Journal of Great Lakes Research 40 (2014) 164–171

Contents lists available at ScienceDirect

Journal of Great Lakes Research

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

Evaluating gonadosomatic index as an estimator of reproductive condition in the invasive round goby, *Neogobius melanostomus*

Jeffrey N. Zeyl *, Oliver P. Love, Dennis M. Higgs

Department of Biological Sciences, University of Windsor, 401 Sunset Avenue, Windsor, Ontario, N9B 3P4, Canada

ARTICLE INFO

Article history:
Received 28 January 2013
Accepted 22 November 2013
Available online 7 January 2014

Communicated by Thomas Pratt

Index words:
Round goby
Gonadosomatic index
Gonad development
11-Ketotestosterone
Testosterone
17β-estradiol

ABSTRACT

Using gonadosomatic index cut-off scores has become a standard protocol for selecting reproductive fish in studies on the reproductive biology of round goby, *Neogobius melanostomus*, a significant invader of the Laurentian Great Lakes, but the validity of this practice has not been validated with histological staging. The goal of the current study was to evaluate the effectiveness of using gonadosomatic index (GSI) cut-off scores to classify reproductive status in male and female round goby by documenting associations between GSI, sex steroids, and gonad development. Gonadal stage was determined in both sexes using hematoxylin and eosin histology. Plasma 11-ketotestosterone and testosterone were measured in males, and testosterone and 17β-estradiol were measured in females. Gonadosomatic index cut-off scores were effective in selecting spawning capable individuals at higher GSI values, but GSI values were limited in the ability to make further distinctions of gonadal stage and missed many spawning capable females. In females, testosterone levels were highest during vitellogenic growth and declined prior to ovulation. 17β-estradiol displayed a similar, but non-statistically significant pattern. Males with developing testes had higher levels of 11-ketotestosterone —but not testosterone —than reproductively immature males, although levels of these androgens were overall positively correlated in males. The findings indicate that the conventional GSI cut-off scores (1% in males, 8% in females) accurately assign spawning capable condition in both sexes; however, they may also exclude some spawning capable females with lower GSIs.

© 2013 International Association for Great Lakes Research. Published by Elsevier B.V. All rights reserved.

Introduction

The round goby, *Neogobius melanostomus* (Pallas 1814), is a significant invader in the Laurentian Great Lakes of North America and several regions in Europe that has become a threat to native aquatic ecosystems worldwide (Corkum et al., 2004; Kornis et al., 2012; Poos et al., 2010). Since its invasion, considerable research has been conducted to assess its impacts and population dynamics by evaluating factors such as age and growth patterns, habitat use, and reproductive potential (Charlebois et al., 1997). One aspect of the round goby management plan that has received less attention is the development of standardized methods for reproductive condition assessment. Although it may be impractical to include detailed reproductive data into routine population assessments, precise measurement of reproductive condition is essential for determining reproductive cohorts to use in population growth models (Lowerre-Barbieri et al., 2011).

A variety of methods are available to assess reproductive condition in fishes, including microscopic gonadal staging, macroscopic gonadal staging, oocyte size–frequency distributions, sex steroid measurement and gonadal indices (Lowerre-Barbieri et al., 2011; West, 1990). Histological gonadal staging provides a direct snapshot of developmental stages of individual gametes within the gonads, while the relationship between other metrics and gonadal gamete stage(s) may depend on the pattern of gonadal development (e.g. synchronous vs. group synchronous vs. asynchronous). However, histological staging is time intensive and impractical in many cases, thus proxies are useful if they can be properly validated (Lowerre-Barbieri et al., 2011; West, 1990).

The gonadosomatic index (GSI = gonad mass / body mass × 100%, or GSI = gonad mass / somatic mass × 100%) (deVlaming et al., 1982) is a common metric of reproductive allocation and reproductive condition in fisheries biology. However, GSI may be an imprecise proxy for gonadal stage because it assumes isometry between somatic mass and gonad mass as well as a tight correspondence between total gonad mass and gonad developmental stage (deVlaming et al., 1982; Packard and Boardman, 1988). Nonetheless, if significant gonad growth occurs at distinct developmental stages (e.g. vitellogenesis), GSI cut-off scores (i.e. designating those above the score as 'reproductive' or 'spawning-capable') may be useful metrics for sorting fish into gross reproductive categories.

