmon (*Oncorhynchus kisutch*): behavioral reaction and the physiological stress response. *Canadian Journal of Zoology* **65**, 481-485.

Reiriz, L., Nicieza, A.G. & Brana, F. 1998. Prey selection by experienced and naïve juvenile Atlantic salmon. *Journal of Fish Biology* **53**, 100-114.

Riddell, B.E. & Leggett, W.C. 1981. Evidence of an adaptive basis for geographic variation in body morphology and time of downstream migration of juvenile Atlantic salmon (*Salmo salar*). *Canadian Journal of Fisheries and Aquatic Sciences* **38**, 308-320.

Rideout, S.G. & Stolte, L.W. 1988. Restoration of Atlantic salmon to the Connecticut and Merrimack rivers. In *Present and Future Atlantic Salmon Management: Measuring Progress towards International Cooperation* (Stroud, R.H. ed.). pp. 67-81.

Marine Recreational Fisheries No. 12. Ipswich, MA: Atlantic Salmon Federation.
Rimmer, D.M., Paim, U. & Saunders, R.L. 1983. Autumnal habitat shift of juvenile Atlantic salmon (*Salmo salar*) in a small river. *Canadian Journal of Fisheries and Aquatic Sciences* **40**, 671-680.

Rodríguez, F., Durán, E. Vargas, J.P., Torres, B. & Salas, C. 1994. Performance of goldfish trained in allocentric and egocentric maze procedures suggests the presence of a cognitive mapping system in fishes. *Animal Learning and Behavior* **22**, 409-420.

Rudenko, G.P. 1970. Biomass and abundance in a roach-perch lake. *Journal of Ichthyology* **11**, 524-535.

Ruxton, G.D., Armstrong, J.D. & Humphries, S. 1999. Modelling territorial behaviour of animals in variable environments. *Animal Behaviour* **58**, 113-120.

Ryan, P.M. 1986. Lake use by wild anadromous Atlantic salmon, *Salmo salar*, as an index of subsequent adult abundance. *Canadian Journal of Fisheries and Aquatic Sciences* **43**, 2-11.

Ryer, C. 2004. Comparison of genetically based antipredator behaviour in hatchery and wild coho fry from a coastal Oregon river: interpreting a negative result. *Journal of Fish Biology* **64**, 1413-1418.

Saglio, P. & Fauconneau, B. 1985. Free amino-acid content in the skin mucus of goldfish, *Carassius auratus* L. – influence of feeding. *Comparative Biochemistry and Physiology A* **82**, 67-70.

Saunders, R.L. & Gee, J.H. 1964. Movements of young Atlantic salmon in a small stream. *Journal of the Fisheries Research Board of Canada* **21**, 27-35.

Savino, J.F. & Stein, R.A. 1989a. Behavioural interactions between fish predators and their prey: effects of plant density. *Animal Behaviour* **37**, 311-321.

Savino, J.F. & Stein, R.A. 1989b. Behavior of fish predators and their prey: habitat choice between open water and dense vegetation. *Environmental Biology of Fishes* **24**, 287-293.

Schmidt-Nielsen, K. 1996. *Animal Physiology: Adaptations and Environment.* pp. 607. Cambridge: Cambridge University Press.

Seaburg, K.G. & Moyle, J.B. 1964. Feeding habits, digestive rates, and growth of some Minnesota warmwater fishes. *Transactions of the American Fisheries Society* **93**, 269-285.

Seely, C.J. & Lutnesky, M.M.F. 1998. Odour-induced antipredator behaviour of the water flea, Ceriodaphnia reticulata, in varying predator and prey densities. *Freshwater Biology* **40**, 17-24.

Shafi, M. & Maitland, P.S. 1971. Comparative aspects of the biology of pike, *Esox lucius* L., in two Scottish lochs. *Proceedings of the Royal Society of Edinburgh* **LXXI B**, 41-60.

Shearer, W.M. 1961. Survival rate of young salmonids in streams stocked with 'green' ova. *ICES, C.M. 1961/98*, Copenhagen.

Shelton, G. 1970. The regulation of breathing In *Fish Physiology Vol IV. The nervous system, circulation and respiration* (Hoar, W. S. & Randall, D. J. eds.). pp. 293-359. London: Academic Press.

Sherman, P.W. 1985. Alarm calls of Belding's ground squirrels to aerial predators: nepotism or self-preservation? *Behavioral Ecology and Sociobiology* **17**, 313-323.

