

**IMPACT OF TEMPERATURE AND TRIMETHYLAMINE OXIDASE ON
TRIMETHYLAMINE OXIDE ACCUMULATION IN SMELT**

(*Osmerus mordax*)

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EXTENDED ABSTRACT ONLY – DO NOT CITE

Introduction

Smelt accumulate trimethylamine oxide (TMAO) when held at ambient winter sea temperatures (approximately 5°C to -1°C) under laboratory conditions (Treberg et al., 2002). This is similar to what has been observed in wild caught smelt from fall and winter conditions (Raymond, 1994). Smelt held at approximately 5°C, after the initial increase in TMAO, subsequently suppressed the TMAO accumulation response; however, TMAO levels remained above what may be considered 'normal' for teleosts for several months (Treberg et al., 2002). Although hypothesised to act as a colligative antifreeze (Raymond 1994), TMAO accumulation began by 5°C. To determine what may be the thermal threshold for the seasonal TMAO accumulation in smelt, a similar experiment to Treberg et al. (2002) was undertaken with an initial sampling temperature of 9°C. Fish were also transiently cooled to 5°C, subsequently rewarmed and maintained at 9°C to determine if a higher temperature would result in further suppression of TMAO accumulation.

The mechanism by which smelt elevate TMAO is unknown. Tissues were examined for trimethylamine oxidase (TMAoxi), the enzyme likely involved

with endogenous synthesis of TMAO. Levels of this enzyme were found to be very high in the kidney, thus kidney TMAoxi activity was followed through the above mentioned experiment to determine if levels of this enzyme may play a role in TMAO accumulation.

Results and Discussion

The relationships between plasma TMAO concentration and kidney TMAoxi activity as well as temperature profiles for the ambient and warm smelt groups are included within Figure 1 A, B and C respectively. Plasma TMAO began to increase by the second sample period in ambient held fish, indicating that if there is a thermal trigger for TMAO accumulation it is likely between 7 and 9°C; however, there was a transient drop in temperature to 4.5°C that can not be discounted from having ‘triggered’ a response. When ambient temperatures approached 5°C, and plasma TMAO concentrations were significantly elevated over the initial sample, a group of fish were switched over to heated seawater maintained at about 9°C (Fig. 1A, and C). Plasma TMAO decreased in the warm group of fish and returned to levels not different from the initial sample period within a few weeks. TMAO concentration in the plasma of the ambient fish was elevated over the warm group at all sample periods. Interestingly, plasma TMAO dropped in the ambient fish at the last sample period when ambient temperatures went from approximately 1 to 2.5°C. Thus, if there is a thermal trigger for TMAO accumulation, the threshold temperature for the decrease is different from the temperature for increase. Of note, other potential seasonal triggers such as photoperiod have yet to be ruled out.

The kidney was the only tissue found to have appreciable TMAoxi activity. No difference was found between warm and ambient kidney TMAoxi activity (Fig. 1 B). Furthermore, no clear trend or correlation with plasma TMAO concentration or temperature was found. Superficially, there was a trend of increased TMAoxi when ambient temperatures were below 0°C; however, no statistical significance was revealed. There was a significant decrease in kidney TMAoxi that precluded the plasma TMAO drop in the ambient smelt but this significance was lost during the sample when TMAO actually decreased. As such, it appears that levels of TMAoxi do not play a key role in the accumulation of TMAO in smelt and that constitutive levels are sufficient for any synthesis that does occur.

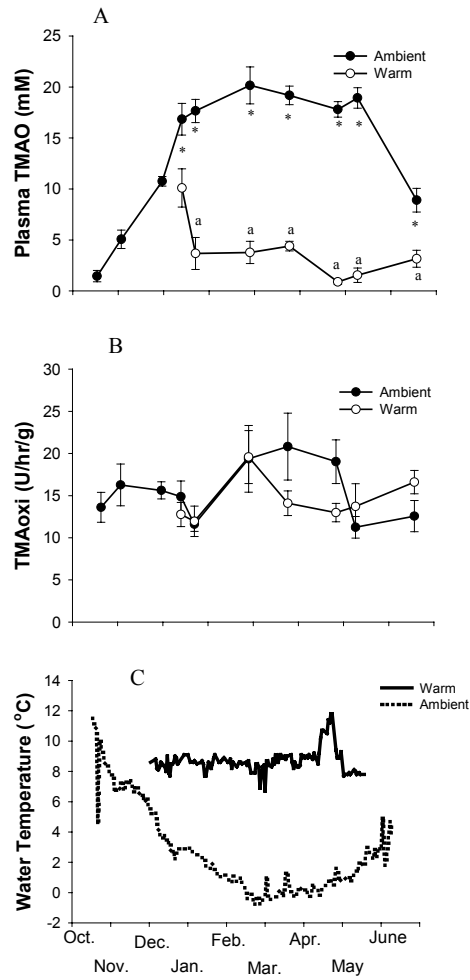


Figure 1: (A) Plasma TMAO concentration (mean±SEM), (B) kidney TMAoxi activity (mean ± SEM), and (C) temperature profiles for ambient and warm smelt groups. *, significant difference between ambient and warm; a, no significant difference from initial October value.

In conclusion, TMAO accumulation in smelt does have a temperature dependent component, possibly for 'triggering' the mechanism involved between 9 and 4.5°C and subsequently turning off the accumulation mechanism as temperatures rise above 0°C. Maintaining fish at temperatures well above ambient winter temperatures suppresses the accumulation mechanism; however, temperatures above 5°C are required for complete suppression of TMAO accumulation. Although smelt have the metabolic machinery for synthesis of TMAO from trimethylamine, levels of TMAoxi do not change during TMAO accumulation and thus constitutive levels appear to be sufficient for any endogenous synthesis that does occur.

References

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- Treberg, J.R., Wilson, C.E, Richards, R.C., Ewart, K.V. and W.R. Driedzic. 2002. The freeze-avoidance response of smelt *Osmerus mordax*: initiation and subsequent suppression of glycerol, trimethylamine oxide and urea accumulation. *J. Exp. Biol.* 205:1419-1427.

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