

CALORIC VALUES OF MARINE INVERTEBRATES WITH AN EMPHASIS ON THE SOFT PARTS OF MARINE BIVALVES

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Abstract Energy values of biological material (expressed in J per unit of weight) are widely used in studies of energy budgets of organisms and of energy transfer in ecosystems. The methods to estimate caloric values are rather cumbersome. In many publications, already published records of the same or a related species are used instead. The present review summarizes records of caloric values in several hundreds of species of marine invertebrates. Estimates in more than 70 species of marine bivalves (Mollusca: Pelecypoda) by about 25 authors are discussed in detail to judge their reliability. Bivalves are a suitable group for a study of the variability in energetic values because they differ widely in the contents of two different types of reserve material: the low-caloric carbohydrates (glycogen) and the high-caloric lipids. Moreover, many bivalve species have been analyzed for these substances. On the basis of their chemical composition, the soft parts of nearly all bivalves must show caloric values within the range of about 5.3–6.3 cal (or 22–26J)mg⁻¹ AFDW (ash-free dry-weight). A high proportion of the published records of direct bomb-calorimetric estimates proved to be below this range as a consequence of methodological errors. Particularly, the omission of separate ash determinations and incomplete drying appear to cause underestimates in energy per unit of ash-free dry-weight. A representative value for marine bivalves appears to be close to 5.5 cal (or 23J)mg⁻¹ AFDW. This proposed value is substantially higher than averages of uncritical compilations. Representative values for other taxa of animals are probably also higher than means of literature records and will exceed this bivalve value of 5.5 cal by 0.1 or 0.2 cal mg⁻¹ AFDW in most major taxa (with organisms using primarily lipids instead of carbohydrates as a reserve material). Pros and cons of the direct and indirect method of estimating caloric values are discussed.

Introduction

Knowledge on energetic values is indispensable in studies of energy flow through organisms, populations and ecosystems. Such values provide a common unit (the Joule or the still often used calorie, 1cal=4.187J) to express such different processes as consumption, respiration, growth and production. Caloric values are also used as a unit to express the nutritive value of food material.

Particularly in the 1960s, when micro or semi-micro bomb calorimeters became available (Phillipson 1964) and studies in the framework of the IBP started on a worldwide scale, a host of publications appeared containing figures on caloric content in a wide variety of species. Pioneer work on marine invertebrates, however, had been done already in the 1930s by Rinke (1938). An early comprehensive study by Brawn et al. (1968) presents many values for marine invertebrates, but unfortunately only on a DW base instead of a more useful AFDW base. The early and at that time almost complete compilation by Cummins & Wuycheck (1971) shows a wide range of caloric estimates, even within the same species. Representative and dependable values cannot simply be derived from such uncritical enumerations. An updated compilation for marine invertebrates, limited to well comparable values on an AFDW base, likewise shows wide ranges of values, both in individual species (compare Table 3) as well as within major taxa (compare Table 1e). Such (by their nature uncritical) compilations do not provide any guide to choose the most representative value for any species or group of related species. The ranges of observed values given in Table 1e point to the inclusion of improbable, probably erroneous values. Therefore, mere averages such as given in Table 1a and 1b are basically unreliable.

In principle, caloric values can also be obtained indirectly by calculation from the chemical composition of the tissues, using appropriate conversion factors for the various main energy-rich components (carbohydrates, proteins and lipids). If the chemical analysis is (almost) complete and realistic conversion factors are available, such calculated values offer an opportunity (a) to check the estimates obtained directly

Table 1 A compilation of published records on average caloric values (cal or Jmg⁻¹) on an AFDW base of aquatic (mostly marine) invertebrates. n=number of species, (a) Published averages of all aquatic invertebrates, arranged in order of increasing values. Such mean values are included only if records were presented for a high number of species from a wide range of major taxa, (b) Averages given separately for major taxonomic groups, calculated from references mentioned under (c), but including also estimates from groups with only one or two records, (c) Detailed records arranged by taxonomic group (and in order of year of publication). Mean values included only if n > 2. Ranges of individual species values are indicated if n > 5.

(a) Invertebrates

mean mg⁻¹ AFDW

(cal)	(J)	n	References
4.83	20.2	30	Wissing et al. (1973)
4.90	20.5	120	Dauvin & Joncourt (1989)
5.09	21.3	82	Steimle & Terranova (1985)
5.32	22.3	44	Thayer et al. (1973)
5.42	22.7		Wacasey & Atkinson (1987)
5.47	22.9	135	Cummins & Wuycheck (1971)
5.49	23.0	229	Brey et al. (1988)
5.70	23.9	12 (taxa)	Norrbin & Båmstedt (1984)
5.95	24.9	42 (species)	Norrbin & Båmstedt (1984)

(b) Major taxa

	mean mg ⁻¹ AFDW		
taxon	(cal)	(J)	n

(a) Invertebrates

mean mg⁻¹ AFDW

(cal)	(J)	n	References
Porifera (sponges)	5.79	24.2	34
Cnidaria (jelly fish, sea anemones, corals)	4.67	19.6	20
Platyhelminthes and other unsegmented worms	5.77	24.2	12
Annelida (segmented worms)	5.32	22.3	26
Mollusca: Bivalvia (clams)	5.30	22.2	115
other (snails, nudibranchs, cephalopods)	5.51	23.1	143
Arthropoda (mainly crustaceans)	5.35	22.4	204
Bryozoa (moss animals) and Brachiopoda (lamp shells)	5.15	21.6	7
Echinodermata (sea stars, sea urchins, etc.)	5.33	22.3	69
Urochordata (tunicates and salps)	4.79	20.1	28

