Journal of Plankton Research Vol. 13 no. 4 pp. 683–689, 1991

Development, growth and egg production of the two copepod species *Centropages hamatus* and *Centropages typicus* in the laboratory

Michael Fryd, Ole Henrik Haslund and Ole Wohlgemuth

Danish Institute for Fisheries and Marine Research, DK-2920 Charlottenlund and Marine Biological Laboratory (University of Copenhagen), Strandpromenaden 5, DK-3000 Helsingør, Denmark

Abstract. Laboratory experiments were set up in order to determine and compare developmental rates, growth rates, generation times and egg production rates for the two calanoid copepod species *Centropages typicus* and *Centropages hamatus*. The nauplii showed a higher developmental rate than the copepodites for both species with quite different individual stage durations, which gave no indication of isochronal development. For *C. typicus* equiproportional development was found. The growth rates were exponential and highest for the largest species *C. typicus*, and for both species the juvenile growth rates were very similar to the egg production rates of the adults.

Introduction

The object of this paper is to investigate whether the growth rate of juveniles is similar to the growth rate of adult females expressed as egg production in two calanoid copepod species, *Centropages hamatus* and *Centropages typicus*, commonly found in Danish waters. This is of great importance, since a growing number of investigators have assumed that the egg production rate is representative of the growth rate of all stages, when secondary production calculations are carried out (e.g. Kiørboe and Johansen, 1986; McLaren, 1986; Kiørboe *et al.*, 1990). Other objectives are to compare developmental rates, growth rates and generation times of the two species.

Growth rates of juvenile copepods and egg production in adult females have been determined for *Acartia clausi hudsonica* by Sekiguchi *et al.* (1980) and for *Acartia tonsa* by Berggreen *et al.* (1988). Both found the growth to be exponential, and that specific growth rates of juveniles were equal to specific female egg production rates.

Isochronal development, which means that all stages have the same duration (Miller *et al.*, 1977; McLaren,1986), has earlier been determined for *A. clausi* by Miller *et al.* (1977), even though they found that isochronality is not necessarily a general rule for marine copepods. Isochronal development was determined for *A. clausi*, *C. hamatus*, *Temora longicornis* and *Pseudocalanus* sp. by Klein Breteler *et al.* (1982), while non-isochronal development was determined for *Calanus marshallae* by Peterson (1986) and for *C. typicus* by Smith and Lane (1985). Landry (1983) found that the term isochronality might not necessarily be right for any species, but might be a reasonable approximation. Growth rates for copepodite stages of *Calanus pacificus* and *Pseudocalanus* sp. were measured by Vidal (1980), who found a sigmoidal increase in body weight with time, while Klein Breteler *et al.* (1982) found exponential growth in *A. clausi*, *C. hamatus*, *Temora longicornis* and *Pseudocalanus* sp.

This work was carried out in order to determine: (i) whether the growth rate of the juveniles is similar to the growth rate of the adult females expressed as egg production; (ii) whether the development of the two species is isochronal; and (iii) whether the growth rate is constant throughout all stages and the growth exponential.

Method

Copepods were collected in Southern Kattegat (Denmark) in September 1989. In the laboratory 200–400 males and females of *C.hamatus* and *C.typicus* were isolated, and transferred to 100 l tanks with 20 μm filtered, 25‰ seawater, 16.5–17.5°C, and were fed with the alga *Rhodomonas baltica* (5–8 μm equivalent spherical diameter, ESD) and the zooflagellate *Oxyrrhis marina* (20–22 μm ESD).

After 2–3 days, eggs were collected for rearing experiments. Nauplii hatched within 12 h were transferred to four 100 l (*C.typicus*) or one 10 l (*C.hamatus*) tanks. For *C.typicus* the number of nauplii was ~10 000 in each 100 l tank (100 nauplii l^{-1}), for *C.hamatus* the total number of nauplii was ~1000 (100 l^{-1}) in each 10 l tank. The hatching percent was calculated as the number of nauplii after a potential hatching period of 22 h compared with the initial number of eggs.

