



Tallow in Atlantic salmon feed

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ABSTRACT

Amongst the various available alternative oils, tallow (TAL) has attracted limited research interest and is not commonly used in commercial aquafeeds. This is likely due to concerns of consumers' perception and its high saturated fatty acid (SFA) content, which raises concerns about its digestibility, particularly for species cultured in cold water conditions, such as the Atlantic salmon (*Salmo salar*). Conversely TAL is conveniently priced and has a very low $n-6$ polyunsaturated fatty acid (PUFA) content, which may be beneficial to the final product quality. In many parts of the world, modern salmon aquafeed commonly contains poultry by-product oil (PbO) as the alternative lipid source to replace fish oil (FO). Accordingly, the control diet used for the present experiment contained 75% PbO and 25% FO, as the added dietary lipid sources. Five additional experimental diets were formulated to progressively increase the level of TAL inclusion substituting PbO in 10% increments (10–50%), with a constant amount of FO (25%). A feeding trial was conducted using triplicate groups of Atlantic salmon over a 14 week time period at 10 °C. No difference in growth performance was recorded between treatments, but TAL substitution impacted lipid and fatty acid digestibility. The apparent *in vivo* β -oxidation of SFA intensified with TAL inclusion, whilst $n-3$ PUFA β -oxidation decreased. TAL inclusion also resulted in increased apparent *in vivo* bioconversion of 22:5 $n-3$ to 22:6 $n-3$. This was also reflected in fillet and whole body $n-3$ long chain PUFA (LC-PUFA) composition. TAL inclusion impacted positively on the fillet $n-3/n-6$ PUFA ratio. This study suggests that TAL appears to be a viable alternative oil, with potential for inclusion in commercial aquafeeds.

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1. Introduction

The limited supply of fish oil (FO) and the increasing demand from the growing aquaculture and nutraceutical industries are reducing its availability and driving up prices (Bimbo, 2007; Mathiesen, 2012; Naylor et al., 2009; Tacon and Metian, 2008, 2009). Thus, the incorporation of alternative lipid sources into commercially compounded aquafeeds for many prominent cultured species is now a common practice (Olsen, 2011; Pickova and Morkore, 2007). Salmonid aquaculture is the largest consumers of FO, though commercial aquafeeds only contain approximately 20–35% FO (% of total added lipid), with the other 65–80% being substituted with alternative lipid sources of terrestrial origin (Tacon and Metian, 2008; Turchini et al., 2009).

Extensive research has focused on the viability of FO substitution with various alternative lipid sources in salmonid diets (Bell et al., 2004; Bureau et al., 2002; Codabaccus et al., 2012; Greene and Selivonchick, 1990; Hardy et al., 1987; Menoyo et al., 2003; Torstensen et al., 2004a; Zheng et al., 2004). In Europe, vegetable oils have been the primary alternative sources used in aquafeeds, likely because of industry's concerns of consumers' perceptions regarding the speculated health hazards linked with the use on terrestrial animal by-products in

animal feeds. However, in North and South America and in Oceania rendered animal fats have been and currently are commonly used (Turchini et al., 2011b), with the most frequently utilised alternate lipid source of animal origin being poultry by-product oil (PbO) (Bureau and Meeker, 2011).

PbO has been regarded as a good alternative lipid source, proven to produce good growth performance, high feed palatability and nutrient digestibility with a final product of acceptable quality (Bowyer et al., 2012; Hatlen et al., 2013; Liu et al., 2004; Rosenlund et al., 2001; Turchini et al., 2003, 2013). However, with the steadily increasing demand for PbO, its availability is currently diminishing and its price is increasing. This, coupled with recent changes in European legislation facilitating the utilisation of some terrestrial animal products in aquafeeds (Regulation (EC) 1069/2009), suggests that soon there will likely be an increase in the use of rendered animal fats, and particularly PbO, which will likely soon result in shortages in its supply and further price hikes. The result of this reduction in PbO economic viability is that, paradoxically, there is now interest in finding potential alternatives to the alternative lipid source PbO, currently used in many countries.