Table 1 *Continued*

(c) Major taxa, detailed.

taxon	calmg ⁻¹ (range)	AFDW (mean)	n	Reference
Porifera		6.04	15	Griffiths (1977)
		6.06	3	Norrbin & Båmstedt (1984)
		5.42	5	Wacasey & Atkinson (1987)
	5.2–6.7	5.46*	8	Brey et al. (1988)
Cnidaria		4.01	3	Wissing et al. (1973)
		4.11	3	Griffiths (1977)
		5.57	3	Norrbin & Båmstedt (1984)
		4.34	6	Steimle & Terranova (1985)
Platyhelminthes	5.1–6.8	5.71	7	Calow & Jennings (1974)
Annelida		6.01	3	Thayer et al. (1973)
	5.1–6.9	6.04	7	Norrbin & Båmstedt (1984)
		5.22	11	Steimle & Terranova (1985)
	5.0–6.1	5.56	18	Wacasey & Atkinson (1987)
	4.7–6.6	5.57*	27	Brey et al. (1988)
	4.2–5.6	5.04	57	Dauvin & Joncourt (1989)
Mollusca: Bivalvia		5.14	3	Kreuzberg & Von Oertzen (1973)

(c) Major taxa, detailed.

taxon	calmg ⁻¹ (range)	AFDW (mean)	n	Reference
Other Mollusca	5.0–6.9	5.83	12	Thayer et al. (1973)
		4.99	3	Chambers & Milne (1979)
		5.73	3	Duarte et al. (1980)
		5.18	10	Steimle & Terranova (1985)
	5.1–6.2	5.46	21	Wacasey & Atkinson (1987)
		5.22	3	Rumohr et al. (1987)
	4.0–7.0	5.44*	38	Brey et al. (1988)
	4.4–5.2	4.71	22	Dauvin & Joncourt (1989)
	4.9–6.7	5.68	17	Cummins & Wuycheck (1971)
		5.38	4	Wissing et al. (1973)
	4.5–5.9***	5.26	11	Thayer et al. (1973)
	5.3–5.6	5.46	6	Duarte et al. (1980)
		6.46	5	Norrbin & Båmstedt (1984)
		5.21	11	Steimle & Terranova (1985)
Arthropoda	4.7–6.2	5.56	27	Wacasey & Atkinson (1987)
	4.5–7.3	5.56*	57*	Brey et al. (1988)
		4.64	5	Dauvin & Joncourt (1989)
	4.4–9.3	5.37	89#	Cummins & Wuycheck (1971)
		6.26	4	Kreuzberg & Van Oertzen (1973)
	3.3–7.5	5.01	19	Wissing et al. (1973)
	3.2–6.4	5.32	10	Thayer et al. (1973)
	4.2–6.6***	5.16	20	Bast & Von Oertzen (1976)
		5.23	5	Hopkins et al. (1978)
	4.7–5.9	5.24	10	Duarte et al. (1980)
	4.1–8.6	6.37	14	Norrbin & Båmstedt (1984)
		5.08	23	Steimle & Terranova (1985)
	4.6–6.1	5.53	19	Wacasey & Atkinson (1987)
	4.3–6.4	5.43*	46#	Brey et al. (1988)
	4.6–5.5	4.97	25	Dauvin & Joncourt (1989)

Table 1 *Continued*

taxon	cal mg ⁻¹ (range)	AFDW (mean)	n	Reference
Bryozoa and Brachiopoda		4.96	3	Norrbin & Båmstedt (1984)

taxon	cal mg ⁻¹ (range)	AFDW (mean)	n	Reference
Echinodermata				
		5.70	5	Griffiths (1977)
		4.83	3	Duarte et al. (1980)
		5.69	5	Norrbin & Båmstedt (1984)
		5.12	13	Steimle & Terranova (1985)
	4.8–6.0	5.46	16	Wacasey & Atkinson (1987)
	4.4–6.4	5.43*	20	Brey et al. (1988)
		4.68	5	Dauvin & Joncourt (1989)
Urochordata				
		4.57	5	Steimle & Terranova (1985)
	3.8–5.0	4.65	9	Wacasey & Atkinson (1987)
	3.8–6.0	4.70*	11	Brey et al. (1988)

* median instead of mean

** Detailed data on Protozoa on a Jmg⁻¹ carbon base can be found in Finlay & Uhlig (1981)

*** Even higher values (up to 8.3 and 6.9, respectively) in egg stages

Includes several freshwater species

by bomb calorimetry, (b) to state the range of realistic values, and (c) to delete unrealistically low or high values. A third method to attain caloric values in an independent way, wet oxidation with dichromate, appears to be used rarely and is not recommended (Henken et al. 1986).

For a comparison of caloric values obtained by different methods, bivalves (Mollusca: Pelecypoda) are a suitable group as these animals use as reserve material large amounts of both low-caloric carbohydrates (with only about 4 cal mg⁻¹) and high-caloric lipids with >9 cal mg⁻¹ (9.45 according to Brody 1945; 9.5 according to Kleiber 1961; and 9.3 according to Morowitz 1968). The sizes of these stores will not only determine the actual caloric values of entire animals, they also set limits to potential values. Supposing a minimal share of 50% of the organic matter of structural proteins (with a caloric content of 5.65 cal mg⁻¹ according to Brody, 5.7 according to Kleiber, and 5.5 according to Morowitz), maximal and minimal caloric values of entire animals would amount to 7.4–7.6 cal (or 31–32J) mg⁻¹ AFDW at 50% lipids and 4.8– 4.9 cal (or about 20J) mg⁻¹ AFDW at 50% carbohydrates, respectively. Genuine values beyond this range will be exceptional, as they are limited to animals with extraordinarily high proportions of one of the two types of reserve material.