The cultures were fed *R.baltica* and, after copepodites appeared, *Oxyrrhis marina*. In order to assure excess food at all times, samples of 100 ml were measured daily on a Coulter Counter model TA II. These samples were taken immediately before feeding, which means that the actual concentrations measured represent the absolute minima. The food concentration was 6–10 p.p.m., well above the critical food concentration (Vidal, 1980). Temperature and salinity were kept constant at all times (16.5–17.5°C, 25‰ salinity) for all cultures. In each culture, subsamples (50–100 animals) were taken daily for length measurements (prosome) and stages were determined after Lawson and Grice (1970). The samples were preserved in formaldehyde solution (3%). The *C.hamatus* culture was observed until C5, at that time no more animals remained. Two cultures of *C.typicus* were observed until adulthood (cultures I and II) and two cultures were observed until N4 (cultures III and IV). The time required to reach a particular stage was defined as the time when 50% of the population had molted into that stage. Body weight was calculated from the length–weight relations of Klein Breteler *et al.* (1982). The growth rates are given as dimensionless quantities for exponential changes in weight in specific indicated time periods, determined as the slope of the regression line of time versus the logarithm of the weight. These are directly comparable with the non-exponential egg production rates.

Since we had no way of knowing at which time 50% of the eggs hatched, due to our collecting of nauplii at T = 24 h, we have no development point for N1. Total mortality was calculated as the difference between the total number of animals picked out from the culture during the experiment and the number of animals that remained in the container at the end of the experiment. When adult

females appeared, individuals for egg production experiments were picked. Single females were incubated with excess food in 600 ml bottles (10 replicates) placed on a plankton wheel (1 1/3 r.p.m.). *Centropages hamatus* was incubated with *R.baltica* and *Ditylum brightwellii* to feed on at 19°C, and *C.typicus* with *R.baltica* and *O.marina* at 16.5–17.5°C. After 24 h of incubation, the eggs in each bottle were counted and the length of the females was measured. The weight of an egg was assumed to be 37.4 ng C egg^{-1} (Kiørboe *et al.*, 1985), the carbon content of the females to be 40% of the dry weight (Parsons *et al.*, 1977) and the weight specific egg production rate (g_f) was calculted after:

$$g_f = W_{egg}/W_f * 24/T$$

where W_{egg} = weight of eggs produced (ng C), W_f = weight of female and T = incubation time (h).

Results

Hatching and mortality

Hatching and mortality were only measured for *C.hamatus* due to a mistake. The hatching percentage was 53% after 22 h. The mortality over the entire experimental period was 16%.

Development

Developmental rate (Figure 1) was on the average higher in nauplii than in copepodites for both species. Average stage duration for nauplii was 35.2 ± 15.1 h $stage^{-1}$ (*C.hamatus*) and 28.5 ± 9.4 h (*C.typicus*) and for copepodites 42.3 ± 7.3 h (*C.hamatus*) and 43.1 ± 12.0 h (*C.typicus*).

