Estimating biovolume of the copepod *Tigriopus brevicornis* from automated imaging in light of changes in shape; a prelude to DEB-based modelling*

DEBtox Research

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Introduction

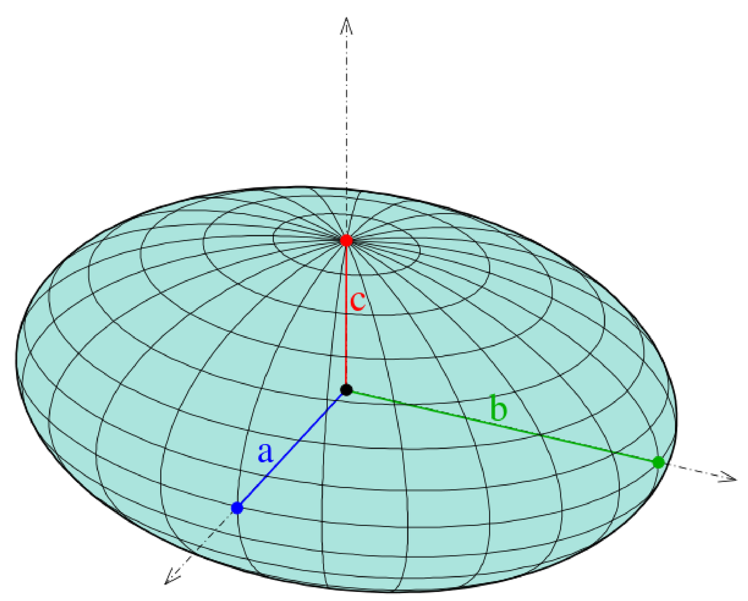
Dynamic Energy Budget (DEB) modelling offers advantages for the analysis and prediction of stressor effects. However, bio-energetic analysis of growth and development requires accurate estimation of an animal's biomass or biovolume. Length measures are often easy to obtain, but are only a useful proxy if the organism retains a constant shape.

Copepods offer several challenges. They have a major change in morphology at metamorphosis from nauplius to copepodite, and for some species, more gradual changes in shape within the nauplius or copepodite phases. Before we can analyse effects of stressors with DEB models, we need to establish rules for estimation of biovolume.

Need for auxiliary hypotheses

DEB models specify body size in terms of biomass or biovolume, often expressed as volumetric length (the cubic root of body volume). Automated image analysis is highly advantageous as it allows following individuals over time, with high temporal resolution. However, it does not provide a measure of biovolume. The measured properties are thus not directly comparable to the modelled ones, requiring auxiliary hypotheses. Here, we assume that the copepod's volume can be approximated by a general ellipsoid (Fig. 1).

Fig. 1. Generalised ellipsoid. This image from https://commons.wikimedia.org/wiki/File:Ellipsoide.svg .



Simplified model

Copepods require modifications of standard DEB models, especially due to their determinate growth (Jager *et al*, 2017). To quantify the growth pattern, we here resort to a simple descriptive model with linear growth, and stops at metamorphosis and adulthood.

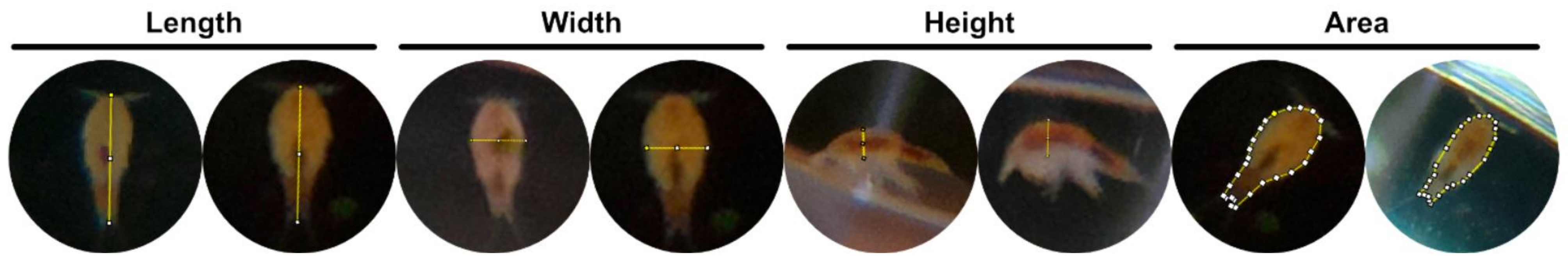


Fig. 2. Example of biometrical measurements on copepodites. ↑

Fig. 3. Fit of the simplified growth model to the volumetric length estimates for a single individual copepod over its life cycle. →

