

**The biochemical
composition of *Saduria*
(*Mesidotea*) *entomon*
(Isopoda) from the
Gulf of Gdańsk
(southern Baltic)***

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Crustacea
Biochemical composition
Saduria entomon

MONIKA NORMANT,
ANNA SZANIAWSKA
Institute of Oceanography,
Gdańsk University,
Gdynia

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Abstract

The biochemical composition of specimens of *S. entomon* from the Gulf of Gdańsk (southern Baltic) has been investigated. The average values found, expressed as a percentage of dry weight, are as follows: 28.8% proteins, 6.7% lipids, 8.0% carbohydrates and 28.0% ash. Seasonal variation in the biochemical composition of *S. entomon* was observed during the study period. The differences in protein, lipid, carbohydrate and ash content between males and females were not statistically significant ($p > 0.05$). The relatively small average contents of individual components compared to those in other crustaceans from the Gulf of Gdańsk may have resulted from the broad food preferences of *S. entomon*, ensuring its access to nutrients throughout the year.

1. Introduction

The isopod *Saduria* (*Mesidotea*) *entomon* is regarded as a glacial relict in the Baltic area and has, according to the definition of a relict, a distribution outside the Baltic (Ekman, 1919). This stenothermal, cold-water and euryhaline benthic crustacean lives most frequently at depths of 30–55 m, where it usually lies buried in the silty-sandy bottom (Mulicki, 1957). *S. entomon* plays an important role in the Baltic ecosystem since, being a scavenger, it contributes to the removal of dead animal organisms from

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the sea bed. At the same time, it represents an essential food component of several demersal fishes, e.g. *Gadus morrhua* or *Platichthys flesus*.

The biochemical composition of an organism, resulting from the types of metabolic processes taking place within it, is optimally adjusted to the requirements of the environment in which it lives (Szaniawska, 1991).

The proteins, lipids and carbohydrates absorbed by organisms in their food are a source of chemical energy necessary for all life processes such as respiration, production and reproduction (Schmidt-Nielsen, 1992). Some of these compounds are accumulated in the organisms and constitute reserves utilised under unfavourable environmental conditions.

The aim of the present study was to determine the biochemical composition of *S. entomon*, i.e. to assess the percentages of organic and inorganic compounds in dry matter. The percentage of organic compounds, comprising proteins, lipids and carbohydrates (including glycogen), was also determined. Another objective was to establish the seasonal variations in the content of individual organic and inorganic components. It was also of interest to discover whether any differences exist in the biochemical composition between males and females belonging to different length classes. The variations in the quantities of organic compounds in *S. entomon* should make it possible to determine the biochemical composition strategy of this crustacean, i.e. to decide which substances can be regarded as energy reserves and which are not essential for metabolic processes in this species.

2. Material and methods

The material for this study was collected once a month from June 1990 to February 1991 from on board r/v 'Oceanograf II' at station GN (54°34'N; 18°54'E) in the Gulf of Gdańsk. The animals were taken with a 66 × 33 cm bottom drag-net of 5 × 5 mm mesh from a depth of about 35 m. The length of the specimens was determined by measuring (cm) from the head depression to the termination of the telson. The animals were then divided according to sex and length on the basis of criteria given by Haahtela (1978). Length classes were assigned every 1 cm, starting from 1.6 cm. Next, the wet weight (g) of specimens belonging to individual length classes was determined. The animals were subsequently dried at 55°C to constant weight. The material was homogenised and its biochemical composition determined.

The total lipid content was measured by Bligh and Dryer's method (1959), which involves extraction with 1:2 chloroform:methanol. The results were recalculated on the basis of a standard curve prepared for tripalmitinic acid. The protein content in *S. entomon* was determined by the method described in Lowry *et al.* (1951) with the aid of Folin reagent, a solution of bovine albumin being used as standard. The total carbohydrate content was

determined using the method described by Dubois *et al.* (1956), involving extraction with 15% trichloroacetic acid. The results were calculated from the standard curve prepared for glucose. The inorganic compound content in the dry matter of *S. entomon* was calculated on the basis of the amount of ash left following the combustion of the samples in a microcalorimetric bomb.

The protein, lipid, carbohydrate and ash contents are expressed as a percentage of dry weight (% d.w.). Results are given as a mean value with a standard deviation (\pm SD).

Comparisons of mean values ($H_0 : \bar{x}_1 = \bar{x}_2$, student's t-test after F-test) were used in the statistical analysis.

3. Results

The *S. entomon* specimens used in this study were divided into six length classes. The total quantity of organic and inorganic compounds determined in the body of *S. entomon* varies from 65.4 to 84.0 % d.w. (Fig. 1), which is probably due to the fact that the total protein content was not measured. The protein content obtained with the aid of the method of Lowry *et al.* (1951) is lower than that calculated by taking the total nitrogen content into account (Giese, 1967).

