Journal of Great Lakes Research 40 (2014) 721–729

Contents lists available at ScienceDirect

Journal of Great Lakes Research

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

Comparison of ovum lipid provisioning among lake whitefish, walleye and northern pike co-habiting in Bay of Quinte (Lake Ontario, Canada)

M.D. Wiegand [a,*], T.A. Johnston [b], L.R. Porteous [a], A.J. Ballevona [a], J.M. Casselman [c], W.C. Leggett [c]

[a] Department of Biology, University of Winnipeg, Winnipeg, MB R3B 2E9, Canada
[b] Ontario Ministry of Natural Resources, Cooperative Freshwater Ecology Unit, Laurentian University, Sudbury, ON P3E 2C6, Canada
[c] Department of Biology, Queen's University, Kingston, ON K7L 3N6, Canada

ARTICLE INFO

Article history:
Received 28 October 2013
Accepted 2 June 2014
Available online 22 July 2014

Communicated by Ed Rutherford

Index words:
Fatty acid
Ova
Pike
Whitefish
Walleye

ABSTRACT

We compared size, total lipid contents, percent of lipids as neutral lipids and fatty acid profiles of ova from Bay of Quinte (Lake Ontario) populations of lake whitefish and northern pike to previously reported data from Bay of Quinte walleye. We also assessed how the relationships between ovum lipid fatty acid composition and maternal size or age varied among these species. Ovum size, total lipid content and percent neutral lipid differed among the three species and in general were not influenced by maternal size or age. The highest percentage of neutral lipid occurred in walleye ova and the lowest in northern pike. Principal components analysis revealed significant separation of fatty acid profiles among the three species, with greater differences in the neutral lipids than in the polar lipids. Lake whitefish were more distinct from the walleye and northern pike than the walleye and northern pike were from each other in the neutral lipids. Lake whitefish ova had higher percentages of eicosapentaenoic acid (EPA) in both lipid fractions than those of the other two species. In direct contrast to the previously observed trends in walleye, percentages of arachidonic acid and docosahexaenoic acid decreased while those of EPA increased with maternal size in the lake whitefish. None of the major fatty acids in northern pike ova varied significantly with maternal size or age. Our study reveals that the Bay of Quinte populations of the three species have different patterns of allocation of fatty acids to ova as they grow and age.

Introduction

Knowledge of the causes and consequences of variability in offspring quality, and ultimately their link to offspring survival, is important to the successful management of exploited fish species and to a broader understanding of their reproductive biology. Fish offspring quality is strongly influenced by the biochemical composition of the egg; of particular significance is the egg lipid fatty acid profile. While manipulation of the fatty acid complement in the maternal diet in culture situations has been shown to influence offspring quality in many studies (Izquierdo et al., 2001; Sargent et al., 2002), the situation in wild fish remains poorly understood. A complicating factor with wild populations is the fact that egg characteristics, including fatty acid profiles and offspring viability, can vary with maternal somatic characteristics, such as body mass, length and age (Berkeley et al., 2004; Johnston et al., 2007). It is therefore worthwhile to investigate possible links between the fatty acid profiles of fish eggs and maternal characteristics.

Lake whitefish, *Coregonus clupeaformis*, are heavily exploited for both commercial and subsistence purposes in the Laurentian Great Lakes. However, the extent, causes and consequences of variability in offspring quality in lake whitefish are largely unknown. The species has experienced nutritional challenges in recent decades as a consequence of changes in the invertebrate communities upon which it feeds (Nalepa et al., 2009; Owens and Dittman, 2003; Pothoven and Nalepa, 2006). The shift in diet from the amphipod *Diporeia* to invasive mussels (*Dreissena* spp.) diminished the condition and energy density of adult lake whitefish (Herbst et al., 2013; Pothoven et al., 2001). Because diet can influence the fatty acid profile of fish eggs (Izquierdo et al., 2001; Sargent et al., 2002), changes in the prey communities could influence offspring viability in this species. However, the relationship between egg composition and offspring viability in lake whitefish is poorly understood. Johnston et al. (2012) described the strategies of resource allocation to reproduction in lake whitefish and compared them to those of sympatric populations of walleye, *Sander vitreus*. Egg size in Bay of Quinte (Lake Ontario) lake whitefish was found to increase with maternal age, but no change in egg lipid concentration was observed.