Extremely high lipid contents can be observed in some planktonic crustaceans and such organisms indeed show high caloric values of around 8 cal mg⁻¹ (Griffiths 1977, Norrbin & Båmstedt 1984). Extremely high carbohydrate contents in animals occur particularly in bivalves (Giese 1966), but their share will rarely exceed about 40% of the AFDW (Wenne & Styczynska-Jurewicz 1987; see also Table 2 of the present paper for an even more exhaustive review of the literature). Therefore, caloric values < 5.0 cal mg⁻¹ AFDW should be rare in animals. A lower-limit value of 5.0 appears to be more realistic than those of 4.1 to 4.2 stated on the base of 100% carbohydrates by some authors (Wacasey & Atkinson 1987, Dauvin & Joncourt 1989). A lower limit of about 4.2 is appropriate only for plant material.

In the literature on bivalves, values <5.0 cal mg⁻¹ AFDW obtained by bomb calorimetry for the soft parts of entire bivalves appear to be rather frequent (compare Table 3). The often wide discrepancy between estimates obtained by this method and values expected from chemical composition poses the question

whether measurements by bomb calorimetry generally produce reliable estimates of caloric values. Are only the extremely low values wrong or should we suspect all bomb-calorimetric values to be underestimates? Bomb calorimetry is not an easily executed method. Several precautions should be met and corrections applied (Paine 1971, Frascchetti et al. 1994). On the other hand, the calculation method that starts from the proximate chemical composition also has its inherent weaknesses: the chemical analysis is usually incomplete (measured components rarely add up to a full 100% of the material) and precise values of the conversion factors are rarely known (as they depend on the exact composition of the group of material determined). Nevertheless, a comparison of the values obtained by the two methods will be useful because it sheds some light on the degree of uncertainty in the estimates obtained by either method.

Because the determination of caloric contents is time-consuming and difficult, students of energy transfer in biological systems often search for shortcuts, accepting general conversion factors from AFDW or organic C to Joules (as summarized in, e.g. Brey et al. 1988) for macrobenthic aquatic invertebrates. Such compilations generally incorporate all available records, without discrimination against unrealistic values. A more selective compilation might result not only in more precise conversion factors from AFDW to energy units but also to different conclusions, for example, in the case of the skewed distribution of caloric values in animals discussed by Slobodkin (1962) or the relationship between size and caloric values of bivalves (Griffiths 1977).

Chemical components and their caloric value

Carbohydrates

Glycogen is the major carbohydrate stored in bivalves (and in many other marine invertebrates). In this and the following sections, I refer to Giese (1967) for a review of the methods most frequently used for the determination of the biochemical constituents of marine animals. Whyte & Englar (1982) discuss more fully proper ways to assess glycogen contents. In several studies carbohydrates were not actually determined, but estimated as 100% minus the added proportions of lipids, proteins and ash. For several reasons, this is not a recommended procedure, for example, because of uncertainties on the proper conversion factor of N to proteins and on the proportion of tightly bound water (see below).

If large amounts of carbohydrates are present in animal material, the bulk appears to be glycogen. However, small amounts of free sugars are always present as well. Therefore, to obtain the total share of carbohydrates, a few per cent of the AFDW should be added to the glycogen estimate if this is the only carbohydrate assessed. Ansell & Trevallion (1967) found 90% of the carbohydrates of *Tellina tennisi* to be glycogen (leaving 2–3% of the AFDW to be free sugars). Gabbot & Bayne (1973) found 3–5% of the AFDW of mussels *Mytilus edulis* to consist of free sugars. Whyte & Englar (1982) found glycogen to account for 79% of the total glucose in Pacific oysters *Crassostrea gigas* (the remainder, 6–9% of the AFDW, being glucose oligomers).

A caloric value of 4.1–4.2 cal mg⁻¹ of well dried animal carbohydrates appears to be undisputed. The higher value applies to the absolutely water-free anhydride of glycogen, but in practice values close to 4.1 will be found for dried (but not perfectly waterfree) material (Rinke 1938). The use of this lower value also accounts for a (generally minor) share of free glucose with a caloric value of only 3.7 cal mg⁻¹.

Proteins

Protein contents are usually assessed as total N, either by a Kjeldahl procedure or by an elemental CHN analysis. The multiplication factor of 6.25 to obtain protein has generally been used for the mixtures of proteins as found in animals. Nevertheless, it gives only an approximate value and may overestimate protein content. Gnaiger & Bitterlich (1984) propose a conversion factor of only 5.8 instead of 6.25. Thompson & MacDonald (1990) applied this factor of 5.8 to nitrogen values obtained by using an elemental analyzer in dried tissues of the scallop *Placopecten magellanicus* and could account for 95% of the total dry weight by adding separately determined estimates for carbohydrates, lipids, proteins and ash. A remainder of 5% for bound water does not appear to be too high (see below). The usually applied factor of 6.25 for N determined by the Kjeldahl procedure does not generally lead to too high estimates of total dry material. Totals of fractions of carbohydrates, lipids, protein and ash appear to be more frequently far below 100% than close to or above this value.

Total protein can also be estimated as such by, for example, the Lowry colorimetric method (Lowry et al. 1951). Comparisons reveal that a small part of total N in entire organisms originates from non-protein material (this may be different in such material as detritus). Little appears to be known of the content of non-protein N in dried bivalve tissues (Giese 1967). Several N-containing compounds are characterized by low caloric values (examples can be found in Paine 1971), but the amounts of such substances in animal tissues will generally be negligible.