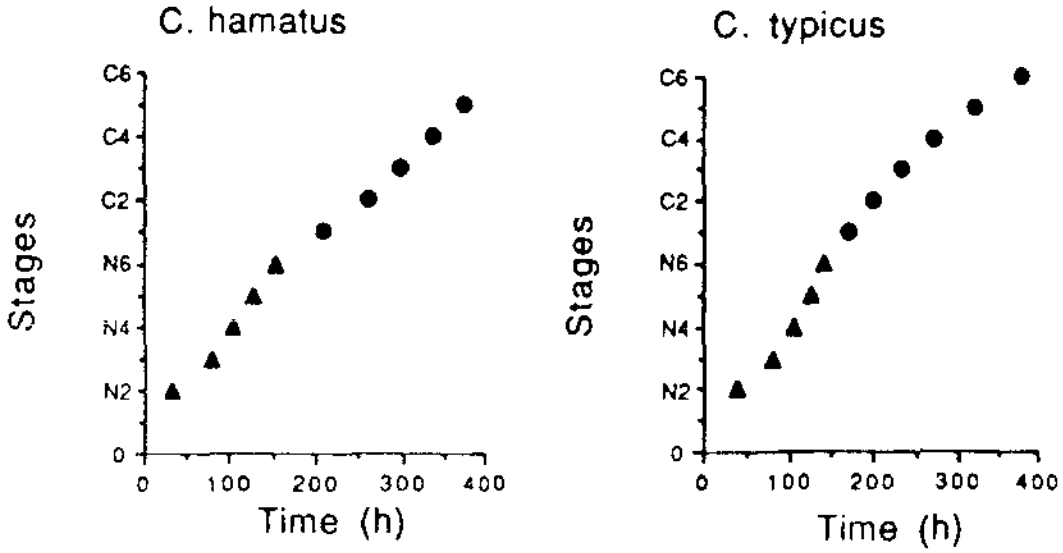

Fig. 1. Developmental rate of *C.hamatus* and *C.typicus* (triangles, nauplii, dots, copepodites). Time required to reach a particular stage is estimated from 50% fractile.

The individual stage durations were quite different (Table I) which gave no indication of isochronal development. The stage durations estimated in the two independent experiments with *C.typicus* were significantly intercorrelated (r^2 = 0.853, $P < 0.01$, Figure 2). The development was equiproportional, as each stage occupied a fixed proportion of total development time (McLaren, 1986). There was no intercorrelation of the stage durations of *C.hamatus* and *C.typicus*. The stage duration of *C.typicus* N2 was significantly longer than for the following nauplii stages (Mann–Whitney *U*-test, $P < 0.05$). The stage durations of the intermediate stages of nauplii had a tendency to be shorter than the later stages, and later copepodites were found to have the longest stage durations. Development time to 50% adults was for *C.hamatus* 418.6 h (extrapolated) and for *C.typicus* 388.0 h.

For *C.typicus* it was observed that the animals began feeding in the second nauplii stage, indicated by *Rhodomonas* cells in the gut.

Table I. Stage duration (h stage^{-1}) for *C.hamatus* and *C.typicus*

	C.ham.	*C.typ.* I	*C.typ.* II	*C.typ.* III	*C.typ.* IV
N2	47.8	41.8	31.5	48.0	44.5
N3	25.3	26.0	31.0	28.0	–
N4	23.8	19.5	26.5	–	–
N5	23.8	15.8	18.0	–	–
N6	55.1	28.5	37.0	–	–
C1	53.8	30.3	31.0	–	–
C2	38.1	33.3	46.0	–	–
C3	37.1	38.0	31.5	–	–
C4	37.1	52.3	49.0	–	–
C5	45.2*	64.8	55.0	–	–

*From extrapolation.

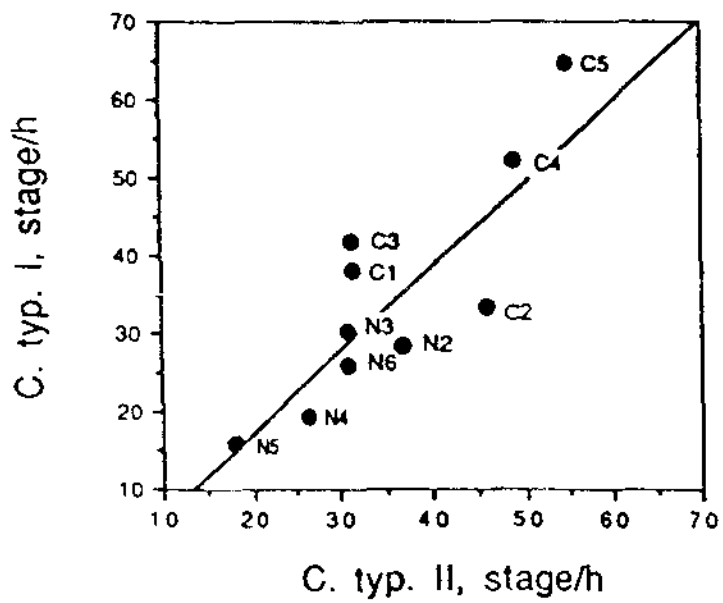

Fig. 2. Relationship between stage durations of *C.typicus* in the two cultures

Growth rate

The growth rate was constant between N1 and C5 for both species, it was not significantly different between naupli and copepodites (Figure 3, Table II). The weight-specific egg production rates of the females of both species corresponded to the growth rates of the juveniles (Table II).