Gonadosomatic cut-off scores of 8% (females) and 1%–1.3% (males) are commonly used to select reproductive individuals in round goby invasion studies (Bowley et al., 2010; Gammon et al., 2005; Kasurak et al., 2012; Marentette et al., 2009; Marentette and Corkum, 2008; Yavno and Corkum, 2011; Young et al., 2010). Despite the widespread use of the GSI cut-off scores in such studies, there are scarce data available to

* Corresponding author at: School of Fisheries, Aquaculture and Aquatic Sciences, 203 Swingle Hall, Auburn University, Auburn, AL 36849, USA. Tel.: +1 334 734 0708.
E-mail addresses: jeffzeyl@gmail.com (J.N. Zeyl), olove@uwindsor.ca (O.P. Love), dhiggs@uwindsor.ca (D.M. Higgs).

0380-1330/$ – see front matter © 2013 International Association for Great Lakes Research. Published by Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.jglr.2013.12.004

validate this practice; neither are there much data on the correspondence between sex hormone levels and gonadal stage (but see Bowley et al., 2010). The goal of the current study was to examine the validity of GSI cut-off scores as reproductive condition classifiers in male and female round goby by examining the correspondence between GSI values, gonadal stage, and sex hormones. In this manner we provide the first direct test of association between these separate measures of reproductive condition in the round goby.

Materials and methods

Subjects

Round gobies were collected by angling from the shoreline of the Detroit River in Windsor, Ontario, Canada (42°18′23.83″N, 83° 4′32.46″W) between April and August 2011. 'Reproductive' males and females ('reproductive' criteria described below) were processed within 1 week of capture, whereas 'non-reproductive' fish were euthanized after being held in the laboratory for various lengths of time (see Supplementary Fig. 1 for GSI and hormone levels of reproductive fish in relation to ordinal date of processing). Gobies were kept in mixed sex tanks, fed with fish flakes (Tetramin Inc, Blacksburg, VA, USA), and kept on a 16L:8D photoperiod. All animal care and experimental protocols complied with the University of Windsor's animal care committee and guidelines of the Canadian Council of Animal Care (CCAC).

Gonadosomatic index was calculated, following fish euthanasia by clove oil overdose (~60 mg L^{-1}), according to the following equation: (GSI = gonad mass / total body mass × 100%) (Gammon et al., 2005; Marentette and Corkum, 2008). Gonad and body mass were measured to the nearest 0.01 g (Scout Pro scale, SP202, Ohaus Corp., Pine Brook, NJ, USA), and male gonad mass included both the testes and seminal vesicles. Fish sex could be easily determined in adults by examining urogenital shape (Charlebois et al., 1997). In accordance with previous round goby studies, females with GSIs greater than or equal to 8% were initially considered 'reproductive' (RFs) and less than 8% 'non-reproductive' (NRFs). Males with GSIs greater than or equal to 1% were scored as 'reproductive' (RMs) and less than 1% as 'non-reproductive' (NRMs). Masses, total lengths, and GSI in each reproductive category are given in Table 1.

No 'sneaker' males (i.e. reproductive males employing a 'parasitic' spawning strategy; Marentette et al., 2009; Taborsky, 1998), were included in the current study. Papilla indices (PI = papilla length / total length × 100%) of NRMs were below that of 'sneaker' males described by Marentette et al., 2009 (current study: 3.07 ± 0.04 SEM; Marentette study: 6.71 ± 0.12).

Steroid assays

Male plasma was assayed for 11-ketotestosterone (11KT) and testosterone (T), and female plasma for 17β-estradiol (E2) and T. Following euthanasia, the caudal peduncle region was severed and blood was drawn from the caudal vein using heparinized capillary tubes. The blood was spun for 10 min at 14,500 rpm (Micro-Hematocrit Centrifuge, LWS-M24, LW Scientific, Lawrenceville, GA, USA) and stored at −80 °C. The typical range of plasma volumes was 5–40 μL. All plasma was collected between 13:00 and 18:00 h to address potential diel influences on hormone levels. Steroids were extracted once with diethyl ether prior to performing enzyme-linked immunosorbent assays (Cayman Chemical, Ann Arbor, MI, USA). All samples were run in triplicate with individuals randomly assigned to plates. Inter-assay variation, calculated from a single control triplicate across plates, was 41.8% (n = 3 plates), 29.1% (n = 4), and 11.1%, (n = 2) for 11KT, T, and E2, respectively. Intra-assay variation was 5.2% ± 8.1%, 5.2% ± 6.1%, and 11.1% ± 13.2% for 11KT, T, and E2, respectively.