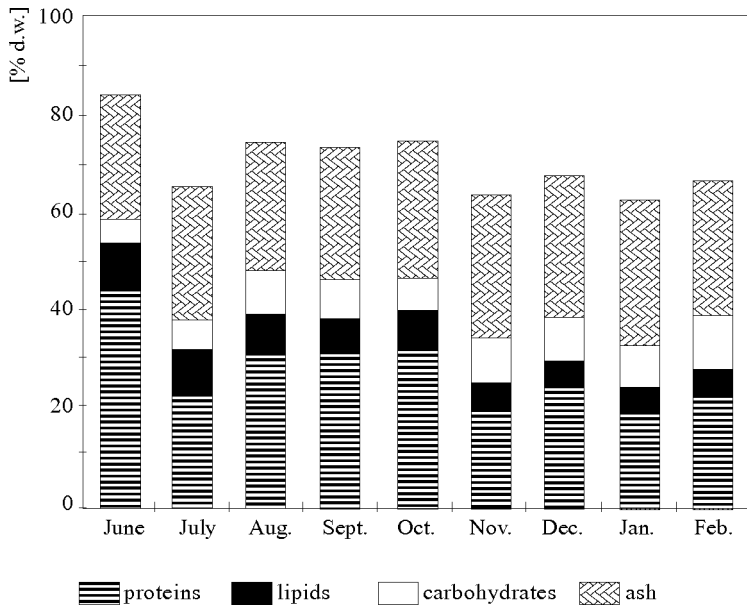


Fig. 1. Seasonal changes in the biochemical composition of *S. entomon*

3.1. Organic compounds

43.5% d.w. of *S. entomon* consists of proteins, lipids and carbohydrates (including glycogen).

Proteins

The average protein content in *S. entomon* is 28.8 (± 9.3) % d.w. ($n = 64$). Females seem to contain more protein than males: 29.9 (± 11.8) % d.w. as against 28.2 (± 7.3) % d.w. respectively (Fig. 2). However, no statistically significant ($p > 0.05$) differences in protein content between *S. entomon* males and females could be determined. The protein content in the body of this species varies during the course of the year, the lowest values being recorded in January (20.0 (± 3.8) % d.w.), the highest ones in June (44.8 (± 10.0) % d.w.) (Fig. 3). The bodies of the smallest specimens, belonging to the 1.6–2.5 cm length class, contain on average the smallest quantities of proteins: 26.5 (± 7.7) % d.w. The protein content increases in subsequent length classes, *i.e.* 27.7 (± 9.8) % d.w. in the 2.6–3.5 cm class, 28.5 (± 10.1) % d.w. in the 3.6–4.5 cm class and 31.3 (± 10.5) % d.w. in the 4.6–5.5 cm class. In the next length class (5.6–6.5 cm), the protein content decreases as compared to the previous class and is 29.3 (± 8.1) % d.w. In the largest animals, 6.6–7.5 cm long, the protein percentage again rises, to 31.6 (± 4.5) % d.w. (Fig. 4a).

Lipids

The average lipid content in *S. entomon* observed over 9 months is 6.7 (± 2.9) % d.w. ($n = 85$). Bodies of males contain fewer lipids (6.3 (± 2.9) % d.w.) than do the bodies of females (7.5 (± 2.7) % d.w.) (Fig. 2), but these differences are not statistically significant ($p > 0.05$). The lowest lipid content was measured in specimens collected in December (4.7 (± 2.1) % d.w.), the highest in those caught in June (9.2 (± 2.8) % d.w.) (Fig. 5). On average, the smallest specimens contain the smallest quantities of lipids (Fig. 4b), *i.e.* 3.8 (± 0.9) % d.w. The dry-weight lipid percentage increases in subsequent length classes to 6.1 (± 2.1) % d.w. in specimens 2.6–3.5 cm long and 7.6 (± 2.4) % d.w. in specimens 3.6–4.5 cm long. The highest values (8.6 (± 3.7) % d.w.) were recorded in animals belonging to the 4.6–5.5 cm length class. In the specimens belonging to the two remaining classes, the content is lower than in the preceding length class and is in fact the same: 6.7 (± 2.5) % d.w. in the 5.6–6.5 cm class and 6.7 (± 1.3) % d.w. in the 6.6–7.5 cm class.

