# EFFECTS OF TEMPERATURE, SALINITY AND DIET ON FATTY ACID COMPOSITION OF THE RAGWORM *HEDISTE DIVERSICOLOR* (OF MÜLLER, 1776) (ANNELIDA: NEREIDAE)

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## Introduction

There is an urgent need in food and feed production to change from linear systems towards circular, recycling based solutions. Many wastes or rest raw materials from linear systems are in fact valuable side streams containing precious compounds. Sludge from land-based aquaculture is such a side stream which comprise faeces and uneaten pellets. A second readily available side stream is the solid phase remaining after biogas production commonly known as "solid biogas digestate" (SBD). Both are nutrient- and energy rich and require handling at the production site, hence the question arises if these side streams can be utilized in a more sustainable way. The common ragworm *Hediste diversicolor* is an omnivorous, burrowing polychaete showing potential as an extractive species in IMTA systems. Both SBD and aquaculture sludge have proven to be suitable feeds for polychaetes, but growth rates can be lower than for worms fed high quality diets such as formulated fish feed (Wang et al., 2019a). It has recently been shown that this species has the capacity for endogenous production of omega-3 long-chain polyunsaturated fatty acids (Kabeya, et al., 2020), however it is yet not understood how environmental cues affects this ability. We conducted two sets of experiments to assess the combined effects of diet, temperature and salinity on total body fatty acid composition in *H. diversicolor* juveniles.

## Materials and Methods

Polychaetes (*H. diversicolor*) were collected at low tide at the mud flat of Leangen Bay, Trondheim, Norway (63°26′24.5″N, 10°28′27.7″E). To investigate the effects of diet and temperature on fatty acid (FA) composition of *H. diversicolor*, worms were fed mixes of solid biogas digestate (SBD) and salmon aquaculture sludge (SS) along a 4-step feed gradient ranging from pure SBD to pure SS, and a 5-step temperature gradient ranging from 5.8 to 17.1 °C, for 15 days, using fish feed (FF) as a control. A second experiment was conducted to investigate the effects of salinity and temperature on the same variables. Here, the worms were fed the diet which yielded the highest growth rates in the first experiment (33:66 % SS:SBD) along 5-step salinity- and temperature gradients ranging from 5 to 40 ppt and 7.7 to 17.9 °C, respectively, for a duration of 28 days. In both experiments, worms were fed isonitrogenous diets equalling 30 % of the worms' total body nitrogen per day (Wang et al., 2019). Both experiments were conducted in a temperature gradient table modified after Thomas et al. (1963) using a 18h:6h light:dark cycle. Worms (n=7-8) were stocked in glass beakers (800 mL) containing an eight cm thick layer of sand and filled with sand- and bag-filtered (1 µm) seawater from the Trondheim fjord collected at 60 m depth. The worms were allowed to evacuate their guts in clean seawater for minimum 4 hours before each sampling and weighing. Water exchange and feeding was conducted every second day. Data analyses were performed using the inbuild statistical package of SigmaPlot v.14.5. Principal component analysis (PCA) on fatty acid composition of polychaetes was performed using PRIMER 5.2. Fatty acid levels were expressed as % of total fatty acids and were arcsine transformed before entering the PCA.

## Results and Discussion

The main differences between the FF and side stream diets were the high concentrations of EPA and DHA in fish feed. EPA accounted for 15% of total fatty acids in FF, and about 1% in both, AS and SBD. DHA accounted for 8.5% of total fatty acids in FF, while it accounted for 2% in AS and 3.5% in SBD. Further, AS was richer in 16:0 (28% *vs.* 18% in SBD and 20% in FF), and it was also richer in 18:1n-9 (25% vs. 21% in SBD and 13% in FF). In sum, FF contained a lower percentage of saturated fatty acids (SFA) compared to AS and SBD (32% *vs.* 41%, respectively), and in turn a higher percentage of unsaturated fatty acids. No temperature- nor salinity-driven segregation patterns could be identified in the FA profiles of the polychaetes, however a clear diet-driven segregation was found between worms fed lipid-rich fish feed (control) and lipid-poor SS and SBD diets. The fatty acid composition of polychaetes fed side stream diets showed high concentrations of EPA (14–19%) comparable to the polychaetes fed fish feed (13%). Further, the percentage of PUFA was high in both (38–40%) also comparable to those fed fish feed (46%). However, DHA was higher in polychaetes fed fish feed (5%) than in polychaetes fed aquaculture sludge and SBD diets (1–2%). The pronounced differences in fatty acid composition found in the different feeds were not as pronounced in the polychaetes reared on these diets (Fig. 1). We here demonstrated that short-term (≤4 weeks) alterations of environmental parameters have neglectable effects on the fatty acid profile in wild caught *H. diversicolor* juveniles. Hence, our results indicate that the major influence on fatty acid composition in *H. diversicolor* is diet.

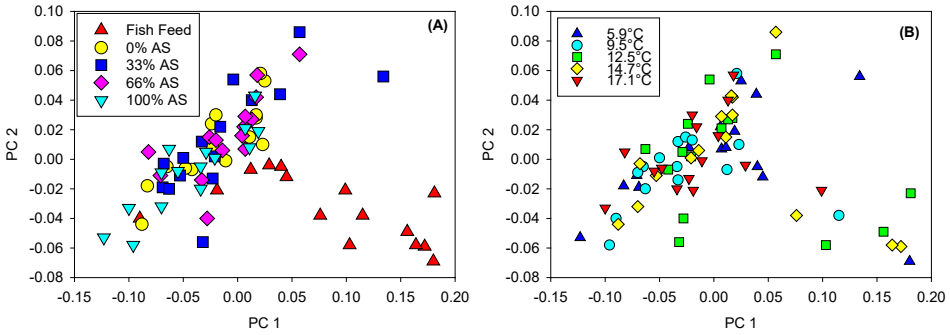


**Figure 1:** Principal Component Analysis (PCA) on total fatty acids composition of *H. diversicolor* fed different diets at different temperature visualized by (A) feed type (% AS in the diet or fish feed and (B) by the five different rearing temperatures (°C). PC1= 56%, PC2= 14%


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## References


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