The caloric value of 5.65 cal mg^{-1} holds for protein mixtures that are completely combusted to gaseous nitrogen and water. In an environment of high temperature and oxygen pressure as applied in bomb calorimeters this will be nearly true, though Gnaiger & Bitterlich (1984) express some doubt. Metabolic oxidation within living animals is far less complete with end products such as ammonia, urea or uric acid. Consequently, only 4–5 cal mg^{-1} are gained as catabolizable energy (Kleiber 1961, Kersting 1972). In studies where realistic food values are needed (McClintock 1986), even lower conversion factors for proteins may be desirable, as also high-caloric Ccontaining material may remain undigested. The conversion factor of 5.1 cal mg^{-1} for proteins, mentioned by Cummins & Wuycheck (1971), apparently represents an average of a wide range of published records, referring to some essentially different types of oxidation. Caloric values for protein of around 4 or 5 cal mg^{-1} should be used only where appropriate (e.g. in studies of food value), but not in comparisons of estimates from bomb calorimetry. Where such values were used (e.g. Platt & Irwin 1973, Hallegraeff 1978, Johnson & Hopkins 1978), the fair match observed between the direct and indirect method is in fact misleading and may have concealed other shortcomings.

Lipids

Lipids are extracted by various solvents that differ in efficiency. Ethyl ether or pentane extracts less lipids than chloroform or chloroform-methanol (Giese 1967). Moreover, the exact procedure of extraction affects the yield (Smedes & Thomasen 1996).

When oxidized in a bomb calorimeter, samples extracted by different solvents did not produce the same amounts of heat. For three solvents, Beukema & De Bruin (1979) show caloric values of lipids extracted from the bivalve *Macoma balthica* varying from 8.4–9.1 cal mg^{-1} . As the higher value belonged to the less complete fraction, more efficient solvents apparently extract a higher proportion of low-energy lipids (such as monoglycerids) than less efficient solvents do. Various types of lipids show significantly different caloric values. The values observed by Krishnamoorthy et al. (1979) in various lipid classes extracted from *Crassostrea virginica* varied from 5.3 (monoglycerides) to 10.3 (free sterols) cal mg^{-1} . Because the proportions of such classes will vary with the species, the season, the age of the animal, etc., there is no

fixed conversion factor for the caloric value for the total-lipids fraction in animal tissue (even if extracted in a standard way). Héral & Deslous-Paoli (1983) found a seasonal range of the lipid fraction of *Crassostrea gigas* from 7.8 to 9.4 with an average of 8.6 cal mg⁻¹. This value is close to the one of 8.4 observed by the same method (chloroform-methanol extraction) by Beukema & De Bruin (1979) in *Macoma balthica*.

A conversion factor that can be applied to indirect determinations of caloric contents depends on the type of extractant, but will often be close to 8.5 and such a value should be preferred above the generally accepted standard value for lipids of 9.45 (Brody 1945). Craig et al. (1978) arrived at a similar conclusion for a fish species and used a value of 8.49 cal mg⁻¹.

Other components

The summed estimates of the above main components usually do not add up to a full 100% of the AFDW. If the main components have been assessed correctly, a major cause of the deficit will be the presence of water tightly bound to various compounds. Such water consistently overestimates the AFDW, but does not contribute any calories, thus causing an error in the determination of the caloric value. At conventional dryingstove temperatures (60–100 °C), a few per cent of the weight of the dried material will still be water. Moreover, some water will almost inevitably be attracted during handling and weighing. The high salt content of dried material of marine origin will cause high attraction and retention of water. The proportion of “undetected” tightly bound water can be high in organisms such as sponges with hydrated skeletal material, explaining the frequently very low estimates of caloric values in such groups (Paine 1964). During prolonged drying (months instead of the usual days), tightly bound water appears to evaporate gradually over several months. Sisula & Virtanen (1977) observed a gradual increase to about 105% of the original value in the energy content of tissues of the snail *Helix pomatia* over a period of about six months of oven storage at 60 °C. Gnaiger & Bitterlich (1984) found a residual water fraction amounting to even 6% in carefully redried tissues of marine bivalves.

Water evaporation by drying temperatures in excess of 100 °C cannot be recommended, because losses of volatile substances would increase. As volatile substances will include such energy-rich material as oils, the resulting estimate of the value may be an underestimate. Freeze-drying and consistent use of desiccators may be the best ways to minimize the water content of “dry” material. So far, however, it appears that a well tested standard procedure has not been developed.

The recommended procedure for assessing the proximate chemical composition of such organic material as dried animal tissues appears to be a full CHN analysis. Gnaiger & Bitterlich (1984) explain in which way such data can be used to estimate not only the contents of carbohydrates, lipids and proteins, but also the amount of residual water. In practice, only a few authors used advanced methods such as elemental CHN analyses. Several authors balance the budget of total AFDW by considering the material not accounted for as carbohydrates (frequently not even estimating them as such). Because organic substances with caloric values even lower than 4.1 cal mg⁻¹ are scarce, the bias introduced will be small. The resulting total caloric value may even be an underestimate if a major part of the unknown material had a higher caloric value.

If such supplementing to a full 100% is applied, the base is no longer an AFDW attainable in actual practice, but rather an idealized organic weight, not including any water. For comparisons of caloric values obtained by bomb calorimetry, one should use organic matter contents of about 95% of the total weight of the dried samples, as the pills combusted in calorimeters will contain about 5% of tightly bound water (or even more if not properly handled).

