PATTERNS AND PROCESSES OF MATING IN ATLANTIC COD (*GADUS MORHUA*)

by

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for the degree of Doctor of Philosophy

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ABSTRACT

Mating systems have received insufficient attention for their relationship with population dynamics and conservation biology. This is particularly true for marine fishes, many of which have recently experienced unprecedented declines. My research explored causes and consequences of variation in the mating system of Atlantic cod (*Gadus morhua*), a species that has been severely over-exploited and is currently at historic population lows.

Using captive groups of Atlantic cod, I examined male reproductive success in relation to morphology, condition, and spawning behaviour. I found that number of offspring sired was highly variable among males and there was a positive association between reproductive success and male body size, as well as aggression.

I also investigated the function of Atlantic cod sound production by quantifying individual variation in mass of the sound-producing musculature and temporal patterns and behavioural contexts of sound production. Males had drumming muscles larger than females and that increased in mass before spawning and declined thereafter. Drumming muscle mass of spawning males showed positive associations with body size, condition, and fertilisation potential, indicating that sound production may reveal information about male quality. Sound production occurred most frequently during the peak of the spawning season and was associated with agonistic interactions and courtship. These findings suggest that male sound production relates to mate competition.

Slow recovery rates and continuing declines of Atlantic cod populations may be attributable to depensation. Using egg fertilisation and male abundance data from captive groups, I explored the basis of reduced mating success as a potential cause of depensation and found that as population size declines, fertilisation rate also declines and variance in fertilisation rate may increase. The former identifies a potential mechanism for depensation in cod and the latter has negative consequences for effective population size.

My results have important implications for conservation of marine fishes, particularly those with mating systems similar to Atlantic cod. There is a need to incorporate knowledge of mating systems in population assessments, to undertake research on spatial and temporal scales of reproduction, and to initiate experiments to test hypotheses about marine fish mating systems, depensation, and correlates of individual reproductive success.

ACKNOWLEDGEMENTS

CHAPTER 1

OVERVIEW

Mating systems are defined by the pattern of matings among individuals in a population, including the number of mates obtained by each female and male, the duration of pair bonds, and all related behavioural activity. Within all mating systems, mate choice, intra-sexual competition, and sperm competition may lead to variation in mating and reproductive success among individuals. Growing evidence indicates that quantification of the patterns and processes of mating may be critical to understanding population dynamics. However, to date, mating systems have received insufficient attention for the indirect but significant consequences they can have for population dynamics and conservation biology (Legendre et al. 1999; Møller and Legendre 2001). This is particularly true for marine fishes, many of which have recently experienced unprecedented declines and yet have very little known about their behaviour during the spawning period (Vincent and Sadovy 1998). One of the best examples of this is seen in Atlantic cod (*Gadus morhua*), a species that has been severely over-exploited and is currently at historic population lows, having declined 90% in the North Sea (ICES 2002) and 99% off northeast Newfoundland (Hutchings 2003) in recent decades. Until recently, very little was known about even basic aspects of cod reproductive behaviour (Brawn 1961a; Hutchings et al. 1999).

Atlantic cod is a marine demersal fish found on both sides of the North Atlantic from inshore regions to the edge of the continental shelf. It is also a bet-hedging species, necessitated by a reproductive strategy in which eggs are released directly into the oceanic environment (broadcasted) and for which no

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parental care is provided (Hutchings 2000a, 2001a). Attaining maturity across a broad range of sizes (35-85 cm) and ages (2-7 years) throughout the species range (Brander 1994; Hutchings 2003), females release their annual egg complement numbering hundreds of thousands, often millions, of 1.25-1.75 mm eggs in batches over 3-6 weeks (Chambers and Waiwood 1996; Kjesbu et al. 1996) within a 6-12 week spawning season (Myers et al. 1993) in water ranging from tens to hundreds of metres in depth (Brander 1994; Morgan et al. 1997; Smedbol and Wroblewski 1997). To date, only two studies have documented Atlantic cod spawning behaviour (Brawn 1961a; Hutchings et al. 1999). As well as demonstrating complex mating patterns, these studies suggest the occurrence of behavioural and acoustic displays by males, mate choice by females, and alternative reproductive tactics among males. However, there is little information on the selective causes and consequences of these behaviours, nor the structure of the mating system (Nordeide and Folstad 2000).

My research employed a quantitative approach to explore causes and consequences of variation in the mating system of Atlantic cod. I incorporated detailed behavioural studies in the laboratory, genetic analyses of individual reproductive success, and studies of the phenotypic variability of reproductive and non-reproductive cod captured in the commercial fishery. Generally, my research involved the following components: (i) determining correlates of reproductive success in male Atlantic cod from two Northwest Atlantic populations; (ii) assessing the function of sound production by Atlantic cod as inferred from patterns of variation in drumming muscle mass; (iii) examining temporal patterns and behavioural contexts of sound production by Atlantic cod

during spawning; (iv) exploring links between depensation, probability of fertilisation, and the mating system of Atlantic cod; and (v) evaluating the influence of mating systems on the conservation of commercially exploited marine fishes.

CHAPTER 2

PHENOTYPIC AND BEHAVIOURAL CORRELATES OF

REPRODUCTIVE SUCCESS IN MALE ATLANTIC COD

INTRODUCTION

Mating systems have received insufficient attention for the indirect but significant relationship they can have with population dynamics and conservation biology (Legendre et al. 1999; Møller and Legendre 2001). This is particularly true of marine fishes, many of which are at historic population lows and for which very little is known of their behaviour during the spawning period (Vincent and Sadovy 1998). Fish live in an environment that makes it challenging to observe reproductive behaviour and patterns of parentage. Nevertheless, many fishes are known to exhibit external fertilisation which provides an opportunity for intense sperm competition (Stockley et al. 1997) and promotes a variety of male mating strategies (Taborsky 1998). Often, this takes the form of individual males either investing in attracting and monopolizing females, or in sperm production and the parasitizing of pair spawnings, although more complex strategies may also occur (Taborsky 1998).

Molecular tools are increasingly being used to investigate the mating systems of numerous taxa (Birkhead and Møller 1998). In fishes, DNA-based parentage inference has primarily been applied to examine paternity contributions from males employing alternative reproductive strategies; most of this work has focused on freshwater species that deposit gametes in nests (e.g. Rico et al. 1992; DeWoody et al. 2000; Jones and Hutchings 2002). By comparison, few studies have addressed the relative contribution of competing males in broadcast spawning marine species (Avise et al. 2002).

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The Atlantic cod (*Gadus morhua*) is a bet-hedging species, releasing multiple batches of eggs throughout multiple breeding seasons. The eggs are released directly into the oceanic environment (broadcasted) and no parental care is provided for them (Hutchings 2000a, 2001a). Attaining maturity across a broad range of sizes (35-85 cm) and ages (2-7 years) throughout the species range (Brander 1994; Hutchings 2003), females release their annual egg complement numbering hundreds of thousands, often millions, of 1.25-1.75 mm eggs in batches over 3-6 weeks (Chambers and Waiwood 1996; Kjesbu et al. 1996) within a 6-12 week spawning season (Myers et al. 1993) in water ranging from tens to hundreds of metres in depth (Brander 1994; Morgan et al. 1997; Smedbol and Wroblewski 1997).

Despite Atlantic cod having been harvested for hundreds of years and being of tremendous economic importance, its mating system, like that of most broadcast spawning fishes, is not well understood (Avise et al. 2002; Chapter 6). Recent studies of captive individuals have shown that successful reproduction in Atlantic cod is not a behaviourally vacuous process of haphazard mating as once depicted (Harris 1998), but instead involves complex behaviour within and between sexes (Brawn 1961a; Hutchings et al. 1999). Release of sperm and eggs occurs during a 'ventral mount' in which the male, whilst grasping the female with his pelvic fins and matching her swimming speed, positions himself beneath her. Upon extrusion of the female's batch of eggs, the spawning couple may be joined by 'satellite males', which swim among the eggs and also release sperm (Hutchings et al. 1999). Accordingly, genetic work indicates that eggs from single reproductive bouts are fertilised by more than one male (Hutchings et al.

1999; Rakitin et al. 2001; Bekkevold et al. 2002) and it is reasonable to assume that alternative mating tactics may exist in male Atlantic cod.

Agonistic behaviour preceding mounting attempts is hypothesized to reflect competition among males for mates and might influence access to females (Brawn 1961a; Hutchings et al. 1999). Primary determinants of success appear to include agonistic interactions, particularly chases of one male by another, and body size, larger individuals dominating smaller ones (Hutchings et al. 1999). Agonistic interactions also supposedly enable some males to defend 'territories' on the bottom of about 2-4 m² in which they generally reside (Brawn 1961a).

Interactions between sexes are consistent with the hypothesis that females, and possibly males, exercise mate choice (Hutchings et al. 1999). In particular, spawning events appear to be preceded by courtship behaviour, which has been described as an intense 'flaunting' display during which the courting male moves alongside and in front of the female with median fins fully erect while "swimming with an excited, jerky, undulating movement with many unnecessary circles" (Brawn 1961a). Because fecundity and sperm volume increase with body size (Oosthuizen and Daan 1974; Trippel and Morgan 1994a), individuals of both sexes might be expected to prefer large individuals as mates, resulting in size-based positive assortative mating, a pattern commonly observed in salmonids (Foote 1988). In addition, among broadcast spawners such as Atlantic cod, mate preference for size complementarity between spawning partners might be important to ensure that the urogenital openings of both fish are aligned opposite one another and that high fertilisation success is achieved (Hutchings et al. 1999; Rakitin et al. 2001; Bekkevold et al. 2002).

Like many fishes, cod can produce and detect sound (Hawkins 1993). The calls of Atlantic cod are produced using three pairs of drumming muscles exterior to the swimbladder wall (Brawn 1961b; Hawkins 1993) and consist of short 'grunts' with peak sound amplitudes at frequencies ranging from 50 to 500 Hz (Brawn 1961b; Hawkins and Rasmussen 1978; Midling et al. 2002). Although both sexes can produce sounds throughout the year, only males seem to do so during the spawning season, typically during agonistic interactions and courtship (Brawn 1961b; Chapter 4). Sound production may reinforce threat displays directed towards other males (Brawn 1961b) and has been predicted to be a sexually selected character upon which females base an active choice of spawning partner (Engen and Folstad 1999; Hutchings et al. 1999). Drumming muscle mass is sexually dimorphic and may reflect male size, condition, and fertilisation potential (Engen and Folstad 1999; Chapter 3).

Based on observations in the laboratory and field, it has been suggested that the mating system of Atlantic cod resembles that of a conventional lekking system (Hutchings et al. 1999; Nordeide and Folstad 2000). Mature males aggregate near the bottom on spawning grounds where they may actively defend small mating territories and females are typically found peripherally to male aggregations (Brawn 1961a; Morgan and Trippel 1996; Nordeide 1998; Hutchings et al. 1999; S. Rowe, personal observation). When ready to spawn a batch of eggs, females enter male aggregations, returning to peripheral areas after spawning (Brawn 1961a; Hutchings et al. 1999). In lekking systems, males cannot monopolize resources to gain matings with females and females typically discriminate among potential mates based on their dominance and sexually

selected traits (Höglund and Alatalo 1995). A common feature of leks is high variance in reproductive success among males, with more attractive and dominant individuals gaining a disproportionate share of matings (Höglund and Alatalo 1995). Based on the available data for Atlantic cod, this situation would lead to the prediction that larger, more aggressive males acquire a larger proportion of matings and achieve greater reproductive success than smaller, less aggressive males. Observations of cod spawning and parentage to date have indicated that paternity frequencies are indeed highly skewed among males, but the evidence for a link between male size or aggression and reproductive success is conflicting (Hutchings et al. 1999; Rakitin et al. 2001; Bekkevold et al. 2002).

Atlantic cod have been reported to spawn in aggregations spanning several kilometres and at average densities of 0.004 fish/m^3 (Morgan et al. 1997) but unfortunately, studies of cod spawning to date have involved only small groups of 3-16 captive individuals. Furthermore, none has directly examined reproductive success in relation to aspects of male spawning behaviour thus limiting the conclusions that can be drawn (Brawn 1961a; Hutchings et al. 1999; Rakitin et al. 2001; Bekkevold et al. 2002). In the present study, I examined reproductive success of male Atlantic cod spawning in much larger captive groups in relation to individual aspects of morphology, condition, and spawning behaviour. Specifically, my objectives were to: (i) describe variation in seasonal reproductive success among males; and (ii) quantify associations between seasonal reproductive success and male body size, age, drumming muscle investment, condition, aggressiveness, and mating attempts.

METHODS

Examination of parental fish and sampling of eggs

To assess mating system structure and identify correlates of reproductive success, I examined Atlantic cod from two spatially distinct areas in the Northwest Atlantic: Western Scotian Shelf and Southern Gulf of St. Lawrence, identified by the Northwest Atlantic Fishery Organization (NAFO) as divisions 4X and 4T, respectively. Fish from each stock were collected by baited hook two to three weeks prior to their annual spawning season and taken to a 684 m^3 tank at Dalhousie University where spawning occurred. Unfortunately, I was unable to control the size or sex of individuals captured. Prior to spawning, cod were anaesthetized and individually marked with 7.5-cm-long, coloured T-bar anchor tags. Every fish received one or two tags, each of which was inserted into the dorsal musculature in a location that could be described by a three-digit alphanumeric code. The first digit of the code indicated the number of the dorsal fin (1 = anterior fin, 2 = middle fin, 3 = posterior fin), the second indicated whether the tag was on the left (L) or right (R) side of the fish, and the third indicated whether the tag was positioned near the anterior (1 = origin) or posterior (2 = insertion) fin rays. For example, the code 2L1 indicates that the tag was positioned near the origin of the second dorsal fin on the left side of the fish. Cod were maintained at densities similar to those reported in the wild (Rose 1993; Morgan et al. 1997), experienced water temperatures of approximately 8°C and ambient photoperiods, and fed Shurgain® 8 mm fish feed daily.

Cod from the Western Scotian Shelf (25 females and 27 males, ranging 42 - 78 cm in length) spawned 29 December 2000 – 6 March 2001 and cod from the

Southern Gulf of St. Lawrence (41 females and 18 males, ranging 50 - 70 cm in length) spawned 5 May – 11 July 2001. Behaviour of individuals was monitored throughout the spawning periods. A platform above the tank permitted behaviour to be recorded by direct observation. During direct observations, each individual fish was monitored continuously for 3 minutes within the last 8 hours of daylight each day (Western Scotian Shelf: 08:00 - 16:00; Southern Gulf of St. Lawrence: 12:00 - 20:00), and the incidence of five agonistic behaviours was documented: (i) chase (swimming toward a swimming fish), (ii) approach (swimming toward a stationary fish), (iii) bite (physical contact between one fish's mouth and another fish's body or fin), (iv) nip (an attempted bite, i.e., an opening and closing of the mouth near another fish's body or fin), and (v) prod (contact between one fish's snout and another fish's body). I made direct observations of every fish from the Western Scotian Shelf on 54 occasions and of every fish from the Southern Gulf of St. Lawrence on 34 occasions. Individuals were examined in varying order each day. Courtship behaviour was infrequent relative to agonistic behaviour and seldom observed during 3-minute focal fish watches. Therefore, in order to examine courtship activity, behaviour of fish from the Southern Gulf of St. Lawrence was recorded by four video cameras mounted above the tank and a colour videotape recorder 12:00 - 20:00 from 15 May 2001 through 14 July 2001. Subsequent to the spawning season, all videotapes were reviewed (totaling 488 hours of video coverage) and the frequency of occurrence of ventral mounts and spawning events (as indicated by the appearance of a cloud of milt in the water around a pair of fish engaged in a ventral mount) by each individual in the group were quantified. This camera

array was not available for data collection for Western Scotian Shelf fish during 2001.

Seawater was continuously pumped into the tank and out flowing surface water was sieved through a plankton net that collected all spawned eggs, which are buoyant in seawater. Each batch of spawned eggs was collected from the net daily at 16:00. Examination of the developmental stage of random samples of eggs collected indicated that almost all had been freshly spawned and fertilised within the previous 24 hours. Each batch of eggs was incubated at approximately $8 \degree C$ in 20 L containers of seawater. When eggs were < 24 hours from hatching, a random sample of about several thousand eggs from each batch was collected and stored in ethanol for subsequent genetic analysis. Random egg samples were obtained by ensuring that eggs were well mixed before making a single sweep up through the water column with a small net.

Experiments were terminated and fish sacrificed when no eggs had been collected for five days. Blood samples were obtained from each individual and preserved in ethanol for pedigree analysis. I recorded total length of each individual to the nearest millimeter, as well as total mass, liver mass, and gonad mass to the nearest 0.5 g. Somatic mass was calculated as total mass less the gonad mass. I also determined sex and stage of maturity by observation of gross morphology of the gonad (Morrison 1990) to verify that each individual was reproductively mature. The three pairs of drumming muscles were extracted from the surrounding tissue using forceps and their combined dry mass was measured to the nearest 0.0001 g for each fish. In addition, saggital otoliths were collected and ages later determined by sectioning the otoliths and counting

annuli. Unfortunately, for technical reasons, I was unable to determine liver mass of eight individuals from the Western Scotian Shelf or age of one individual from the Southern Gulf of St. Lawrence.

Genetic analysis

DNA was collected from blood of parental fish using a Qiagen genomic extraction kit according to the manufacturer's protocol and from whole eggs, using the extraction method of Jones and Hutchings (2002). Incubation of eggs produced samples of 38 egg batches from Western Scotian Shelf and 22 egg batches from Southern Gulf of St. Lawrence. From each of the batches, 11-171 offspring were genotyped to include in the parentage analyses for a total of 2931 offspring genotyped for the full season from Western Scotian Shelf and 1633 for the full season from Southern Gulf of St. Lawrence. From a variety of available cod microsatellite DNA loci, three tetranucleotide repeat loci, *Gmo8*, *Gmo19*, and *Gmo37*, and one trinucleotide repeat locus, *Gmo35*, were chosen to be run on all samples based on their high levels of heterozygosity and genotyping reliability (Miller et al. 2000). In cases where these loci proved insufficient in determining parentage, two tetranucleotide repeat loci, *Gmo34* (Miller et al. 2000) and *Mae9* (a haddock (*Melanogrammus aeglefinus*) microsatellite DNA locus; O'Reilly et al. 2002), were also examined so that paternity could be unambiguously established. Loci were amplified using polymerase chain reaction conditions specified by Miller et al. (2000) and O'Reilly et al. (2002), samples were run on 6% denaturing acrylamide gels, and microsatellite alleles or bands were visualized using an FMBIO II (Hitachi, Tokyo, Japan). For parental fish, all loci were amplified and

run three times to ensure correct genotyping. Standard individuals were run on all gels to check consistency in fragment lengths.

Parentage was determined by the program PROBMAX (Danzmann 1997), which assigns progeny to parents from a mixture of potentially contributing parents when parental genotypes are known. This program also allows manipulation of the data set to identify potential mis-genotyping, such as failure to recognize a heterozygote due to stuttering or mis-genotyping by one allele length (see PROBMAX program manual).