Because limited plasma volumes precluded measurement of extraction recoveries on all samples, representative recoveries were determined for each GSI-based reproductive group using cold spike recoveries on plasma pools comprised of equal volumes from at least 10 individuals (Bowley et al., 2010; 10 RM, 12 NRM, 10 RF, 17 NRF). The extraction recoveries in each reproductive group were as follows: RM: T = 13%, 11KT = 18%; NRM: T = 49%, 11KT = 56%; RF: T = 90%, E2 = 95%; NRF: T = 31%, E2 = 115%. Although androgen recoveries clearly varied across the representative plasma pools, recoveries were similar between 11KT and T within both RM and NRM pools. The variation in extraction recoveries was addressed in two ways: (1) androgen values were adjusted omnibus to 100% according to the recovery value of the sample's reproductive group plasma pool, and (2) tests for differences in androgen levels across gonad stage were performed separately for 'non-reproductive' and 'reproductive' GSI groups.

Gonad histology

Following GSI measurements, gonadal tissue was fixed in 10% neutral buffered formalin (after Marentette et al., 2010) and transferred to 70% ethanol within 3 months for long-term storage before histological processing (2–8 months). Testes were dehydrated, cleared with xylene, embedded in paraffin wax (Fisher Scientific, www.fishersci.com), and sectioned at 5–7 μm using a microtome (Leica RM2125, RT, Leica Microsystems Nussioch GmbH, Nussloch, Germany). Ovaries were embedded in a glycol methacrylate resin immuno-bed kit (Model #17324, Polysciences, Inc., Warrington, PA) and sectioned at 10 μm. Large gonads were cut in half along the medial–lateral axis before embedding, and multiple sections were taken near the middle of each gonad to minimize potential variation in cell type composition due to section location. Testis and ovary sections were mounted on Superfrost Plus slides (VWR International, LLC, Radnor, PA) and placed on a slide warmer to adhere tissue to slides. Slides were stained with hematoxylin and eosin (Fisher Scientific, www.fishersci.com) and viewed under a light microscope (Leica DME, Buffalo, NY).

Gonad staging criteria followed Brown-Peterson et al. (2011), and were based on the most advanced germ cell stage present (Table 2). Representative light micrographs of ovaries and testes in each stage are shown in Figs. 1 and 2, respectively. We used coalescence of the ooplasm yolk globules as our marker of maturation. Because degree of yolk globule coalescence varies among species (Patiño and Sullivan, 2002), this marker must be validated on a species-specific basis. We found this marker to be valid for work in female round goby since (1) all ovulated oocytes possessed fused yolk globules, (2) there was an increase in GSI in association with yolk globule fusion, and (3) T declined in association with this fusion.

Statistical analyses

All data were examined for normality using Kolmogorov–Smirnoff tests, and both hormone and male GSI data were log-transformed to meet normality assumptions. As indicated earlier, tests for differences in androgen levels across gonad stage were performed separately for 'non-reproductive' and 'reproductive' GSI groups due to differences in extraction recovery results. Differences in hormone level and GSI across

Table 1
Number of fish from each GSI category used for testosterone (T), 11-ketotestosterone (11KT) and 17β-estradiol (E2) steroid hormone assays as well as gonad histology with corresponding GSI and body size metrics (all mean ± SEM).

	T	11KT	E2	Gonad histology	GSI (%)	Mass (g)	Total length (cm)
NRM	22	24	NA	21	0.15 ± 0.03	15.3 ± 1.79	10.8 ± 0.32
RM	18	19	NA	19	1.8 ± 0.10	20.4 ± 2.50	11.8 ± 0.38
NRF	22	NA	22	22	1.9 ± 0.27	6.4 ± 0.60	8.1 ± 0.18
RF	17	NA	19	21	12.9 ± 0.59	6.8 ± 0.38	8.1 ± 0.17

Download English Version:

https://daneshyari.com/en/article/6305374

Download Persian Version:

https://daneshyari.com/article/6305374

Daneshyari.com