Fisheries Research 176 (2016) 76–85

Contents lists available at ScienceDirect

Fisheries Research

journal homepage: www.elsevier.com/locate/fishres

Contrasting batch fecundity estimates of albacore (*Thunnus alalunga*), an indeterminate spawner, by different laboratory techniques

Sámar Saber[a,*], David Macías[b], Josetxu Ortiz de Urbina[b], Olav Sigurd Kjesbu[c,d]

[a] Departamento de Biología Animal, Universidad de Málaga, 29071 Málaga, Spain
[b] Instituto Español de Oceanografía, CO Málaga, Puerto Pesquero s/n, 29640 Fuengirola, Spain
[c] Institute of Marine Research (IMR) and Hjort Centre for Marine Ecosystem Dynamics, P.O. Box 1870 Nordnes, NO 5817 Bergen, Norway
[d] Centre for Ecological and Evolutionary Synthesis (CEES), Department of Biosciences, University of Oslo, P.O. Box 1066 Blindern, NO 0316 Oslo, Norway

ARTICLE INFO

Article history:
Received 19 August 2015
Received in revised form
22 December 2015
Accepted 25 December 2015
Available online 5 January 2016

Keywords:
Assumption-based stereology
Physical Disector
Oocyte Packing
Density
Indeterminate species
Fecundity

ABSTRACT

A range of methods can be applied to estimate the batch fecundity of species with an indeterminate reproductive strategy. The traditional Hydrated Oocyte (HO) method based on direct counts of hydrated oocytes is the easiest and most accurate method but the main problem with this method is the shortage of hydrated ovaries in sampled fish such as tuna species. Batch fecundity estimates of albacore *Thunnus alalunga* resulting from counts of migratory nucleus (MG) oocytes using the application of the Weibel and Gomez (W&G), Physical Disector (PD), Oocyte Packing Density (OPD), and HO methods were compared using the last method as "control". Postovulatory follicles (POFs) were also counted using the PD method. Correction factors due to shrinkage were considered in the application of the different methods. Our results showed the highest batch fecundity estimates were obtained with the design-based PD method. The outputs from the assumption-based W&G and the theoretical OPD methods were closest to the HO method. Annotations of POFs instead of MG oocytes gave markedly lower values. The new OPD method was used to estimate batch and relative fecundity on a larger sample of fish (selected according to their length). The relationships between batch and relative fecundity estimates of albacore and the associated biological metrics (length, body weight and ovary weight) were investigated. Batch fecundity estimates ranged from 0.42 to 2.16 million oocytes with a mean relative batch fecundity of 136 oocytes per gram of body weight. The batch fecundity was shown to increase with fish size (length and weight) and gonad weight, while relative batch fecundity (g^{-1}) was related only to gonad weight.

© 2015 Elsevier B.V. All rights reserved.

1. Introduction

Fecundity is a biological parameter of interest to fisheries researchers as a basic aspect of population dynamics (Hunter et al., 1992). The estimation of fecundity for species with an indeterminate reproductive strategy ("indeterminate species" from now on) is particularly complicated because annual fecundity is not fixed prior to the start of the spawning season (Hunter et al., 1992). In these species, potential annual fecundity should be estimated from the number of eggs spawned per batch (batch fecundity) and the number of spawning events per season (Hunter et al., 1985; Murua et al., 2003). Nowadays a variety of methods can be applied to estimate the batch fecundity (BF) and the method chosen by researchers may depend upon the available resources (see Ganias et al., 2014; Kjesbu, 2009; Murua et al., 2003).