Carbohydrates

During the study period, the average carbohydrate content in *S. entomon* was 8.0 (± 2.9) % d.w. ($n = 66$). There appeared to be smaller quantities of these compounds in males than in females: 7.5 (± 2.7) % d.w. and

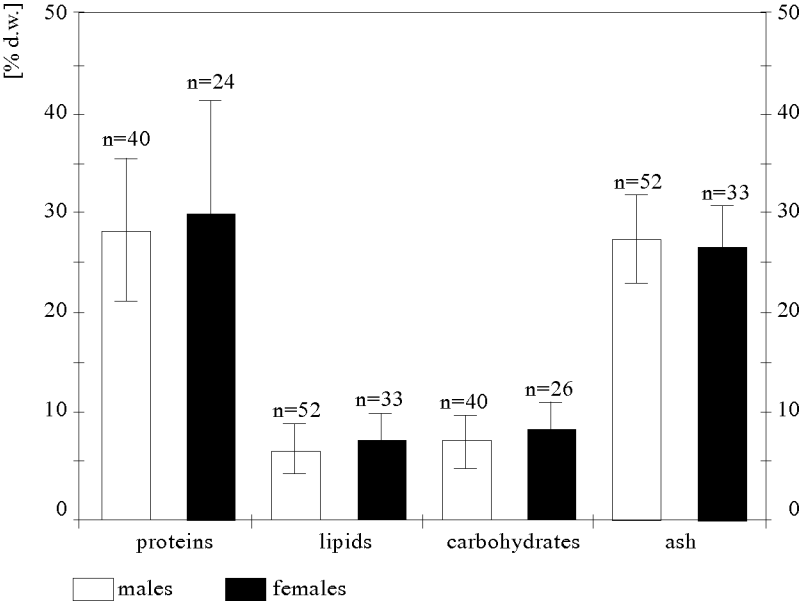


Fig. 2. Comparison of the biochemical composition of male and female *S. entomon*. Values are expressed as a mean (\pm SD)

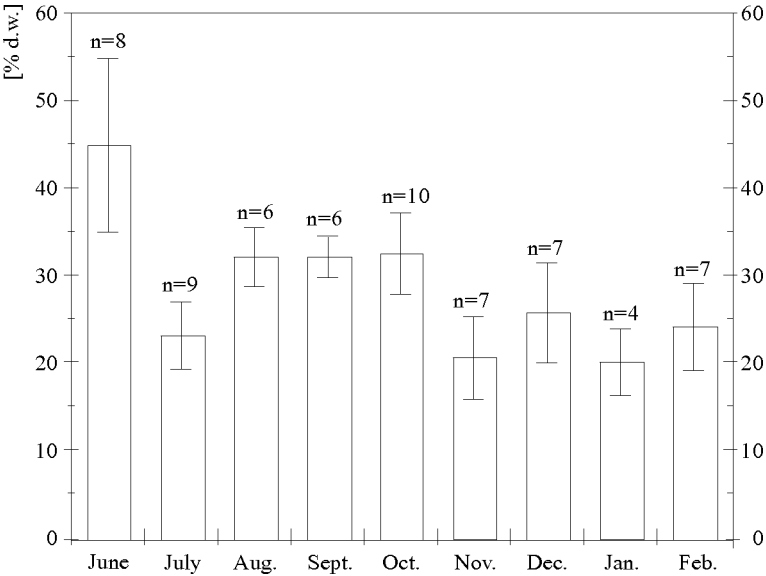


Fig. 3. Variation in the protein content (mean \pm SD) of *S. entomon* during the year

