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# Walleye Sander vitreus performance, water quality, and waste production in replicated recirculation aquaculture systems when feeding a low phosphorus diet without fishmeal versus a traditional fishmeal-based diet



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#### ABSTRACT

Walleye Sander vitreus is a popular sport- and food-fish in areas surrounding the Great Lakes. Walleye are mainly provided as food-fish by limited capture fisheries, but have potential for profitable production to market-size in recirculation aquaculture systems (RAS). Walleye are piscivorous with a supposed requirement for fishmeal in artificial diets, thus little information is available regarding the effects of feeding fishmeal-free diets to walleye. During this study, the health and growth performance of juvenile walleye cultured in RAS were compared between groups fed either a traditional fishmeal-based diet (FM) or a low phosphorous, fishmeal-free (FMF) diet. Water quality and waste production rates resulting from feeding each diet were evaluated. The FM diet contained fishmeal, poultry meal, soybean meal, wheat flour, and blood meal proteins; and the FMF diet used poultry meal, wheat flour, soy protein concentrate, and corn protein concentrate proteins. The only lipid source used in the FM diet was fish oil from menhaden, whereas the FMF diet used menhaden oil and poultry oil. Each diet was formulated with a protein: fat ratio of approximately 42/18. Fish (initial weight 85 g fish<sup>-1</sup>) were cultured in 6 replicated RAS for 9 months, each operated with 99.9% water recycle on a flow basis, a mean system hydraulic retention time of 135 days, and a mean feed loading rate of 3.5 kg feed/m<sup>3</sup> of daily makeup water. At study's end, mean weights  $\pm$  standard error of fish fed the FM and FMF diets were  $571 \pm 26$  and  $589 \pm 15$  g, respectively (P > 0.05). Cumulative survival for both diet treatments was  $\geq 98.5\%$ . Average thermal growth coefficient (TGC), condition factor (CF), and feed conversion ratio (FCR) were similar (P > 0.05) for the FM and FMF diets, respectively: TGC was  $0.82 \pm 0.01$  and  $0.83 \pm 0.02$ ; CF was  $1.05 \pm 0.02$  and  $1.03 \pm 0.02$ ; and FCR was  $1.32 \pm 0.02$  and  $1.27 \pm 0.03$ . Water color index and UV transmittance values (P < 0.05) indicated slightly clearer water in RAS where the FMF diet was fed. Total nitrogen (TN) was greater (P<0.05) in the culture water of RAS associated with the FM diet; however, TN production per unit feed was similar between treatments;  $0.031 \pm 0.010 \, \text{kg}$  TN/kg feed for the FM diet and  $0.030 \pm 0.009 \, \text{kg}$  TN/kg feed for the FMF diet. Total phosphorous (TP) concentration in the culture water of RAS associated with the FMF diet was 48% of that measured for the FM diet; TP produced per unit feed reflected this trend,  $0.107 \pm 0.003$ vs.  $0.0049 \pm 0.006$  kg TP/kg feed for the FM and FMF diets, respectively. Average fillet yield (skin and scales on) of fish harvested at the end of the trial was 47-49% (P>0.05). Whole-body and fillet proximate composition was similar between treatments; however, gonadosomatic index and the ratio of omega 6: 3 fatty acids was greater (P<0.05) for walleye fed the FMF diet. This was the first study of its kind to report comparable walleye growth when feeding a specially formulated diet devoid of fishmeal and while culturing this species in RAS. Reduced phosphorous discharge resulting from feeding this fishmeal-free diet formulation increases the feasibility of meeting stringent effluent requirements and possibly reduces the capital investment required for waste treatment.

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#### 1. Introduction

In 2014, the European Union was the largest single market for imported fish and fishery products in the world. The United States followed as the second largest, but qualified as the largest singlecountry importer of seafood in the world (FAO, 2016). This data continued a multi-decade deficit trend that is in sharp contrast to the record trade surpluses that U.S. farmers produce from terrestrial agriculture (USDA-ERS, 2016). A variety of solutions are being explored that could lead to reduced reliance on imported seafood in the U.S, including the research and development of modern production system technologies. In particular, recirculation aquaculture systems (RAS) could be a viable alternative to traditional aquaculture methods due to the flexibility to site these systems as a result of reduced water requirements and the ability to minimize pollution, as well as their capability to control and optimize the culture environment and exclude obligate fish pathogens (Summerfelt and Vinci, 2008; Martins et al., 2010; Davidson et al., 2016a). Recently, RAS have been described as a "maturing" technology (Dalsgaard et al., 2015), which is supported by the growth in the number of commercial facilities using RAS in North America and abroad (Dalsgaard et al., 2013; Summerfelt and Christianson, 2014).