Discussion

The egg production rate and the growth rate of juveniles were determined to be similar. This supports the idea that *in situ* estimates of female fecundity may be used for a field estimate of copepod production as suggested by Berggreen *et al.* (1988), as long as the food concentration *in situ* does not limit the egg production.

Two terms describing patterns in copepod development are isochronal and equiproportional development. Isochronality means that the duration of all stages is the same (Smith and Lane, 1985) while equiproportionality means that each development stage is assumed to occupy the same proportionate amount of time relative to other stages at any constant temperature (Corkett *et al.*, 1986).

Our study has shown no clear evidence of isochronal development in either of the two species. The same observation has been made by Smith and Lane (1985). They also suggested that the first feeding stage is likely to be N3. We found *Rhodomonas* in the guts of N2, and the significantly longer stage duration

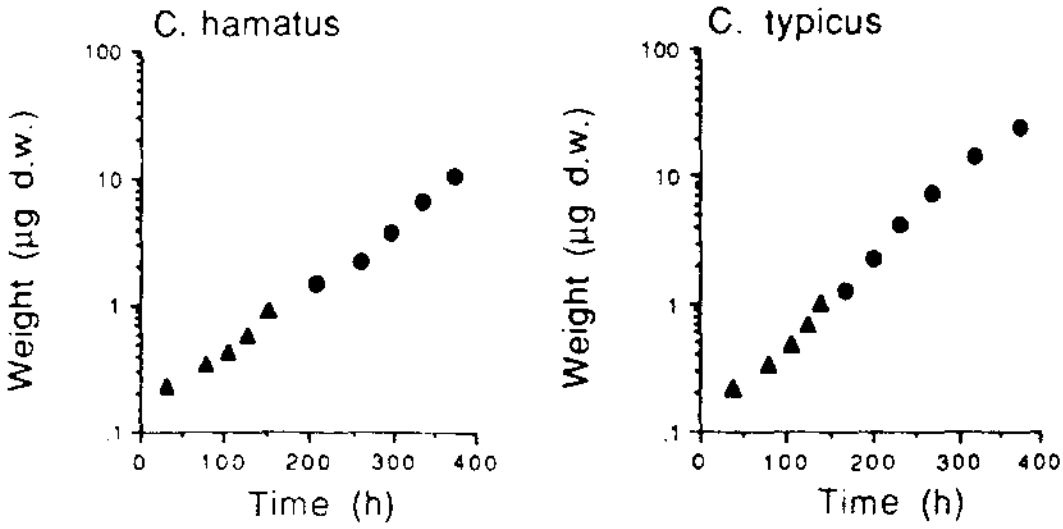

Fig. 3. *Centropages hamatus* and *C. typicus* (triangles, naupli, dots, copepodites) weight increment over time.

Table II. Growth rates (day^{-1} ± one standard deviation)

	C. hamatus	*C. typicus* I	*C. typicus* II
Naupli	0.264 ± 0.010	0.340 ± 0.001	0.351 ± 0.009
Copepodites	0.288 ± 0.008	0.376 ± 0.000	0.376 ± 0.007
All points	0.267 ± 0.004	0.368 ± 0.000	0.347 ± 0.005
Egg production	0.248 ± 0.066	–	0.339 ± 0.088

of N2 (see Landry, 1983), also suggests that N2 is able to feed. The stage durations of *C.typicus* C4 and C5 were longer than the younger stages. In *C.hamatus* only C5 was longer than the other stages. The long stage duration in the late copepodid stages may be due to the formation of sex products before entering the adult stages. This has been suggested for several species (Landry, 1983; Smith and Lane, 1985; Peterson, 1986). We found equiproportional development for *C.typicus*. This has been found for different *Calanus* species by, for example, Corkett (1984) and Peterson and Painting (1990).