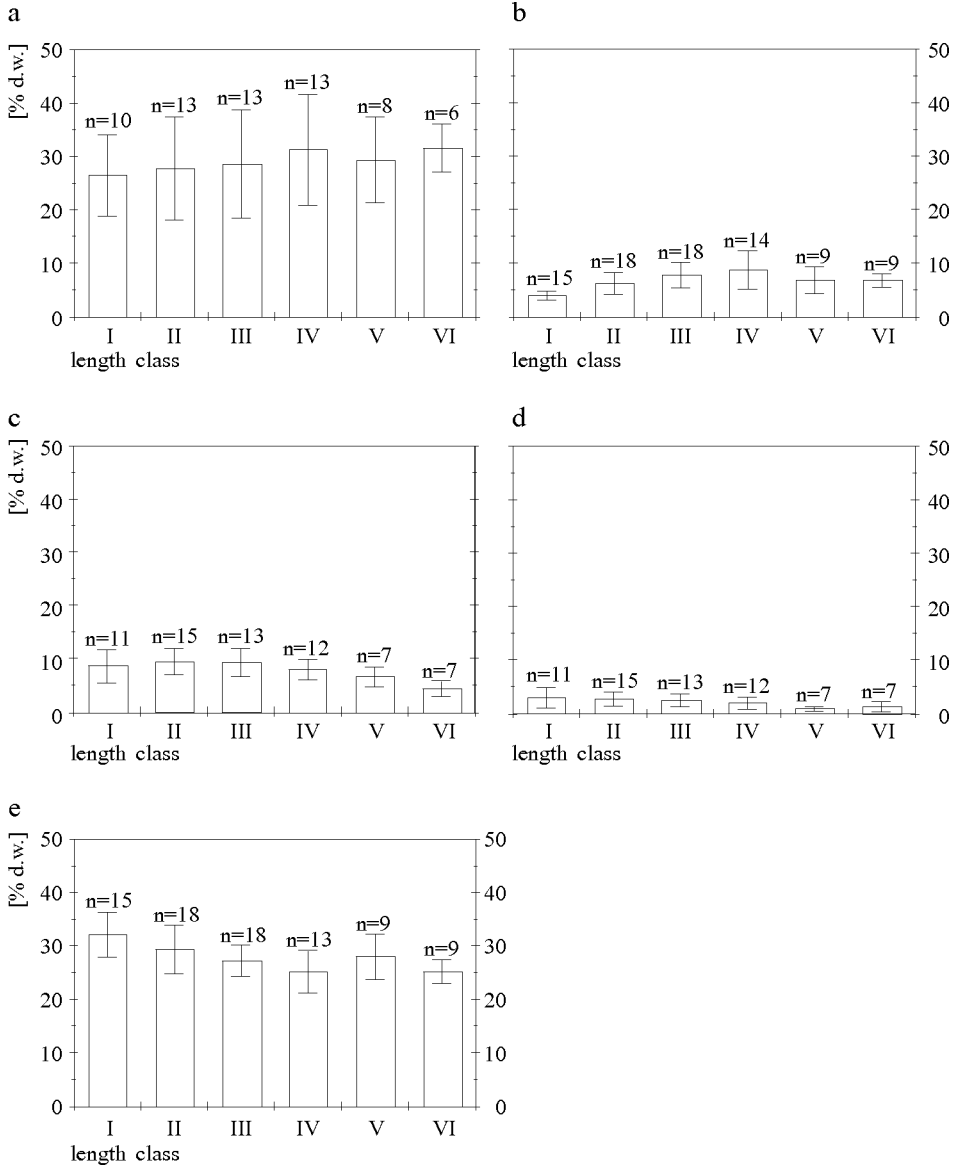


Fig. 4. Percentage of biochemical compounds (mean \pm SD) in the dry weight of *S. entomon*: proteins (a), lipids (b), carbohydrates (c), glycogen (d), ash (e)

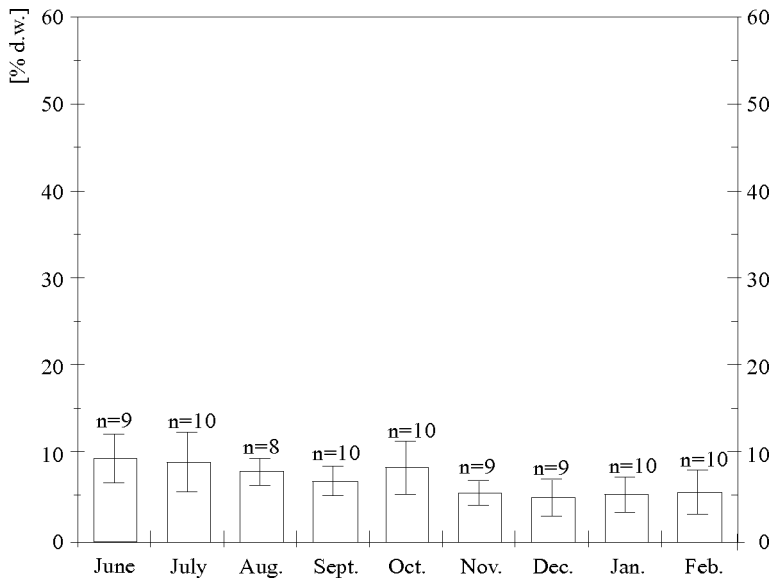


Fig. 5. Variation in the total lipid content (mean \pm SD) of *S. entomon* during the year

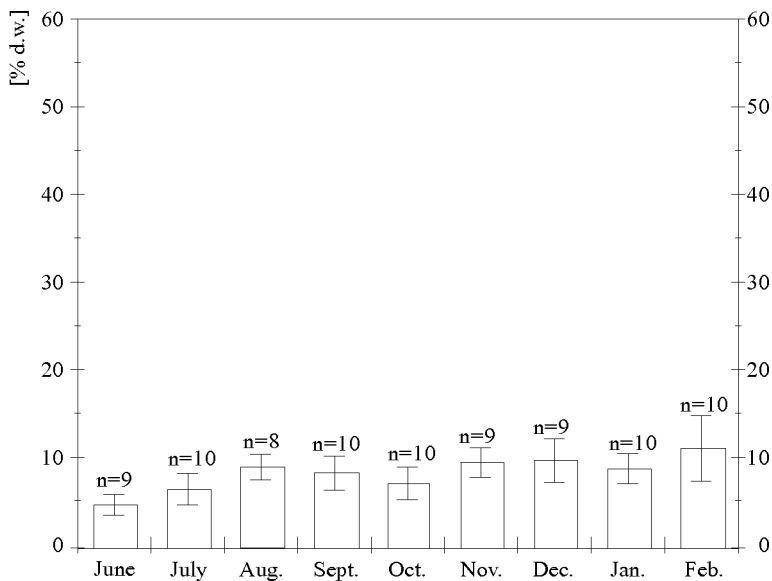


Fig. 6. Variation in the total carbohydrate content (mean \pm SD) of *S. entomon* during the year