The most common method used for estimating the BF in indeterminate species is the Hydrated Oocyte (HO) method developed by Hunter et al. (1985), not only because of its simple application, but also because only ordinary laboratory facilities, e.g. a stereomicroscope, are needed. Initially the procedure for the application of the HO method only included ovaries that contained hydrated oocytes as the most advanced group of oocytes (and no POFs) because these oocytes are easily distinguished from other oocytes and should properly reflect the "batch of eggs" soon to be spawned (Hunter et al., 1985). Later, ovaries that contained migratory nucleus (germinal) (MG) oocytes (the final stage of oocyte maturation before hydration) were also considered for BF estimations (Hunter et al., 1992; Schaefer, 1996). The background for this being that specimens with hydrated ovaries, such as tuna species, are seldom found in the catch (Schaefer, 2001). Batch fecundity estimates obtained by applying methods that required histological sections are also seen in marine laboratories. Although histology is both time-consuming and expensive, microscopic examination of the ovarian tissue makes possible to accurately distinguish the

* Corresponding author. Fax: +34 952463808.
E-mail addresses: samar.saber@uma.es (S. Saber), david.macias@ma.ieo.es (D. Macías), urbina@ma.ieo.es (J.O.d. Urbina), olav.kjesbu@imr.no (O.S. Kjesbu).

http://dx.doi.org/10.1016/j.fishres.2015.12.013
0165-7836/© 2015 Elsevier B.V. All rights reserved.

different oocyte developmental stages, as well as the presence of postovulatory follicles (POFs) and atretic oocytes and thereby, allow an unequivocal characterization of the reproductive phase. The earliest and the most widely used stereological method for estimating oocyte numbers is the assumption-based method of Weibel and Gomez (1962) (W&G). This method requires assumptions to be made regarding particle shape and size distribution. Nevertheless, it has been successfully used to estimate fecundity for many fish species (Coward and Bromage, 1998; Emerson et al., 1990; Haslob et al., 2013; Knapp et al., 2014; Medina et al., 2002, 2007). Two decades later Sterio (1984) introduced the physical disector (PD) method, i.e. a stereological method for design-based particle number estimation, which so far has been in limited use among fish reproductive biologist because of the large work load involved (Aragón et al., 2010; Bucholtz et al., 2013; Kraus et al., 2008). The PD method has, however, recently been applied to reflect realized batch fecundity from counts of postovulatory follicles in bluefin tuna (*Thunnus thynnus*), an indeterminate species (Aragón et al., 2010; Aranda et al., 2011, 2013). Note that these true stereological methods, W&G and PD, do not directly give the absolute number of particles but the numeric density within a reference volume. Finally, Oocyte Packing Density (OPD) theory (Kurita and Kjesbu, 2009) has been used for estimating the numbers of different stages of oocytes per gram of ovary in indeterminate species (Korta et al., 2010; Kurita et al., 2009; Kurita and Kjesbu, 2009; Saber et al., 2015a; Schismenou et al., 2012). This newer method which is strictly speaking not a true stereological method, only a part of it, i.e. the estimation of volume fraction, can, as with the two others, be scaled (by including ovary size) to give batch or potential fecundity (Ganias et al., 2014; Schismenou et al., 2012).

Only a few studies comparing fecundity methods have been published throughout time (Aragón et al., 2010; Cooper et al., 2005; Coward and Bromage, 2002; Emerson et al., 1990; Kjesbu et al., 1998). Before the use of the PD method in fisheries laboratories and the introduction of OPD theory, the most relevant paper was written by Emerson et al. (1990). These authors compared estimates of the batch fecundity using three methods and described the advantages of the novelty of the stereological method in that moment, the W&G method, over the volumetric and automated particle counter ("fish egg counter") methods, while the recent study of Aragón et al. (2010) evaluated the differences in the estimated number of oocytes in each developmental stage obtained by applying the W&G and PD methods.

The present study used albacore *Thunnus alalunga*, a batch spawner with asynchronous oocyte development and indeterminate fecundity (Otsu and Uchida, 1959) to contrast a defined variety of suitable fecundity methods. More specifically, the aim of this study was to compare batch fecundity estimates of spawning albacore resulting from counts of MG oocytes from the W&G, PD, OPD and HO methods. In addition, batch fecundity estimations carried out from counts of POFs using the PD were also compared. Corrections factors due to shrinkage were considered in the application of the different methods and benefits and limitations of the methods applied were discussed. Another objective of this study was to further test the application of the OPD method for estimating batch and relative fecundities for female albacore selected according to their size and then, to investigate the relationships between these reproductive traits and the associated biological metrics (length, body weight and ovary weight).