Sherman, P.W., Reeve, H.K. & Pfennig, D.W. 1997. Recognition Systems. In *Behavioural Ecology Fourth Edition* (Krebs, J.R. & Davies, N.B. eds.). pp.69-96. Oxford: Blackwell Science.

Shively, R.S., Poe, T.P. & Sauter, S.T. 1996. Feeding response by northern squawfish to a hatchery release of juvenile salmonids in the Clearwater River, Idaho. *Transactions of the American Fisheries Society* **125**, 230-236.

Shuter, B.J. 1990. Population level indicators of stress. In *Biological indicators of stress in fish* (Adams, S.M. ed.). pp. 145-166. Bethesda: American fisheries Society.

Sih, A. & Moore, R.D. 1993. Delayed hatching of salamander eggs in response to enhanced larval predation risk. *The American Naturalist* **142**, 947-960.

Sih, A. & Kats, L.B. 1994. Age, experience, and the response of streamside salamander hatchlings to chemical cues from predatory sunfish. *Ethology* **96**, 253-259.

Skov, C. & Berg, S. 1999. Utilization of natural and artificial habitats by YOY pike in a biomanipulated lake. *Hydrobiologia* **408/409**, 115-122.

Skov, C., Berg, S., Jacobsen, L. & Jepsen, N. 2002. Habitat use and foraging success of 0+ pike (*Esox lucius* L.) in experimental ponds related to prey fish, water transparency and light intensity. *Ecology of Freshwater Fish* **11**, 65-73.

Sleeper, H.L., Paul, V.J. & Fenical, W. 1980. Alarm pheromones from the marine opisthobranch *Navanax inermis*. *Journal of Chemical Ecology* **6**, 57-70.

Smith, R.J.F. 1992. Alarm signals in fishes. *Reviews in Fish Biology and Fisheries* **2**, 33-63.

Sneddon, L.U., Braithwaite, V.A. & Gentle, M.J. 2003a. Novel object test: Examining nociception and fear in the rainbow trout. *Journal of Pain* **4**, 431-440.

Sneddon, L.U., Braithwaite, V.A. & Gentle, M.J. 2003b. Do fish have nociceptors: Evidence for the evolution of a vertebrate sensory system. *Proceedings of the Royal Society of London Series B* **270**, 1115-1121.

Snow, H.E. 1974. Effects of stocking northern pike in Murphy flowage. *Technical Bulletin of Wisconsin Department of Natural Resources* **79**, 20. *In* Raat, A.J.P. 1988. Synopsis of biological data on the northern pike, *Esox lucius* Linnaeus, 1758. *FAO Fish. Synopsis 30 Rev. 2.*

Snow, H.E. & Beard, T.D. 1972. A 10-year study of native northern pike in Bucks Lake, Wisconsin, including evaluation of an 18-inch size limit. *Technical Bulletin of Wisconsin Department of Natural Resources* **56**, 2-20. *In* Raat, A.J.P. 1988. Synopsis of biological data on the northern pike, *Esox lucius* Linnaeus, 1758. *FAO Fish. Synopsis 30 Rev. 2.*

Solomon, D.J. 1978. Migration of smolts of Atlantic salmon (*Salmo salar* L.) and sea trout (*Salmo trutta* L.) in a chalkstream. *Environmental Biology of Fishes* **3**, 223-229.

Sovrano, V.A., Bisazza, A. & Vallortigara, G. 2002. Modularity and spatial reorientation in a simple mind: encoding of geometric and nongeometric properties of a spatial environment by fish. *Cognition* **85**, B51-B59.

Stabell, O.B. 1987. Intraspecific pheromone discrimination and substrate marking by Atlantic salmon (*Salmo salar*). *Journal of Chemical Ecology* **13**, 1625-1643.

Stamps, J.A. & Krishnan, V.V. 1994. Territory acquisition in lizards: II. Establishing social and spatial relationships. *Animal Behaviour* **47**, 1387-1400.

Stamps, J.A. & Krishnan, V.V. 1995. Territory acquisition in lizards: III. Competing for space. *Animal Behaviour* **49**, 679-693.

Stevenson-Hinde, J., Stillwell-Barnes, R. & Zung, M. 1980. Individual differences in young rhesus monkeys: Continuity and change. *Primates* **21**, 62-66.

Stewart, I.J., Carlson, S.M., Boatright, C.P., Buck, G.B. & Quinn, T.P. 2004. Site fidelity of spawning sockeye salmon (*Oncorhynchus nerka* W.) in the presence and absence of olfactory cues. *Ecology of Freshwater Fish* **13**, 104-110.