Examples in bivalves

Chemical composition of bivalves

Published reports on the chemical composition of the soft parts of bivalves are numerous. Table 2 summarizes proportions of carbohydrates and lipids on a DW or (preferably) AFDW base. An undoubtedly incomplete survey of the literature revealed 75 records on 36 different species.

Most species show a broad seasonal range in both carbohydrate and lipid contents. Carbohydrate proportions frequently range from a lower limit of 1–10% to an upper limit of 10–40%. Only 5% of the maximal values compiled in Table 2 exceed 40% of the AFDW. All high values (around or above 40%) belong to mussels (*Mytilus*) or oysters (*Ostrea/Crassostrea*). Lipid proportions frequently range from a lower limit of 4–10% to an upper limit of 8–24%. Only one record exceeds 25%. More than half of the seven (out of 65) records of > 20% are from Baltic populations of *Macoma balthica*.

Thus, nearly all values observed in marine bivalves are within the range of 1% to ~40% of AFDW for carbohydrates and within 4% and ~25% in lipids. These limits

Table 2 Ranges of carbohydrate (glycogen or glycogen+glycose) and lipid contents observed in various species of marine bivalves (Mollusca, Pelecypoda), expressed in % of either DW (italicized) or AFDW of the soft parts of entire animals. The percentages of AFDW were taken either directly from the cited papers or (if percentages were based on DW and an ash percentage was stated) the percentages were re-calculated to apply to AFDW. Papers stating the proportions only on a ww basis were omitted. Usually, the ranges indicate the seasonal variation. An * indicates a species with an extremely high proportion of carbohydrates or lipids.

Species	Area	Carboh.	Lipids	Reference
<i>Abra alba</i>	UK	9–20	4–8	Ansell (1974a)
<i>Argopecten irradians</i>	USA	22–29	11–16	Epp et al. (1988)
<i>Astarte montagui</i>	UK	9–20	5–8	Ansell (1975)
<i>Astarte sulcata</i>	UK	13–14	7–10	Ansell (1975)
<i>Astarte elliptica</i>	UK	15–16	5	Ansell (1975)
<i>Cerastoderma edule</i>	UK	8–31		Newell & Bayne (1980)
<i>Cerastoderma edule</i>	Denmark	6–31	6–22	Ivell (1981)
<i>Cerastoderma edule</i>	Netherlands		1–25	Hummel et al. (1988)
<i>Cerastoderma edule</i>	Spain	3–27	7–11	Navarro et al. (1989)
<i>Cerastoderma glaucum</i>	Italy	12–35	10–16	Ivell (1979a)
<i>Cerastoderma glaucum</i>	UK		3–10	Ivell (1979b)
<i>Chlamys opercularis</i>	UK	4–16	4–8	Taylor & Venn (1979)
<i>Chlamys septemradiata</i>	UK	4–9	5–14	Ansell (1974b)
<i>Chlamys varia</i>	France	1–10	5–17	Shafee (1981)
<i>Crassostrea angulata</i>	Spain	11–23	8–13	Establier (1966)
<i>Crassostrea corteziensis</i>	Mexico	5–37	10–24	Paez-Osuna et al. (1993)
<i>Crassostrea gigas</i>	Japan	1–22	7–14	Masumoto et al. (1934)
<i>Crassostrea gigas</i>	UK	2–28		Walne & Mann (1975)
<i>Crassostrea gigas</i>	USA	29–42	9–11	Whyte & Englar (1982)
<i>Crassostrea gigas</i>	France	3–9	9–19	Héral & Deslous-Paoli (1983)
<i>Crassostrea iridescens</i> *	Mexico	37–53	10–16	Paez-Osuna et al. (1993)

Species	Area	Carboh.	Lipids	Reference
<i>Crassostrea virginica</i>	USA		9–15	Lee et al. (1960)
<i>Donax incarnatus</i>	India	3–16	8–14	Ansell et al. (1973)
<i>Donax incarnatus</i>	India		6–15	Balasubramanian et al. (1979)
<i>Donax spiculum</i>	India	2–8	6–6	Ansell et al. (1973)
<i>Donax trunculus</i>	Algeria		5–8	Ando et al. (1976)
<i>Donax trunculus</i>	France	4–12	3–10	Ansell & Bodoy (1979)
<i>Donax trunculus</i>	Algeria	6–16	4–9	Ansell et al. (1980)
<i>Donax vittatus</i>	UK	8–23	5–17	Ansell (1972)
<i>Donax vittatus</i>	UK	9–18	6–11	Ansell & Bodoy (1979)
<i>Lima hians</i>	UK	2–8	6–10	Ansell (1974d)
<i>Macoma balthica</i>	Netherlands	9–35	8–16	Beukema & De Bruin (1977)
<i>Macoma balthica</i>	Germany: Baltic	10–22	8–21	Graf et al. (1982)
<i>Macoma balthica</i>	Finland	9–32	14–24	Pekkarinen (1983)
<i>Macoma balthica</i>	Poland: IPB	8–15	10–18	Wenne & Styczynska-Jurewicz (1985,1987)
<i>Macoma balthica</i> *	Poland: H-35	8–14	23–38	Wenne & Styczynska-Jurewicz (1985,1987)
<i>Macoma balthica</i>	Alands	7–12	14–21	Bonsdorff & Wenne (1989)
<i>Modiolus modiolus</i>	N. Ireland	3–6	3–6	Brown & Seed (1977)
<i>Modiolus modiolus</i>	UK	9–22	7–11	Comely (1981)
<i>Mytilus edulis</i>	UK	14–39	4–12	Daniel (1921)
<i>Mytilus edulis</i>	Netherlands	6–33	4–10	Gerritsen & Van Pelt (1945)
<i>Mytilus edulis</i>	Poland	1–39	13–18	Drzycimski (1961)
<i>Mytilus edulis</i>	UK	2–25	6–19	Williams (1969)
<i>Mytilus edulis</i>	France		7–12	Lubet & Le Feron de Longcamp (1969)
<i>Mytilus edulis</i>	Netherlands	10–35		De Zwaan & Zandee (1972)
<i>Mytilus edulis</i>	UK	5–41	4–11	Dare & Edwards (1975)
<i>Mytilus edulis</i>	Netherlands	3–33	7–12	Pieters et al. (1979)
<i>Mytilus edulis</i>	Netherlands	2–36	7–12	Zandee et al. (1980)
<i>Mytilus edulis</i>	Netherlands	1–26		Hummel et al. (1988)