Lipid and fatty acid allocation to ova in northern pike has been studied from the perspective of energy and fatty acid budgets (Medford and MacKay, 1978; Schwalme and MacKay, 1992; Schwalme et al., 1993).

* Corresponding author.
E-mail address: m.wiegand@uwinnipeg.ca (M.D. Wiegand).

http://dx.doi.org/10.1016/j.jglr.2014.07.004
0380-1330/

Depletion of ovum fatty acid reserves during development in this species has also been examined (Desvilettes et al., 1997b). As is the case with lake whitefish, however, the influence of maternal somatic traits on the biochemical composition of northern pike ova is unknown. Northern pike are of particular interest because of their position in the Bay of Quinte ecosystem (Taraborelli et al., 2010) and also because of the relation between ova production and clearance of lipophilic environmental contaminants in this species (Larsson et al., 1993).

In this study, we determined the dry mass, total lipid content, and percent neutral lipid of ova of lake whitefish and northern pike from the Bay of Quinte in Lake Ontario and compared these findings with those of an earlier investigation of the same traits in walleye (Wiegand et al., 2004). We assessed and compared the influences of maternal size and age on neutral and polar lipid fatty acid composition in these three species. We also employed principal components analysis (PCA) on pooled data from the three species to compare the neutral and polar lipid fatty acid profiles of their ova.

Based on these species' trophic ecologies, we predicted that the neutral lipid fatty acid profiles of the walleye and northern pike ova would be more similar to each other than to those of the lake whitefish. Walleye and northern pike are piscivores while lake whitefish are primarily benthivores. In particular, we predicted comparatively higher proportions of eicosapentaenoic acid (EPA; 20:5(n−3))[1] and lower ratios of docosahexaenoic acid (DHA; 22:6(n−3)) to EPA in the lake whitefish ova, especially in the neutral lipids, compared to those in the other two species because of the high levels of EPA and low levels of DHA or DHA/EPA ratios in many freshwater invertebrates (Bell et al., 1994; Desvilettes et al., 1997a; Ghioni et al., 1996), including *Dreissena* (Czesny et al., 2011; Lazzara et al., 2012). Ovum neutral lipids are more heavily influenced by diet than ovum polar lipids (Almansa et al., 1999; Johnson, 2009; Lewis et al., 2011; Wiegand, 1996), and whole body fatty acid signatures in other Laurentian Great Lakes fishes can be separated on the basis of benthic vs. pelagic diets (Czesny et al., 2011). We further predicted, based on trends previously reported for Lake Ontario walleye (Wiegand et al., 2004, 2007), that the proportions of DHA and arachidonic acid (ARA; 20:4(n−6)) in ovum lipids would increase and proportions of EPA, and thus the EPA/ARA ratio would decline with female length and/or age in lake whitefish and northern pike. These predicted trends are consistent with the hypothesis that larger, older females produce superior ova (Johnston, 1997; Johnston and Leggett, 2002). DHA has critical roles in membrane structure and function, especially in neural tissue (Bell and Dick, 1991, 1993; Bell et al., 1995). ARA enhances a number of physiological functions, including resistance to infection, growth, egg quality, stress response and ion regulation (Ackman and Takeuchi, 1986; Bell and Sargent, 2003) because eicosanoids derived from ARA are more potent than those derived from EPA (Sargent et al., 2002; Tocher, 2003). Finally, we predicted that, as is the case with walleye (Wiegand et al., 2004), the percentages of essential fatty acids, especially DHA, would be more variable in the ova neutral lipids than the polar lipids of the northern pike and lake whitefish because of the greater influence of diet on the neutral lipids (above) and the likely strong selection pressure to maintain DHA in ova polar lipids within a narrow range (Wiegand, 1996).

