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Improved astaxanthin utilization in Atlantic salmon fed sulphuric acid extracted fish bone compounds

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In Atlantic salmon, digestive and absorptive processes, and metabolic turnover of astaxanthin (Ax), influence the utilization and flesh deposition of carotenoids. Usually, less than 10 % of ingested Ax is retained in the flesh of Atlantic salmon. In salmon 0^+-smolt fed sulphuric acid extracted fish bone compounds, significantly increased plasma, liver and whole body Ax concentrations were observed. A high Ax deposition rate in the flesh may be a result of increased Ax uptake in the intestine, decreased metabolic turnover of digested Ax or increased Ax uptake in the tissues. All of these potential altered Ax uptake and distribution mechanisms were studied in large salmon (1.7 kg), and the results used to understand the physiological impacts of sulphuric acid extracted fish bone compounds on Ax utilization.

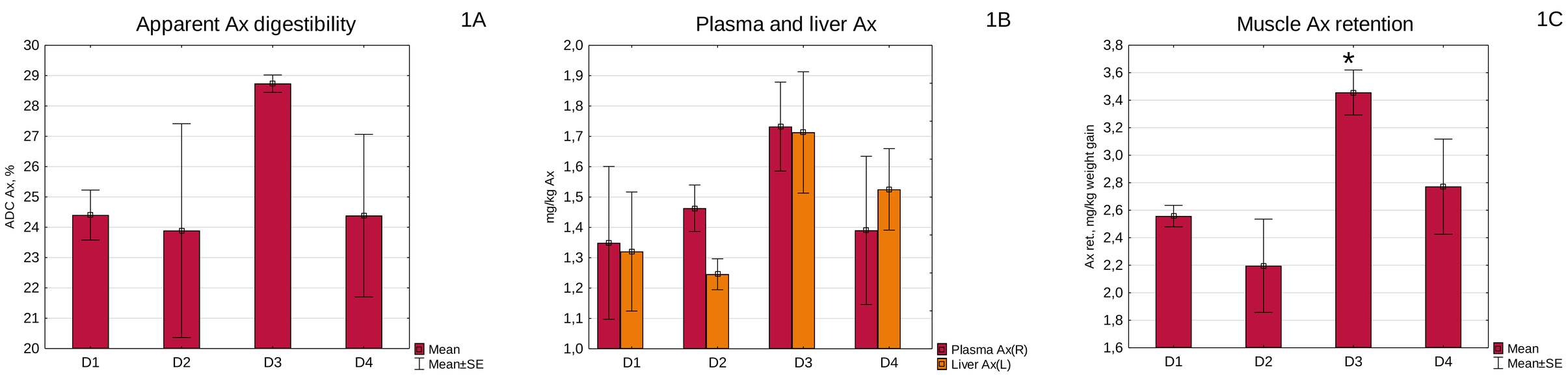


*Fig. 1 Increased astaxanthin (Ax) digestibility (1A), followed by a tendency (ns) towards increased plasma and liver Ax deposition (1B), may explain the significant improved muscle Ax retention (1C) observed in Atlantic salmon fed a sulphuric acid (H_2SO_4) extracted fish bone ingredient provided as a feed additive at 4.2 % (D3), compared to fish fed a control diet without the fish bone ingredient (D1). Mean values and SEM for each diet are presented, n=3. Significant differences are marked with an asterix, *, P < 0.05.*

Materials and Methods

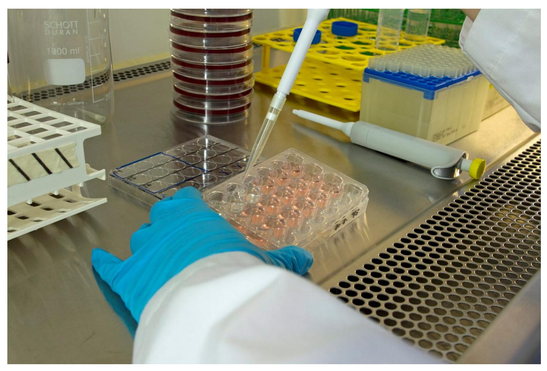
Atlantic salmon (1.7 kg) were reared in 12 net-pens and fed a practical formulated control diet (D1), experimental diets added sulphuric acid (H_2SO_4) extracted fish bone hydrolysate (FBH) at respectively 2.1 % (D2) and 4.2 % (D3), and a diet added K_2SO_4 to study potential impacts of the chemicals used for mineral extraction, for a feeding period of 78 days. All diets were balanced to meet dietary phosphorus (P) requirement (8 g kg^{-1} P). An *in vitro* cell culture study with hepatocytes collected from salmon (600 g) was performed to evaluate effects of the FBH ingredient on hepatic Ax uptake.