Table 2 *Continued*

Species	Area	Carboh.	Lipids	Reference
<i>Mytilus edulis</i>	Poland		11–21	Pazikowska & Szaniawska (1988)
<i>Mytilus edulis</i>	Norway	18–30	5–9	Barkati & Ahmed (1990)
<i>Nucula sulcata</i>	UK	2–12	5–11	Ansell (1974c)
<i>Nucula turgida</i>	Ireland	8–18	9–19	Davis & Wilson (1983)
<i>Ostrea edulis</i>	Norway	30–39	7–12	Gaarder & Alvsaker (1941)
<i>Ostrea edulis</i>	Yugo-Slavia	15–28	8–12	Krvaric (1953)
<i>Ostrea edulis</i>	UK	28–36		Walne (1970)
<i>Ostrea edulis</i>	UK	10–30	4–9	Holland & Hannant (1974)

Species	Area	Carboh.	Lipids	Reference
<i>Ostrea edulis</i>	UK	3–24		Walne & Mann (1975)
<i>Ostrea edulis</i>	Netherlands	1–37		Hummel et al. (1988)
<i>Patinopecten yessoensis</i>	Japan	2–18	5–15	Takahashi & Mon (1971)
<i>Pinctada martensii</i>	Japan	1–15	4–13	Tanaka & Hatano (1952)
<i>Placopecten magellanicus</i>	Canada	5–13	7–12	Thompson (1977)
<i>Placopecten magellanicus</i>	Canada	10–24	1–15	Thompson & MacDonald (1990)
<i>Spisula subtruncata</i>	France	3–16	5–9	Bodoy (1980)
<i>Tapes decussatus</i>	France	2–26	8–10	Beninger & Lucas (1984)
<i>Tapes philippinarum</i>	USA	4–30	5–8	Mann (1979)
<i>Tapes philippinarum</i>	France	4–27	8–12	Beninger & Lucas (1984)
<i>Teredo pedicellata</i>	USA	11–34		Greenfield (1953)
<i>Tellina tenuis</i>	UK: Kames Bay	14–30	10–16	Ansell & Trevallion (1967)
<i>Tellina tenuis</i>	UK: Loch Ewe	2–35	5–12	Trevallion (1971)
<i>Tellina tenuis</i>	UK: Loch Ewe	11–28	8–13	Trevallion (1971)
<i>Tellina tenuis</i>	UK: Loch Ewe	12–27	10–18	Trevallion (1971)
<i>Venus gallina</i>	France	4–17	4–9	Bodoy (1983)
<i>Venus mercenaria</i>	UK	16–32	2–9	Ansell et al. (1964)
<i>Venus mercenaria</i>	UK	16–52		Ansell & Lander (1967)

can be used to predict a range of probable values of caloric value of marine bivalve material (see below).

Caloric values based on chemical composition

The relation between the share of the main components and the resulting heat of combustion can be shown in a simple chart showing isoenergetic lines (Fig. 1). In a somewhat similar plot, Morowitz (1968) shows that most animals should have higher caloric values (per unit of AFDW) than plants, because carbohydrate contents are generally much higher in plants than in animals.

Of course, the position of the isoenergetic lines in Fig. 1 depends on the exact value of the conversion factors. Relatively high factors (as stated by Brody 1945) cause a downward shift in the position of the lines compared with relatively low conversion factors (as stated by Morowitz 1968): the same energetic value is reached at a lower lipid content if the conversion factor for lipids is higher. If a more realistic value of 8.5 cal mg⁻¹ is used for the mixture of lipids extracted by commonly used extractants, the position of the lines is again higher than those based on the Brody conversion factors (Fig. 1).

Figure 1 conveniently shows the influence of changes in proportions of lipids or carbohydrates on the caloric value: with each 1% increase in lipids it rises by about 0.03 cal mg⁻¹ and with each 1% increase in carbohydrates it declines by about 0.015 cal mg⁻¹.

Because their proportions of lipids are rarely <4% and >25% and those of carbohydrates rarely <1% and >40% (see above), the caloric values of bivalves will usually be restricted to a range between ~5.2 and ~6.2 cal mg⁻¹ organic matter. The window for probable caloric values of bivalve organic material has been indicated in Fig. 2, using values for carbohydrate and lipid proportions compiled in Table 2. The inner (hatched) part of the window contains >90% of the values, the larger window includes all but a few outliers. The position of these two outliers is indicated by an * both in Fig. 2 and in Table 2.