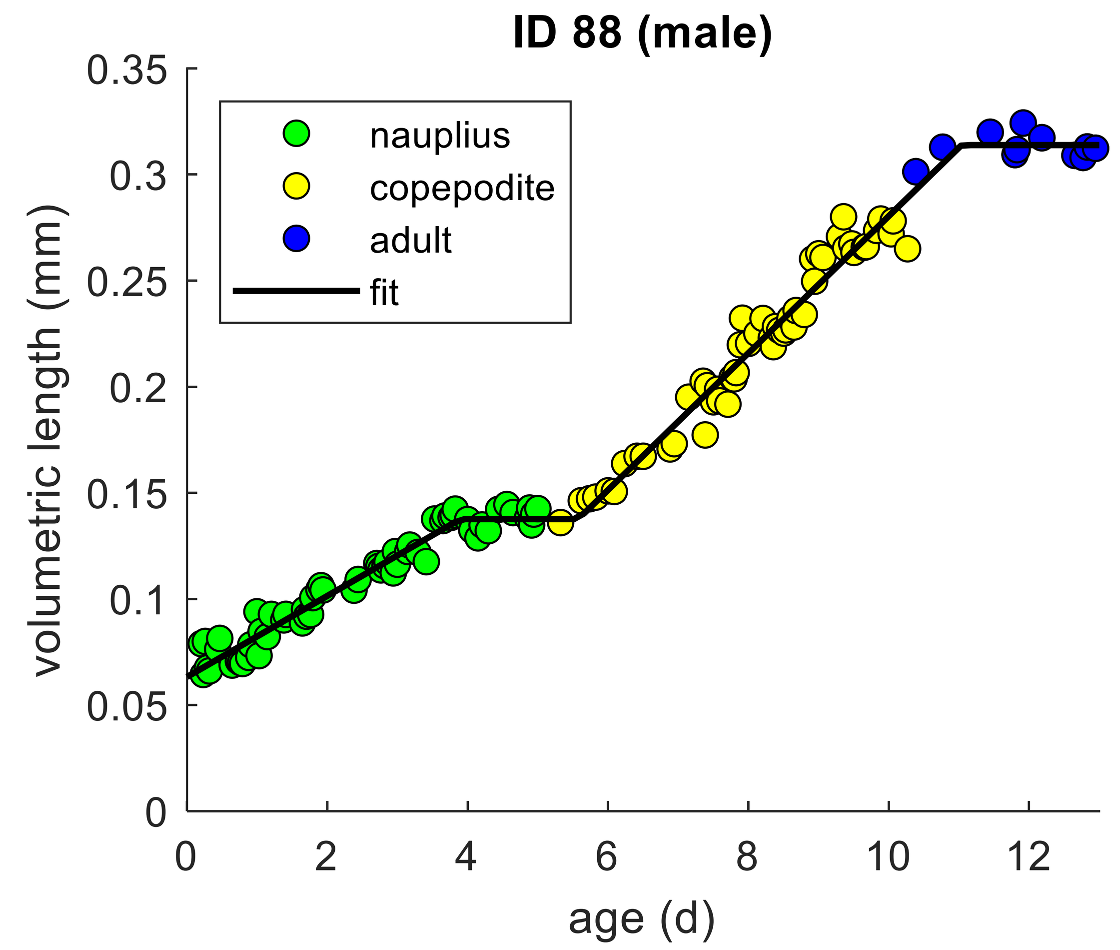


Fig. 4. Model parameters from the fits for several individual copepods. Error bars are 95% confidence intervals. ↓