Materials and methods

Field sampling and fish processing

Mature adult females were sampled at the time of spawning from the Bay of Quinte, Lake Ontario (44° 09′ N, 77° 16′ W). Walleye were sampled in spring 2000 from the Salmon River, a tributary entering the central part of the bay, as described previously (Wiegand et al., 2004). Northern pike were sampled in spring 2003 by trap-net from a nearshore area along the south side of the bay near Big Island. Lake whitefish were sampled in fall 2004, also near Big Island, as described previously (Johnston et al., 2012). Selected females were killed by a blow to the head. Walleye were processed at the sampling site whereas northern pike and lake whitefish were packed on wet ice and taken to the laboratory for processing. Fish processing information was described previously for the walleye (Johnston et al., 2005; Wiegand et al., 2004) and lake whitefish (Johnston et al., 2012). For each female, fork length was measured (±10 mm) and a sample of ova (~60 mL) was stripped from ripe fish or an ovary sample was excised from the ovary mid-section in unovulated fish. The ova or ovary sample was then divided into subsamples that were frozen in small plastic bags at −70 °C for fatty acid analyses and at −20 °C for egg size and total lipid determinations. For walleye only, an additional aliquot of ova was retained fresh for egg size determinations (see below). Aging structures were removed from walleye (opercles) and lake whitefish (otoliths) but not from northern pike.

Laboratory analyses

Egg size was estimated as mean ovum dry mass. For walleye, this was estimated by counting two subsamples (~30 each) of fresh ova into pre-weighed glass vials, oven drying at 60 °C for 24 h, desiccating for 1 h, and weighing (Wiegand et al., 2004). For all three species, ova and ovary samples frozen at −20 °C were freeze-dried for seven days. For lake whitefish and northern pike, two subsamples of 30 freeze-dried ova were weighed to determine mean dry mass per ovum for each female. A comparison of duplicate ovum mass determinations for a subset of the walleye indicated that oven-drying and freeze-drying techniques provided nearly identical estimates of mean ovum size. All freeze-dried ova samples were ground to a powder in a ball mill. Total lipids were then extracted from the powder using a chloroform: methanol procedure as outlined previously (Kaufman et al., 2007; Moles et al., 2008) and expressed on a dry mass basis. Total lipid estimates were performed in duplicate and a third extraction was performed if the coefficient of variation between the first two exceeded 10%. Female ages were determined by counting annuli on opercle bones for walleye (Johnston et al., 2005) and on transverse sections of otoliths for lake whitefish (Johnston et al., 2012).

Determinations of the fatty acid profiles of neutral and polar ova lipids using flame ionization gas chromatography have been reported previously for these walleye (Wiegand et al., 2004). The same procedures were employed for lake whitefish and northern pike ova with the exception that neutral and polar lipid fractions were separated using Sep-Pak 6 cm^3 (500 mg) silica cartridges (Waters Corporation, Milford, MA, USA) rather than hand-made silica gel columns. Fatty acids were expressed on a relative abundance basis (i.e., percentage of total fatty acids). A second set of extractions and separations was performed to determine the percentage of ovum total lipids as neutral lipid using a charring assay (Marsh and Weinstein, 1966) with tripalmitin as the standard. Polar lipid percentage was determined by difference. Percent neutral lipid was also measured gravimetrically for subsamples of each species in order to determine correction factors for the differential responses of total and neutral lipids of each species in the charring assay (Wiegand et al., 2004).

Statistical analysis

All statistical analyses were performed using SAS procedures (SAS Institute Inc., 2009). Proportional data were arcsine-square-root transformed prior to analyses. Relationships between primary ovum traits (size, total lipid content, neutral lipid content) and maternal size and age were first examined by correlation analyses. These traits were then compared among species using Kruskal–Wallis tests. We assessed variation in the neutral and polar lipid fatty acid profiles of both species

[1] Fatty acids are designated a:b(n−x) where a is the number of carbon atoms, b is the number of methylene-interrupted double bonds and (n−x) indicates the position of the first double bond relative to the methyl end of the molecule.