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Evaluation of Atlantic bluefin tuna reproductive potential in the western Mediterranean Sea

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ABSTRACT

Ovarian tissue samples of Atlantic bluefin tuna (ABFT) spawners (n = 49) caught by purse seine in the Balearic Sea (western Mediterranean) were used to assess the stock reproductive characteristics. The frequency of spawning females estimated by the postovulatory follicle method was 84% and the spawning periodicity 1.2 days. Using an unbiased stereological method, the realized batch fecundity was estimated from counts of postovulatory follicles (POFs), whereas the batch fecundity of the subsequent spawn was estimated by quantification of the number of follicles containing oocytes at maturation stage (OMFs). The number of POFs was used as a reliable proxy of the realized batch fecundity, as it represents the actual number of eggs released in the last spawning event. The average relative realized batch fecundity was estimated to be approximately 48 eggs g^{-1} of total body mass. While the absolute batch fecundity was isometrically related to the fork length, the relative batch fecundity was not dependent on fish size, which leads to the assumption that all length classes contribute proportionally to their size, towards the total number of eggs spawned by the broodstock. Size-related variations in the sex ratio were observed in the study area and in other Mediterranean locations; females were more abundant in mid-size classes while males predominated in large-size classes.

1. Introduction

To guarantee the persistence of exploited fish populations over time, the stocks of progenitors should be adequately preserved to allow egg production rates that ensure sustainable recruitment (Mace and Sissenwine, 1993). A reliable estimation of the population productivity is, therefore, essential for fisheries management, for it determines the maximum sustainable levels of fishing to allow conservation of the resource or recovery of depleted populations (Morgan et al., 2009). The relationship between spawning stock and recruitment thus becomes a key issue in fisheries management. Many stock assessment studies assume that the spawning stock biomass (SSB) represents a direct measure of the reproductive potential of the stock (SRP). However, that egg production is proportionally related to the SSB in fishes is not always assumable in a fisheries resource management context (Lambert, 2008; Marshall et al., 1998, 2003; Marteinsdottir and Begg, 2002; Mehault et al., 2010; Scott et al., 1999; Trippel et al., 1997). As SSB is a measure that does not implicitly comprise the demographic structure of the reproductive stock, it provides population assessments that do not regard potential variations in egg production between individuals. For this reason, Marshall et al. (1998, 2006) proposed the total egg production of stocks as an improved index of recruitment potential over spawning biomass estimates.

There exist a number of biotic factors related to the reproductive potential of the spawning stock – e.g., fecundity-size relationship, spawning frequency, age-related shifts in sex-ratio, or egg viability – that may markedly affect recruitment but are often overlooked in fish stock assessments (Hoyle et al., 2009; Marteinsdottir and Begg, 2002; Trippel, 1999). Consequently, the incorporation of these features into management protocols results in significant improvement of population productivity estimations over the use of SSB as the sole index of SRP (Marshall et al., 2003). The non-genetic contribution of progenitors (especially females) to the offspring performance (parental effect) is of great importance in the formation of future year classes; hence the demographic structure of adults in fish populations has recently drawn the interest of fisheries scientists.

A widespread source of bias in stock productivity assessments based on SSB is the assumption that the proportion of females in the reproductive stock remains constant over time and through all age classes (Marshall et al., 2006). Baglin (1982) showed that ABFT females from the western stock were more prevalent than males in spawning aggregations (April–May), while males were more frequent in feeding schools. More recent studies carried out in Mediterranean spawning grounds have shown a male predominance in young and old age classes (El Tawil et al., 2004; Fenech et al., 2003; Mèlich et al., 2011). In the light of these observations, variations in the sex ratio with size should be

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addressed and eventually incorporated into stock recruitment predictive models.

Assessments of the ABFT eastern stock have generated large uncertainties due to poor fisheries data reporting (Fromentin, 2003; Fromentin and Ravier, 2005). The stock reproductive potential (SRP), recruitment rates and the effects of commercial catches on SSB trends remain uncertain and arouse intense debate (Fromentin and Powers, 2005). For this reason, the search for alternative, fisheries-independent methodologies have been encouraged to improve the scientific advice in fisheries management (ICCAT, 2010). In the same way that SSB data serve to estimate the stock egg production as an indicator of SRP, the reverse pathway can also be explored with a view to provide fisheries-independent stock abundance estimations using ichthyoplankton scientific surveys (Lasker, 1985; Somarakis et al., 2002; Stratoudakis et al., 2006). Recent work has increased our knowledge of the reproductive biology of ABFT in the west Mediterranean Sea (Abascal et al., 2004; Aragón et al., 2010; Corriero et al., 2005; Gordoa et al., 2009; Medina et al., 2002; Mourente et al., 2002), but many questions still remain to be answered in terms of interrelations between breeding stock, SRP and recruitment. In precedent papers, histological qualitative and quantitative analyses of ABFT gonads were undertaken to assess a) maturation changes in eastern populations during reproductive migrations (Medina et al., 2002), and b) the influence of different capture methods in the perception of the reproductive performance of ABFT broodstocks (Medina et al., 2007). In the present study, an increased number of samples of active spawners caught by purse-seine, which targets tuna schools at peak reproduction, is analyzed to estimate reproductive parameters that are essential for fisheries assessments. The application of the disector stereological principle (Sterio, 1984) allows unbiased (assumption-free) estimations of numbers of maturation oocytes and postovulatory follicles, and is therefore preferred over the model-based techniques used previously (Medina et al., 2002, 2007).