Stoddard, P.K. 2002. The evolutionary origins of electric signal complexity. *Journal of Physiology Paris* **96**, 485-491.

Stein, R.A. & Magnusson, J.H. 1976. Behavioral response of prey to a fish predator. *Ecology* **57**, 751-761.

Steingrímsson, S.O. & Grant, J.W.A. 1999. Allometry of territory size and metabolic rate as predictors of self-thinning in young-of-the-year Atlantic salmon. *Journal of Animal Ecology* **68**, 17-26.

Stolte, L.W. 1994. Atlantic salmon restoration in the Merrimack River Basin. In *Proceedings of the New England Atlantic Salmon Management Conference on a Hard Look at some Tough Issues* (Calabi, S. & Stout, A. eds.). pp. 22-35. Silver Quill Brook, Camden, Maine: New England Salmon Association.

Stummer, L.E., Weller, J.A., Johnson, M.L. & Côté, I.M. 2004. Size and stripes: how fish clients recognize cleaners. *Animal Behaviour* **68**, 145-150.

Suboski, M.D. 1990. Releaser-induced recognition learning. *Psychology Review* **97**, 271-284.

Suboski, M.D. & Templeton, J.J. 1989. Life skills training for hatchery fish: social learning and survival. *Fisheries Research* **7**, 343-352.

Sundström, L.F. & Johnsson, J.I. 2001. Experience and social environment influence the ability of young brown trout to forage on live novel prey. *Animal Behaviour* **61**, 249-255.

Sutherland, W.J. 1992. Game-theory models of functional and aggregative responses. *Oecologia* **90**, 150-152.

Swain, D.P., Riddell, B.E. & Murray, C.B. 1991. Morphological differences between hatchery and wild populations of coho salmon (*Oncorhynchus kisutch*): environmental versus genetic origin. *Canadian Journal of Fisheries and Aquatic Sciences* **48**, 1783-1791.

Symons, P.E.K. 1974. Territorial behaviour of juvenile Atlantic salmon reduces predation by brook trout. *Canadian Journal of Zoology* **52**, 677-679.

Symons, P.E.K. 1979. Estimated escapement of Atlantic salmon (*Salmo salar*) for maximum smolt production in rivers of different productivity. *Journal of the Fisheries Research Board of Canada* **36**, 132-140.

Symons, P.E.K. & Heland, M. 1978. Stream habitats and behavioral interactions of underyearling and yearling Atlantic salmon (*Salmo salar*). *Journal of the Fisheries Research Board of Canada* **35**, 175-183.

Thompson, J.D. 1991. Phenotypic plasticity as a component of evolutionary change. *Trends in Ecology and Evolution* **6**, 246-249.

Thompson, R.B. 1966. *Effects of predator avoidance conditioning on the post-release survival rate of artificially propagated salmon.* Ph.D thesis, University of Washington, Seattle. *In* Berejikian, B.A. 1995. The effects of hatchery and wild ancestry and experience on the relative ability of steelhead trout fry (*Oncorhynchus mykiss*) to avoid a benthic predator. *Canadian Journal of Fisheries and Aquatic Sciences* **52**, 2476-2482.

Thorpe, J.E. 1977. Bimodal distribution of length of juvenile Atlantic salmon (*Salmo salar* L.) under artificial rearing conditions. *Journal of Fish Biology* **11**, 175-184.

Thorpe, J.E. & Metcalfe, N.B. 1998. Is smolting a positive or a negative developmental decision? *Aquaculture* **168**, 95-103.

Thorpe, J.E., Morgan, R.I.G., Ottaway, E.M. & Miles, M.S. 1980. Time of divergence of growth groups between potential 1+ and 2+ smolts among sibling Atlantic salmon. *Journal of Fish Biology* **17**, 13-21.

Thorpe, J.E., Ross, L.G., Struthers, G. & Watts, W. 1981. Tracking Atlantic salmon smolts, *Salmo salar* L., through Loch Voil, Scotland. *Journal of Fish Biology* **19**, 519-537.

Treasurer, J.W., Owen, R. & Bowers, E. 1992. The population dynamics of pike, *Esox lucius*, and perch, *Perca fluviatilis*, in a simple predator-prey system. *Environmental Biology of Fishes* **34**, 65-78.

Tulley, J.J. & Huntingford, J.A. 1987. Age, experience and the development of adaptive variation in anti-predator responses in three-spined stickleback, *Gasterosteus aculeatus*. *Ethology* **75**, 285-290.