The two species, *C.hamatus* and *C.typicus*, both showed exponential growth from egg to adult, which was obviously higher in the larger species, *C.typicus*, which has a slightly shorter generation time (388 h) than *C.hamatus* (418 h). For both species the growth rate of the copepodites is similar to that of the nauplii. Only minor differences in the slope of the regression lines were seen between the nauplii and the copepodites (Figure 3), although there is a tendency to a higher growth rate for the copepodites in both species. This was also found by Klein Breteler *et al.* (1982). The specific growth rate calculated by Klein Breteler *et al.* (1982) for *C.hamatus* was 0.31 day^{-1} at 15°C, which is slightly higher than the growth rate we found; 0.27 day^{-1} (16.5–17.5°C). This difference might be due to the methods used to determine the development time to a specific stage. We used the 50% fractile while Klein Breteler *et al.* used first appearance of a single stage, which estimates the developmental time ~10% faster (Peterson and Painting, 1990). Berggreen *et al.* (1988) found a specific growth rate for *A.tonsa* of 0.45 day^{-1} at 15°C fed on excess food. The development time for *C.typicus* was found to be 34–51 days at 10°C by Smith and Lane (1985), while we found 18 days at 17°C. The difference in temperature of 8°C between the two experiments probably accounts for some of the differences in development time. Another explanation might be the kind of food offered to the cultures. Smith and Lane used *Thalassiosira weissflogii*, whereas we used a mixture of *R.baltica* and *O.marina*. Another study has been made by Lawson and Grice (1970). They found a development time of 22 days at 18–19°C, which is much closer to our findings. They fed the culture with a mixture of crysomonads and diatoms. Klein Breteler *et al.* (1982) found a growth rate for *C.hamatus* of 0.312 day^{-1}, at 15°C when fed on *Isochrysis galbana*, *R.baltica* and *O.marina*. For this species we found 0.267 day^{-1} at 17°C.

We never found any crumpled and empty egg membranes, which could indicate egg cannibalism, leading to underestimated egg production rates. Dagg (1977) correlated his egg production results with counting of such empty membranes and deduced that up to 40% of all produced eggs were eaten over 24 h. Probably due to better food quality and quantity and a much larger incubation-volume our animals did not obviously eat their eggs to such an extent, although we cannot entirely exclude this source of error.

According to Lawson and Grice (1970), our eggs should hatch at somewhere between 29 and 36 h at the temperature we used, which might mean that the effective hatching percent in our experiment would be somewhat higher. The total mortality over the entire experimentation period corresponds to ~1% day^{-1}, which is less than found by Støttrup *et al.* (1986) with *A.tonsa*, and it

indicates that *C.hamatus* throughout the experiment have been in good condition, and as all cultures were treated in the same way, this should also be valid for *C.typicus*.

Acknowledgements

We would like to thank Thomas Kiørboe and William T.Peterson for constructive help and criticism of the manuscript. This work was supported by a grant from the Danish Environmental Agency (HF-90, 2-03 B).

References

Berggreen,U., Hansen,B. and Kiørboe,T. (1988) Food size spectra, ingestion and growth of the copepod *Acartia tonsa* during development: implications for determination of copepod production. *Mar. Biol.*, **99**, 341–352.

Corkett,C.J. (1984) Observations on development in copepods. *Crustaceana*, suppl., **7**, 150–153.

Corkett,C.J., McLaren,I.A. and Sevigny,J.-M. (1986) The rearing of the marine calanoid copepods *Calanus finmarchius* (Gunnerus), *C.glacialis* Jaschnov and *C.hyperboreus* Kroyer with comment on the equiproportional rule. *Syllogeus (Natl. Mus. Can.)*, **58**, 539–546.

Dagg,M. (1977) Some effects of patchy food environment on copepods. *Limnol. Oceanogr.*, **22**, 99–107.

Kiørboe,T. and Johansen,K. (1986) Studies of a larval herring (*Clupea harengus* L.) patch in the Buchan area. IV. Zooplankton distribution and productivity in relation to hydrographic features. *Dana*, **6**, 37–51.

Kiørboe,T., Møhlenberg,F. and Riisgård,H.U. (1985) *In situ* feeding rates of planktonic copepods: a comparison of four methods. *J. Exp. Mar. Biol. Ecol.*, **88**, 67–81.