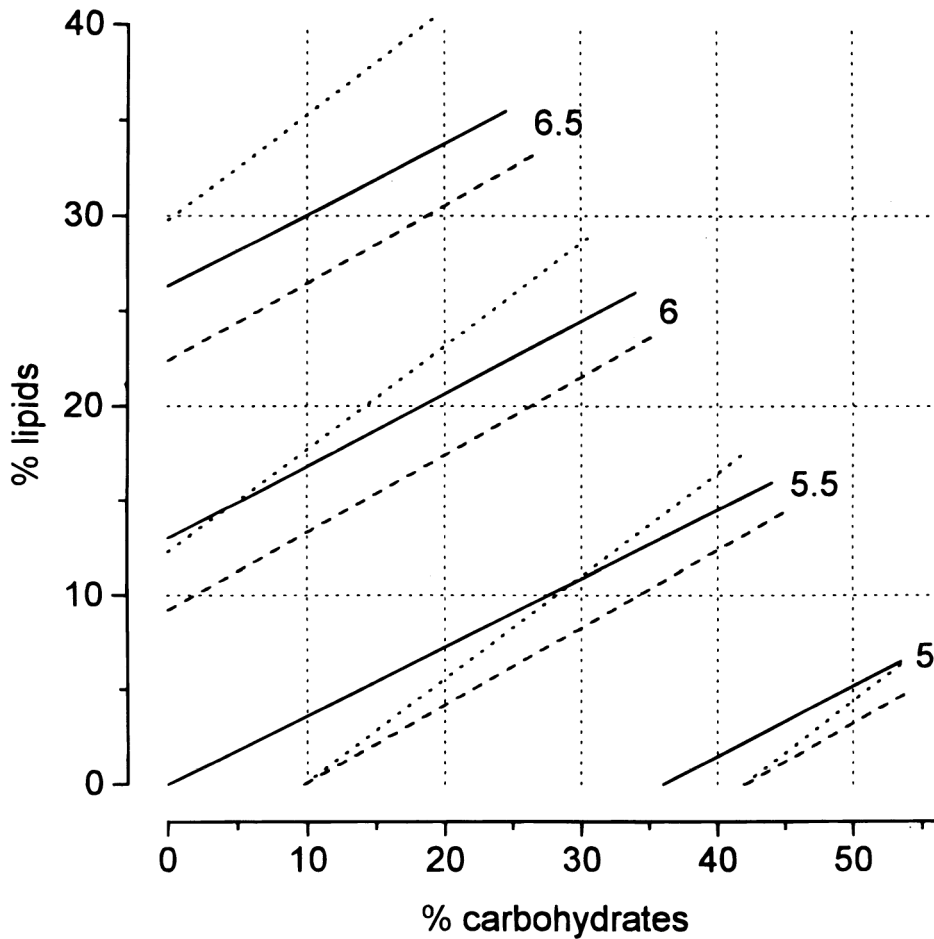


Figure 1 The relation between the proportion (in % of total organic material) of the main components (horizontal axis: carbohydrates and vertical axis: lipids) and the heat of combustion (isoenergetic lines in cal mg^{-1}). The isoenergetic lines end at a maximum summed share of carbohydrates plus lipids of 60% (leaving a minimum share of 40% for proteins). Broken lines: all conversion factors relatively high (after Brody 1945), solid lines: slightly lower conversion factors for both lipids (9.3 instead of 9.45) and proteins (5.5 instead of 5.65) (after Morowitz 1968), and punctuated lines: conversion for lipids at a realistic level for most frequently used way of extraction (8.5 instead of 9.3 or 9.45) and for proteins at 5.65. In all cases a conversion factor of cal mg^{-1} is used for carbohydrates.

Caloric values beyond the window shown in [Fig. 2](#) must be rare. Values < 5.0 can occur only if the proportion of carbohydrates exceeds 40% (at 0% lipids) or 50% at a normal minimal share of lipids. Similarly, values of $> 6.4 \text{ cal mg}^{-1}$ can occur only at lipid contents $> 25\%$. Such extreme values of lipid and carbohydrate contents have rarely been observed in bivalves ([Table 2](#)).

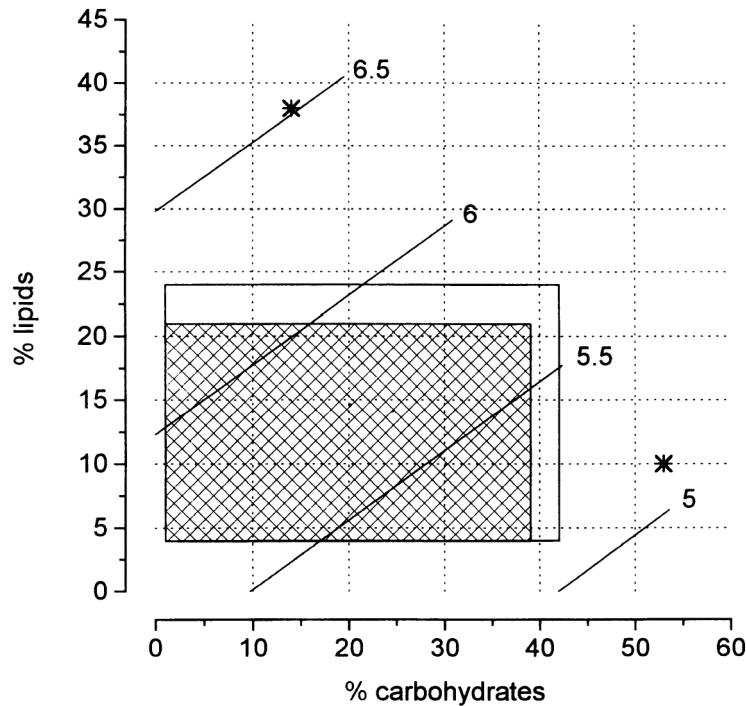


Figure 2 The window of probable caloric values (cal mg⁻¹ organic matter) of bivalves as indicated by commonly occurring ranges of carbohydrate and lipid contents. Isoenergetic lines calculated according to the most realistic conversion factors, viz. the punctuated lines in Figure 1. The outer border of the window includes all but two (indicated by *) published combinations of carbohydrate and lipid proportions of organic material of marine bivalves (Table 2).