With the adoption of RAS technology, other important industry trends must be considered. For example, the supply of fishmeal from marine resources for use in fish diets remains a concern (Hardy, 2010). Thus, expansion of intensive fish culture in RAS requires feed formulations that utilize less fishmeal, but still result in optimal fish performance and product quality, while also remaining compatible with the production system and increasingly stringent effluent requirements. Traditional diets high in fishmeal can increase discharge of P in fish hatchery effluents (Ketola and Harland, 1993; Hernandez et al., 2004; Brinker and Reiter, 2011; Davidson et al., 2013). Because P is the major nutrient associated with eutrophication of freshwater systems, feeding experiments have been underway for more than 25 years to find means to produce low P diets (Ketola and Harland, 1993). This effort continues to date with a focus on accessing novel diets using alternatives to fishmeal as a protein source (Davis et al., 1995; Luzier et al., 1995; Barrows et al., 2007; Weeks et al., 2010; Hixson et al., 2014). Recent on-site research at the Conservation Fund's Freshwater Institute (TCFFI) demonstrated that a grain-based diet significantly reduced P discharge from RAS (primarily in the system overflow) without compromising rainbow trout performance compared to a traditional fishmeal-based diet (Davidson et al., 2013). Diets that reduce phosphorous, as well as nitrogen, solids, and other nutrient discharge, will help RAS facilities meet regulatory requirements and thus increase the potential for placement of RAS throughout the U.S.

Commercial success of RAS operations also requires the selection of high-value finfish species that can be effectively cultured in these systems (De Ionno et al., 2006). Walleye Sander vitreus represent a niche-market species that has potential for profitable production in RAS. Walleye are a popular sport- and food-fish species in states and provinces bordering the Great Lakes. In the U.S., walleye are only available as food-fish from wild-caught product, predominantly provided by First Nations tribes from the U.S. and Canada (Summerfelt et al., 2010). Walleye are typically cultured to fingerling-size for recreational fishing and for stock supplementation in many Northern states (Summerfelt et al., 2011); however, limited research has been published that has evaluated walleye cultured to food size (≥570 g; Summerfelt et al., 2010), particularly using RAS (Summerfelt and Summerfelt, 1996). However, walleye are congeneric to pikeperch (a.k.a. zander) Sander lucioperca (Stepien and Haponski, 2015) which are intensively cultured in Europe (Steffens et al., 1996; Steenfeldt et al., 2010; Dalsgaard et al., 2013; Pyanov et al., 2014), in a few cases in commercial RAS to

market size up to 1 kg (Dalsgaard et al., 2013). In 2011, total aquaculture production of pikeperch by European countries was 329 metric tons, 68% from Denmark and the Netherlands; one Danish producer of food-size pikeperch using RAS reported plans to expand production to 500 metric tons in a new RAS facility (Steenfeldt et al., 2015). Production of food-sized pikeperch in RAS in Europe points to the potential for walleye production in the United States.

The present study was designed to evaluate production of food-sized walleye in recirculation systems when feeding a low phosphorus diet devoid of fishmeal and compared to that of walleye fed a traditional fishmeal-based diet. The research was focused on evaluating the effects of these diets on walleye performance, water quality, waste production, and food quality metrics using relevant scale (9.5 m<sup>3</sup>) replicated RAS.

#### 2. Methods

#### 2.1. Fish

Walleye used for the study were 45 g, 8 months posthatch to begin. Fish were obtained from the University of Wisconsin-Stevens Point, Northern Aquaculture Demonstration Facility, Bayfield, Wisconsin, USA. A commercially licensed hauling company transported fish from Wisconsin to TCFFI in Shepherdstown, WV, USA. After temperature acclimation, fish were equally distributed into six identical RAS. Following a 3-week interval to accommodate for post-transportation stress and mortality, the number of fish was rebalanced among the 6 RAS (90 walleye/tank) to begin the diet study.

#### 2.2. Recirculation aquaculture systems

Six replicated recirculation aquaculture systems were used (Fig. 1). Each RAS contained a water volume of  $9.5\,\mathrm{m}^3$  and recirculated water through a  $5.3\,\mathrm{m}^3$  dual drain culture tank, a radial flow settler, a microscreen drum filter with  $60\,\mu\mathrm{m}$  screens, a  $1.65\,\mathrm{m}^3$  fluidized sand biofilter ( $3.66\,\mathrm{m}$  tall  $\times\,0.76\,\mathrm{m}$  dia.) loaded with approximately 1 m of static silica sand, a geothermal heat exchanger, a carbon dioxide stripping column, and a low head oxygenator (LHO) (Fig. 1). For purposes of the study, small in-line heaters (Aqualogic, San Diego, CA, USA) were installed within the recycle loop to achieve system water temperatures optimal for walleye culture of  $23-24\,^{\circ}\mathrm{C}$  (Summerfelt and Summerfelt, 1996).

#### 2.3. Water exchange

Recirculating water flows for each RAS were measured and adjusted approximately biweekly using an ultrasonic liquid flowmeter (Digital Flow DF868, GE Panametrics, Waltham, MA, USA). The total (adjusted) recirculating flow was  $343 \pm 0.5$  Lpm for each RAS, including a continuous flow of 228 ± 0.3 Lpm through the fluidized sand biofilter, and  $115 \pm 0.4$  Lpm through the heater which bypassed the biofilter and was directed to the top of the carbon dioxide stripping column (Fig. 1). Makeup water was not continuously added to the systems; therefore, the RAS were operated at a 99.9% recycle rate on a flow basis. Makeup water was automatically added by a float valve located in the pump sump to account for water lost through evaporation and flushing of settled biosolids from the base of the radial flow settler. Radial flow settlers were flushed for 3 s daily (about 18.9 L/day) to remove captured solids from each system. The same settlers were completely drained and sprayed out once weekly, accounting for 341 L/week (90 gal/week) of water exchange. Approximately, 10% of the drum filter backwash spray, which was new spring water, was found to enter the RAS as makeup water due to some spray splashing off of

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