Kiørboe,T., Kaas,H., Kruse,B., Møhlenberg,F., Tiselius,P. and Ærtebjerg,G. (1990) The structure of the pelagic food web in relation to water column structure in the Skagerrak. *Mar. Ecol. Prog. Ser.*, **59**, 19–32.

Klein Breteler,W.C.M., Fransz,H.G. and Gonzalez,S.R. (1982) Growth and development of four calanoid copepod species under experimental and natural conditions. *Neth. J. Sea Res.*, **16**, 195–207.

Landry,M.R. (1983) The development of marine calanoid copepods with comment on the isochronal rule. *Limnol. Oceanogr.*, **28**, 614–626.

Lawson,T.J. and Grice,G.D. (1970) The developmental stages of *Centrophages typicus* Kroyer (Copepoda, Calanoida). *Crustaceana*, **18**, 187–208.

McLaren,I.A. (1986) Is "structural" growth of *Calanus* potentially exponential. *Limnol. Oceanogr.*, **31**, 1342–1346.

Miller,C.B., Johnson,J.K. and Heinle,D.R. (1977) Growth rules in the marine copepod genus *Acartia*. *Limnol. Oceanogr.*, **22**, 326–335.

Persons,T.R., Takahashi,M. and Hargrave,B. (1977) *Biological Oceanographic Processes*. 2nd edn. Pergamon Press, Oxford, pp. 40–64.

Peterson,W.T. (1986) Development, growth, and survivorship of the copepod *Calanus marshallae* in the laboratory. *Mar. Ecol. Prog. Ser.*, **29**, 61–72.

Peterson,W.T. and Painting,S.J. (1990) Developmental rates of the copepods *Calanus australis* and *Calanoides carinatus* in the laboratory, with discussion of methods used for calculation of development time. *J. Plankton Res.*, **12**, 283–293.

Peterson,W.T., Tiselius,P. and Kiørboe,T. (1991) Copepod egg production, moulting and growth rates, and secondary production, in the Skagerrak in August 1988. *J. Plankton Res.*, **13**, 131–154.

Sekiguchi,H., McLaren,I.A. and Corkett,C.J. (1980) Relationship between growth rate and egg production in the copepod *Acartia clausi hudsonica*. *Mar. Biol.*, **58**, 133–138.

Smith,S.L. and Lane,P.V.Z. (1985) Laboratory studies of the marine copepod *Centropages typicus*: egg production and development rates. *Mar. Biol.*, **85**, 153–162.

Støttrup,J.G., Richardson,K., Kirkegård,E. and Pihl,N.J. (1986) The cultivation of *Acartia tonsa* Dana for use as a live food source for marine fish larvae. *Aquaculture*, **52**, 87–96.

Vidal,J. (1980) Physioecology of zooplankton. I. Effects of phytoplankton concentration, temperature, and body size on the growth rate of *Calanus pacificus* and *Pseudocalanus* sp. *Mar. Biol.*, **56**, 111–134.

Received on August 20, 1990; accepted on December 13, 1990

Danmarks Fiskeri & Havundersøgelser

Charlottenlund, april 1991

FORFATTERERKLÆRING

Michael Fryd, Ole Henrik Haslund and Ole Wohlgemuth : Development, growth and egg production of the two copepod species Centropages hamatus and Leptopages typicus in the laboratory.
Journal of Plankton Research, 1991.

Ovenstående artikel er et resultat af en fælles indsats fra alle tre forfattere. Eksperimenterne er lavet i fællesskab og det samme er gældende for udarbejdelse af artikeludkast, senere rettelser til endelig udgave samt kommunikation til tidsskriftet. Navnerækkefølgen er valgt tilfældigt og følger blot efternavnene alfabetisk.

Michael Fryd | Ole Henrik Haslund | Ole Wohlgemuth

Danmarks Fiskeri- og Havundersøgelser
Charlottenlund Slot
DK-2920 Charlottenlund

Tel 31 62 85 50
Fax 31 62 85 35
Telex 19960 dfh dk