8.9 (± 2.9) % d.w. respectively (Fig. 2). Statistical tests showed, however, that these differences are not statistically significant ($p > 0.05$). The lowest carbohydrate levels were recorded in animals collected in June (4.5 (± 1.2) % d.w.), the highest in those caught in February (11.0 (± 3.7) % d.w.) (Fig. 6). The smallest specimens contain an average of 8.5 (± 3.1) % d.w. carbohydrates. The highest carbohydrate content (9.4 (± 2.5) % d.w.) was found in animals from the next length class, *i.e.* 2.6–3.5 cm long. In specimens from the subsequent length classes, the proportion of carbohydrates gradually decreased, reaching a minimum value (4.3 (± 1.5) % d.w.) in the largest *S. entomon* (Fig. 4c).

Glycogen

Ca 26% of the total carbohydrate content is glycogen. The average glycogen content in the *S. entomon* body over the nine months' study period amounted to 2.1 (± 1.5) % d.w. ($n = 66$), and did not differ significantly between males (2.1 (± 1.7) % d.w.) and females (2.2 (± 1.3) % d.w.) (Fig. 2).

In the summer months, the percentage of glycogen turned out to be the smallest, falling to minimum values in September (0.7 (± 0.6) % d.w.). Starting from October, the glycogen content in dry weight increased gradually, rising to a maximum value (4.1 (± 2.1) % d.w.) in February (Fig. 7).

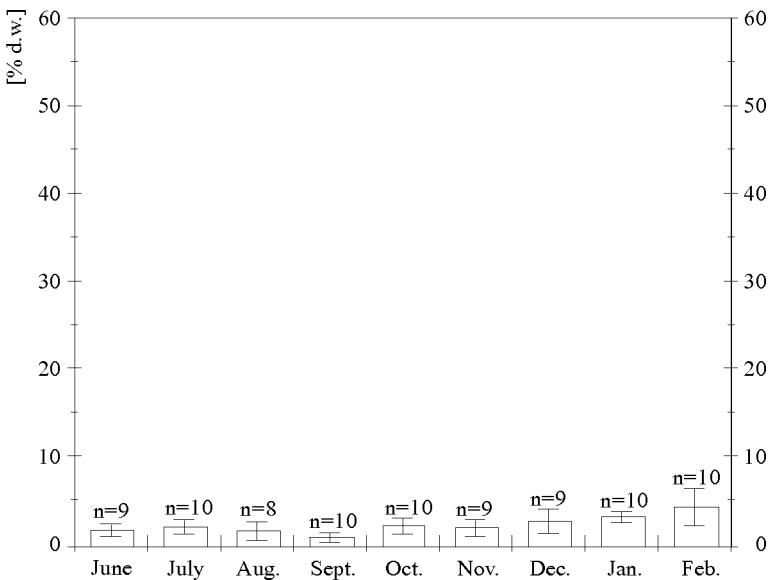


Fig. 7. Variation in the glycogen content (mean \pm SD) of *S. entomon* during the year

The glycogen content was found to decrease with specimen length. The highest values ($2.9 (\pm 1.9) \% \text{ d.w.}$) were recorded in the smallest animals from the 1.6–2.5 cm length class. The lowest values ($0.8 (\pm 0.4) \% \text{ d.w.}$) were measured in animals 5.6–6.5 cm long (Fig. 4d).

3.2. Inorganic compounds

Ash

On average, *S. entomon* contains as much as $28.0 (\pm 4.5) \% \text{ d.w.}$ ($n = 85$) of ash. This quantity seems to be larger in males than in females – $28.3 (\pm 4.6) \% \text{ d.w.}$ and $27.5 (\pm 4.4) \% \text{ d.w.}$ respectively (Fig. 2) – although these differences are not statistically significant ($p > 0.05$).