The Balearic archipelago is long known as an important breeding area for ABFT eastern populations (Alemany et al., 2010; García et al., 2005; Medina et al., 2002; Rodríguez-Roda, 1975). Other Mediterranean spawning grounds have been identified in the south Tyrrhenian Sea and the Levantine Sea (Corriero et al., 2003; Heinisch et al., 2008; Karakulak et al., 2004; Oray and Karakulak, 2005). The ABFT Balearic spawning ground does not appear to be excessively broad (Alemany et al., 2010), which suggests that assessment of this Mediterranean spawning population based upon egg production methods would be technically feasible.

Incorporation of detailed reproductive data into ABFT stock assessments is indeed necessary to understand how reproductive biology affects population productivity. In this paper, we provide information about reproductive traits that influence the SRP of ABFT using ovary histological samples from broodstock schools. A number of particular reproductive traits of tunas (indeterminate fecundity, asynchronous oocyte development, multiple spawning) render it difficult to make accurate estimations of total egg production. An additional drawback in the case of ABFT is the current reduced catches due to limited management quotas, and most importantly the unavailability to collect tissue and organ samples of active ABFT spawners from purse-seine catches, since this fishery is fully aimed at supplying living fish to fattening farms. Yet, ABFT ovarian samples collected between 1999 and 2005 from purse-seiners in the Balearic Sea were available to derive data for SRP estimations that may be useful in fisheries management.

2. Material and methods

2.1. Animals and biometry

ABFT spawners (n = 49) were sampled from commercial purse-seine catches around the Balearic archipelago between 1999 and 2005 (Table 1). The curved fork length (CL_F) of each individual was measured to the lowest 1 cm and then transformed into the straight fork length (L_F) using the formula: $L_F = 0.955\ CL_F$ (ICCAT conversion factors; http://www.iccat.int/Documents/Stats/convers.pdf). The ovaries were immediately removed and weighed onboard. Body mass (M_B) was calculated from the equation $M_B = 1.9607 \cdot 10^{-5}\ L_F^{3.0092}$, and the volume of the pair of ovaries (V_O) was estimated from their mass (M_O) according to the equation $V_O = 0.9174\ M_O$ (Medina et al., 2007). Regression analysis was used to evaluate the relationship between V_O and L_F.

Table 1
Data of ABFT sampled by purse seine on years 1999–2001 and 2005. L_F, fork length; *GSI*, gonadosomatic index ($GSI = 100\ M_O M_B^{-1}$).

Year	n	L_F (cm) Mean (±SD)	L_F range (cm)	*GSI* (%) Mean (±SD)
1999	11	209 (8.4)	191–219	4.29 (1.80)
2000	17	180 (27.7)	114–213	3.76 (1.64)
2001	14	141 (54.1)	92–207	3.75 (1.20)
2005	7	168 (47.2)	98–208	2.59 (1.18)

2.2. Tissue samples, spawning frequency and fecundity estimations

Ovarian tissue samples were fixed in buffered 4% formaldehyde (10% formalin) and processed as described elsewhere (Medina et al., 2007). The frequency of spawning in the Balearic spawning ground was estimated by the postovulatory follicle method (Hunter and Macewicz, 1985), which calculates the mean spawning fraction as the total number of spawning females whose ovaries show postovulatory follicles (POFs) divided by the total number of mature females sampled.

For batch fecundity estimations, only histological samples that contained a suitable amount of tissue (~2 cm^3) and produced high quality sections were considered. Potential batch fecundity estimations were carried out from counts of ovarian follicles that contained oocytes at maturation stage (oocyte maturation follicles, OMFs), whether migratory-nucleus or hydrating oocytes. The realized fecundity was estimated as number of POFs. Samples where very old POFs (>12 h) occurred were also excluded from the analysis as they could be confounded with atretic follicles; moreover, some of them could be highly degraded or already fully reabsorbed, leading to underestimations of the realized fecundity.

Unlike model-based stereological procedures that have been widely used for estimating numbers of intraovarian oocytes in many fishes, including the ABFT (Medina et al., 2002, 2007), the disector principle (Sterio, 1984) provides unbiased estimates of the number of objects in a given volume of tissue as it does not rely on assumptions, models, or correction factors. Using 10-μm serial paraffin sections, the physical disector method (Sterio, 1984) was applied to estimate the numerical density (number of particles mm^{-3}, N_V) of OMFs and POFs (Aragón et al., 2010; Aranda et al., 2011). The batch fecundity, as represented by the total number of OMFs (potential fecundity) or POFs (realized fecundity) contained in the pair of ovaries (N), was obtained by raising the value of N_V to the whole ovarian volume ($N = N_V\ V_O$). From this estimate, the relative batch fecundity (number of eggs produced per gram of total body mass, Ng^{-1}) was calculated as $Ng^{-1} = N M_B^{-1}$.

Comparisons of means of the stereological estimates (N_V, N and Ng^{-1}) among years were carried out by Kruskal–Wallis test. Potential and realized fecundity values were compared by Mann–Whitney *U*-test. Regression analyses were performed to evaluate the relationships between fecundity stereological parameters and fork length. In all statistical tests used, a significance level of $\alpha = 0.05$ was considered.

2.3. Sex ratio

Over 10,000 ABFT caught by purse seine in the Balearic spawning ground between 1995 and 2009 were checked for sex determination by visual identification of the gonads during harvesting in commercial