Turner, L.J. & Mackay, W.C. 1985. Use of visual census for estimating population size in northern pike (*Esox lucius*). *Canadian Journal of Fisheries and Aquatic Sciences* **42**, 1835-1840.

Utne-Palm, A.C. 2001. Response of naïve two-spotted gobies *Gobiusculus flavescens* to visual and chemical stimuli of their natural predator, cod *Gadus morhua*. *Marine Ecology Progress Series* **218**, 267-274.

VanDamme, R. & Castilla, A.M. 1996. Chemosensory predator recognition in the lizard *Podarcis hispanica*: Effects of predation pressure and relaxation. *Journal of Chemical Ecology* **22**, 13-22.

Verbeek, M.E.M., Boon, A. & Drent, P.J. 1996. Exploration, aggressive behaviour and dominance in pair-wise confrontations of juvenile male great tits. *Behaviour* **133**, 945-963.

Verbeek, M.E.M., De Goede, P., Drent, P.J. & Wiepkema, P.R. 1999. Individual behavioural characteristics and dominance in aviary groups of great tits. *Behaviour* **136**, 23-48.

Vigg, S., Poe, T.P., Prendergast, L.A. & Hansel, H.C. 1991. Rates of consumption of juvenile salmonids and alternative prey fish by northern squawfish, walleyes, smallmouth bass, and channel catfish in John Day reservoir, Columbia River. *Transactions of the American Fisheries Society* **120**, 421-438.

Vilhunen, S. & Hirvonen, H. 2003. Innate antipredator responses of Arctic charr (*Salvelinus alpinus*) depend on predator species and their diet. *Behavioral Ecology and Sociobiology* **55**, 1-10.

Vilhunen, S., Hirvonen, H. & Laakkonen, M.V-M. 2005. Less is more: social learning of predator recognition requires a low demonstrator to observer ratio in Arctic charr (*Salvelinus alpinus*). *Behavioral Ecology and Sociobiology* **57**, 275-282.

Vøllestad, L.A., Skurdal, J. & Qvenild, T. 1986. Habitat use, growth, and feeding of pike (*Esox lucius* L.) in four Norwegian lakes. *Archiv für Hydrobiologie* **108**, 107-117.

Vøllestad, L.A., Varreng, K. & Poléo, A.B.S. 2004. Body depth variation in crucian carp *Carassius carassius*: an experimental individual-based study. *Ecology of Freshwater Fish* **13**, 197-202.

von Frisch, K. 1941. Über einen Schreckstoff der Fischaut und seine biologische Bedeutung. *Z. Vergl. Physiol.* **29,** 46-146.

Wahle, R.A. 1992. Body-size dependent antipredator mechanisms of the american lobster. *Oikos* **65**, 52-60.

Waldman, B. 1982. Quantitative and developmental analyses of the alarm reaction in the zebra danio, *Brachydanio rerio*. *Copeia* **1982**, 1-9.

Waldman, B. 1985. Olfactory basis of kin recognition in toad tadpoles. *Journal of Comaparative Physiology A* **156**, 565-577.

Warburton, K. 1990. The use of local landmarks by foraging goldfish. *Animal Behaviour* **40**, 500-505.

Warburton, K. 2003. Learning of foraging skills by fish. *Fish and Fisheries* **4**, 203-215.

Warburton, K., Retif, S. & Hume, D. 1998. Generalists as sequential specialists: diets and prey switching in juvenile silver perch. *Environmental Biology of Fishes* **51**, 445-454.

Ward, A.J. & Hart, P.J. 2003. The effects of kin and familiarity on interactions between fish. *Fish and Fisheries* **4**, 348-358.

Ward, J.F., Macdonald, D.W., Doncaster, C.P. & Mauget, C. 1996. Physiological response of the European hedgehog to predator and nonpredator odour. *Physiology & Behavior* **60**, 1469-1472.

Ware, D.M. 1971. Predation by rainbow trout (*Salmo gairdneri*): the effect of experience. *Journal of the Fisheries Research Board of Canada* **28**, 1847-1852.

Watt, P.J. & Young, S. 1994. Effect of predator chemical cues on Daphnia behaviour in both horizontal and vertical planes. *Animal Behaviour* **48**, 861-869.