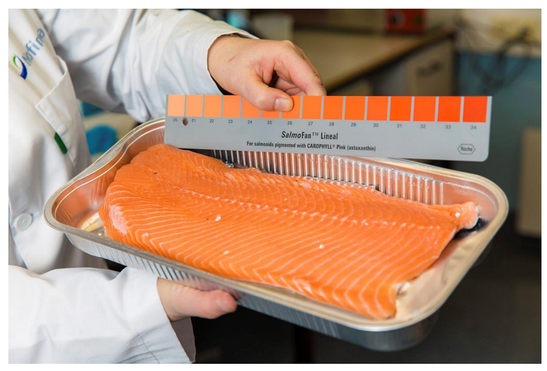
Results

The body weights increased from 1.7 to 2.5 kg and mean specific growth rate (SGR) was 0.52 ± 0.03 % for all diets. The FBH ingredient significantly increased specific Ax retention in muscle by 35 % in fish fed D3 (P < 0.05), as compared to fish fed D1 (Fig.1 C). A tendency towards improved Ax digestibility (Fig.1 A), and increased plasma and tissue Ax deposition (ns) was found in fish fed D3 (Fig.1 B), compared to fish fed D1. Reduced metabolic turnover of Ax was indicated, with about 10 % more of absorbed Ax retained in the flesh of fish fed D3. The *in vitro* study showed no significant impacts of the fish bone compounds on hepatic Ax uptake (P < 0.05), Table 1.

	Control	FBH Blue whiting	Ax	FBH Blue whiting + Ax	ANOVA P < 0.05
Ax uptake					
µg Ax/µg protein	nd	nd	$5.2*10^{-3}$ ($0.5*10^{-3}$)	$4.5*10^{-3}$($0.6*10^{-3}$)	ns
% of added Ax	nd	nd	53.9 (4.7)	46.3 (6.0)	ns
Catalase (U/g protein)	0.79 (0.15)	0.90 (0.18)	0.70 (0.19)	0.94 (0.19)	ns

Table 1 Astaxanthin (Ax) uptake and Catalase enzyme activity measured in salmon hepatocytes. The growth media (Control) consisted of 2 % foetal calf serum, 1 % bicarbonate, 5mM Hepes and 1 % PenStrep, and was added acid extracted fish bone hydrolysate (FBH) produced from Blue whiting (FBH Blue whiting), Ax or FBH Blue whiting + Ax.





Nofima AS, illustration pictures

Improved tissue Ax deposition in A. salmon 0^{+}- smolt

Preliminary results in A. salmon 0^+-smolt showed significant improved tissue Ax deposition in fish fed H_2SO_4 extracted fish bone compounds. The fish were fed a control diet with inadequate available P (D1: 0.33 g kg^{-1}), adequate available P from inorganic P (D2: 7 g kg^{-1}) or from HCl (D3: 5 g kg^{-1} diet) and H_2SO_4 (D4: 7 g kg^{-1}) extracted fish bones. The growth was significantly highest in fish fed D2 and D4 (P < 0.05). Diet D4 increased Ax in plasma and liver with 55 and 29 % (P < 0.05), and in whole body with 22 % (ns), as compared to Diet 2, Fig. 4. Diets D1 and D3 resulted in low plasma and tissue Ax deposition.

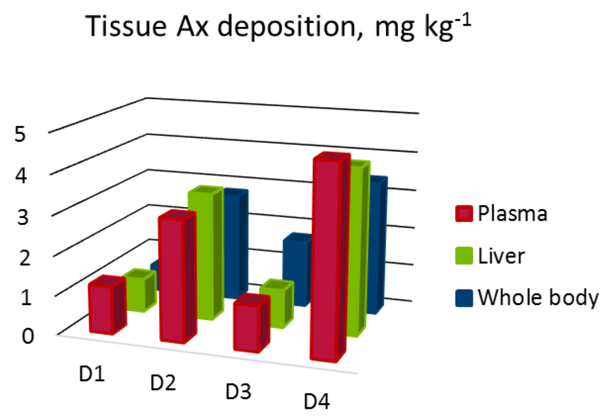


Fig. 4 Significant improved plasma, liver and whole body Ax in 0^+-smolt fed H_2SO_4 extracted P from fish bones (D4), compared to inorganic P (D2) providing similar available P.

Summary and conclusion

- Sulphuric acid (H_2SO_4) extracted fish bone compounds significantly increased muscle Ax retention by 35 % in Atlantic salmon (2.5 kg), possibly explained by a tendency towards improved Ax digestibility followed by increased circulating Ax and tissue Ax deposition.
- Slightly reduced metabolic turnover of Ax was indicated *in vivo*, while no effect of H_2SO_4 extracted fish bone compounds on hepatic Ax uptake was found *in vitro*.
- Results obtained in Atlantic salmon reared at different life stages indicate that dietary addition of H_2SO_4 extracted fish bone compounds may improve Ax utilization.

Acknowledgment
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