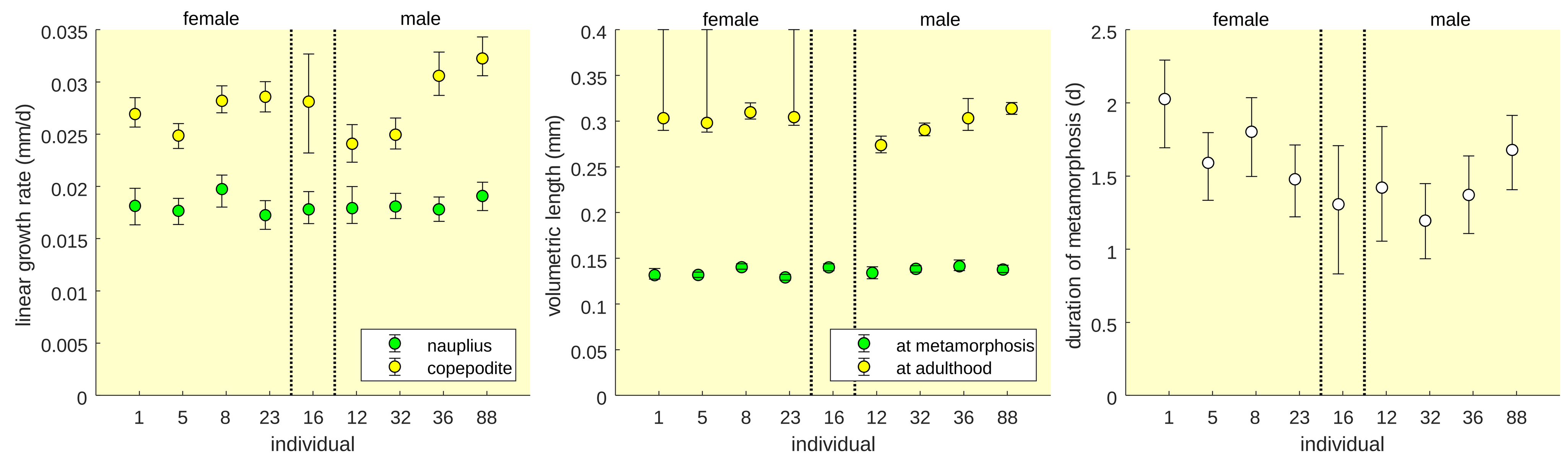
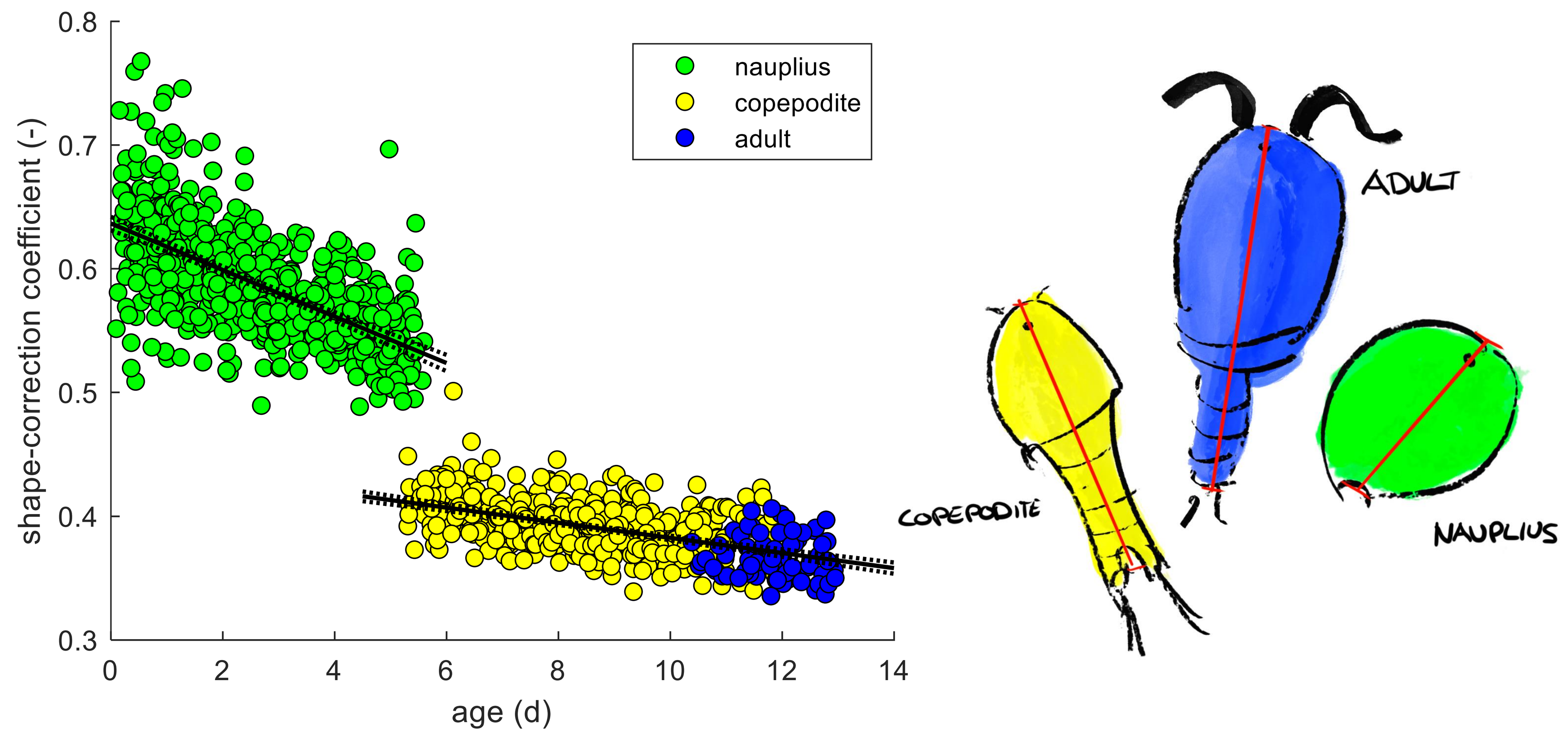