One lipid source that is envisaged to be a possible substitute for PbO is tallow (TAL; rendered beef and/or lamb fat). TAL is abundantly produced globally (6.69 million tonnes in 2011) (FAO, 2012), and, even if only few studies focusing on TAL inclusion in aquafeed are available, they suggest it may have good potential (Bureau et al., 2008; Gause

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and Trushenski, 2013; Guderley et al., 2008; Hardy et al., 1987; Xue et al., 2006b). Nevertheless, concerns over its utilisation have also been made (Bureau and Meeker, 2011; Trushenski and Lochmann, 2009), fundamentally for its overall fatty acid composition and the possible resulting effects on lipid digestibility. In fact, the digestibility of lipids can be influenced by the lipid source, its fatty acid composition and more specifically the fatty acid chain length, degree of saturation and the resulting melting point. Thus, the digestibility of a dietary lipid source is fundamentally influenced by its relative amounts of saturated fatty acids (SFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) (Bureau and Meeker, 2011). Additionally, lipid digestibility can also be influenced by water temperature, as shown by Cho and Kaushik (1990); when fed to rainbow trout (*Oncorhynchus mykiss*), lipid sources high in MUFA and PUFA produced higher apparent digestibility coefficient (ADC) values than lipid sources high in SFA, and the decreases in ADC due to reductions in water temperature were disproportionately greater for high SFA lipid sources. TAL is high in SFA relative to other lipid sources (including PbO), and this raises concerns over nutrient utilisation and growth performance outcomes as this could negatively affect lipid digestibility, which could also be exacerbated at low temperatures (Hua and Bureau, 2009; Ng et al., 2007).

From a final product quality point of view, whilst the total content of health promoting (Arab-Tehrany et al., 2012; Williams and Burdge, 2006) omega-3 long chain PUFA ($n-3$ LC-PUFA) in cultured fish is only minimally affected by the type of alternative oil used to substitute FO (Greene and Selivonchick, 1990; Izquierdo et al., 2005; Thomassen and Rosjo, 1989; Torstensen et al., 2004b; Xue et al., 2006a), the $n-3/n-6$ PUFA ratio can be largely affected by the $n-6$ PUFA content of the alternative oil used (Turchini et al., 2013). An unbalanced $n-3/n-6$ PUFA ratio in fish fillet is considered to be detrimental to the overall nutritional quality of the final product (Pickova and Morkore, 2007; Rosenlund et al., 2011), impacting directly the potential health benefits delivered to the consumer (Simopoulos, 2008, 2011). Current commonly utilised alternative oils, such as soybean, sunflower and PbO, are relatively rich in $n-6$ PUFA, and in comparison TAL has very little. Thus, the limited content of $n-6$ PUFA in TAL could be considered advantageous from a final product quality standpoint.

This study focused specifically on better understanding the viability of substituting PbO with TAL in commercial-like aquafeed for Atlantic salmon (*Salmo salar*) reared at a low temperature in respect to growth, feed utilisation, fatty acid metabolism and final product composition. If TAL is proven to be a viable alternative to PbO, this will assist the aquaculture industry in formulating new and more economically viable aquafeed, and thus allowing the industry to further expand.

2. Materials and methods

2.1. Ethics statement

All animals and procedures used in this experimentation were approved by the Deakin University Animal Welfare Committee (Number A46-2011). All possible steps towards minimizing animal suffering were taken.

2.2. Animals, experimental design and sampling

Atlantic salmon (*S. salar*) sourced from a private aquaculture farm (Yarra Valley Salmon, Thornton, Victoria, Australia) were transported to Deakin University's Aquaculture Research Facility at the Warrnambool campus. Fish were acclimated to the experimental conditions for 2 weeks and maintained on a commercial salmon diet (Ridley), at a fixed ration of 2% of their body weight daily preceding the commencement of the experiment. The experiment was conducted in a freshwater, closed loop, thermostatically (10.0 ± 0.2 °C) and photoperiod (12L:12D cycle) controlled recirculating aquaculture system. The system consisted

of 18 (1000 L) rearing tanks with biological and physical filtration (drum filter with 60-mm screen; Hydrotech, Vellinge, Sweden) and ultra violet disinfection. The system was maintained on a 12:12 hour light:dark cycle. Temperature was set at 10 °C using a chilling/heating unit, and remained at 10.0 ± 0.2 °C for the duration of the trial. The levels of metabolic waste, total ammonium and nitrite, were monitored bi-weekly using Aquamerck test kits (Merck, Darmstadt, Germany), and they remained below 0.50 and 0.05 mg L, respectively, for the duration of the trial. Dissolved oxygen was measured daily, recording values above 7 mg L for the duration of the trial.