Data analyses

Differences among males in the number of offspring sired during the spawning season were assessed in relation to aspects of morphology, condition, and spawning behaviour using a generalized linear model (GLM) of count data with poisson errors (Crawley 2002). Using S-Plus (Version 6.1), I fitted a maximal model and then simplified it by sequentially removing non-significant explanatory variables to obtain a minimal adequate model. To adjust for overdispersion of the data (Western Scotian Shelf: dispersion parameter for minimal adequate model = 169; Southern Gulf of St. Lawrence: dispersion parameter for minimal adequate model = 41), model simplification was conducted using an F test instead of a chi-square test (Crawley 2002). To examine the association between reproductive success and drumming muscle investment, I controlled for the influence of body size on drumming muscle mass by using the residuals of the regression of drumming muscle mass on somatic mass (Chapter 3). Two standard condition indices that have been identified as reflecting the total available energy reserves of cod (Lambert and Dutil 1997)

were calculated for each fish. Fulton's K was calculated by dividing the somatic mass by body length cubed and then multiplying by 100. The hepatosomatic index, HSI, was calculated by dividing liver mass by total body mass and multiplying by 100. These indices are based on the generally accepted assumption that a heavier body mass for a given length or a heavier liver for a given body mass, corresponds to better condition. Fulton's K and HSI have been shown to be related to feeding intensity, growth, maturation, fecundity, and survival, thus making them potentially good indicators of the general well-being of individuals (Lambert and Dutil 1997). The aggressiveness of each male was determined based upon the number of agonistic interactions observed initiated by that male towards other fish during the sum of all daily 3-minute focal fish watches for which he was monitored. I considered mating attempts to be reflected by the total number of ventral mounts initiated by individual males in the video recordings.

RESULTS

The number of offspring sired during the spawning season was highly variable across males in both groups of fish examined. The most successful male sired approximately 51% of the offspring in the Western Scotian Shelf group (Figure 2.1). In the Southern Gulf of St. Lawrence group, the most successful male sired approximately 26% of the offspring (Figure 2.1). In both groups, more than 80% of offspring were sired by the top five most successful males while some males had no fertilisation success (Western Scotian Shelf: 7 males or 26%

sired no offspring; Southern Gulf of St. Lawrence: 1 male or 6% sired no offspring).

I found strong positive associations among the various metrics of body size for both groups of fish examined (Table 2.1). Similarly, there was a strong positive association between Fulton's K and HSI, the two metrics of condition (Western Scotian Shelf: $R = 0.87$, $P < 0.0001$, $n = 19$; Southern Gulf of St. Lawrence: $R = 0.91$, $P < 0.0001$, $n = 18$). Therefore, I elected to incorporate only a single metric of body size and a single metric of body condition in the maximal model examining number of offspring sired in relation to aspects of morphology, condition, and spawning behaviour. For the maximal model, I selected somatic mass as the metric of male body size (to maximize variation among males) and Fulton's K as the metric of male body condition (to maximize the number of data points available for analysis).

For both groups examined, the minimal adequate model describing the number of offspring sired included only somatic mass and the number of agonistic interactions initiated (Tables 2.2-2.5). The number of offspring sired tended to increase with increased somatic mass (Figures 2.2 and 2.3; Tables 2.4 and 2.5) and increased number of agonistic interactions initiated by individuals during the spawning season (Figures 2.4 and 2.5; Tables 2.4 and 2.5). The variation explained by the minimal adequate model was approximately 58% for the Western Scotian Shelf group and 76% for the Southern Gulf of St. Lawrence group. Other morphological traits (post-spawning body condition and drumming muscle investment) and the number of ventral mounts initiated (i.e. mating attempts) appeared to show little association with male reproductive

success (Tables 2.2 and 2.3).

DISCUSSION

Reproductive behaviour has significant consequences for population ecology and genetic structure of fishes. An example of this is when a small number of males control a large number of females, which leads to a reduction in the genetically effective population size (N_e), a parameter of considerable import in conservation biology because of its negative association with rate of loss of genetic variation (Nunney 1993). In broadcast spawning marine fishes, large differences between estimates of N_e and census population sizes are often reported (e.g. Hauser et al. 2002; Turner et al. 2002), and high variance in reproductive success among individuals has been proposed as a cause of these differences (Hedgecock 1994). Unfortunately, reproductive success of individuals, particularly among broadcast spawning marine species, is extremely difficult to measure and few studies have dealt with this issue (Avise et al. 2002).

I examined variation in reproductive success of male Atlantic cod and its relation to patterns of individual morphology and social behaviour using large captive groups from two Northwest Atlantic populations. It is reasonable to assume that spawning behaviour and reproductive success could have been influenced by stress associated with captivity. However, I do not believe that stress had a severe impact on the observed patterns of behaviour and paternity because the cod in my study were not subject to disturbance after their arrival at Dalhousie and were not visibly stressed. Furthermore, in a comparison of captive groups of experimentally stressed cod with groups of unstressed cod,

although stressed cod performed slightly altered courtship sequences, their spawning and fertilisation rates remained similar to those of unstressed individuals (Morgan et al. 1999).

While spawning groups under natural conditions are open systems with potentially millions of individuals free to move around in response to social or environmental cues, my experimental protocol involved a closed system with relatively few individuals present. Captivity may have altered female availability or male reproductive strategies relative to those found in nature, although I have no direct information regarding whether this is actually the case. Very little is known about cod spawning behaviour in the wild. Atlantic cod spawn over vast areas of the continental shelf and slope (Hutchings et al. 1993) in aggregations that may span several kilometres. Average densities of 0.004 fish/m^3 have been reported for spawning aggregations in nature (Morgan et al. 1997) which does not preclude the possibility that males establish territories of a size similar to those that have been described in captivity (Brawn 1961a). Field recordings at natural spawning grounds when large numbers of spawning cod are present have revealed increased sound activity within the frequency range of sounds produced by cod in captivity (Nordeide and Kjellsby 1999), suggesting that acoustic communication is also an important part of spawning behaviour in the wild. However, in the absence of detailed information about spawning patterns outside of captivity, I may only cautiously assume that the patterns observed in my study reflect those under natural spawning conditions. Fortunately, the tank used to conduct my experiments has the largest volume (684 m^3) of any ever used to study cod spawning behaviour. In addition, because

I used a closed system, I was able to collect all eggs produced by spawning females in my study populations and randomly sample fertilised eggs to determine parentage and estimate relative reproductive success of individuals.

Past observations of cod spawning behaviour and reproductive success have suggested that paternity frequencies may be highly skewed (Brawn 1961a; Hutchings et al. 1999; Rakitin et al. 2001; Bekkevold et al. 2002). In the present study, I examined male reproductive success in captive groups of 52-59 individuals, group sizes that were more than three times larger than any examined in the past. My observations of these large groups confirm the high skew in male reproductive success suggested by previous studies. Reproductive skew appeared particularly high in the Western Scotian Shelf group where one male sired approximately 51% of the offspring and 26% of males sired no offspring. This large skew in reproductive success may have been related to the size range of males involved. In the Western Scotian Shelf group, most males ranged 44-55 cm in length while the three most successful males that collectively achieved 78% of the fertilisation success were substantially larger, measuring greater than 70 cm in length. In contrast, all males in the Southern Gulf of St. Lawrence group were more similar in body size (ranging 54-66 cm in length) and reproductive success was more evenly distributed.

In both of my experimental groups, male reproductive success was positively associated with male body size and aggression. Previous investigators have hypothesized that agonistic interactions and large body size may enable some males to exclude other individuals from small areas in which they preferentially engage in courtship and spawning with females (Brawn 1961a; Hutchings et al.

1999; Bekkevold et al. 2002). Consequently, one interpretation of the paternity patterns in my study is that the male siring the most offspring was the male that initiated the most ventral mounts and most often elicited egg release in the female. However, this could not be verified by the video observations. Only six spawning events were recorded on video and although all clearly involved satellite males (defined as individuals that quickly joined the spawning couple and also released sperm), it was not often possible to determine the identities of individuals involved and thus, link male behaviour during spawning to parentage data. Unfertilised cod eggs and sperm remain viable for more than one hour in seawater (Kjørsvik and Lønning 1983; Trippel and Morgan 1994b) potentially enabling sperm to compete for fertilisation during a long period following ejaculation, and ejaculating even when not engaged in a ventral mount with a spawning female may enable males to acquire some reproductive success. Therefore, it is possible that larger males may have sired more offspring simply by having larger ejaculates which contained more sperm than those of smaller males, rather than because they acquired more matings with females. A considerable amount of variation remained unexplained by the minimal adequate models suggesting that additional unidentified factors may be of considerable import to cod reproductive success.

Behavioural and genetic data in the present study are consistent with the hypothesis that alternative mating tactics exist among male Atlantic cod (Hutchings et al. 1999). Multiple males appeared to release sperm in all spawning events even though typically only one male was engaged in the ventral mount with the spawning female. Accordingly, the number of ventral

mounts initiated (i.e. mating attempts) did not appear to be a strong predictor of reproductive success (Table 2.3) and several males in my spawning groups achieved fertilisation success seemingly without having initiated much agonistic activity (although I cannot exclude the possibility that these individuals also engaged in courtship and aggressive activity but outside my periods of daily observation; Figures 2.4 and 2.5). Unlike some other fishes that exhibit 'bourgeois' and 'parasitic' male forms (Taborsky 1998), there do not appear to be morphological differences between male Atlantic cod employing alternative mating tactics. Estimates of reproductive success between 1-5% for individual satellite males are consistent with those in salmonids (Jones and Hutchings 2001, 2002). The ability of some males to sire offspring without courting females or aggressively interacting with other males may partially explain weak relationships apparent between reproductive success and morphological traits examined.

Experiments by Rakitin et al. (2001) involving pairs of males differing in body size and spawning with a single female during an entire spawning season failed to find a correlation between male reproductive success and body size. However, their experimental set-up offered very little opportunity for mate choice and at this scale, factors other than body size may have been more important in determining the proportion of offspring sired by each male (Rakitin et al. 2001).

High skew in male reproductive success with larger, more aggressive males siring a larger number of offspring than smaller, less aggressive males as reported here, is consistent with the hypothesis that cod reproduce within the

behavioural constructs of a lekking mating system. Although, the degree to which the reproductive skew may be a function of intrasexual competition among males or female choice of mate based upon sexually selected characters, as documented in other lekking species, remains unknown. Previous studies of Atlantic cod spawning behaviour have also suggested that this species might exhibit a lekking mating system (Hutchings et al. 1999; Nordeide and Folstad 2000). Specifically, the mating characteristics of species showing conventional lekking behaviour are similar to those known or proposed for Atlantic cod: (i) absence of male parental care, (ii) inability of males to monopolise resources to gain matings with females, (iii) defence by males of small, clustered mating territories that females visit solely for mating, (iv) self-advertisement by males in the form of visual, acoustic, or olfactory displays, and (v) non-random mating by females (Höglund and Alatalo 1995; Hutchings et al. 1999; Nordeide and Folstad 2000). With the exception of some cichlids, leks have rarely been described in fishes (Höglund and Alatalo 1995).

Together with previous studies of Atlantic cod spawning behaviour and reproductive success, my study indicates high skew in paternity frequencies. However, to date, all investigations have only lasted for a single spawning season or less and may not reflect lifetime reproductive success. Although male reproductive success may be highly skewed within one year, it is often age- and experience-dependent so that variation in lifetime reproductive success is likely to be much lower than variation within years (Andersson 1994). Accordingly, I found that larger, more aggressive males sired a larger number of offspring. Further studies of cod spawning behaviour and parentage that consider both

sexes and address alternative male mating tactics and temporal variation in reproductive success are warranted.

Table 2.1. Correlation coefficients between various metrics of body size of male Atlantic cod from the Western Scotian Shelf and Southern Gulf of St. Lawrence.

	Western Scotian Shelf			Southern Gulf of St. Lawrence		
	R	P	n	R	P	n
Somatic mass vs. length	0.94	< 0.0001	27	0.78	0.0001	18
Somatic mass vs. age	0.59	0.001	27	0.67	0.003	17
Length vs. age	0.69	< 0.0001	27	0.54	0.03	17

Table 2.2. Models describing number of offspring sired by male Atlantic cod from the Western Scotian Shelf in relation to morphological and behavioural traits.

Model	Deviance	Change in deviance	P
somatic mass + Fulton's K + drumming muscle investment + agonistic interactions	3785.0		
somatic mass + Fulton's K + agonistic interactions	3837.4	52.4	0.61
somatic mass + agonistic interactions	4044.7	207.3	0.29
somatic mass	5162.2	1117.5	0.02
agonistic interactions	4903.0	858.3	0.05

Table 2.3. Models describing number of offspring sired by male Atlantic cod from the Southern Gulf of St. Lawrence in relation to morphological and behavioural traits.

Model	Deviance	Change in deviance	P
somatic mass + Fulton's K + drumming muscle investment + agonistic interactions + ventral mounts	524.3		
somatic mass + Fulton's K + agonistic interactions + ventral mounts	539.7	15.4	0.59
somatic mass + agonistic interactions + ventral mounts	565.3	25.6	0.47
somatic mass + agonistic interactions	610.3	45.0	0.32
somatic mass	2220.8	1610.5	< 0.0001
agonistic interactions	955.5	345.2	0.01

Table 2.4. Parameter estimates for the minimal adequate model describing number of offspring sired by male Atlantic cod from the Western Scotian Shelf in relation to morphological and behavioural traits.

	Value	SE
Intercept	2.63	0.0521
Somatic mass	0.000639	0.0000208
Agonistic interactions	0.151	0.00471

Table 2.5. Parameter estimates for the minimal adequate model describing number of offspring sired by male Atlantic cod from the Southern Gulf of St. Lawrence in relation to morphological and behavioural traits.

	Value	SE
Intercept	1.93	0.117
Somatic mass	0.00109	0.0000577
Agonistic interactions	0.0482	0.00115
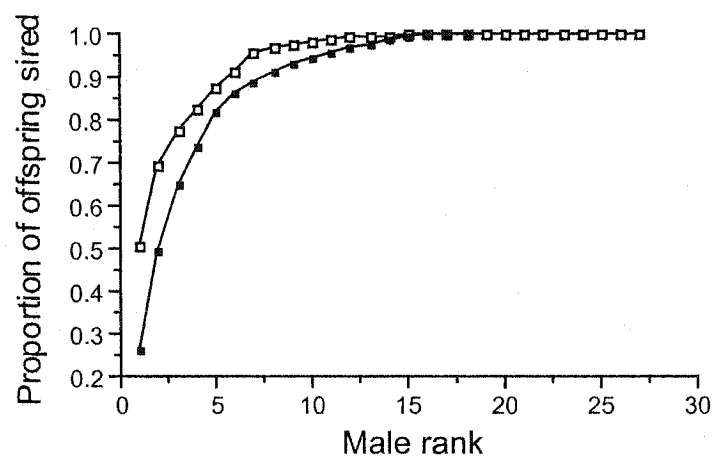
Figure 2.1. Cumulative proportion of offspring sired by male Atlantic cod ranked from most to least successful. Open squares, Western Scotian Shelf cod; closed squares, Southern Gulf of St. Lawrence cod.
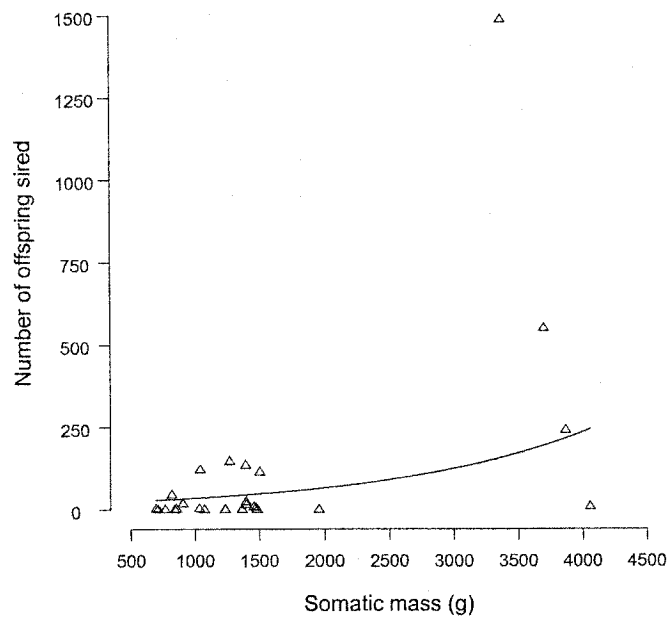
Figure 2.2. Variation in number of offspring sired in relation to somatic mass of male Atlantic cod from the Western Scotian Shelf. Line indicates values predicted by the minimal adequate model with number of agonistic interactions initiated held constant at the mean observed value (1.8).
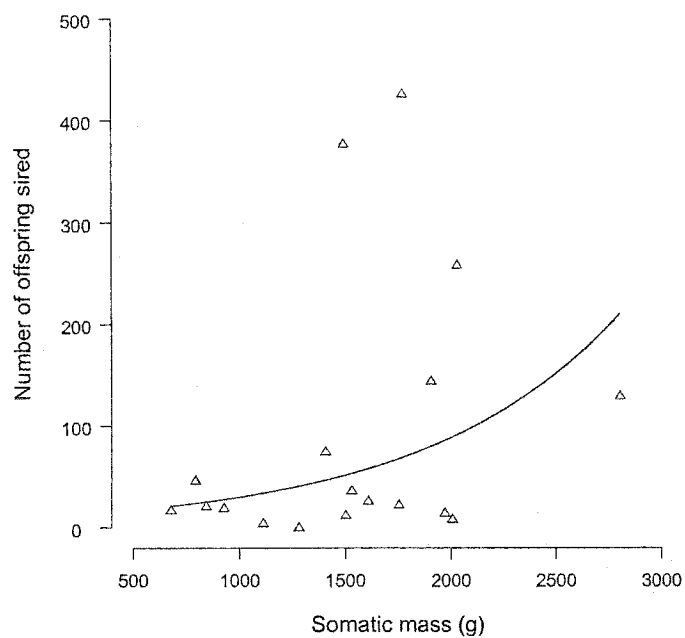
Figure 2.3. Variation in number of offspring sired in relation to somatic mass of male Atlantic cod from the Southern Gulf of St. Lawrence. Line indicates values predicted by the minimal adequate model with number of agonistic interactions initiated held constant at the mean observed value (7.9).
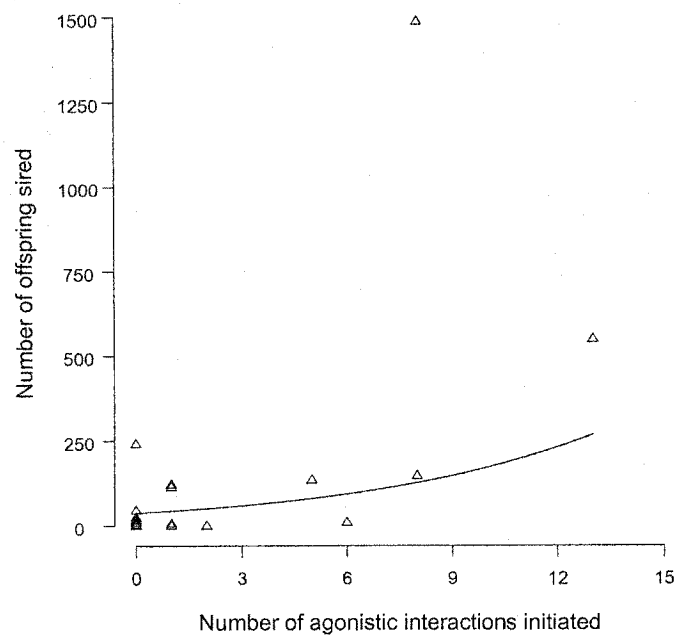
Figure 2.4. Variation in number of offspring sired in relation to number of agonistic interactions initiated towards other individuals by male Atlantic cod from the Western Scotian Shelf. Line indicates values predicted by the minimal adequate model with somatic mass held constant at the mean observed value (1559 g).
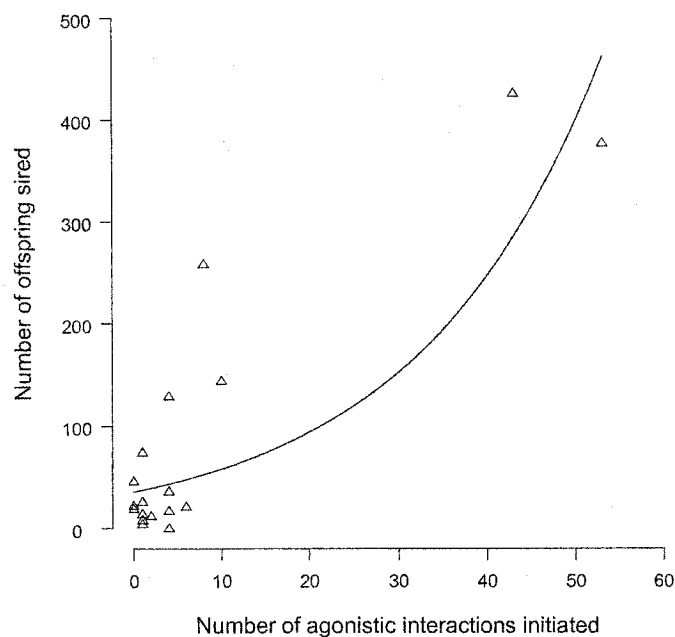
Figure 2.5. Variation in number of offspring sired in relation to number of agonistic interactions initiated towards other individuals by male Atlantic cod from the Southern Gulf of St. Lawrence. Line indicates values predicted by the minimal adequate model with somatic mass held constant at the mean observed value (1522 g).