Fig. 5. Shape coefficient as function of age. Lower values imply more elongated body shape. →



Measurements

We re-analysed measurements from automated imaging on *Tigriopus brevicornis* (Heuschele *et al*, 2019), where individuals were followed over their entire development (nauplius stage I to adult copepodite stage VI). From the images, we obtained total body length, body width, and projected area (Fig. 2). In addition, some images allowed estimation of the organism's height.

We first established the general width:height ratio of the animals: for nauplii, 1.82 (SE 0.034) and for copepodites 1.42 (SE 0.017). All stages are thus significantly flattened, but the nauplii more so than the copepodites. Next, we approximate the volume of the copepods by an ellipsoid, using these ratios and measurements for body length and projected body area.

Results and discussion

Figure 3 shows an example fit for a single individual on estimated volumetric length over time. Growth is linear in between two growth stops. Figure 4 shows the model parameters for a range of individuals. Copepodites grow at a faster rate than nauplii, but there are no obvious differences between the sexes. Growth stops around metamorphosis for several days.

Figure 5 shows shape coefficients (volumetric length divided by total body length) for all individuals. *T. brevicornis* changes shape abruptly at metamorphosis, but within the nauplius and copepodite phases there is a more gradual change in shape.

Conclusions

- Body length is a poor proxy for body volume; volumetric length provides a more realistic picture of growth.
- The results will help us analyse automated imaging results in later stages of the project, studying multi-stress by combined predation risk and copper exposure.
- For DEB-based modelling, the growth stop at metamorphosis and the differential growth of nauplii and copepodites are challenging.

References

Heuschele *et al* (2019). An affordable and automated imaging approach to acquire highly resolved individual data - an example of copepod growth in response to multiple stressors. PeerJ 7, e6776.

Jager *et al* (2017). Modelling the dynamics of growth, development and lipid storage in the marine copepod *Calanus finmarchicus*. Mar Biol 164:15.

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