Webb, J.H. & Hawkins, A.D. 1989. The movements and spawning behaviour of adult salmon in the Girnock Burn, a tributary of the Aberdeenshire Dee, 1986. *Scottish Fisheries Research Reports* **No. 40.**

Webb, P.W. 1977. Effects of median fin amputation on fast-start performance of rainbow trout. *The Journal of Experimental Biology* **68**, 123-135.

Webb, P.W. 1982. Avoidance responses of fathead minnows to strikes by four teleost predators. *Journal of Comparative Physiology A* **147**, 371-378.

Weber, E.D. & Fausch, K.D. 2003. Interactions between hatchery and wild salmonids in streams: differences in biology and evidence for competition. *Canadian Journal of Fisheries and Aquatic Sciences* **60**, 1018-1036.

West-Eberhard, M.J. 1989. Phenotypic plasticity and the origins of diversity. *Annual Review of Ecology and Systematics* **20**, 249-278.

Wheeler, A. 1977. The origin and distribution of the freshwater fishes of the British Isles. *Journal of Biogeography* **4**, 1-24.

Whiteman, E.A. & Côté, I.M. 2004. Dominance hierarchies in group-living cleaning gobies: causes and foraging consequences. *Animal Behaviour* **67**, 239-247.

Whitehouse, M.E.A. & Lubin, Y. 1999. Competitive foraging in the social spider *Stegodyphus dumicola*. *Animal Behaviour* **58**, 677-688.

Wiley, R.W., Whaley, R.A., Satake, J.B. & Fowden, M. 1993. An evaluation of the potential for training trout in hatcheries to increase post-stocking survival in streams. *North American Journal of Fisheries Management* **14**, 1-13.

Williams, D.D. & Moore, K.A. 1985. The role of semiochemicals in benthic community relationships of the lotic amphipod *Gammarus pseudolimnaeus*: A laboratory analysis. *Oikos* **44**, 280-286.

Wilson, D.J. & Lefcort, H. 1993. The effect of predator diet on the alarm response of red-legged frog, *Rana aurora*, tadpoles. *Animal Behaviour* **46**, 1017-1019.

Wilson, D.S., Coleman, K., Clark, A.B. & Biederman, L. 1993. Shy-bold continuum in pumpkinseed fish (*Lepomis gibbosus*): An ecological study of a psychological trait. *Journal of Comparative Psychology* **107**, 250-260.

Wilson, P. 1987. Substrate selection and decorating behavior in *Acanthonyx petiveri* related to exoskeleton color (Brachyura, Mandae). *Crustaceana* **52**, 135-140.

Wisenden, B.D. 2000. Scents of danger: the evolution of olfactory ornamentation in chemically-mediated predator-prey interactions. In *Animal signals: signalling and signal design in animal communication* (Espmark, Y., Amundsen, T. & Rosenqvist, G. eds.). pp. 365-386. Trondheim: Tapic Academic Press.

Wisenden, B.D. 2001. Chemically-mediated strategies to counter predation. In *Sensory Processing in Aquatic Environments* (Collin, S.P. & Marshall, N.J. eds.). pp. 236-243. New York: Springer-Verlag.

Wisenden, B.D., Vollbrecht, K.A. & Brown, J.L. 2004. Is there a fish alarm cue? Affirming evidence from a wild study. *Animal Behaviour* **67**, 59-67.

Wright, R.M. 1990. The population biology of pike, *Esox lucius* L., in two gravel pit lakes, with special reference to early life history. *Journal of Fish Biology* **36**, 215-229.

Wright, R.M. & Giles, N. 1987. The survival, growth and diet of pike fry, *Esox lucius* L., stocked at different densities in experimental ponds. *Journal of Fish Biology* **30**, 617-629.

Yasukawa, K. & Bick, E.I. 1983. Dominance hierarchies in dark-eyed juncos (*Junco hyemalis*), a test of game-theory model. *Animal Behaviour* **31**, 439-448.

Ydenberg, R.C. 1998. Behavioral decisions about foraging and predator avoidance. In *Cognitive Ecology* (Dukas, R. ed.). pp. 343-378. Chicago: University of Chicago Press.

Ydenberg, R.C. & Dill, L.M. 1986. The economics of fleeing from predators. *Advances in the study of Behaviour* **16**, 229-249.

Youngson, A.F., Buck, R.J.G., Simpson, T.H. & Hay, D.W. 1983. The autumn and spring emigrations of juvenile salmon, *Salmo salar* L., from the Girnock Burn, Aberdeenshire, Scotland: environmental release of migration. *Journal of Fish Biology* **23**, 625-639.