CHAPTER 3

THE FUNCTION OF SOUND PRODUCTION BY ATLANTIC COD
AS INFERRED FROM PATTERNS OF VARIATION IN
DRUMMING MUSCLE MASS

INTRODUCTION

Conspicuous song and other acoustic displays produced by animals during the breeding season may vary in expression between females and males, across size or age classes, and among individuals in ways that reveal much about their function (Andersson 1994). In many species, experimental evidence has demonstrated that song and other acoustic displays by males are favoured by sexual selection because females prefer to mate with males with the most exaggerated features, such as the largest song repertoire or the loudest, longest, or most low pitch acoustic displays (Andersson 1994). For example, in playback experiments using loudspeakers, female tungara frogs (*Physalaemus pustulosus*) preferred low-pitched calls characteristic of large males, which were capable of higher fertilisation success than small males (Ryan 1983). Song and other acoustic displays may also be favoured by sexual selection through their role in intrasexual contests for access to mates. For instance, possession of a good territory is crucial for mating success in many birds (Andersson 1994) and song helps in territory defense as demonstrated by muted male red-winged blackbirds (*Agelaius phoeniceus*) which are less likely to gain and more likely to lose territories than control males (Peek 1972; Smith 1976, 1979). Consequently, an important aspect of display trait variation is sexual dimorphism, which reflects differences in the force of sexual selection on males versus females. Several studies have also documented high levels of variation in sexually selected

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display traits relative to morphological traits thought to be due to other forms of natural selection (Alatalo et al. 1988; Jones and Montgomerie 1992; Evans and Barnard 1995; Jones et al. 2000). Wide variation in display traits has been hypothesized to result from lack of stabilizing selection (Alatalo et al. 1988) and the tendency for their expression to be costly and thus correlated with individual health or condition (Grafen 1990; Iwasa et al. 1991; Møller 1991).

Most studies of song and acoustic displays have focused on birds, insects, and frogs (Andersson 1994). Fishes also perform acoustic displays that may be critical to mating success (Hawkins 1993) but these have received comparatively little attention. Atlantic cod (*Gadus morhua*) is a broadcast spawning marine demersal fish found on both sides of the North Atlantic that reproduces in water depth ranging from tens to hundreds of metres (Brander 1994; Morgan et al. 1997; Smedbol and Wroblewski 1997). Like many fishes, cod are capable of producing and detecting sound (Hawkins 1993). The calls of Atlantic cod are produced by three pairs of drumming muscles exterior to the swimbladder wall (Brawn 1961b) and consist of short 'grunts' with peak sound amplitudes at frequencies ranging between 50 and 500 Hz (Brawn 1961b; Hawkins and Rasmussen 1978; Midling et al. 2002). Recent studies have shown that successful reproduction in Atlantic cod involves complex behaviour within and between sexes and it has been hypothesized that acoustic displays are used in mate competition (Brawn 1961a, 1961b; Hutchings et al. 1999).

Release of sperm and eggs by Atlantic cod involves a 'ventral mount' in which the male, while grasping the female with his pelvic fins and matching her swimming speed, positions himself beneath the female. Prior to mounting

attempts, males exhibit aggression towards other fish, typically in the form of fast approaches and threat displays that are often accompanied by grunting sounds (Brawn 1961a, 1961b; Hutchings et al. 1999). Agonistic interactions are believed to reflect competition among males for mates and may influence access to females (Brawn 1961a, 1961b; Hutchings et al. 1999). Interactions between sexes are consistent with the hypothesis that females, and possibly males, exercise mate choice (Hutchings et al. 1999). One prominent behaviour observed in large tanks is the 'flaunting' display exhibited by males as they circle around individual females on or near the bottom with all the median fins fully erect (Brawn 1961a; Hutchings et al. 1999). Brawn (1961a, 1961b) reported that while giving this display before the female during courtship, the male also makes grunting sounds which increase the 'excitement' of the female as indicated by a momentary increase in speed, turn towards the male or tensing of the median fins. Hutchings et al. (1999) hypothesised that females may be choosing males on the basis of these acoustic displays.

Preliminary observation has suggested that Atlantic cod produce sound most frequently during the spawning period and although both sexes are capable of producing sounds throughout the year, only males seem to do so during the spawning season (Brawn 1961b). This pattern of sound production strongly suggests a sexually selected function but sporadic observations at variable times of day and examination of only a small group of individuals that differed in number and sex composition limits the conclusions that can be drawn from observations to date (Brawn 1961a, 1961b). Furthermore, there has been no attempt to quantitatively relate characteristics of the sounds produced to

seasonal or individual variation.

To understand the function of a trait, it is essential to understand its patterns of variation. However, measuring individual variation in the acoustic displays of fishes has proven challenging. Because sounds propagate rapidly and effectively in water, it is often difficult to identify which individual emitted a particular sound when fish occur in groups. Furthermore, fish rarely produce acoustic displays when alone (Bremner et al. 2002). Nonetheless, mass of the drumming muscles, the acoustic display structure, has a large influence on the characteristics of sound produced (Connaughton et al. 1997, 2000) and can be used as a metric of acoustic display expression. For instance, male weakfish (*Cynoscion regalis*) also produce sounds using drumming muscles exterior to the swimbladder wall. In weakfish, the mass of these muscles is as seasonal as their use, with the muscles hypertrophying early in the spring spawning season and atrophying markedly late in the summer in response to changes in blood androgen levels (Connaughton and Taylor 1994, 1995). Sound amplitude mirrors this pattern, becoming more intense when the muscle is hypertrophied and decreasing in amplitude coincident with muscle atrophy (Connaughton et al. 1997).

In the present study, I sampled Atlantic cod captured in the commercial fishery to quantify individual variation in the size of the drumming muscles (the acoustic display structure) for comparison with other evidence on the role of sexual selection in the evolution of sound production (acoustic display expression) by cod. Specifically, my objectives were to: (i) evaluate potential differences in drumming muscle mass in relation to sex, spawning status, and

body size; (ii) quantify the amount of variation expressed in drumming muscles by each sex, as well as the relative amount of variation expressed in drumming muscles versus other morphological traits; and (iii) test whether drumming muscle mass reflected body condition or fertilisation potential of spawning males.

METHODS

To quantify variation in drumming muscle mass, I examined random samples of Atlantic cod caught on the Western Scotian Shelf (Northwest Atlantic Fishery Organization Division 4X). Samples ranging between 60-139 individuals were collected on nine occasions between 21 March 2001 and 25 February 2002 (Table 3.1). Fishing trips would typically last 3-5 days. Fish were captured using an otter trawl and placed on ice until arrival in port where they were measured and processed. For each individual examined, I recorded total length to the nearest centimeter, as well as total mass and gonad mass to the nearest gram. Somatic mass was calculated as total body mass less the gonad mass. I also determined sex and maturity (immature, ripening, ripe/spawning, and spent) of each individual by observation of gross morphology of the gonad (Morrison 1990). Saggital otoliths were collected and ages later determined by sectioning the otoliths and counting annuli. The three pairs of drumming muscles cover the second, third, and fourth lobes of the swimbladder and are attached to the pleural rib and the swimbladder wall (Brawn 1961b). Considerable time is required to separate drumming muscles from the surrounding tissue using forceps so the section of vertebral column on which they are found was removed

from the body and frozen for later dissection. After extraction, combined dry mass of the three pairs of drumming muscles was measured to the nearest 0.0001 g.

I used ANCOVA with somatic mass (a metric of body size) as the covariate to test for differences in drumming muscle mass in relation to sex and spawning status. Initial models included all interaction terms although non-significant interaction terms were removed from subsequent models via model simplification (Crawley 2002). Mass data were log-transformed to meet the assumptions of normality.

Based upon observation of gross morphology of the gonad, I found that it was sometimes difficult to distinguish between 'immature' and 'spent' maturity stages, as well as between 'ripening' and 'ripe/spawning' maturity stages (Morrison 1990). Therefore, for the purpose of my analyses, I considered individuals to be 'spawning' if they were initially scored as ripening or ripe/spawning and 'non-spawning' if they were initially scored as immature or spent.

Controlling for fish age, I used sign tests to examine sexual dimorphism in the coefficients of variation of drumming muscle mass and other morphological traits. In addition, controlling for fish sex and age, I used sign tests to compare the coefficients of variation of drumming muscle mass to those of other morphological traits. For all sign tests, only age classes in which at least 10 individuals were sampled were included in the analysis.

To test whether drumming muscle mass reflected body condition of spawning males, I regressed the residuals of the regression of drumming muscle

mass on somatic mass against the residuals of the regression of somatic mass on length. I selected somatic mass for use in this analysis because it excluded the mass of the gonad, which can vary significantly and independently of fish condition between seasons and within stocks (Lambert and Dutil 1997). In addition, because available energy reserves will be located in somatic tissues as opposed to germ cells, somatic mass may be considered a good reflection of condition (Lambert and Dutil 1997). My technique produced a standard metric of condition on the x-axis, and by using the residuals of the regression of drumming muscle mass on somatic mass on the y-axis, I removed any possibility that the ensuing association was a spurious consequence of drumming muscle mass being associated with somatic mass. Similarly, to test whether drumming muscle mass reflected fertilisation potential of spawning males, I examined whether there was any association between residuals of the regression of drumming muscle mass on somatic mass and residuals of the regression of gonad mass on somatic mass.

RESULTS

During 2001-2002, I measured 913 Atlantic cod from the Western Scotian Shelf and drumming muscle mass was highly variable among individuals. Using ANCOVA with somatic mass as the covariate to test for differences in drumming muscle mass in relation to sex and spawning status, I found a significant interaction between sex and somatic mass (Two Factor ANCOVA: $F_{1, 908} = 13.93$, $P = 0.0002$; Figure 3.1; Table 3.2). Specifically, controlling for body size, males generally displayed larger drumming muscles than females although the

magnitude of this difference tended to increase with increasing body size considered. Further ANCOVA involving only males revealed a significant association between drumming muscle mass and somatic mass (One Factor ANCOVA: $F_{1, 412} = 1099.08$, $P = 0.0001$), as well as a significant difference in drumming muscle mass in relation to spawning status (One Factor ANCOVA: $F_{1, 412} = 6.61$, $P = 0.01$). Similarly, involving only females, I observed a significant association between drumming muscle mass and somatic mass (One Factor ANCOVA: $F_{1, 495} = 1355.10$, $P = 0.0001$), although there was no difference in drumming muscle mass in relation to spawning status (One Factor ANCOVA: $F_{1, 495} = 0.07$, $P = 0.79$).

Sign tests provided no evidence for sexual dimorphism in the coefficients of variation for drumming muscle mass ($P = 0.38$), somatic mass ($P = 1.00$), or body length ($P = 1.00$; Table 3.3). However, this may have been a result of small sample sizes, particularly in the case of drumming muscle mass where differences between females and males in the coefficient of variation within age classes were large. Generally, drumming muscles exhibited more variation than other morphological traits examined (Table 3.3). The coefficients of variation for drumming muscle mass were greater than those for body length for both females ($P = 0.02$) and males ($P = 0.03$). I also found greater coefficients of variation for drumming muscle mass than somatic mass among males ($P = 0.03$), but there was no such difference among females ($P = 0.13$).

Residual analyses revealed that, among spawning males, drumming muscle mass was weakly, but significantly, associated with both my metric of body condition ($F_{1, 110} = 6.61$, $P = 0.01$, $R^2 = 0.06$; Figure 3.2a) and fertilisation potential

($F_{1, 110}$ = 5.24, P = 0.02, R^2 = 0.05; Figure 3.3). Controlling for differences in age and, to some degree, differences in lifetime breeding events, associations between drumming muscle mass and body condition were positive for spawning male cod 3, 4, and 5 years of age (Figure 3.2b-d), and statistically significant for all but the 4-year-old spawning males (spawning males aged 3 years: $F_{1, 47}$ = 4.83, P = 0.03, R^2 = 0.09; spawning males aged 4 years: $F_{1, 18}$ = 0.80, P = 0.38, R^2 = 0.04; spawning males aged 5 years: $F_{1, 28}$ = 4.22, P = 0.05, R^2 = 0.13).

DISCUSSION

The first description of sound production by spawning cod was based primarily on observations of seven females and six males held in captivity (Brawn 1961a, 1961b). This important preliminary investigation suggested that Atlantic cod produce sound most frequently during the spawning season, typically during agonistic interactions and courtship display by males. However, Brawn (1961a, 1961b) was unable to quantitatively relate characteristics of the sounds produced to seasonal or individual variation and there has been no definitive study subsequent to her work to elucidate the function of sound production by Atlantic cod.

In the present study, I elected to address this problem by examining patterns of variation in mass of the sound-producing musculature, which can be precisely measured, comparing among individuals. Although there are no data confirming that the mass of drumming muscles determines the loudness or other characteristics of sounds produced by Atlantic cod, for a number of reasons, I believe drumming muscle mass to be an appropriate metric for potential sound

production by individuals. Firstly, it makes intuitive sense that the muscles powering cod sound production should directly determine the nature of the sound. One would predict, for example, that larger muscles should produce greater sound energy; evidence for this is forthcoming in weakfish for which sound pressure level increases with increasing sonic muscle mass (Connaughton et al. 1997). Secondly, individual drumming muscle mass should demonstrate more consistency over time than the characteristics of sounds they produce, thus making it a more reliable indicator of individual sound-producing ability. For example, sound characteristics including sound pressure level and dominant frequency have been shown to vary in relation to environmental conditions, particularly water temperature, on a day-to-day basis, making an individual's sounds difficult to measure and compare over time (Connaughton et al. 2000; Midling et al. 2002). Furthermore, drumming muscle mass is easy to quantify as a characteristic of individuals because the tissue can be dissected and dried for very precise measurements.

Controlling for the influence of body size, I found that Atlantic cod males have relatively larger drumming muscles than females and that among males, drumming muscles increase in mass before spawning and decline thereafter. Because spawning status and time of year were confounded in my study (i.e. most spawning occurred November - February), I cannot exclude the possibility that variation in drumming muscle mass is a seasonal occurrence unrelated to spawning activity. However, I believe that the most parsimonious explanation for the patterns observed is that sound production by males during the spawning season has fitness benefits, perhaps through a role in mate competition. Sexual

dimorphism and seasonal variation in sound-producing musculature have been reported for several other fishes, including haddock (*Melanogrammus aeglefinus*) in which males have drumming muscles larger than females and that increase to nearly twice their normal size during the pre-spawning and spawning period (Templeman and Hodder 1958). This also matches the patterns of display trait variation of numerous other animals including insects, amphibians, reptiles, birds and mammals that relate to their primary use in intrasexual contests and courtship during the breeding season (Andersson 1994). The coefficients of variation observed for Atlantic cod drumming muscles indicate that they are generally more variable than other morphological traits, a pattern consistent with a widespread pattern in animals whereby traits believed to be influenced by sexual selection are more variable than other traits (Andersson 1994).

Why should male cod have larger drumming muscles than females and invest in their development during the breeding season? An accumulating variety of albeit circumstantial evidence suggests it is because cod acoustic displays are produced mostly by males to re-enforce threat displays directed primarily towards other males, as well as to attract and communicate spawning readiness to females (Brawn 1961a, 1961b; Chapter 4).

I found that drumming muscle mass showed a strong positive relationship with body size, indicating that sound production is likely to be a good indicator of the size of the signaler. This can be seen in weakfish for which there is also a strong positive relationship between the mass of the sound-producing musculature and overall body size, which results in increased sound pressure level and decreased dominant frequency with increasing fish size (Connaughton

et al. 2000). Similarly, many sound-producing animals show decreasing dominant sound frequency with increasing animal size (e.g. tungara frog, Ryan 1983). Atlantic cod are broadcast spawners that release eggs and sperm during a ventral mount and there is evidence to suggest that size complementarity between spawning partners might be important to ensure that the urogenital openings of both fish are aligned opposite one another and that high fertilisation success is achieved (Rakitin et al. 2001). Consequently, assessment of male sound characteristics may provide useful information to females regarding size and hence suitability of potential mates. Although it is conceivable that cod could visually assess the size of conspecifics, most spawning occurs at night (Chambers and Waiwood 1996) when visual signals are less effective (Anthony 1981).

Elaborate animal displays are often thought to convey not only size information but also details about the condition or quality of the signaler to potential mates (Andersson 1994). I found a weak but statistically significant positive relationship between relative drumming muscle mass and an index of body condition. Given that male Atlantic cod provide neither parental care nor physical resources to females, the possible benefits to female fitness associated with mate preferences are limited. Males in good condition may be less likely to be affected by parasites and diseases that could be acquired by the female during reproduction. In addition, there is recent evidence to suggest that male condition is a potentially critical factor determining sperm fertilisation potency in Atlantic cod (Rakitin et al. 1999a). I also found evidence that males with relatively large drumming muscles have larger gonads, even after controlling for body size, thus allowing them to produce large amounts of sperm. In broadcast spawners such

as Atlantic cod, it is seldom that all eggs are fertilised and sperm limitation may be a significant issue (Chapter 5). Thus, females would be expected to benefit directly by mating with males that possess large quantities of sperm. The only possibility for longer-term benefits to females would be if males differ in their genetic quality, which would be reflected in the genotypes provided by their gametes. These kinds of genetic qualities might also be reflected by male condition. This suggests that by assessing characteristics of sound production by males, females might also be able to gather information relating to the ability of the individual to provide 'good genes' and consequent benefits to offspring viability. In my study, as in many studies (e.g. Jones et al. 2000; Veit and Jones 2003), the relationship between display trait expression and condition or fertilisation potential was quite weak, suggesting that sound production may only be a weak reflection of individual quality and that other processes may contribute to mate selection in Atlantic cod.

An important study by Engen and Folstad (1999) also examined whether male Atlantic cod drumming muscles could provide information to females about male quality. They found no association between drumming muscle mass and parasite intensity or leukocyte density when controlled for body mass. However, they did find a negative correlation between spermatocrit (i.e. the percentage of the total volume of milt composed by sperm cells) and residual mass of drumming muscles, after controlling for fish body size, which they inferred to be the result of attractive males having frequent ejaculations. This interpretation by Engen and Folstad (1999) was based on an observation that many batch spawners exhibit a decrease in spermatocrit level with increasing

number of batches spawned, a finding that has subsequently been called into question for Atlantic cod which have been reported to show an increase in spermatocrit during the spawning season (Rakitin et al. 1999b). Given the current uncertainty about what spermatocrit represents, I believe that my estimate of gonad mass might be a more useful indicator of male ability to provide sperm. Overall, my results are reasonably consistent with those of Engen and Folstad (1999), although it is unfortunate that our studies are not more readily comparable.