2. Material and methods

Albacore (*T. alalunga*) ovaries were collected from fish caught by commercial and recreational fisheries in the western Mediterranean Sea between 2005 and 2012 (Saber et al., 2015b). Fork length (cm), fresh total body weight (kg) and gonad weight (GW, g) were recorded. As part of this large sampling program, histology was applied for detailed maturity staging finding that 27% of the females showed MG oocytes as the most advanced group of oocytes within their ovaries, for more detail see Saber et al. (2015b). In the current study 61 out of these 157 females were selected according to their fork length (*FL*) to estimate batch fecundity (BF), i.e. the total number of eggs (here substituted by MG oocytes) released in a single spawning event, using advanced oocyte packing density (OPD) theory (Kurita and Kjesbu, 2009). Prior to initiating this work, a total of 12 ovaries, selected out of the 61 ovaries, were used to directly compare these theoretical results with those given from the application of the classical Weibel and Gomez (W&G) method (Weibel and Gomez, 1962) and the modern physical disector (PD) method (Sterio, 1984), both being fully stereological in nature. Given that small POFs, which represent the follicular remains in the ovary after the eggs are *de facto* spawned, coexisted with MG oocytes in all ovaries, the PD method was also applied to estimate the BF based on counts of POFs. Hence the PD method was split into two: PD_{MG} and PD_{POF}. The traditional Hydrated Oocyte (HO) method (Hunter et al., 1985) was also applied to estimate the number of MG oocytes. For proper comparisons, the HO method was defined as "control" since this method is based on direct counting of oocytes in subsamples using gravimetric principles to indicate raising factor of gonad weight to analyzed subsample weights. An additional sample of 16 fresh ovaries, all subsequently classified histologically as "spawning", was collected in the same area in July 2013 to estimate loss in ovarian tissue weight and volume during histological processing (see below).

The following sections give more background information on methods used in the detailed comparative analysis on fecundity estimation for the 12 specimens but also address the subsequent OPD study on the 61 specimens.

2.1. Approaches and methods used to enumerate oocytes using histology

Assuming a homogeneous distribution of oocytes within and between ovarian lobes, as documented earlier for tunas (Otsu and Uchida, 1959; Stéquert and Ramcharrun, 1995), a 2 – 3 cm wide cross-section from the central part of one of the lobes was fixed in Bouin's fluid for four hours and then preserved in 70% ethanol. Then, a representative subsample (from the ovarian wall to the lumen) was taken from each of preserved ovary, dehydrated through increasing concentrations of ethanol series, cleared, and embedded in paraffin. Sections were cut at 10 μm and stained with either Mallory's trichrome stain or haematoxylin-VOF (Gutiérrez, 1967). All images were taken on a Nikon photomicroscope with either ×4 (W&G and OPD methods) or ×2 (PD method) objective magnifications. Significant differences in the distribution of oocytes among the centre, mid-region and periphery of the same albacore ovarian section has been found (Otsu and Uchida, 1959). Thus, in order to avoid biased estimations due to these transversal structural differences affecting oocyte distribution, the set of micrographs of each ovary was taking randomly but representing the whole ovarian section. The resulting area of each micrograph was 3.302 mm^2 and 13.552 mm^2 for ×4 and ×2 objective magnifications, respectively.

2.1.1. Assumption-based stereological method: Weibel and Gomez

The numerical density (N_v) of the particles of interest, i.e. the number of MG oocytes per unit volume, was calculated according to principles first developed by Weibel and Gomez (1962) but using here the further modified formula in Weibel et al. (1966). The W&G stereological method includes "the Delesse principle", which states that the fractional volume (V_i) of a component *i* in a given tis-

Download English Version:

https://daneshyari.com/en/article/4542756

Download Persian Version:

https://daneshyari.com/article/4542756

Daneshyari.com