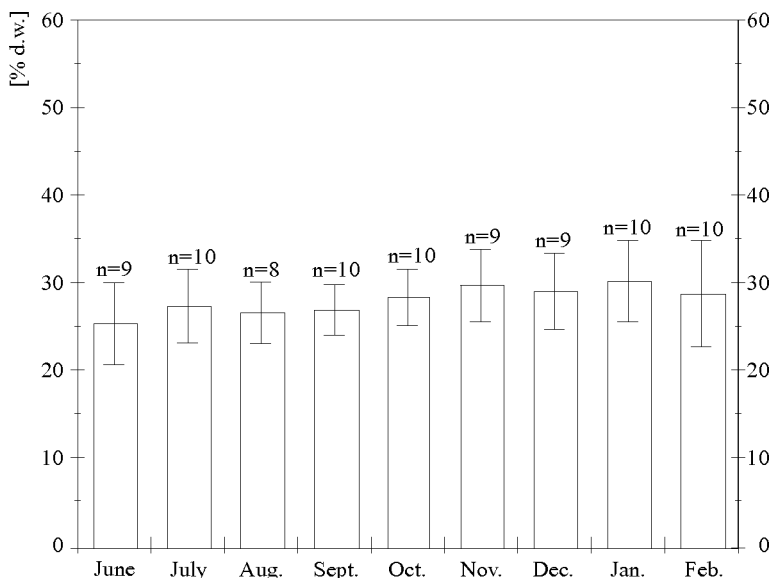


Fig. 8. Variation in the ash content (mean \pm SD) of *S. entomon* during the year

The smallest amounts of ash were determined in June ($25.3 (\pm 4.7) \% \text{ d.w.}$), the largest in January ($30.1 (\pm 4.7) \% \text{ d.w.}$) (Fig. 8). The highest ash levels ($32.1 (\pm 4.2) \% \text{ d.w.}$) were found in the smallest animals, *i.e.* those in the 1.6–2.5 cm length class (Fig. 4e). In the next two length classes, a decrease in the ash content was found. This value reached a minimum in animals 4.6–5.5 cm long ($25.2 (\pm 4.0) \% \text{ d.w.}$). In the 5.6–6.5 cm length class, the inorganic compound content was again found to have increased, to $28.0 (\pm 4.3) \% \text{ d.w.}$ In the largest animals, the ash content decreased to $25.2 (\pm 2.2) \% \text{ d.w.}$

4. Discussion

The present study has demonstrated that in *S. entomon* the protein, lipid and carbohydrate level, as well as the ash content vary over a period of nine months. The content of individual compounds also varies among specimens belonging to different length classes. The protein and lipid contents are least in the smallest specimens and increase with the animals' length to reach maximum values in the 4.6–5.5 cm length class. The high protein and lipid contents in mature specimens may have resulted from the accumulation of these compounds prior to gametogenesis. From earlier studies (Haahtela, 1990) it follows that although this species undergoes reproduction all the year round, the process intensifies during the summer months. Proteins and lipids are utilised during the formation of sperm cells as well as during the development of egg cells and embryos (Giese, 1966, 1969). The situation is similar in other crustaceans, e.g. *Monoporeia affinis* and *Pontoporeia femorata*, where the youngest specimens accumulate fewer lipids than mature ones (Hill, 1992). In the largest *S. entomon*, the protein and lipid contents again decline. In turn, the largest quantities of carbohydrates (including glycogen) and ash are found in the smallest specimens and these amounts decrease in subsequent length classes.

Proteins are the most abundant compounds in *S. entomon*. Moreover, the protein content fluctuates widely in individual months and can be as high as 21.8% d.w. (the difference between the mean June and July values). In November, the protein content decreases by as much as 11.9% d.w. as compared to October. The large quantity of proteins in July, before *S. entomon* reproduction, is due to the fact that apart from lipids, proteins constitute the major building material of egg cells.

In typical benthic species to which, among others, *S. entomon* belongs, the lipid level is generally rather low compared to that in planktonic organisms (Clarke *et al.*, 1985). In comparison with other crustaceans from the Gulf of Gdańsk, e.g. *Crangon crangon* or *Palemon adspersus*, the lipid content in dry weight of *S. entomon* is ca 1.5% higher (Szaniawska, 1991). However, *P. adspersus* caught off the Greek coast contained twice as many lipids, i.e. 13.6% d.w. (Papoutsoglou and Papaparaskeva-Papoutsoglou, 1976).

In numerous marine invertebrates energy is stored in the form of lipids (Lawrence, 1976). In *S. entomon*, the highest lipid level is recorded during the summer months, while in winter this content decreases. Furthermore, a relationship exists between the lipid and carbohydrate contents. When the quantity of lipids in dry weight increases, that of carbohydrates falls. Lipids are utilised in the course of egg cell development (Thorson, 1950) and this probably explains the high level of these compounds in *S. entomon* during the summer, i.e. in the period when reproduction intensifies. In autumn, this

species accumulates lipids, as energy reserves are needed to ensure survival during the unfavourable environmental conditions of winter. The fact that *S. entomon* does not normally accumulate large amounts of lipids is typical for *e.g.* bivalves (Wołowicz, 1991). *S. entomon* feeds on different kinds of food, and even engages in cannibalism (Leonardsson, 1990), so it is fairly independent of temporary food shortages and does not need to accumulate large stores of energy before the onset of winter. In *Mesidotea entomon*, which originates from the western Arctic, the lipid content in females caught in July amounted to 13.8% d.w. (Korczyński, 1989), hence it was about 4% higher than in females from the Gulf of Gdańsk. The lipid content in male bodies differed only slightly and was about 0.5% higher in specimens from the Gulf of Gdańsk. It is possible that the difficult thermal and trophic conditions in the Arctic restrict reproduction in this species. In this case, the egg cells produced are fewer in number but are larger and contain more nutrients, as is usual in other crustaceans (Thorson, 1950; Steele and Steele, 1975).