In addition to its potential use as a short range signal related to agonistic and courtship behaviour (Brawn 1961a, 1961b; Chapter 4), sound production by male Atlantic cod may serve as a long range signal to attract females to the spawning area (Nordeide and Kjellsby 1999). It has been suggested that during the spawning season, mature male Atlantic cod aggregate near the bottom on spawning grounds where they actively defend small mating 'territories' and females only enter male aggregations for a brief period when ready to spawn a batch of eggs (Brawn 1961a; Morgan and Trippel 1996; Nordeide 1998; Hutchings et al. 1999). Nordeide and Kjellsby (1999) estimated that the chorus of sound produced by a large aggregation of Atlantic cod males may be heard by females up to several kilometres away and hypothesized that it may draw them to the spawning area.

The highest catch rates in many commercial fisheries, including those for Atlantic cod, are achieved by mobile fleets that target spawning aggregations (Hutchings 1996) and the biological consequences of such fishing practices has been an issue of recent concern (Hutchings and Myers 1994; Myers et al. 1997).

Aside from the obvious issues related to direct removal of spawning individuals by fishing gear and consequent damage to populations, the growing evidence for the importance of sound production in cod spawning behaviour presents a new concern relating to interference caused by noise produced during fishing activity (from engines, trawl doors, gear contact with substrate, and trawl cables) which overlaps the frequencies of sounds produced by Atlantic cod (Brawn 1961b; Hawkins and Rasmussen 1978; Nordeide and Kjellsby 1999; Midling et al. 2002).

Together with previous studies of sound production by Atlantic cod, my study underscores the potential importance of sound production to cod spawning behaviour, hypothesizing that acoustic displays act as a sexually selected indicator of male size, condition, and fertilisation potential. Experiments should be undertaken to assess the proximate mechanism for seasonal and sex-related variation in Atlantic cod drumming muscles (e.g. the role of blood androgens) and to quantify the relationship between drumming muscle mass and sound characteristics. Further studies of cod sound production in which individual females and males are identifiable are required to elucidate the precise role of acoustic signals in reproductive behaviour. Specifically, we need to determine whether there are temporal patterns of variation in sound production, identify the behavioural contexts in which sounds are produced, and quantify the extent to which cod populations differ in drumming muscle investment and in the characteristics of sounds they produce.

Table 3.1. Summary of Atlantic cod sampled from the Western Scotian Shelf, 2001-2002.

Date	n Females	Males	% spawning Females	Males
21 March 2001	39	41	2.56	2.44
4 May 2001	68	67	0.00	0.00
18 June 2001	34	29	0.00	0.00
27 July 2001	52	52	0.00	0.00
20 September 2001	31	29	0.00	6.90
27 November 2001	69	55	21.74	65.45
17 December 2001	37	49	97.30	100.00
14 January 2002	76	46	11.84	19.57
25 February 2002	92	47	6.52	31.91
Total	498	415	13.45	26.99

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Table 3.2. Regression equations describing drumming muscle mass (y) as a function of somatic mass (x) for Western Scotian Shelf Atlantic cod females and males in non-spawning and spawning condition. Both variables were log-transformed.

	Regression equation	n	R^2	P
Non-spawning females	$y = 1.08x - 4.22$	431	0.74	< 0.0001
Non-spawning males	$y = 1.17x - 4.45$	303	0.73	< 0.0001
Spawning females	$y = 0.91x - 3.56$	67	0.64	< 0.0001
Spawning males	$y = 1.21x - 4.54$	112	0.72	< 0.0001

Table 3.3. Variation of drumming muscles and morphological traits of Western Scotian Shelf Atlantic cod in relation to gender.

			CV					
			Drumming muscle mass (g)		Somatic mass (g)		Body length (cm)	
	n							
Age	Females	Males	Females	Males	Females	Males	Females	Males
2	20	18	0.50	0.63	0.45	0.43	0.14	0.14
3	162	138	0.61	0.61	0.45	0.40	0.14	0.13
4	150	128	0.57	0.66	0.49	0.49	0.17	0.16
5	106	79	0.64	0.75	0.49	0.51	0.17	0.18
6	32	23	0.73	0.62	0.50	0.42	0.17	0.13
7	12	13	0.31	0.55	0.39	0.41	0.11	0.17
8	11	6	0.58	0.58	0.48	0.28	0.19	0.08
9	3	3	0.82	0.64	0.60	0.72	0.23	0.24
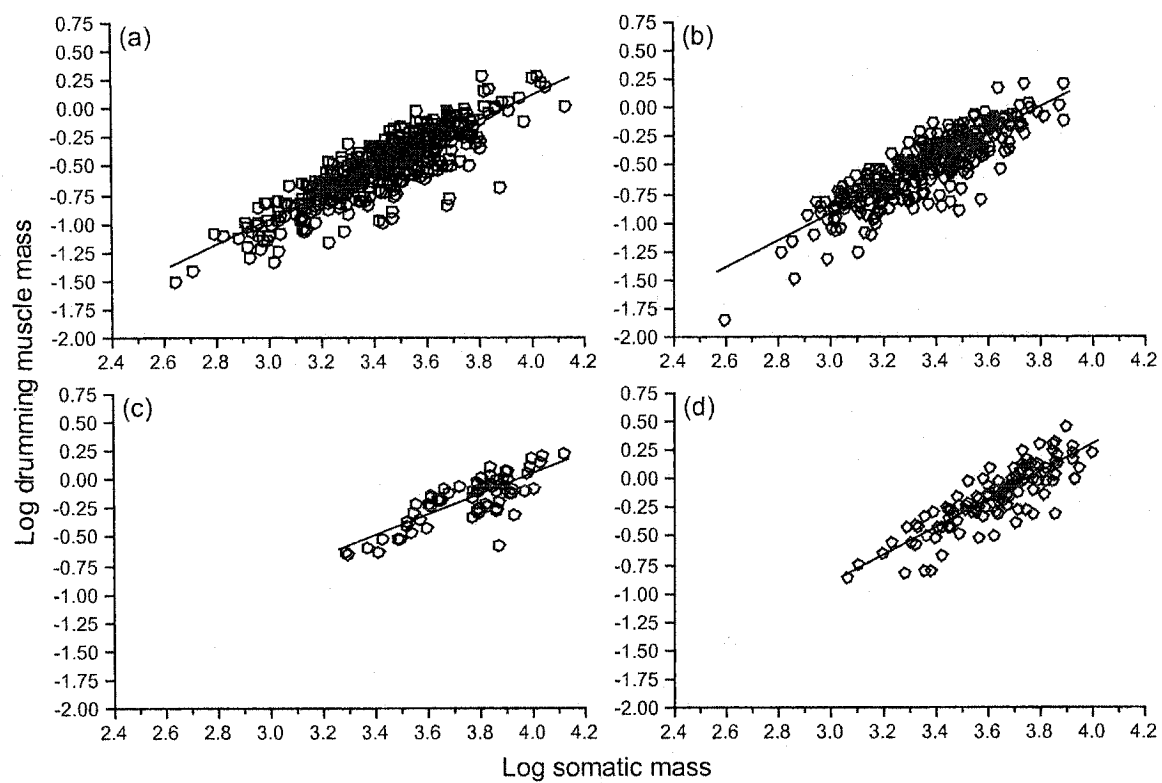
Figure 3.1. Variation in drumming muscle mass in relation to somatic mass for (a) non-spawning female, (b) non-spawning male, (c) spawning female, and (d) spawning male Atlantic cod from the Western Scotian Shelf.
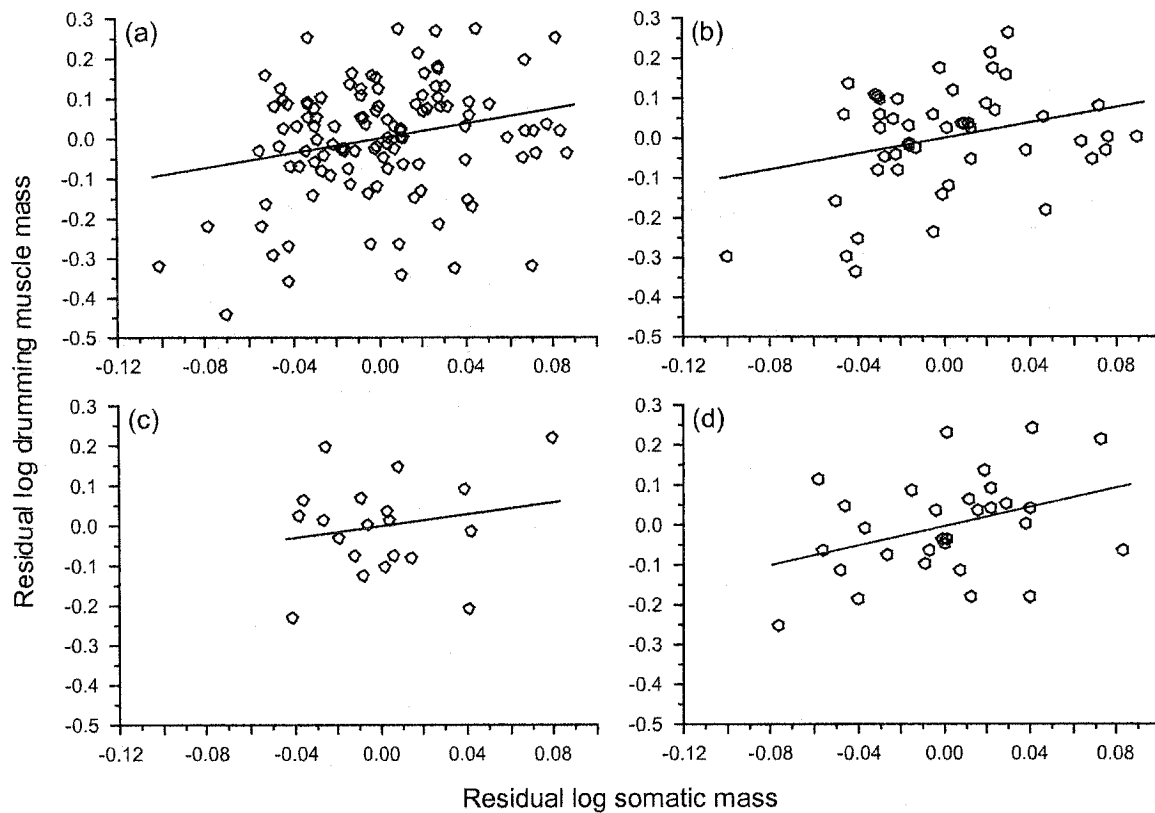
Figure 3.2. Association between residual drumming muscle mass after controlling for somatic mass and residual somatic mass after controlling for length in spawning male Atlantic cod (a) ages 3-9, (b) age 3, (c) age 4, and (d) age 5, from the Western Scotian Shelf.
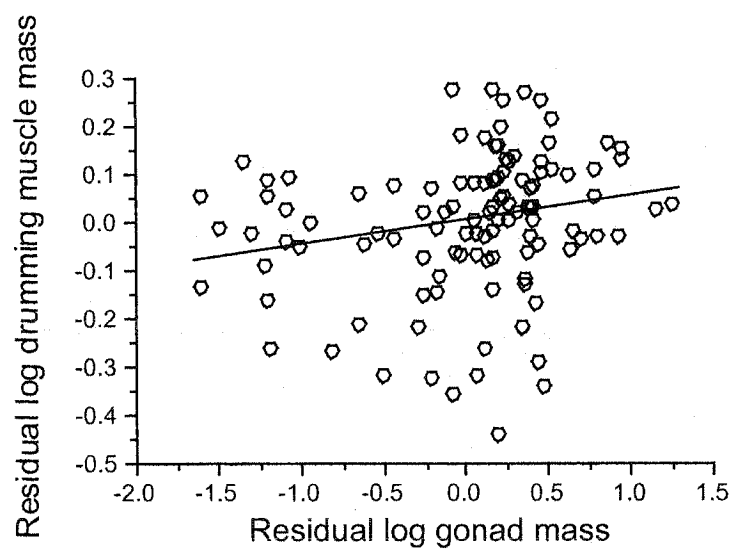
Figure 3.3. Association between residual drumming muscle mass and residual gonad mass after controlling for somatic mass in spawning male Atlantic cod from the Western Scotian Shelf.

CHAPTER 4

SOUND PRODUCTION BY ATLANTIC COD DURING SPAWNING

INTRODUCTION

Many fishes produce sounds to communicate information to other individuals as part of their social behaviour, and one of the most common contexts of sound production is during reproduction (Hawkins 1993). Sounds are typically produced by males, often during agonistic interactions that may ultimately reflect competition among males for access to females. For example, territorial male bicolor damselfish (*Pomacentrus partitus*) produce 'pops' when approached by another (Myrberg 1972), and high amplitude sounds are produced by male croaking gourami (*Trichopsis vittatus*) during agonistic encounters (Ladich et al. 1992). In both cases, sounds are typically accompanied by agonistic behaviours such as chases, lateral displays, and circling, and may induce withdrawal of the opponent.

Reproductive sounds may also serve to advertise presence and reproductive readiness of males to females, and may even arouse reproductive activity in the latter. During the spawning season, male toadfish (*Opsanus tau*) occupy well-defined areas on the seabed and emit 'boatwhistle' sounds that attract gravid females (Gray and Winn 1961). Similarly, male haddock (*Melanogrammus aeglefinus*) occupy territories during the spawning period and produce long trains of repeated 'knocks' that are believed to be attractive to females, drawing them to male territories to engage in courtship and mating (Hawkins et al. 1967; Hawkins and Amorim 2000). When a female haddock approaches a male, and as courtship proceeds, the rate of knock repetition increases so as to appear as a continuous 'hum' in the courtship stages immediately prior to gamete release.

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The progressive increase in rate of sound production as males become increasingly aroused may indicate to females a readiness to mate and help synchronize release of eggs and sperm (Hawkins and Amorim 2000). Sound production by males might also provide a basis for mate choice by females. Female bicolor damselfish use courtship sounds of conspecific males to locate nest sites (Myrberg et al. 1986) and acoustically-mediated individual recognition has been documented for this species (Myrberg and Riggio 1985), thus providing a basis for mate assessment.

Atlantic cod (*Gadus morhua*), a broadcast spawning marine demersal fish found on both sides of the North Atlantic, reproduces in water depths ranging from tens to hundreds of metres (Brander 1994; Morgan et al. 1997; Smedbol and Wroblewski 1997). Like many fishes, cod are capable of producing and detecting sound (Hawkins 1993). Atlantic cod produce sound using three pairs of drumming muscles exterior to the swimbladder wall (Brawn 1961b). Their sounds have been described as short 'grunts' with peak sound amplitudes at frequencies ranging between 50 and 500 Hz, and the acoustic repertoire is believed to be simple, with the same call used in a variety of contexts (Brawn 1961b; Hawkins and Rasmussen 1978; but see Midling et al. 2002).

Recent studies have shown that successful reproduction in Atlantic cod involves complex behaviour within and between sexes and it has been hypothesized that acoustic displays are involved in mate competition (Brawn 1961a, 1961b; Hutchings et al. 1999). Release of sperm and eggs by Atlantic cod involves a 'ventral mount' in which the male, while grasping the female with his pelvic fins and matching her swimming speed, positions himself beneath her

with the urogenital openings of both fish aligned to each other. Before mounting attempts, agonistic interactions (especially chases) are often seen among males and are believed to influence access to females (Brawn 1961a; Hutchings et al. 1999). Successful ventral mounts also appear to be preceded by courtship behaviour, which has been described as an intense 'flaunting' display during which the courting male moves alongside and in front of the female with median fins fully erect while "swimming with an excited, jerky, undulating movement with many unnecessary circles" (Brawn 1961a). Preliminary observation has suggested that Atlantic cod produce sound most frequently during the spawning period and although both sexes are capable of making sounds throughout the year, only males appear to do so during the spawning season, typically during agonistic interactions and courtship display (Brawn 1961b).

Atlantic cod have been harvested for hundreds of years and are now at historic population lows, having decreased by almost 90% since the 1970s in the North Sea (ICES 2002), and by more than 99% off the coast of Newfoundland since the 1960s (Hutchings 2003). Despite being of theoretical interest and practical importance, the spawning behaviour of Atlantic cod is poorly understood; spawning has rarely been observed and earlier descriptions of behaviour and sound production have been brief and anecdotal. Having recently re-examined the behaviour of Atlantic cod in some detail, here I quantify temporal patterns of sound production by cod within large captive spawning groups and relate sounds to patterns of fish behaviour. Specifically, my objectives were to: (i) measure seasonal and diurnal variation in the rate of sound production; and (ii) establish the behavioural contexts in which sounds are

produced. Furthermore, because I had the opportunity to examine groups of cod from two distinct populations, I assessed whether they differed in rate of sound production or drumming muscle investment.

METHODS

To assess patterns of sound production by Atlantic cod, I examined fish from two spatially distinct areas in the Northwest Atlantic: Western Scotian Shelf and Southern Gulf of St. Lawrence, identified by the Northwest Atlantic Fishery Organization (NAFO) as divisions 4X and 4T, respectively. Fish from each population were collected by baited hook two to three weeks prior to their annual spawning season, individually tagged, and placed in a 684 m³ tank at Dalhousie University where spawning occurred. Unfortunately, I was unable to control the size or sex of individuals captured. Cod from the Western Scotian Shelf typically spawn during December-March and their behaviour was observed in two years: 2001 (25 females and 27 males, ranging 42-78 cm in length) and 2002 (25 females and 29 males, ranging 54-105 cm in length). By comparison, cod from the Southern Gulf of St. Lawrence spawn during May-July and they were observed only in 2001 (41 females and 18 males, ranging 50-70 cm in length). Cod were maintained at densities similar to those reported for spawning individuals in the wild (Rose 1993; Morgan et al. 1997), they experienced water temperatures of approximately 8°C, as well as natural photoperiods, and were fed Shurgain® 8 mm fish feed daily. A plankton net covered the tank outflow and was checked daily for the presence of eggs, the volume of which I quantified during 2002 as an indicator of spawning activity.

Fish were sacrificed at the end of the spawning periods so that they could be sexed and measured. I recorded total length of each individual to the nearest millimeter, as well as total mass and gonad mass to the nearest 0.5 g. Somatic mass was calculated as total mass less the gonad mass. In addition, in the 2001 protocol, the three pairs of drumming muscles were extracted by forceps from the surrounding tissue and their combined dry mass was measured to the nearest 0.0001 g for each fish.

Sounds were recorded on Maxell T160 cassettes using a hydrophone (Vemco VHLF-10: built-in preamplifier, sensitivity of –147 dB re 1V for a sound pressure of 1 μPa, and a flat frequency response from 10 Hz to 20 kHz) suspended in midwater at the centre of the tank and a Sony SVO-1630 videotape recorder. For Western Scotian Shelf and Southern Gulf of St. Lawrence fish in 2001, sounds were recorded continuously from approximately two to three weeks after the start of spawning until the time when no eggs had been collected for five days. For Western Scotian Shelf fish in 2002, I only recorded sounds during the last eight hours of daylight each day (08:00-16:00) but started to monitor sound production approximately two weeks before spawning began. In addition, behaviour was documented daily during the last eight hours of daylight for fish from Western Scotian Shelf in 2002 (08:00-16:00) and Southern Gulf of St. Lawrence in 2001 (12:00-20:00) by four video cameras mounted above the tank and the videotape recorder that also recorded audio information. Unfortunately, this camera array was not available for data collection during 2001 for Western Scotian Shelf fish.