Six iso-proteic and iso-lipidic experimental diets were formulated to contain 220 mg/g of lipid and 490 mg/g of protein, varying only in lipid source (Table 1). Three lipid sources, fish oil, poultry by-product oil and tallow (FO, PbO and TAL, respectively), were used and blended to obtain six experimental diets characterised by increasing percentage of TAL inclusion. The first diet consisted of 25% FO and 75% PbO as the added dietary fat, and this was regarded as representative of current practices in the aquaculture industry and therefore considered the control diet (CD). In the 5 remaining experimental diets the inclusion level of FO remained constant (25%), whilst increasing amounts of PbO were replaced with TAL to achieve a decreasing percentage of PbO and an increasing percentage of TAL inclusion. These five diets had a TAL inclusion rate of 10, 20, 30, 40 and 50% of the total added dietary fat and were named as follows: 10TAL, 20TAL, 30TAL, 40TAL and 50TAL, respectively. Thus, the only difference between experimental diets was the relative percentage of PbO and TAL inclusion. The experimental diets were manufactured and stored as previously described (Brown et al., 2010).

Following the acclimation period, an initial sample of 10 fish were euthanized in excess anaesthetic (AQUI-S, 0.5 ml/l) and stored at -20 °C until subsequent analysis. Three hundred and sixty fish (mean weight 138 g) were randomly distributed into 18 tanks (20 fish per tank). Each of the tanks was then randomly assigned one of the six

Table 1
Formulation and proximate composition of the experimental diets.

	Experimental treatments ¹					
	CD	10TAL	20TAL	30TAL	40TAL	50TAL
<i>Diet formulation (g/kg)</i>						
Fish meal ²	427.7	427.7	427.7	427.7	427.7	427.7
Wheat ²	115.0	115.0	115.0	115.0	115.0	115.0
Soybean protein concentrate ²	100.0	100.0	100.0	100.0	100.0	100.0
Blood meal ²	60.0	60.0	60.0	60.0	60.0	60.0
Poultry meal ²	50.7	50.7	50.7	50.7	50.7	50.7
Wheat gluten 75% ³	50.0	50.0	50.0	50.0	50.0	50.0
Pregelatinized maize starch ²	40.0	40.0	40.0	40.0	40.0	40.0
Vitamin & mineral premix ⁴	6.0	6.0	6.0	6.0	6.0	6.0
Monosodium phosphate ²	4.7	4.7	4.7	4.7	4.7	4.7
Celite ⁵	5.0	5.0	5.0	5.0	5.0	5.0
DL-Methionine ²	0.9	0.9	0.9	0.9	0.9	0.9
Stay C-35% Vit C ²	0.6	0.6	0.6	0.6	0.6	0.6
Fish oil ²	43.2	43.2	43.2	43.2	43.2	43.2
Poultry oil ²	129.6	112.3	95.0	77.8	60.5	43.2
Tallow ⁶	0.0	17.3	34.6	51.8	69.1	86.4
<i>Proximate composition (mg/g)</i>						
Moisture	45.3	45.1	38.9	42.6	48.7	46.0
Protein	488.9	500.1	499.2	481.4	500.4	493.5
Lipid	204.3	214.8	219.9	201.4	198.0	199.8
Ash	89.1	86.5	88.6	88.5	88.5	90.1
Nitrogen free extract	172.3	153.4	153.4	186.2	164.5	170.5
Energy (kJ/g) ⁷	22.6	22.9	23.1	22.5	22.5	22.5

¹ Experimental diet abbreviations and oil sources: all diets contained 25% fish oil. CD (Control Diet), 75% poultry by-product oil; 10TAL, 10% substitution with tallow; 20TAL, 20% tallow; 30TAL, 30% tallow; 40TAL, 40% tallow; 50TAL, 50% tallow.

² Ridley Agriproducts, Narangba, Queensland, Australia.

³ Agrifood Ingredients, Kew East, Victoria Australia.

⁴ DSM Nutritional Products, Wagga Wagga, New South Wales, Australia.

⁵ Merck KGaA, Darmstadt, Germany.

⁶ The Midfield Group, Warrnambool, Victoria, Australia.

⁷ Calculated on the basis of 23.6, 39.5 and 17.2 kJ/g of protein, fat and carbohydrate, respectively.

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