It is interesting that the bodies of *S. entomon* contain on average more carbohydrates than are usually found in the other crustaceans inhabiting the Gulf of Gdańsk (Szaniawska, 1991). These differences are no more than *ca* 7%. Similar observations were made when the amounts of carbohydrates in *M. entomon* from the western Arctic were compared (Korczyński, 1989). In July, the carbohydrate level was about 5% higher in the specimens from the Gulf of Gdańsk. As in the bivalves from the Gulf of Gdańsk (Szaniawska, 1991) or the crab *Chasmagnatus granulata* (Kucharski and Da Silva, 1991), the carbohydrate and glycogen contents in *S. entomon* are low in summer and higher in autumn and winter. The lack of data for March, April or May specimens means that it is impossible to determine whether the glycogen content decreases before reproduction, as is the case in *e.g.* bivalves from the Gulf of Gdańsk (Wołowicz, 1991). Carbohydrates, and glycogen in particular, are utilised during the anaerobic respiration of organisms. It is possible that the environmental conditions to which *S. entomon* has recently been exposed, *i.e.* oxygen deficiency, especially at greater depths, resulting from the eutrophication of Baltic waters (Cedrewall and Elmgren, 1990) and the declining frequency of water influxes from the North Sea, have forced this species to accumulate larger amounts of glycogen than is the case in other crustaceans. During unfavourable conditions, these reserves can then be consumed in anaerobic respiration. *S. entomon* displays considerable tolerance to changes in seawater oxygenation and is capable of surviving for even several days in water devoid of oxygen (Hagerman and Szaniawska, 1990).

Being a storage compound with a relatively simple chemical structure, glycogen undergoes rapid metabolic transformations. It can also be

metabolised to fatty acids, which are energy reserves used under difficult thermal and trophic circumstances. Nevertheless, only 26% of the carbohydrates in *S. entomon* consist of glycogen; bivalves from the Gulf of Gdańsk contain 38% glycogen (Wołowicz, 1991).

Inorganic compounds are the second most abundant constituent after proteins. This is probably due to their incorporation in the structure of the carapace, which in *S. entomon* makes up 50–70% of a specimen's wet weight (author's own data). This proportion is twice as high as that in other crustaceans from the Gulf of Gdańsk (Szaniawska, 1991). In July, *M. entomon* from the northern Arctic contains about 8% more ash in dry weight. This can be explained by the smaller accessibility of nutrients in the latter region. The differences in ash content in specimens from the Gulf of Gdańsk during the nine-month study period were of the order of 5.5%.

Constituting the highest proportion of organic compounds in specimen dry weight, proteins play a significant part in reproduction and are the last to be used in metabolic processes, as can be implied from their high content compared to that of the other compounds studied. In an organism, the function of proteins is mainly structural. Their energy value is low compared to that of lipids or carbohydrates, so they are rather a poor source of metabolic energy, to be used only after other storage compounds have been consumed. This was confirmed by studies on *S. entomon* starvation (Hagerman and Szaniawska, 1990). Being high-energy compounds, lipids provide most of the energy for the metabolic processes, and are used in egg cell production during the reproductive period. The feeding mode of *S. entomon*, which guarantees constant access to food, means that the accumulation of large amounts of these compounds is not necessary. As in bivalves, carbohydrates are used as storage materials during non-reproductive periods. It is possible that the larger amounts of glycogen are needed in anaerobic respiration processes, which take place mainly during the summer months, when the oxygen concentration in near-bottom waters is at its lowest.

Earlier studies (Szaniawska, 1991) showed that in the case of bivalves from the Gulf of Gdańsk such as *Mytilus edulis* or *Cardium glaucum*, carbohydrates and lipids act as energy reserves, while in crustaceans, e.g. *C. crangon* and *P. adspersus*, this function is fulfilled by lipids and proteins.

This analysis has demonstrated that the biochemical composition of *S. entomon* differs from that of other crustaceans from the Gulf of Gdańsk, and is probably due to the characteristics of this species.