More than two hours was required to extract information on sound production from each hour of videotape. Therefore, I examined only a subset of the recordings to investigate patterns of sound production. In order to examine seasonal variation in sound production by fish from the Western Scotian Shelf in 2002, I reviewed two hours (11:00-12:00 and 15:00-16:00) of audio recordings approximately every fourth day during the spawning season and quantified the number of grunts heard. This pattern of sub-sampling provided rather limited coverage on any particular day but increased the number of days examined throughout the spawning period. To examine diurnal variation in sound production by Western Scotian Shelf and Southern Gulf of St. Lawrence fish in 2001, I reviewed six hours (03:00-04:00, 07:00-08:00, 11:00-12:00, 15:00-16:00, 19:00-20:00, and 23:00-24:00) of audio recordings approximately every fourth day (starting two to three weeks after the start of spawning and continuing until no eggs had been collected for five days) and quantified the number of grunts heard. Again, this pattern of sub-sampling allowed me to examine numerous days spread across the spawning season but also provided detailed daily coverage. Using the total number of grunts heard on each day of observation during 2001, I compared the rate of sound production between groups from Western Scotian Shelf and Southern Gulf of St. Lawrence, controlling for the number of males in the tank (most sound production during the spawning season is supposed to be by males (Brawn 1961b)). An ANCOVA with somatic mass (a metric of body size) as the covariate was used to test for differences in drumming muscle mass in relation to population origin and sex. Initial models included all interaction terms although non-significant interaction terms were

removed from subsequent models via model simplification (Crawley 2002). Both drumming muscle and somatic mass were log-transformed to meet the assumptions of normality.

I examined videos recorded during the last 30 minutes of daylight every fifth day during the spawning season and documented any behavioural interactions visible in the group when sounds were produced. I focused my observation effort on the time close to dusk because cod activity levels appeared slightly higher during this period than other daylight hours. Nonetheless, behavioural interactions and sound production were infrequent events and the chance of them occurring simultaneously solely by chance was very small. Interactions considered included five agonistic behaviours: (i) chase (swimming toward a swimming fish), (ii) approach (swimming toward a stationary fish), (iii) bite (physical contact between one fish's mouth and another fish's body or fin), (iv) nip (an attempted bite, i.e., an opening and closing of the mouth near another fish's body or fin), and (v) prod (contact between one fish's snout and another fish's body). I also considered courtship behaviours (flaunting and ventral mounts), as well as spawning events (as indicated by the appearance of a cloud of milt in the water around a pair of fish engaged in a ventral mount). As part of a related study, all video recordings of Southern Gulf of St. Lawrence fish in 2001 were reviewed to document the incidence of ventral mounts and spawning events in the group. In this case, I noted the identities of the fish involved (whenever possible) and whether the behaviour was accompanied by sound production.

RESULTS

Although I recorded sounds beginning approximately two weeks after the onset of egg production and did not quantify the volume of eggs collected during 2001, qualitative observations suggested that sound production occurred most frequently during the peak of the spawning season. This hypothesis was supported when I monitored sounds starting approximately two weeks before spawning began and quantified the volume of eggs collected daily for Western Scotian Shelf cod during 2002 (Figure 4.1). During the spawning season, there was considerable variation in the occurrence of sound production in relation to time of day. For both populations, sound was produced at all times of day but was most frequent shortly after sunset, continuing at high levels throughout the night, and tapering off during daylight hours (Figure 4.2).

There was a dramatic difference in the number of sounds produced by groups of cod from the Southern Gulf of St. Lawrence and the Western Scotian Shelf (Figure 4.2). Differences between the two groups in the number of sounds produced per hour persisted after controlling for number of males present (Southern Gulf of St. Lawrence: 0.05 ± 0.01 SE grunts/hour/male; Western Scotian Shelf: 0.42 ± 0.11 SE grunts/hour/male; ANOVA: $F_{1,25} = 11.75$, $P = 0.002$). Data available for Western Scotian Shelf fish in 2002 showed similar high rates of sound production to those observed for Western Scotian Shelf fish in 2001 during sessions recorded both 11:00-12:00 (2001: 0.18 ± 0.07 SE grunts/hour/male; 2002: 0.23 ± 0.06 SE grunts/hour/male; ANOVA: $F_{1,31} = 0.35$, $P = 0.56$) and 15:00-16:00 (2001: 0.34 ± 0.15 SE grunts/hour/male; 2002: 0.67 ± 0.31 SE grunts/hour/male; ANOVA: $F_{1,31} = 0.95$, $P = 0.34$). Differences in the rate of sound production by the

groups of cod representing different populations corresponded to differences in drumming muscle mass; Western Scotian Shelf fish had larger drumming muscles than Southern Gulf of St. Lawrence fish (Figure 4.3; Table 4.1). Using ANCOVA with somatic mass as the covariate to test for differences in drumming muscle mass in relation to population origin and sex, I found a significant interaction between population origin and somatic mass (Two Factor ANCOVA: $F_{1, 106} = 4.64$, $P = 0.03$). An additional ANCOVA involving only individuals from Western Scotian Shelf revealed a significant association between drumming muscle mass and somatic mass (One Factor ANCOVA: $F_{1, 49} = 280.95$, $P = 0.0001$), as well as a significant difference in drumming muscle mass in relation to sex (One Factor ANCOVA: $F_{1, 49} = 9.66$, $P = 0.003$). Similarly, involving only individuals from Southern Gulf of St. Lawrence, I observed a significant association between drumming muscle mass and somatic mass (One Factor ANCOVA: $F_{1, 56} = 94.20$, $P = 0.0001$), although there was no significant difference in drumming muscle mass in relation to sex (One Factor ANCOVA: $F_{1, 56} = 3.20$, $P = 0.08$).

Sound production in cod was associated with both agonistic interactions and courtship behaviour. For example, among 425 grunts recorded during the last 30 minutes of daylight every fifth day during the spawning seasons, 26 (6.1%) were concurrent with an agonistic display (including 23 accompanying chases and 3 accompanying approaches). A total of 139 (32.7% of total) grunts were concurrent with courtship display (male flaunting, 132) and mounting attempts (7). No discernable behavioural interactions were noted for the remaining 260 grunts. In addition, I found no evidence for a difference in the behavioural

context of sound production between populations (Western Scotian Shelf: 5.8% agonistic, 33.0% courtship, 61.2% no behaviour, $n = 379$; Southern Gulf of St. Lawrence: 8.7% agonistic, 30.4% courtship, 60.9% no behaviour, $n = 46$; $G_2 = 0.59$, $P = 0.75$).

Among 106 ventral mounts involving Southern Gulf of St. Lawrence fish, 104 (98.1%) were initiated by males and in the remaining two cases (1.9%), the initiator could not be identified. These 106 ventral mounts included 91 (85.8%) with no accompanying grunt(s) and 15 (14.2%) with grunt(s). Interestingly, among the 88 ventral mounts where the recipient could be identified, 33 (37.5%) involved a male recipient and 55 (62.5%) involved a female recipient. However, I found no evidence for a difference in the occurrence of sound production during ventral mounts in relation to sex of the recipient (female recipient: 14.5% involved sound production, 85.5% did not involve sound production, $n = 55$; males: 15.2% involved sound production, 84.8% did not involve sound production, $n = 33$; $G_1 = 0.01$, $P = 0.94$). Nevertheless, sound production was more likely to occur during ventral mounts that were associated with spawning (mounts with sound: 37.5% were associated with spawning, 62.5% were not associated with spawning, $n = 8$; mounts without sound: 6.4% were associated with spawning, 93.6% were not associated with spawning, $n = 47$; Fisher's Exact test, $P = 0.03$). In three of the six spawning events, no sounds were heard after the start of gamete release. In all cases where sounds were heard after the start of gamete release, intense male-male competition for access to the spawning female was apparent in the form of four or more males moving rapidly into the gamete cloud, sometimes displacing the male involved in the ventral mount. For

instance, on one occasion, the male that initiated courtship and spawning was displaced from the female early in the spawning event, whereafter the spawning female was mounted by a second male. At this moment, two grunts were heard while the original male counterattacked the second male. I believe the original male produced these grunts during his counterattack; immediately after emission of the second grunt, the second male abandoned the female, as did other satellite males when a third grunt was emitted.

Almost all sounds produced by cod in my study matched the short grunt type sounds described by other investigators (Brawn 1961b; Hawkins and Rasmussen 1978; Midling et al. 2002). However, on two occasions, I heard a sound that seemed like a very low-pitched grunt lasting several seconds and that may be best described as a 'growl'. In both cases, the growl was emitted concurrent with a ventral mount, in the final moments immediately preceding gamete release.

DISCUSSION

The first investigation of sound production by cod was provided by Brawn (1961b). This important study described deep grunting sounds that were produced most frequently during the spawning season, typically in agonistic interactions and courtship display by males. However, this description was based on sporadic observations at variable times of day and on an examination of a small group of captive individuals that varied in number and sex composition, thus limiting the conclusions that could be drawn. To date, there has been no definitive study subsequent to Brawn's (1961b) work to elucidate patterns of

sound production by Atlantic cod or to determine whether they differ among populations.

I examined sound production in large groups of spawning cod from two Northwest Atlantic populations, focusing in particular on the behavioural contexts in which sounds were produced. Although sound production studies on large groups of individuals can be challenging, I found it useful to combine the recording of fish sounds with videotapes of fish behaviour so that they could be observed repeatedly. Indoor concrete aquarium tanks, such as the one used in this study, are not ideal for recording fish sounds; the tank walls form reflecting boundaries that distort sounds produced and aquarium machinery can result in persistent low levels of background noise. Consequently, I was unable to make detailed measurements of individual sound characteristics of a nature similar to those undertaken on cod held in netting enclosures at sea (Hawkins and Rasmussen 1978; Midling et al. 2002). Nonetheless, sounds produced by fish in my study were audible (even against background noise caused by machinery) and there did not appear to be discernable differences in the characteristics of grunts produced between my two populations of cod and those documented by other investigators (although very few 'clean' waveforms could be obtained in my study; Brawn 1961b; Hawkins and Rasmussen 1978; Midling et al. 2002).

Brawn's (1961b) description of sound production by spawning cod was based primarily on observations of seven females and six males held in captivity. She reported being able to identify the sound-producing individual within the group by observing sudden movements of the body that often accompanied sound production, particularly bending of the head and tail ventrally on the trunk.

Using this technique, she concluded that only males seemed to produce sound during the spawning period, and that sounds were produced during agonistic encounters with other males and unripe females and during the flaunting display characteristic of courtship of ripe females. Unfortunately, I did not observe any such external body movements that would allow me to identify the sound producer in the tank (perhaps because only an overhead view of the tank was available with the camera set-up). However, consistent with Brawn's (1961b) observations, I found that sounds were associated with agonistic interactions among males and courtship although sound production seemed to occur more often during courtship (as opposed to during agonistic encounters as reported by Brawn (1961b)) for both of the populations that I examined. Curiously, no behavioural interactions were apparent during most instances of sound production and it is difficult to ascertain the function of calls in these situations. Brawn (1961b) did not report sound production during the ventral mount (except when a male was mistakenly mounted and he was interpreted to have then grunted to break up the pairing). In contrast, I found that 14.2% of ventral mounts were accompanied by sounds and I found no evidence for a difference in the occurrence of sound production during ventral mounts in relation to sex of the recipient. Although Brawn's (1961b) observations were irregularly distributed throughout time, they are consistent with my work indicating that most sound production occurs during the spawning period, particularly after sunset. Increased use of acoustic signals during hours of darkness may not be surprising given the reduced effectiveness of visual signals at this time (Anthony 1981). The behavioural context of sound production during hours of darkness

remains unknown.

There has been much speculation about the function of sound production by Atlantic cod (Brawn 1961b; Engen and Folstad 1999; Hutchings et al. 1999; Nordeide and Kjellsby 1999; Nordeide and Folstad 2000). Both sexes are capable of producing sounds throughout the year and often do so during agonistic behaviour, causing threatened individuals to flee (Brawn 1961b). Sound production by males becomes especially frequent during the spawning period, at which time agonistic behaviour accompanied by sound production may indicate competition among males for space, perhaps even particular areas on the seabed, or for females themselves. Brawn (1961b) reported that approximately three weeks before spawning, males became aggressive and using grunting sounds to reinforce threat displays, the most aggressive male was able to establish the largest 'territory' in which all observed spawning occurred. Its prevalence during male flaunting also suggests that sound might be used in mate assessment by females.

Acoustic communication may be an important criterion by which females discriminate among males from different cod populations. The spawning periods of migratory and stationary populations of cod off Norway overlap and reproducing individuals from both populations mingle spatially at the same spawning grounds (Nordeide 1998). In addition, artificial crossings of eggs and sperm from the two populations have been successfully conducted in the laboratory (Godø and Moksness 1987). Nonetheless, migratory and stationary individuals do not seem to interbreed in the wild and female assessment of potential population differences in male acoustic displays has been suggested as

a premating mechanism enabling this separation (Nordeide and Folstad 2000). Tests should be conducted to assess whether cod from these populations differ in the characteristics of sounds they produce and if so, whether females exhibit preferences for calls produced by males originating from their own natal population.

Acoustic displays may also allow females to discriminate among males for assessment purposes at an individual level (Myrberg et al. 1986; Andersson 1994). Greater investment by females than males in each zygote means that females have more to lose in an unsuccessful reproductive event, a cost that should make them more cautious in their choice of mate (Clutton-Brock and Vincent 1991; Andersson 1994). In Atlantic cod, females receive nothing from males but gametes; there is no parental care and males do not occupy an area with resources necessary for female survival or reproduction. Therefore, should direct benefits result from mate choice, they would most likely come in the form of high-quality ejaculates that assure fertilisation of eggs. Indirect benefits may also be realized if females can obtain information about heritable qualities of mates. Accordingly, patterns of variation in Atlantic cod drumming muscle mass are consistent with the hypothesis that acoustic displays represent a sexually selected indicator of male size, condition, and fertilisation potential (Chapter 3). In the present study, the importance of sound production to male mating and reproductive success might be reflected by the fact that ventral mounts accompanied by sound were more likely to lead to successful spawning than those that were not accompanied by sound. The occurrence of sound production also seemed to increase as courtship behaviour by males intensified and a sound

best described as a growl was sometimes heard immediately prior to gamete release. Assuming that these sounds were produced by the courting male, it is possible that the increase in rate of sound production as the male becomes increasingly aroused provides an indication to the female of male readiness to spawn, resulting in a synchronization of egg and sperm release, thus achieving high fertilisation success.

It has been suggested that during the spawning season, mature male Atlantic cod aggregate near the bottom on spawning grounds where they actively defend small mating territories and females are typically found peripherally to male aggregations (Brawn 1961a; Morgan and Trippel 1996; Nordeide 1998; Hutchings et al. 1999). When ready to spawn a batch of eggs, females enter male aggregations, returning to peripheral areas after spawning (Brawn 1961a; Hutchings et al. 1999). In addition to its apparent use as a short range signal related to agonistic and courtship behaviour, it is not unreasonable to assume that the chorus of sound produced by large aggregations of Atlantic cod males on spawning grounds may serve as a long range signal to attract females to the spawning area (Nordeide and Kjellsby 1999).

My study is the first to examine sound production in more than one cod population. For fish from the Western Scotian Shelf and the Southern Gulf of St. Lawrence, I found no evidence for a difference in the temporal patterns, nor behavioural contexts of sound production. Nonetheless, the rate of sound production was much higher for Western Scotian Shelf cod compared to those from the Southern Gulf of St. Lawrence. Furthermore, the greater sound production was reflected by differences in the mass of drumming muscles

relative to body size. Unfortunately, I am unable to identify the specific factors responsible for group differences in rate of sound production and drumming muscle investment. Most sound production occurs at night and fish from the Western Scotian Shelf might have been expected to produce more sound than those from Southern Gulf of St. Lawrence solely because they experienced longer periods of darkness by spawning in winter. However, a less than two-fold difference in the period of darkness seems an improbable explanation for the more than eight-fold difference observed in rate of sound production per male between groups. Similarly, although differences in sex ratio between my experimental groups might have influenced male mating strategies and display rates, these are unlikely to explain related differences in drumming muscle mass. While it is possible that the observed differences in rate of sound production and drumming muscle investment between cod from the Western Scotian Shelf and the Southern Gulf of St. Lawrence were solely a result of chance, they may also reflect inherent differences between these cod populations.

The highest catch rates in many commercial fisheries, including those for Atlantic cod, are achieved by mobile fleets that target spawning aggregations (Hutchings 1996) and the biological consequences of such fishing practices have been of recent concern (Hutchings and Myers 1994; Myers et al. 1997). Aside from the obvious issues related to direct removal of spawning individuals by fishing gear and consequent damage to populations, the growing evidence for the importance of sound production in cod spawning behaviour presents a new concern relating to interference caused by noise produced during fishing activity (e.g. from engines, trawl doors, gear contact with substrate, and trawl cables)

which overlaps the frequencies of sounds produced by Atlantic cod (Brawn 1961b; Hawkins and Rasmussen 1978; Nordeide and Kjellsby 1999; Midling et al. 2002). Disruption of acoustic signaling associated with mating might prolong intervals between egg batch releases because of increased time required by males to aggressively compete for females and increased time used by females to evaluate male quality (Hutchings et al. 1999). For batch-spawning fishes such as Atlantic cod, delays in the release of eggs after ovulation of just a few hours can dramatically reduce egg viability (Kjørsvik and Lønning 1983; Kjørsvik et al. 1990). Unfortunately, at present, there are no data to directly assess the impact of anthropogenic noise on spawning behaviour and reproductive success of Atlantic cod.

Together with previous work, my study underlines the importance of sound production to Atlantic cod spawning behaviour. Further studies involving playback experiments are required to elucidate the precise role of acoustic signals in reproductive behaviour. In addition, research should be undertaken to better understand the causes and consequences of variation in rates of sound production and drumming muscle investment at both the individual and population levels.

Table 4.1. Regression equations describing drumming muscle mass (y) as a function of somatic mass (x) for female and male Atlantic cod from the Southern Gulf of St. Lawrence and Western Scotian Shelf. Both variables were log-transformed.

	Regression equation	n	R^2	P
Southern Gulf of St. Lawrence				
Females	$y = 1.76x - 6.54$	41	0.58	< 0.0001
Males	$y = 2.16x - 7.68$	18	0.74	< 0.0001
Western Scotian Shelf				
Females	$y = 1.43x - 5.31$	25	0.88	< 0.0001
Males	$y = 1.48x - 5.36$	27	0.82	< 0.0001
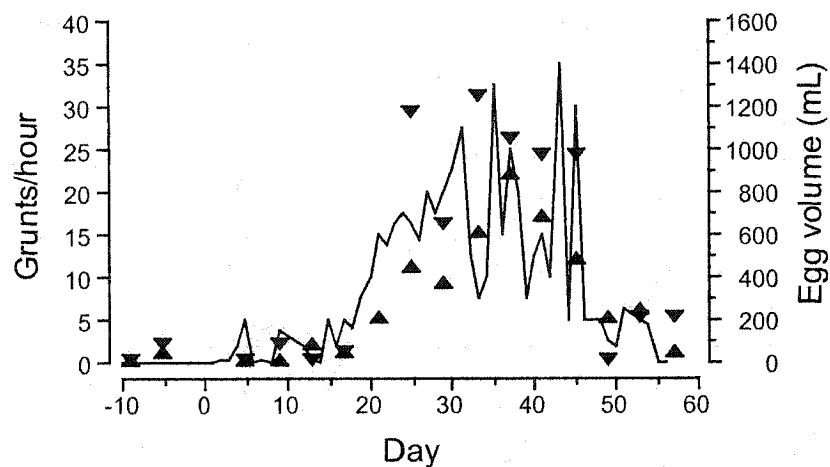
Figure 4.1. Variation in sound and egg production by 25 female and 29 male Atlantic cod from the Western Scotian Shelf during the 2002 spawning period. Day 1 represents the first day that fish spawned. Upward triangles, number of grunts recorded 11:00-12:00; downward triangles, number of grunts recorded 15:00-16:00; line, volume of eggs collected. On Day 21, 146 grunts were recorded between 15:00-16:00 but this point was excluded from the graph.
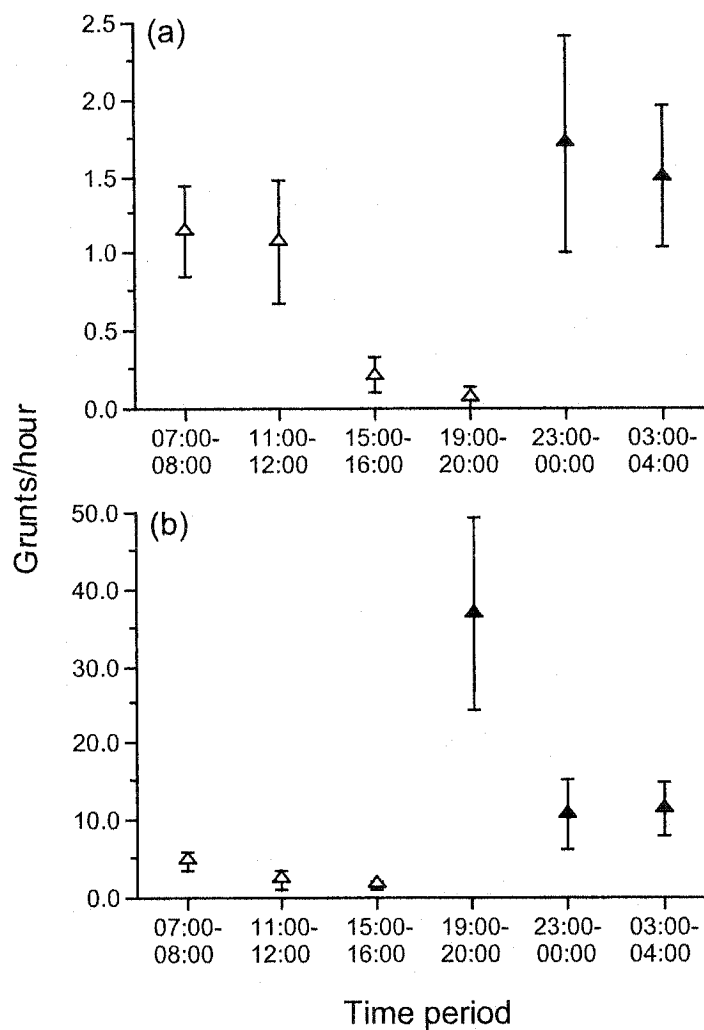
Figure 4.2. Variation in sound production in relation to time of day for Atlantic cod from (a) Southern Gulf of St. Lawrence and (b) Western Scotian Shelf. Mean ± 1 SE shown. Southern Gulf of St. Lawrence: 14 days sampled; Western Scotian Shelf: 13 days sampled. Open triangles, recorded during daylight; closed triangles, recorded during darkness.
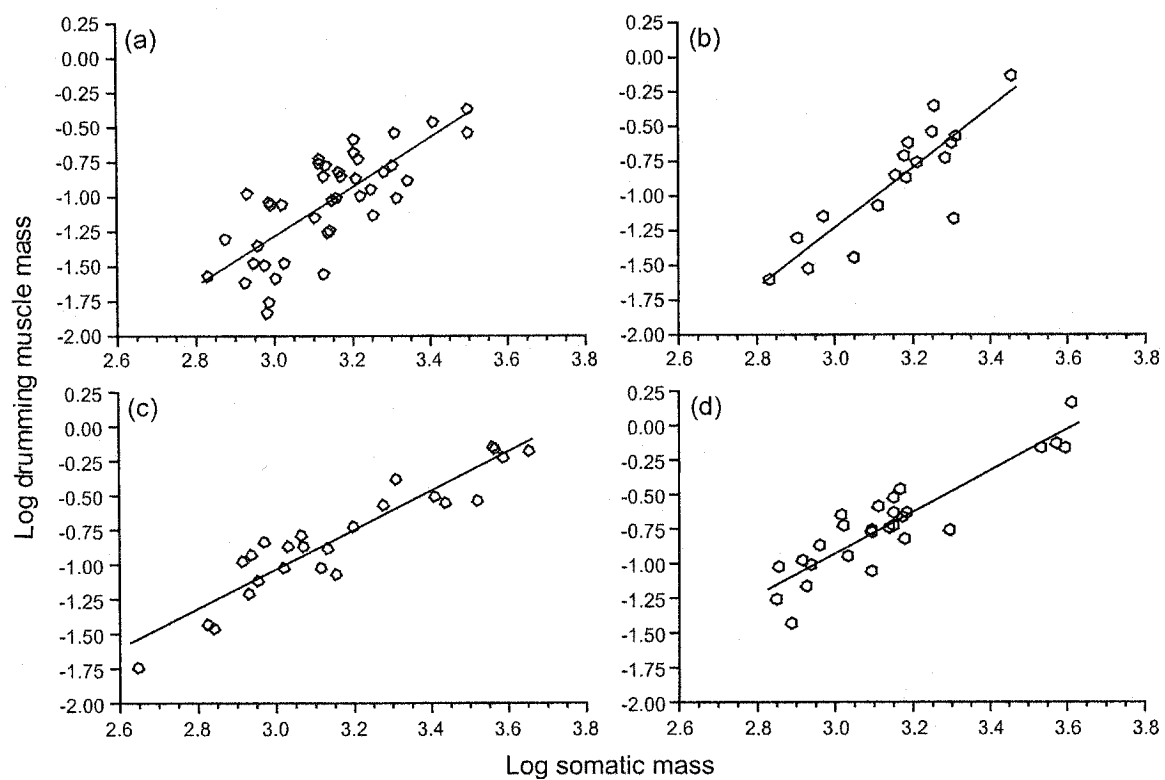
Figure 4.3. Variation in drumming muscle mass in relation to somatic mass for
(a) Southern Gulf of St. Lawrence female, (b) Southern Gulf of St. Lawrence male,
(c) Western Scotian Shelf female, and (d) Western Scotian Shelf male Atlantic
cod.

CHAPTER 5

DEPENSATION, PROBABILITY OF FERTILISATION, AND THE MATING SYSTEM OF ATLANTIC COD

INTRODUCTION

Severe over-exploitation has driven Atlantic cod (*Gadus morhua*) to historic low levels of abundance (Hutchings and Myers 1994; Myers et al. 1997; Smedbol et al. 2002; Hutchings 2003). In the North Sea and adjacent waters, cod have declined 90% since the early 1970s (ICES 2002), a loss of more than 200 million individuals. Even more dramatic is the collapse experienced by Newfoundland's northern cod which is estimated to have declined 99% between 1962 and 1992, a loss of approximately 1.5 billion breeding individuals (Hutchings 2003). Once supporting the largest cod fishery in the world (McGrath 1911), commercial fishing of northern cod was closed in 1992 for the first time in its 500-year history. The stock has since shown no signs of recovery (DFO 2003). Slow or negligible rates of recovery among cod populations have been attributed to a reduction in population growth rate concomitant with reduced population size (Shelton and Healey 1999; Frank and Brickman 2000), an association termed depensation in the fisheries literature and Allee Effect, or positive density dependence, in the ecological literature. Despite increased attention to depensatory mechanisms as factors responsible for slow recovery rates in Atlantic cod, a dearth of empirical work stands as a significant barrier to assessing the potential for Allee Effects in this species.

The Atlantic cod is a bet-hedging species, necessitated by a reproductive strategy in which eggs are released (broadcasted) directly into the oceanic environment and for which no parental care is provided. Attaining maturity

across a broad range of sizes (35-85 cm) and ages (2-7 years) throughout the species range (Brander 1994; Hutchings 2003), females release their annual egg complement numbering hundreds of thousands, often millions, of 1.25-1.75 mm eggs in batches over 3-6 weeks (Chambers and Waiwood 1996; Kjesbu et al. 1996) within a 6-12 week spawning season (Myers et al. 1993) in water ranging from tens to hundreds of metres in depth (Brander 1994; Morgan et al. 1997; Smedbol and Wroblewski 1997). Successful reproduction appears to involve complex mating interactions, including behavioural and acoustic displays by males and mate choice by females (Brawn 1961a; Hutchings et al. 1999). Release of sperm and eggs by Atlantic cod occurs during a 'ventral mount' in which the male, while grasping the female with his pelvic fins and matching her swimming speed, positions himself beneath the female. Upon extrusion of the female's egg batch, the spawning couple is often joined by nearby satellite males who swim amongst the eggs and also release sperm. Accordingly, genetic work indicates that eggs from single reproductive bouts can be fertilised by more than one male (Hutchings et al. 1999; Rakitin et al. 2001; Bekkevold et al. 2002). It is, thus, reasonable to assume that alternative mating tactics exist in male Atlantic cod.

When population sizes were large, Atlantic cod were reported to spawn over vast areas of the continental shelf and slope (Hutchings et al. 1993) in aggregations spanning several kilometres. Densities within spawning aggregations have been reported to range from an average of 0.004 fish/m³ (Morgan et al. 1997) to a maximum of about 3 fish/m³ (George Rose, Memorial University, St. John's, Canada, personal communication). It is not improbable that there have been changes in density within spawning aggregations

concomitant with documented changes in population size (ICES 2002; Hutchings 2003), but there are no data available to assess either this issue or the question of whether the precise locations of spawning grounds have shifted over time (detailed spawning maps do not exist for Atlantic cod, past or present). Analyses by Myers and Cadigan (1995) and Hutchings (1996) suggest that as population numbers decreased, population density may have increased. However, these apparent increases in density are based on research survey data aggregated at spatial scales considerably larger than those at which spawning takes place. Although many cod populations are only small fractions of their former sizes, they still contain millions of breeding individuals (ICES 2002; Hutchings 2003), and it might be argued that if reductions in density of individuals in spawning aggregations have also occurred, these changes may be more important in determining cod reproductive success in the wild than absolute abundance.

If populations fall below some critical size or density threshold, individuals may experience problems that negatively affect mating success and/or offspring production (Møller and Legendre 2001). At low population density, individuals may experience delays in seasonal reproduction as more time might be required to find a mate. For batch spawning fishes such as Atlantic cod, delays in the release of eggs after ovulation of just a few hours can result in over-ripening of gametes and dramatically reduce viability (Kjørsvik and Lønning 1983; Kjørsvik et al. 1990). Scarcity of potential mates also lessens the potential for mate choice, negatively affecting reproductive success (Jennions and Petrie 1997). At the very least, size complementarity between spawning partners may be important for Atlantic cod to ensure the urogenital openings of both fish are aligned opposite

one another. If population size and/or density are reduced to low levels (low density potentially resulting in prohibitively large increases in search time), there may be less scope or breadth of choice and individuals may resort to mating with partners of nonpreferred phenotypes or genotypes. In Atlantic cod, pairings involving size-mismatched mates may negatively affect fertilisation rates (Hutchings et al. 1999; Rakitin et al. 2001). As population density declines, there may be fewer individuals in the vicinity of a spawning couple and thus, the number of satellite males per spawning female may also decline, resulting in reduced sperm concentration (Marconato et al. 1997). This would provide another mechanism by which egg fertilisation rates might decrease with declining adult density. Multiple paternity may also be important as a means for females to maximize their chance of producing viable offspring (Zeh and Zeh 1996, 1997) and increased genetic variation can also improve the ability of a population to respond to future selection pressures.

Despite being of theoretical interest and practical importance, many aspects of the spawning behaviour of Atlantic cod are poorly understood and there is no information regarding the way in which mating or reproductive success may be affected by changes in adult numbers or density (Nordeide and Folstad 2000). Using data obtained from 21 laboratory populations, collected under the auspices of 3 separate research programmes (1 by S. Rowe and J.A. Hutchings, 1 by D. Bekkevold (Bekkevold et al. 2002), 1 by A. Rakitin (Rakitin et al. 2001); Table 5.1), I explored the empirical basis of two potential causes of depensation in Atlantic cod. Firstly, I tested the hypothesis that fertilisation rate declines with abundance. Reduced fertilisation success at lower population sizes, leading

potentially to reduced per capita recruitment, could produce an Allee effect. Secondly, I tested the hypothesis that variance in fertilisation rate increases as abundance declines. Increased variance in individual reproductive success would reduce effective population size (N_e), increasing the rate of loss of genetic variation at low levels of abundance.

While density of spawning individuals may be a more important factor influencing cod fertilisation success than absolute number of individuals present in the wild, the opposite may be true within the spatial confines of a tank. In captive situations, the maximum distance between individuals and thus, the potential search time required to find a suitable mate, is very small relative to that which might occur in nature and consequently, the absolute number of mates available from which to choose may be considered more critical. Therefore, for the purpose of this study, I focused my attention on the influence of male abundance (as opposed to male density) on egg fertilisation success.

METHODS

The experimental protocols associated with 2 of the 3 aforementioned research programmes are described in detail elsewhere (Rakitin et al. 2001; Bekkevold et al. 2002) and will not be repeated here. I will, however, describe the methods employed by S. Rowe and J.A. Hutchings to examine the relationship between abundance and fertilisation rate.

Atlantic cod were captured by baited hook approximately two to three weeks before their annual spawning periods in 2001 and 2002 and transported to Dalhousie University, where the fish were allowed to spawn undisturbed. Upon

arrival at Dalhousie in late April 2001, cod collected from the Southern Gulf of St. Lawrence were assigned either to the 684 m³ Pool Tank or to one of five 2.9 m³ tanks where they would remain for the duration of the study (Table 5.1). In 2002, cod were collected from both the Southern Gulf of St. Lawrence (late April/early May) and the Western Scotian Shelf (January) and placed in the Pool Tank during their temporally distinct spawning periods (Table 5.1). In all cases, cod experienced water temperatures of approximately 8°C, as well as natural photoperiods, and fed Shurgain® 8 mm fish feed daily.

To obtain fertilisation data, each tank was outfitted with an egg collector. In the Pool Tank, eggs were collected by a plankton net covering the single tank outflow located at the surface. In the 5 smaller tanks, I employed an air-lift assisted egg collector located at the surface. All egg collectors were checked at approximately 16:00 every day and any eggs present were extracted. A random sample of at least 100 eggs from each daily collection was examined under a microscope to estimate the proportion of eggs fertilised (fertilisation rate) and to confirm that all eggs had been spawned within the previous 24 hours, based on descriptions by Hardy (1978), Davenport et al. (1981), and Kjørsvik and Lønning (1983). Experimental animals were sacrificed when no spawning had occurred in the tank for at least 5 days. Each fish was then measured and its sex and stage of maturity determined by observation of gross morphology of the gonad (Morrison 1990) to verify that each individual was reproductively mature.

For each experimental population (Table 5.1), I report the median fertilisation rate for the entire spawning period based upon measurements of fertilisation rate in daily egg collections. To control for small differences among research

programmes in the timing of initial fertilisation measurements, I report fertilisation data for those days between the first day of the second week of spawning to the end of each spawning experiment. I initially use data from all research programmes to examine median seasonal fertilisation rate in each experimental population in relation to the number of reproductive males present. Unfortunately, this relationship may be confounded by differences within or among research programmes. To address this issue, I proceeded to examine a subset of the data collected by Rakitin et al. (2001) and Bekkevold et al. (2002) in which, based on parentage analyses of daily egg collections, it was believed that only a single female and a known number of males had participated in spawning on a particular day. In these experiments, eggs were collected daily and a random sample was incubated until around the time of hatching, at which point it was preserved, along with tissue samples from parental fish, for parentage analysis. Only daily egg batches in which a single female had spawned and for which the number of males contributing sperm could be reliably determined are included in this component of my study. Therefore, I was able to reduce the variation in spawning density and experimental tank size (Table 5.1) and, based on genetic information, control for number of spawning females.

RESULTS

Pooling all data from the three experiments, the proportion of eggs fertilised increased with total number of males (Figure 5.1). In addition to a positive association between proportion of eggs fertilised and number of males, the pooled data suggest that the among-brood variance in fertilisation rate might

decline with increased male abundance.

Similar associations between fertilisation rate and male abundance appear to be evident after controlling for experimental tank size and for number of females to have contributed to daily egg batches collected. Based on the experiments conducted by Bekkevold et al. (2002), as the number of males known to spawn with a single female in a spawning event increased from 1 to 4, fertilisation rate increased concomitantly (Figure 5.2). The data fit an asymptotic relationship with egg fertilisation rate increasing rapidly with male number before levelling off at approximately 97%. The fitted equation, being forced through the origin, was fertilisation rate = $0.97*(1-e^{-2.02*\text{number of males}})$. The convex nature of the relationship, which is of primary interest in the present study, remains if the fitted relationship is not forced through the origin. Data from Rakitin et al. (2001), in which either 1 or 2 males spawned with a single female in a spawning event, are also indicative of an increase in fertilisation rate with increasing male abundance in an asymptotic manner (Figure 5.3; fertilisation rate = $0.96*(1-e^{-2.54*\text{number of males}})$).

The increased among-brood variance in fertilisation rate with declining male abundance suggested by the pooled data (Figure 5.1) was also evident from the daily egg batches for which only a single female and a known number of spawning males contributed (Figures 5.2 and 5.3). Comparing 3 metrics of variability (variance, coefficient of variation, inter-quartile range), the variability in fertilisation rate among batches fertilised by a single male appeared to be consistently greater than the variability among batches fertilised by 2 males (Table 5.2). However, variance ratio tests involving the arcsine-transformed data

revealed that this difference was statistically significant only for the data provided by Bekkevold (Bekkevold et al. (2002): $F_{6,6} = 5.30$, $P = 0.03$; Rakitin et al. (2001): $F_{11,28} = 1.33$, $P = 0.26$).

DISCUSSION

Despite a pressing need to evaluate the potential for Allee Effects to affect population growth in Atlantic cod, an empirical vacuum faces those trying to comprehend why most collapsed populations have failed to recover. In the present study, I attempted to address hypotheses relating to this issue by exploring the way that fertilisation success might be affected by changes in male abundance. Because captive cod require large tanks for spawning and reproduce over a 6-12 week annual spawning season (Myers et al. 1993), it is difficult to replicate spawning experiments and I opted to use data from three independent research programmes. One drawback to this approach is that experimental population differences within or among research programmes may have confounded differences observed in relation to number of males present. Nonetheless, I found a similar pattern when I controlled for number of spawning females and reduced variation in spawning density and experimental tank size by separately analysing fertilisation rates in daily egg batches for which only a single female and a known number of spawning males contributed.

The experimental work on egg fertilisation rate and male abundance presented here suggests two mechanisms by which depensation might be realised within populations of Atlantic cod. Firstly, there is evidence to suggest that the proportion of eggs fertilised declines with number of spawning males. If

manifested as reduced numbers of recruits per spawner, this would provide one means by which per capita growth rate would decline, rather than increase, with reduced abundance. Secondly, the data suggest that variance in fertilisation success may increase as male abundance declines. This means that there may also be increased variance in individual reproductive success with declining abundance, something that would negatively affect both effective population size (N_e) and the number of individuals contributing genes to future generations relative to the estimated census number of spawning individuals (N_c), a ratio (N_e/N_c) that is already estimated to be quite small (10^{-3} to 10^{-5}) in broadcast spawning marine fishes (Hauser et al. 2002; Turner et al. 2002).