5. Conclusions

- The differences in the biochemical component content between males and females of *S. entomon* are not statistically significant ($p > 0.05$).
- The organic compound content in *S. entomon* increases with length, which indicates that the animals gain proteins, lipids and carbohydrate during growth.
- Changes in biochemical composition were recorded throughout the year. The largest quantity of organic compounds in *S. entomon* were noted during the summer, when the maximum reproduction output occurs.
- The carbohydrate content was higher in *S. entomon* than in other crustaceans from the Gulf of Gdańsk.
- Being a scavenger, *S. entomon* has food available throughout the year and does not need to store large amounts of organic compounds.

References

- Bligh E. G., Dyer W. J., 1959, *A rapid method of total lipid extraction and purification*, Can. J. Biochem. Physiol., 37, 911–917.
- Cedrewall H., Elmgren R., 1990, *Biological effects of eutrophication in the Baltic Sea*, AMBIO, 19, 109–112.
- Clarke A., Skadsheim A., Holmes L. J., 1985, *Lipid biochemistry and reproductive biology in two species of Gammaridae (Crustacea: Amphipoda)*, Mar. Biol., 88, 247–263.
- Dubois M., Gilles K. A., Hamilton J. K., Bebecs P. A., Smith F., 1956, *Calorimetric methods for determination of sugar and related substances*, Anal. Chem., 28, 350–356.
- Ekman S., 1919, *Studien über die marinen Relikte der nordeuropäischen Binnengewässer, VI. Die morphologischen Folgen des Reliktwerdens*, Int. Rev. Hydrobiol. Hydrogr., 8, 477–528.
- Giese A. C., 1966, *Lipids in the economy of marine invertebrates*, Physiol. Rev., 46, 244–298.
- Giese A. C., 1967, *Some methods for study of the biochemical constitution of marine invertebrates*, Oceanogr. Mar. Biol. Ann. Rev., 5, 159–189.
- Giese A. C., 1969, *A new approach to the biochemical composition of the molluscs body*, Oceanogr. Mar. Biol. Ann. Rev., 7, 175–229.
- Haahtela I., 1978, *Morphology as evidence of maturity in Isopod Crustacea, as exemplified by Mesidotea entomon (L.)*, Ann. Zool. Fenn., 15, 186–190.
- Haahtela I., 1990, *What do Baltic studies tell us about the isopod Saduria entomon?*, Ann. Zool. Fenn., 27, 269–279.

- Hagerman L., Szaniawska A., 1990, *Anaerobic metabolic strategy of the glacial relict isopod Saduria (Mesidotea) entomon*, Mar. Ecol. Prog. Ser., 59, 91–96.
- Hill C., 1992, *Seasonal changes in lipid content and composition in the benthic amphipods Monoporeia affinis and Pontoporeia femorata*, Limnol. Oceanogr., 37, 1280–1289.
- Korczyński R. E., 1989, *Biochemical composition of the isopod Mesidotea entomon (Linnaeus) from the Western Arctic*, Polar Biol., 9, 391–395.
- Kucharski L. C. R., Da Silva R. S. M., 1991, *Seasonal variations in the energy metabolism in an estuarine crab Chasmagnatus granulata (Dana, 1851)*, Comp. Biochem. Physiol., 100 (A), 599–602.
- Lawrence J. M., 1976, *Patterns of lipid storage in post-metamorphic marine invertebrates*, Am. Zool., 16, 747–762.
- Lowry D. H., Roseborough I., Farrand A. L., Rondall R. J., 1951, *Protein measurement with Folin phenol reagent*, J. Biol. Chem., 193, 263–275.
- Leonardsson K., 1990, *Cannibalism as a population-regulating mechanism in the brackish-water isopod Saduria entomon (L.)*, Ann. Zool. Fenn., 27, 285.
- Mulicki Z., 1957, *Ecology of selected benthic invertebrates from the Baltic Sea*, Pr. Mor. Inst. Ryb., 9 (A), 313–377, (in Polish).
- Papoutsoglou S. E., Papaparaskeva-Papoutsoglou E. G., 1976, *The chemical composition of Palemon adspersus*, Boll. Pesca Pisc. Idrobiol., 31, 55–58.
- Schmidt-Nielsen K., 1992, *Animal physiology. Adaptation to the environment*, PWN, Warszawa, (in Polish).
- Steele D. H., Steele V. J., 1975, *Egg size and duration of embryonic development in Crustacea*, Int. Rev. ges. Hydrobiol., 60, 711–715.
- Szaniawska A., 1991, *Energy management in benthic invertebrates from the Baltic Sea*, Ph. D. thesis, Wyd. UG, Gdańsk, (in Polish).
- Thorson G., 1950, *Reproductive and larval ecology of marine bottom invertebrates*, Biol. Rev., 25, 1–45.
- Wołowicz M., 1991, *Geographical diversification of the Cardium glaucum Bruguiere (Bivalvia) population*, Ph. D. thesis, Wyd. UG, Gdańsk, (in Polish).