A reduction in fertilisation rate with reductions in breeding population size might be a product of the mating system of Atlantic cod, in which satellite males compete with primary males for egg fertilisation. During spawning, a female's brood is fertilised by one primary male and by several secondary males (Hutchings et al. 1999; Rakitin et al. 2001; Bekkevold et al. 2002). As population size declines, the number of satellite males per spawning female may decline as well, a consequence that may be particularly deleterious to fertilisation success if the number of eggs fertilised depends on sperm concentration. Consistent with this hypothesis is the observation that sperm numbers (both total number of sperm released and number released per male) and fertilisation rates in multiple-male spawnings are higher than those in single-male spawnings in the bluehead wrasse (*Thalassoma bifasciatum*; Shapiro et al. 1994; Marconato et al. 1997).

A second proximate cause of the positive association between fertilisation rate and abundance may be attributable to reduced opportunities for mate choice

afforded by reduced numbers of males. Both genetic and behavioural data are consistent with the hypothesis that mate choice is an integral part of cod spawning. Although unfertilised cod eggs and sperm remain viable for more than one hour in seawater (Kjørsvik and Lønning 1983; Trippel and Morgan 1994b), the sperm cells swim slowly (Trippel and Neilson 1992; Litvak and Trippel 1998) and Brawn (1961a) suggested that close alignment of the urogenital openings during the ventral mount was necessary to achieve high fertilisation rate. Hutchings and Myers (1993) predicted that this should favour assortative mating, a hypothesis later supported by the genetic analyses conducted by Rakitin et al. (2001). In experimental competitive mating situations, they found that only males of similar length (within 13%) to the female were able to sire entire batches of offspring. Mate choice is also suggested by various behavioural interactions between spawning males and females (Brawn 1961a; Hutchings et al. 1999; Jon Egil Skjaeraasen, University of Bergen, Bergen, Norway, personal communication).

Genetic complementarity may also be important insofar as various agents of intragenomic conflict and other forces acting at the suborganismal level may result in genetic incompatibility between some female and male gametes. Such genetic incompatibilities may prevent fertilisation (reviewed by Wirtz 1997) or successful development of offspring to sexual maturity (reviewed by Zeh and Zeh 1996, 1997). Using genetic markers, Rakitin et al. (1999a) found that when sperm from two males was combined in vitro with eggs of a single female, the relative success of males varied depending on which female was the egg donor suggesting female 'choice' at the gamete level or perhaps genetic

incompatibilities. All else being equal, I would expect genetic complementarity to increase with population size. In other words, increased opportunities for mate choice may result in increased fertilisation rate because of increased phenotypic and genetic variability among potential mates.

Increased variance in individual reproductive success, reflected by increased variance in fertilisation rate, will result in a reduction in effective population size (Nunney 1993; Sugg and Chesser 1994). Although abundance can appear high in severely depleted populations of Atlantic cod, it is important to acknowledge that census estimates of mature individuals (N_c) can poorly reflect the number of individuals that contribute genes during spawning, as represented by the effective population size (N_e). For broadcast spawning species such as Atlantic cod, it has been estimated that N_e may be 2 to 5 orders of magnitude lower than N_c because of the high variance in individual reproductive success associated with this type of mating system (Hedgecock 1994).

Empirical support for this range in N_e/N_c has recently been forthcoming from studies of marine fishes. Based on estimates of N_e from declines in heterozygosity and temporal fluctuations in allele frequency over 46-48 years, Hauser et al. (2002) concluded that effective population size in the New Zealand snapper (*Pagrus auratus*) was 5 orders of magnitude less than census population sizes. The effective number of female red drum (*Sciaenops ocellatus*) in the Gulf of Mexico, estimated from both mitochondrial (Turner et al. 1999) and microsatellite DNA (Turner et al. 2002), has been estimated to be 10^{-3} that of the female census population size. Increased variation in fertilisation rate concomitant with reductions in population size may increase the rate at which N_e declines with

abundance, which would increase the rate at which genetic variation is lost from the population.

In summary, my data are consistent with the hypotheses that (a) fertilisation rate declines with abundance and that (b) variance in fertilisation rate increases as population size declines. The former identifies one potential mechanism underlying depensation in Atlantic cod; the latter may have negative genetic consequences for effective population size (N_e). However, it needs to be stressed that these patterns between fertilisation rate and abundance are the product of exploratory analysis, and that considerably more work needs to be done before the veracity of the conclusions drawn here can be fully assessed. Consequently, it is urgent that we undertake further experiments to explore the relationship between sperm competition and fertilisation success in the laboratory and quantify cod density and fertilisation rates in the wild.

Table 5.1. Experimental tank volumes and sample sizes from which egg fertilisation and spawner abundance data were obtained from each of the research programmes.

Research programme	Tank volume (m³)	Number of females	Number of males	Spawning density (fish/m³)	Number of egg batches
Rowe/Hutchings	2.9	3	2	1.73	11
Rowe/Hutchings	2.9	6	2	2.77	23
Rowe/Hutchings	2.9	7	4	3.81	33
Rowe/Hutchings	2.9	6	5	3.81	27
Rowe/Hutchings	2.9	3	2	1.73	16
Rowe/Hutchings	684.0	41	18	0.09	55
Rowe/Hutchings	684.0	40	27	0.10	44
Rowe/Hutchings	684.0	25	29	0.08	39
Rakitin	2.1	1	2	1.43	4
Rakitin	3.9	1	2	0.76	5
Rakitin	3.9	1	2	0.76	11
Rakitin	3.9	1	2	0.76	7
Rakitin	3.9	1	2	0.76	3
Rakitin	2.1	1	2	1.43	7
Rakitin	3.9	1	2	0.76	8
Bekkevold	8.5	4	4	0.94	22
Bekkevold	8.5	4	4	0.94	14
Bekkevold	8.5	4	4	0.94	11
Bekkevold	8.5	5	2	0.82	18
Bekkevold	8.5	4	6	1.18	11
Bekkevold	8.5	5	3	0.94	18

Table 5.2. Metrics of variability (variance, coefficient of variation (CV), inter-quartile range (IQR)) in arcsine-transformed fertilisation rate in single-female egg batches fertilised by either 1 or 2 males.

Reference	Spawning males	Number of egg batches	Variance	CV	IQR
Bekkevold et al. (2002)	1	7	171.3	0.19	21.7
	2	7	32.3	0.07	9.2
Rakitin et al. (2001)	1	12	98.2	0.13	15.7
	2	29	73.7	0.11	9.7
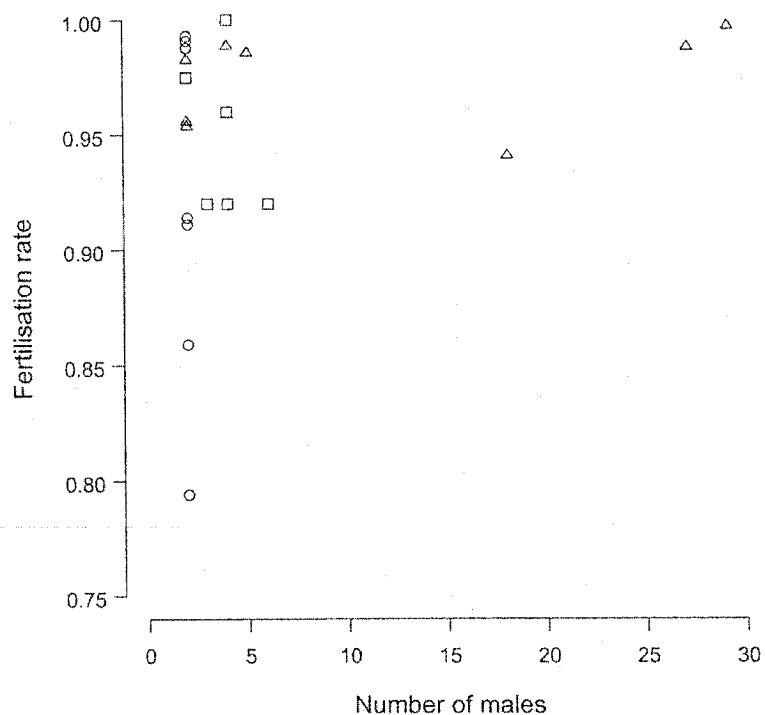
Figure 5.1. Median daily egg fertilisation rate measured across the spawning season in captive Atlantic cod in relation to the number of males present in each tank. Triangles represent data collected at Dalhousie University, circles represent data provided by Rakitin et al. (2001), and squares represent data provided by Bekkevold et al. (2002).
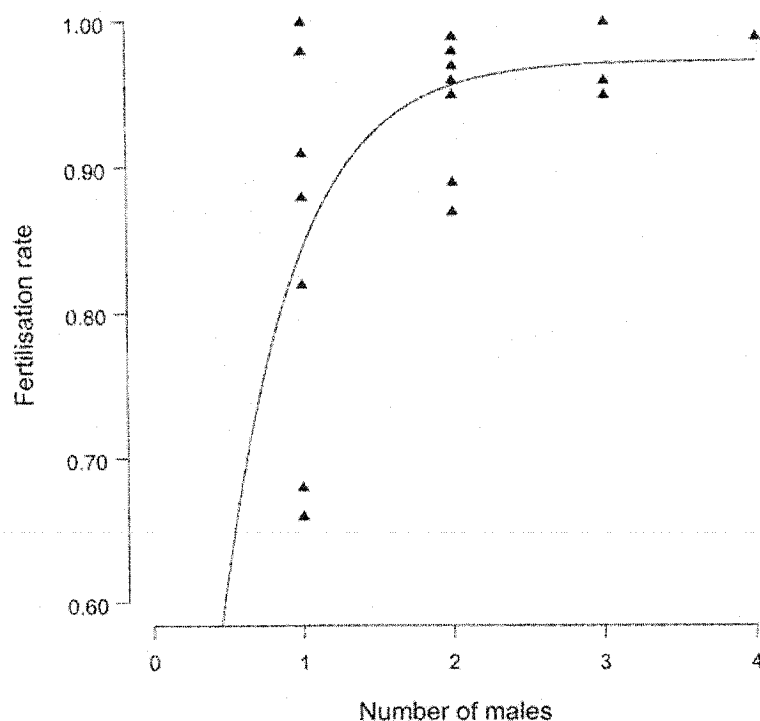
Figure 5.2. Egg fertilisation rate in Atlantic cod spawning events involving a single female in relation to the number of males that contributed sperm to the event. Closed triangles represent data provided by Bekkevold et al. (2002) and the line indicates the exponential asymptotic function that best described the data: fertilisation rate = $0.97*(1-e^{-2.02*\text{number of males}})$.
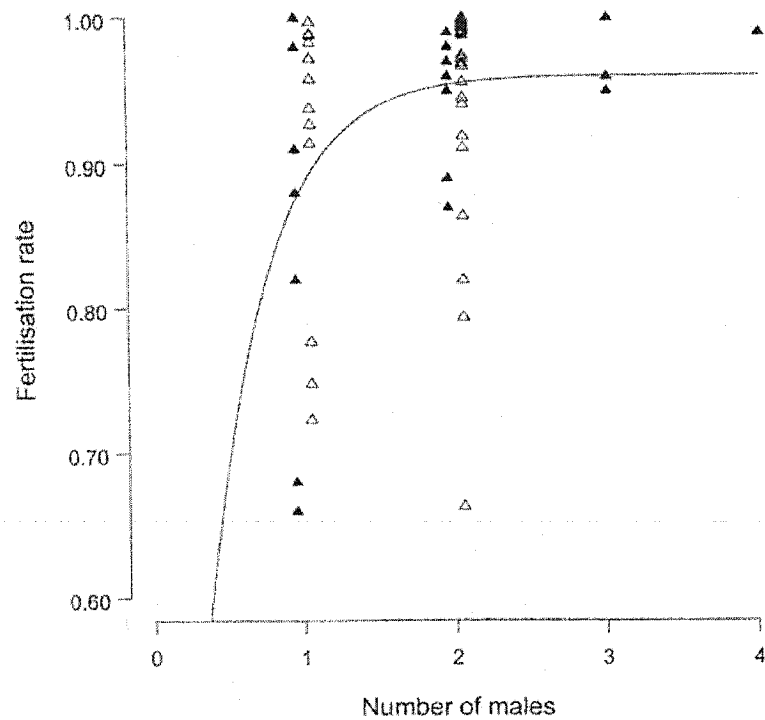
Figure 5.3. Egg fertilisation rate in Atlantic cod spawning events involving a single female in relation to the number of males that contributed sperm to the event. Closed triangles represent data provided by Bekkevold et al. (2002), open triangles represent data provided by Rakitin et al. (2001), and the line indicates the exponential asymptotic function that best described the data: fertilisation rate = $0.96*(1-e^{-2.54*number\ of\ males})$.

CHAPTER 6

MATING SYSTEMS AND THE CONSERVATION OF COMMERCIALLY EXPLOITED MARINE FISHES

INTRODUCTION

Worldwide declines in marine fishes and the uncertain consequences of biological changes to trophic structures and food-webs have left ocean ecosystems in a fragile state. Most dramatically, these collapses are reflected by the extirpation of more than 50 species of fish in the past century (Dulvy et al. 2003). Rates of decline for many species have been staggering. Large coastal and oceanic North Atlantic sharks, including threshers (*Alopias* spp.), great whites (*Carcharodon carcharias*), and hammerheads (*Sphyrna lewini*), have declined by > 75% since 1986 (Baum et al. 2003). Atlantic cod (*Gadus morhua*) throughout the North Atlantic are also at historic lows, having decreased by almost 90% since the 1970s in the North Sea (ICES 2002), and by more than 99% off the coast of Newfoundland since the 1960s (Hutchings 2003). Throughout Canadian waters, reproductive cod currently number ~ 200 million, considerably less than the 2.5 to 3 billion that existed in the 1960s (Hutchings 2003).

Assessments of conservation status of exploited fish populations by fishery management agencies are usually based on the premise that temporal estimates of abundance, age-specific metrics of commercial catch and geographical limits of distribution are all that are required to manage fish populations in a sustainable manner (Hutchings 2000a). When fish stocks are healthy and catches high, these data are often sufficient to address the majority of questions asked of fisheries science. But when fish stocks collapse, the main questions asked of fisheries science are questions pertaining to the life history, behaviour and ecology of the

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collapsed species.

An empirical vacuum faces those trying to understand why most collapsed marine fish populations have failed to recover. A long-standing premise was that recovery would immediately follow reductions in fishing, but it has now become evident that fishing is only one of a number of factors that determine the growth of collapsed populations (Hutchings 2001b; Dulvy et al. 2003). Within this context, it is now appropriate to explore the utility of incorporating behavioural ecology in conservation assessments of marine fishes. I believe that the traditional lack of attention to basic elements of fish ecology and evolution has limited the effectiveness of management plans and weakened the empirical basis of recovery strategies.

Although knowledge about the mating systems of most marine fishes is poor, with many gaps to fill, enough is known to examine the potential consequences for population extinction of exploited species that possess mating systems that are more complex than assumed by fishery managers and most fisheries scientists. I focus on commercially exploited fishes, most of which have been severely overexploited (Pauly et al. 2002) and show little signs of recovery (Hutchings 2000b, 2001a). Atlantic cod, a species that has experienced extraordinary declines in spite of enormous effort to prevent them from happening, merits particular attention. In addition to being the species whose declines precipitated debate concerning extinction risks faced by marine fishes (Hutchings 2001a; Dulvy et al. 2003), cod are one of the few marine fishes for which data about their mating system are available.

I argue that mate competition, mate choice and other components of the

mating systems of commercially exploited marine fishes can negatively influence population growth rate at declining and low levels of abundance. Knowledge of mating systems, incorporated into fisheries population assessments, can be used to assess the conservation risks posed by various management strategies. I conclude by identifying the questions that now need to be addressed and the research required to answer them.

IMPLICATIONS OF MATING SYSTEMS FOR COLLAPSE AND RECOVERY OF EXPLOITED MARINE FISHES

Mating systems have received little attention for the indirect but significant impact that they can have on population dynamics and conservation biology (Legendre et al. 1999; Møller and Legendre 2001). This is particularly true for marine fishes (Vincent and Sadovy 1998). Removal of breeding individuals from any population has important consequences, regardless of the mating system. However, as I discuss below, complex mating behaviour might inadvertently accelerate rates of population decline owing to harvesting and reduce subsequent rates of recovery.

Sex-biased changes to phenotype, genotype and operational sex ratio

The effects of exploitation on a fished population are rarely random. Fisheries tend to target the largest, fastest-growing individuals in a population rather than the smallest, slowest-growing individuals (Stokes and Law 2000; Conover and Munch 2002). In the short term, such differential harvesting will reduce phenotypic, and potentially genetic, variation; in the longer term, if the

characteristics that make some phenotypes more vulnerable to harvesting than others are heritable, harvesting will produce a genetic response to such differential selection. Fishing is also more likely to capture active individuals, whose more frequent movements render them more likely to come in contact with stationary and possibly mobile fishing gear, than less active individuals. Another means by which exploitation can effect differential selection of individuals within a population occurs when fish utilize different habitats, such as different depths or bottom substrate, and when fishing gear is preferentially deployed over some habitats rather than others.

If one sex is more likely to be caught by fishing than the other, exploitation will produce changes to the operational sex ratio, the relative number of males and females on the breeding grounds. In fish, either sex can be more vulnerable to exploitation, depending on the mating system and on the magnitude of sex biases in factors that affect vulnerability to exploitation, such as growth rate and behaviour. For example, among protogynous hermaphrodites, such as the graysby (*Epinephelus cruentatus*), males are often more susceptible to fisheries because of their larger size, greater aggression and higher activity levels (Côté 2003). Although females can respond to differential loss of males by changing sex, significant changes to sex ratio can still result because of temporal constraints on the rate at which sex change can take place under intense fishing pressure (Vincent and Sadovy 1998).

Changes in operational sex ratio can negatively affect reproductive success by reducing the probability of encountering potential mates or by reducing fertilisation success (Møller and Legendre 2001). The latter may be particularly

important in broadcast spawning species in which satellite males contribute significantly to egg fertilisation. If the number of breeding males per female declines with reductions in overall abundance (as would be expected if males are more vulnerable to exploitation than females), the reduced concentration of sperm (Marconato et al. 1997) can lead to reduced fertilisation success, notably among organisms that spawn in aggregations and release their gametes directly into the ocean (Côté 2003; Chapter 5).

Consequences of exploitation for mate choice

Seasonally and spatially synchronous reproduction provides an important means by which mating behaviour can significantly influence population vulnerability to, and recovery from, exploitation. Many marine fishes are exploited most heavily when they aggregate in large numbers for breeding; seasonally high densities enable fishers to maximize catch rates during this period. Sustained disruptions of mate choice throughout the breeding period can be expected to reduce fitness among members of the sex that is exerting choice. This argument is based on the reasonable premise that the expression of preferences has fitness benefits, assuming that selection has favoured genotypes that exert preferences over those that do not.

The probability that mate choice will be negatively affected will depend on the degree to which spawning behaviour is disrupted during and after fishing. This may be particularly true among species for which agonistic interactions among males (resulting in differential access to females) and defence of breeding territory are integral to the mating system. Constant reshuffling of social ranks

during extensive periods of fishing might prolong intervals between egg batch releases because of increased time required by males to aggressively compete for females and increased time used by females to evaluate male quality (where quality is measured either by the quality of breeding resources provided by the male or by the quality of genes that the male will pass on to offspring). For batch-spawning fishes, delays in the release of eggs after ovulation of just a few hours can dramatically reduce egg viability (Kjørsvik and Lønning 1983; Kjørsvik et al. 1990).

If the sex more vulnerable to exploitation is the sex upon which choice is being exerted, then reductions in phenotypic variation and in operational sex ratio generated by selective fishing will reduce the scope or breadth of choice. As a consequence, the choosier sex may resort to mating with partners of nonpreferred phenotypes or genotypes. Mating with a nonpreferred individual can lead to a reduction in reproductive investment, deleteriously influencing fertilisation rates and offspring viability (Sheldon 2000). For instance, male bluehead wrasse (*Thalassoma bifasciatum*) and bucktooth parrotfish (*Sparisoma radians*) alter sperm release depending on female size or fecundity (Shapiro et al. 1994; Marconato and Shapiro 1996). Similarly, female Banggai cardinalfish (*Pterapogon kauderni*) differentially allocate more resources to eggs when paired with a preferred male (Kolm 2002). Thus, individual reproductive decisions related to the phenotype of a mate can influence reproductive rate and hence the survival prospects of a population.

Density-dependent variation in mating behaviour and reproductive success

Slow rates of recovery among some marine fishes can be attributable to a reduction in population growth rate that is concomitant with reduced population size, an association termed an Allee effect in the ecological literature and depensation in the fisheries literature (Figure 6.1). The logistic model of density-dependent population regulation indicates that per capita growth rate is positive below and negative above the stable equilibrium, or carrying capacity of the population (i.e. competition for resources is the sole factor influencing population growth). However, in the presence of an Allee effect, the per capita growth rate is reduced at low population levels relative to the logistic model and it can even become negative if levels fall below the unstable equilibrium or critical population threshold. Reductions in per-capita growth can be attributable to increased per-capita death rate and/or decreased per-capita birth rate (Roff 2002). Populations that decline to the unstable equilibrium are unlikely to persist.

In spite of increased attention to Allee effects as factors responsible for slow recovery rates (Hutchings 2000a, 2001b, 2003; Reynolds et al. 2001), a dearth of empirical work is a significant barrier to assessing the potential for Allee effects in marine fishes. Population growth could be negatively affected by factors such as reduced mate availability, lower fertilisation success, changes to operational sex ratios, and reduced intensity of social interactions during spawning. If a population falls below some threshold density, individuals might have problems encountering potential mates and might either cease breeding or alter their reproductive behaviour in such a way that there are adverse effects on fertilisation success or offspring viability. Individuals unable to gain access to

mates might reallocate energetic resources from present reproduction to future growth, which might improve mating chances in subsequent seasons, e.g., deep-snouted pipefish (*Syngnathus typhle*; Berglund 1991). At low population density, individuals may experience delays in seasonal reproduction as more time might be required to find a mate. Delays in spawning might cause a mismatch between the timing of peak abundance of fish larvae and their planktonic prey, causing decreased larval survival and eventual recruitment. Scarcity of potential mates also lessens the potential for mate choice, negatively affecting reproductive success in a manner similar to that caused by reduced genetic and phenotypic variability. For broadcast spawners, particularly those species for which substantive numbers of eggs are fertilised by satellite males, an Allee effect might also be realized by reduced egg fertilisation rate and decreased genetic variation among offspring. The increased genetic variation effected by multiple paternity might also allow females to maximize their chance of producing viable offspring. Increased genetic variation can also improve the ability of a population to respond to future selection pressures.

MATING SYSTEM OF ATLANTIC COD: IMPLICATIONS FOR COLLAPSE AND RECOVERY

The Atlantic cod is a bet-hedging species, necessitated by a reproductive strategy in which eggs are released directly into the oceanic environment (broadcasted) and for which no parental care is provided (Hutchings 2000a, 2001a). Attaining maturity across a broad range of sizes (35-85 cm) and ages (2-7 years) throughout the species range (Brander 1994; Hutchings 2003), females

release their annual egg complement numbering hundreds of thousands, often millions, of 1.25-1.75 mm eggs in batches over 3-6 weeks (Chambers and Waiwood 1996; Kjesbu et al. 1996) within a 6-12 week spawning season (Myers et al. 1993) in water ranging from tens to hundreds of metres in depth (Brander 1994; Morgan et al. 1997; Smedbol and Wroblewski 1997).

Breeding groups of broadcast spawning marine fishes, particularly those that do not inhabit coral reefs, have often been described as disorganized aggregations lacking the behavioural complexity of bird and mammal mating systems (Nordeide and Folstad 2000). By depicting cod spawning as a behaviourally vacuous process of haphazard mating, the Canadian Department of Fisheries and Oceans (DFO) in the 1980s and early 1990s was able to defend its decision to permit fishing of spawning aggregations (Harris 1998).

However, recent studies have shown that successful reproduction in Atlantic cod involves complex behaviour within and between sexes (Brawn 1961a; Hutchings et al. 1999). Release of sperm and eggs by Atlantic cod involves a 'ventral mount' in which the male, whilst grasping the female with his pelvic fins and matching her swimming speed, positions himself beneath her (Figure 6.2a). This behaviour might be typical for gadid fishes as similar patterns have been described for haddock (*Melanogrammus aeglefinus*; Hawkins and Amorim 2000). Upon extrusion of the female's batch of eggs, the spawning couple are typically joined by satellite males, which swim among the eggs and also release sperm (Chapter 2; Figure 6.2b). Accordingly, genetic work indicates that eggs from single reproductive bouts are fertilised by more than one male (Hutchings et al. 1999; Rakitin et al. 2001; Bekkevold et al. 2002) and it is reasonable to assume that

alternative mating tactics exist in male Atlantic cod.

Agonistic behaviour preceding mounting attempts is hypothesized to reflect competition among males for mates and might influence access to females (Brawn 1961a; Hutchings et al. 1999; Chapter 2). Primary determinants of competitive ability include agonistic interactions, particularly chases of one male by another, and body size, larger individuals dominating smaller ones (Hutchings et al. 1999; Chapter 2). Agonistic interactions also enable some males to defend 'territories' in which they generally reside (Brawn 1961a).

Interactions between sexes are consistent with the hypothesis that females, and possibly males, exercise mate choice (Hutchings et al. 1999). One prominent behaviour observed in large tanks of cod is the circling of individual females by males on or near the bottom of the tank. Several factors associated with circling behaviour are suggestive of female choice. First, circling bouts are initiated and terminated by females. Second, by restricting circling to occasions when they are on the bottom, females can prevent ventral mounts by males. Third, circling provides females the opportunity to be in close physical contact with, and assess the quality of, several males before spawning.

Similar to many fishes, cod can produce and detect sound (Hawkins 1993). The calls of Atlantic cod comprise short 'grunts' with peak sound amplitudes at frequencies ranging from 50 to 500 Hz (Brawn 1961b; Hawkins and Rasmussen 1978; Midling et al. 2002). Although both sexes can produce sounds throughout the year, only males seem to do so during the spawning season, typically during agonistic interactions and courtship (Brawn 1961b; Chapter 4). Acoustic displays by male Atlantic cod during the spawning season may function as short range

signals to re-enforce threat displays directed primarily towards other males, as well as to attract and communicate spawning readiness to females (Brawn 1961a, 1961b; Chapter 4). Sound production by males might be important to females for mate assessment (Hutchings et al. 1999; Chapters 3 and 4). Grunting is achieved by Atlantic cod through the contraction of paired, striated drumming muscles surrounding three external lobes of the swimbladder wall (Brawn 1961b; Hawkins 1993). Drumming muscle mass is sexually dimorphic and is related to male fertilisation potential (Engen and Folstad 1999; Chapter 3), a quality of direct importance to females at risk of not having all their eggs fertilised. Consequently, there might be benefits to females who can discriminate among males by evaluating their courtship song. Calls produced by large aggregations of male gadids, such as cod and haddock, on spawning grounds might also be a long range signal to attract females to the spawning area (Nordeide and Kjellsby 1999; Hawkins and Amorim 2000); at, however, a reduced population size, the chorus might not be sufficiently loud to attract all individuals to the spawning grounds.

The consequences of removing the largest or most active individuals in a population to operational sex ratios have not yet been explored for broadcast spawning, non-hermaphrodites. Based on observations in the laboratory and field, Atlantic cod have been hypothesised to form leks during reproduction (Hutchings et al. 1999; Nordeide and Folstad 2000). Mature males aggregate near the bottom on spawning grounds where they actively defend small mating territories and females are typically found peripherally to male aggregations (Brawn 1961a; Morgan and Trippel 1996; Nordeide 1998; Hutchings et al. 1999; S.

Rowe, personal observation). When ready to spawn a batch of eggs, females enter male aggregations, returning to peripheral areas after spawning (Brawn 1961a; Hutchings et al. 1999). Spatial and vertical separation of the sexes can result in skewed sex ratios among fish harvested and skewed operational sex ratios within spawning shoals that persist after harvesting. Evidence that males might be more vulnerable to fishing gear deployed on or near the bottom during spawning has been forthcoming from reports of male-dominated catches in fisheries-independent surveys during the spawning period (Morgan and Trippel 1996).

Fishing might negatively affect the quality of males that can be chosen by females, where quality is measured by the quality of genes that the male will pass on to offspring, given that male cod provide no resources other than sperm. This might occur through the break-up of shoal structure and selective removal of the largest and most active of males which, are most likely to be caught. If the largest and most active males are the highest quality males, their removal can be expected to have a negative influence on reproductive success and offspring viability. Furthermore, noise produced during fishing activity (e.g. from engines, trawl doors, gear contact with substrate, and trawl cables) often overlaps the frequencies of sounds produced by Atlantic cod (Brawn 1961b; Hawkins and Rasmussen 1978; Nordeide and Kjellsby 1999; Midling et al. 2002) and might interfere with acoustic signaling associated with mating, thus also affecting female mating decisions and reproductive success.

Exploitation can reduce phenotypic and genetic variability. If female mate choice is part of the Atlantic cod mating system, reduced variation might

negatively influence recovery because of a reduced range in the phenotypic and genetic quality of individuals from which females can choose. Overfishing can have particularly dramatic consequences for the phenotypic variability in this regard, reflected best by severe truncation in the size distribution of breeding individuals. For example, within Newfoundland's collapsed northern cod population, mean individual mass declined by 50% between 1962 and 1991, the last full year of harvesting before the fishery's closure (based on 1990 commercial fishery statistics (Lilly et al. 2001)). More importantly, from a mate choice perspective, the coefficient of variation in body size declined by 55% during this 30-year period.

Fishing can also reduce phenotypic variability if some genotypes are more vulnerable to exploitation than are others. Fishing mortality, which often exceeds natural mortality by two- or threefold, might generate high selection differentials (Stokes and Law 2000). Given that body size is heritable (Roff 2002), prolonged fishing can be expected to effect a genetic change that narrows the range of body sizes within affected populations. Similarly, intensive size-selective fishing against the largest individuals in a population will almost certainly result in selection against the largest individuals of the same cohort or year class (Stokes and Law 2000; Conover and Munch 2002). As a consequence, faster growing individuals will be more vulnerable to exploitation than are slower growing individuals, as borne out by data on cod in the Southern Gulf of St Lawrence, Canada (Sinclair et al. 2002).

Size-selective fishing reduces the diversity of potential mates, and individuals might resort to mating with partners of nonpreferred phenotypes or genotypes.

Size complementarity between spawning partners could be important for broadcast spawners such as Atlantic cod to ensure that the urogenital openings of both fish are aligned opposite one another and that high fertilisation success is achieved (Hutchings et al. 1999; Rakitin et al. 2001). The outcome of pairings involving less suitable mates may be low fertilisation success because of the reduced probability of obtaining mates of complementary size.

As a consequence of disruptions in mate choice at low population density, individuals might experience delays in seasonal reproduction because more time might be required to find a mate (Hutchings et al. 1999). Spawning delays can result in over-ripening of cod gametes, which reduces the ability of eggs to be fertilised, as well as affecting the developmental success of fertilised eggs and larvae (Kjørsvik and Lønning 1983; Kjørsvik et al. 1990).

As population density declines, so might the number of satellite males per spawning female, resulting in a decrease in egg fertilisation rate if the latter is dependent upon sperm concentration (Marconato et al. 1997; Chapter 5). Furthermore, if it is difficult for a male to signal, and for a female to detect, that he carries genes that would enhance fitness, multiple paternity might be selectively advantageous to females to minimize the risk of egg fertilisation by genetically incompatible sperm (Zeh and Zeh 1996). Involvement of fewer males in spawning events would decrease genetic variation among offspring and increase the risk of developmental failure owing to non-complementarity between eggs and sperm.

POPULATION STATUS ASSESSMENT AND MATING SYSTEM RESEARCH FOR EXPLOITED MARINE FISHES

My primary aim has been to promote the idea that a lack of knowledge about the patterns and processes of mating systems has hindered our understanding of how anthropogenic activities influence the resistance of commercially exploited marine fishes to catastrophic decline and their resilience thereafter. Although limited, available data suggest that mate competition, mate choice and other processes related to mating systems are almost certain to have an impact on population growth rate at declining and low levels of abundance (Reynolds and Jennings 2000; Reynolds et al. 2001; Côté 2003). Under intense fishing pressure, I conclude that characteristics of the mating system of some marine fishes, such as Atlantic cod, increase the rate of population decline and diminish the rate of recovery relative to collapse and recovery rates that would have been predicted under the assumption that mating system was of no importance to population status. Broadly speaking, I would predict that:

1. The probability that fishing negatively influences reproductive success increases with fitness benefits of territorial behaviour and mate choice to mating systems.

2. Rates of population decline and time to recovery increase with: (i) intensity of mate competition; (ii) importance of mate choice to fitness; and (iii) sex bias in commercial catches.

Knowledge of mating systems can be profitably incorporated into fisheries population assessments in a manner similar to the means by which environmental data are considered. Although they can be applied in predictive

models, environmental data are usually used heuristically to evaluate current and future prospects for population growth. For example, if water temperatures are above long-term averages, current and short-term prospects for individual growth, condition, survival or recruitment of offspring might be considered favourable. Similarly, knowledge of a commercially exploited species' mating system could be used (i) to assess the conservation risks posed by various management options, (ii) to minimize the probability of fishery-induced changes to operational sex ratio, (iii) to minimize or negate anthropogenic disturbances during spawning, and (iv) to ensure that fishing does not significantly reduce phenotypic and genetic variability.

I envisage research programmes on the mating systems of commercially exploited marine fishes as having three components. The first, and least expensive, would involve the partitioning *by sex* of the variables routinely measured from commercial and research catches (e.g., length, mass, condition, age). The second would be the identification of spawning grounds and delimitation of spawning periods (detailed spawning maps do not exist for Atlantic cod, despite its historical socio-economic and ecological significance). The third component would involve laboratory experiments designed to test hypotheses of mate competition, mate choice and other components of the mating system, and to identify the phenotypic and genetic correlates of individual variation in reproductive success. This component could also involve manipulation experiments designed to evaluate the effects of changes to density, operational sex ratio and sex-biased changes in phenotypic variation to metrics of population growth such as mating success, egg fertilisation rate and genetic

variation among offspring.

Although knowledge of the reproductive behaviour of most marine fishes is limited, I maintain that enough is known to evaluate the potential consequences for population extinctions of commercially exploiting species with complex mating systems. Indeed, our general inability to prevent or halt serious declines in many populations underscores a pressing need to act cautiously on the basis of incomplete information about the nature of mating systems of exploited species.
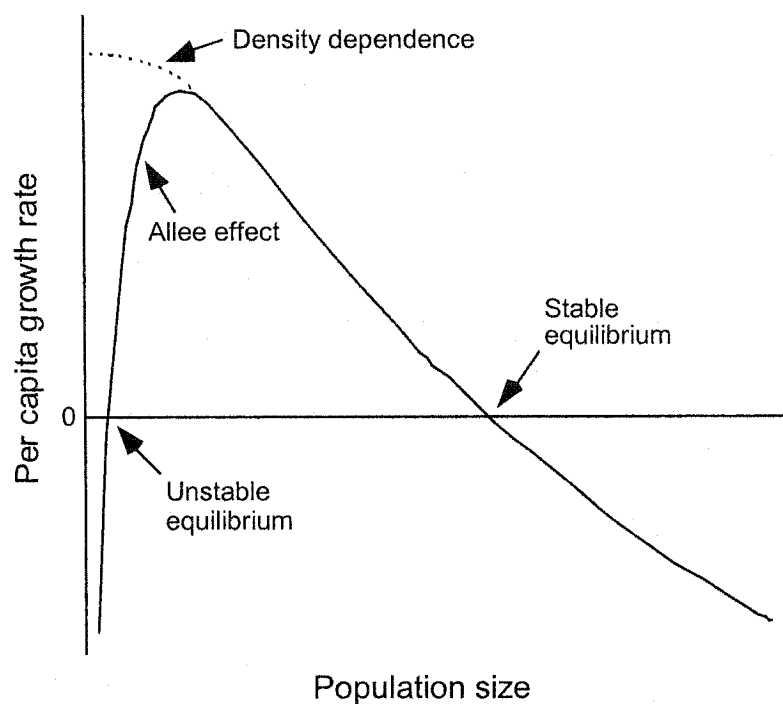
Figure 6.1. Relationship between per capita growth rate and population size in the presence (solid line) and absence (dashed line) of an Allee effect. An Allee effect is reflected by an increase in per capita growth with increasing population size.
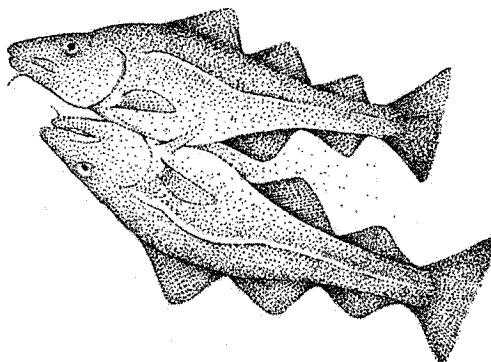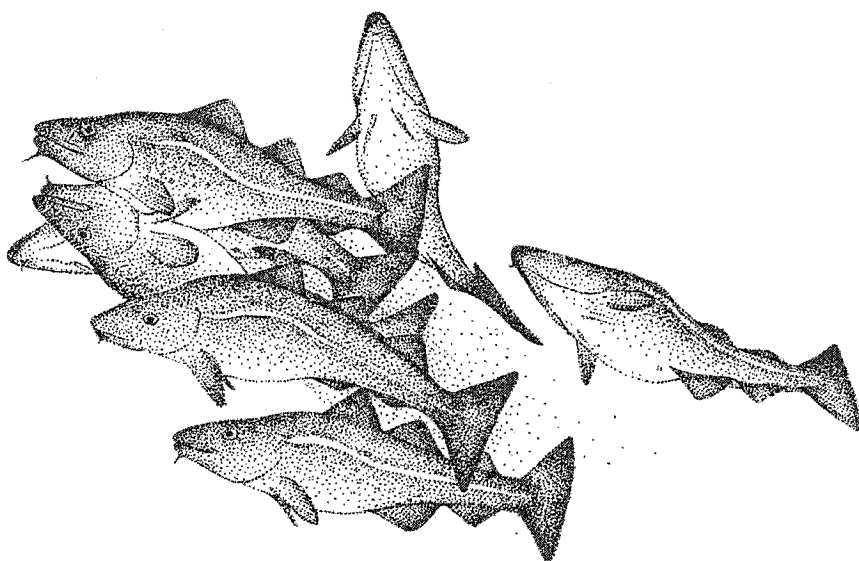
Figure 6.2. The spawning behaviour of a female Atlantic cod spawning with one or more males. (a) A female (top) and a male (bottom) Atlantic cod engaging in spawning. (b) Atlantic cod spawning events are initiated by a female-male pair who is often joined by satellite males that also release sperm.

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