Seasonal variation within species

Seasonal changes in biochemical composition are generally related to the annual cycles of growth and reproduction. Moreover, there are year-to-year differences due to variability in such conditions as food availability and temperature (e.g. Trevallion 1971).

Bivalves generally store carbohydrates in large amounts during their growing season and use them up over the remainder of the year. Part of the carbohydrates are usually transformed to lipids to build up eggs. Female reproductive tissues contain large proportions of lipids (e.g. Martinez 1991) to supply the energy needs of future larvae. Thus, there is generally a sudden drop in lipid content at spawning.

Frequently (but depending on species and geographic area), the seasons of growth and maturation more or less coincide. Lipid accumulation in gonads at the expense of glycogen is then hidden in the general increase of these components. In most species studied, proportions of carbohydrates and lipids rise and decline in parallel over most of the year. The resulting positive correlations between the proportions of carbohydrates and lipids appear the rule rather than the exception. Walne (1970) shows several examples in oyster species.

If carbohydrate and lipid proportions vary in parallel, seasonal variation in caloric values is reduced. Such values are then close to the diagonal from the lower left to the upper right corner of the window shown in Fig. 2. Indeed, Hughes (1970) observed little seasonal variation in the caloric content of the soft parts of *Scrobicularia plana*. Zwartz & Wanink (1993), however, found significantly lower values in winter than in

spring and summer in this species. In *Macoma balthica*, caloric values (as calculated from the biochemical composition (see Fig. 3A) hardly changed during the initial part of the growing season (both glycogen and lipid contents increased) and again during the non-growing season (both compounds declined), but caloric values significantly declined between spawning and the end of the growing season, when glycogen continued to rise but lipids declined (Fig. 3A).

Species using only lipids as reserve material will show much stronger seasonal fluctuations in energy per unit of weight, e.g. the crustacean *Mysis relicta* studied by Hakala (1979).

Geographic variation within species

Geographic variation in chemical composition may arise as a response to differences in environmental conditions and from different genetic backgrounds. In the *Cerastoderma glaucum*-*C. lamarcki* complex, differences in chemical composition between populations of these cockles were related to genetic differences (Brock & Wolowicz 1994). These differences were particularly high in carbohydrate contents. At similar maturity stages, carbohydrate proportions were high along the French west coast and low in Baltic and Mediterranean areas. Baltic populations showed relatively high lipid contents.

In five species, data could be compiled from at least four different areas (Table 2). In *Cerastoderma edule*, *Mytilus edulis*, *Ostrea edulis* and *Tellina tenuis*, seasonal ranges of carbohydrate and lipid contents were more or less similar in the various areas. However, in *M. balthica* substantial variation in minimal and particularly maximal values of carbohydrate and lipid contents were present (Table 2). In this species, the proportions of carbohydrates were <15% of AFDW in parts of the Baltic (Poland, Alands) year round, but rose to values > 30% at the end of the growing season in other parts of the Baltic (Finland) and in the Wadden Sea (The Netherlands). Hummel et al. (1996) observed very low carbohydrate proportions (<5% of the AFDW) near the southern edge (estuaries of Gironde and Loire in France) of the area of distribution of *M. balthica*. In samples of this species collected simultaneously in more northern areas in France and The Netherlands, they found carbohydrate proportions between 10% and 20%. Proportions of lipids in *M. balthica* varied significantly from place to place even within the rather small area of the Gulf of Gdansk: around 15% of AFDW in the inner bay (where growth is slow and weights are low) and 20–40% outside where growing conditions are better (Wenne & Styczynska-Jurewicz 1985, 1987, Bonsdorff & Wenne 1989). As in *Cerastoderma glaucum* (Brock & Wolowicz 1994) and *Mytilus edulis* (Pazikowska & Szaniawska 1988), lipid contents of *M. balthica* appear to be generally higher in the Baltic than elsewhere (Table 2). Lipid contents and consequently also caloric values in bivalves such as *Macoma*, *Cerastoderma* and *Mytilus* appear to be generally larger in the Gulf of Gdansk and some other Baltic areas than in the North Sea and adjacent areas (Table 2). So far, it is not clear whether this difference is an adaptation of bivalves to brackish conditions or a consequence of a possible difference in the composition of the local phytoplankton used as food (Wenne & Styczynska-Jurewicz 1987).

As a consequence of the large local variation in lipid proportions, the caloric contents of *M. balthica* varied significantly from place to place (Fig. 3B). This variation was even larger than the seasonal variation in caloric value within any area. Thus, caloric values for a certain species observed in one place cannot be used indiscriminately for other areas. Particularly in the Baltic, caloric values will be higher than in other areas (compare Fig. 3B with Fig. 3A).

A value of lipids of 38% of the AFDW, as observed in *Macoma balthica* at the station with optimal growth in the Gdansk area, appears to be the highest value ever recorded in bivalves (compare Table 2). At the same time carbohydrate contents were as low as 14% of the AFDW. According to Fig. 1, a caloric value

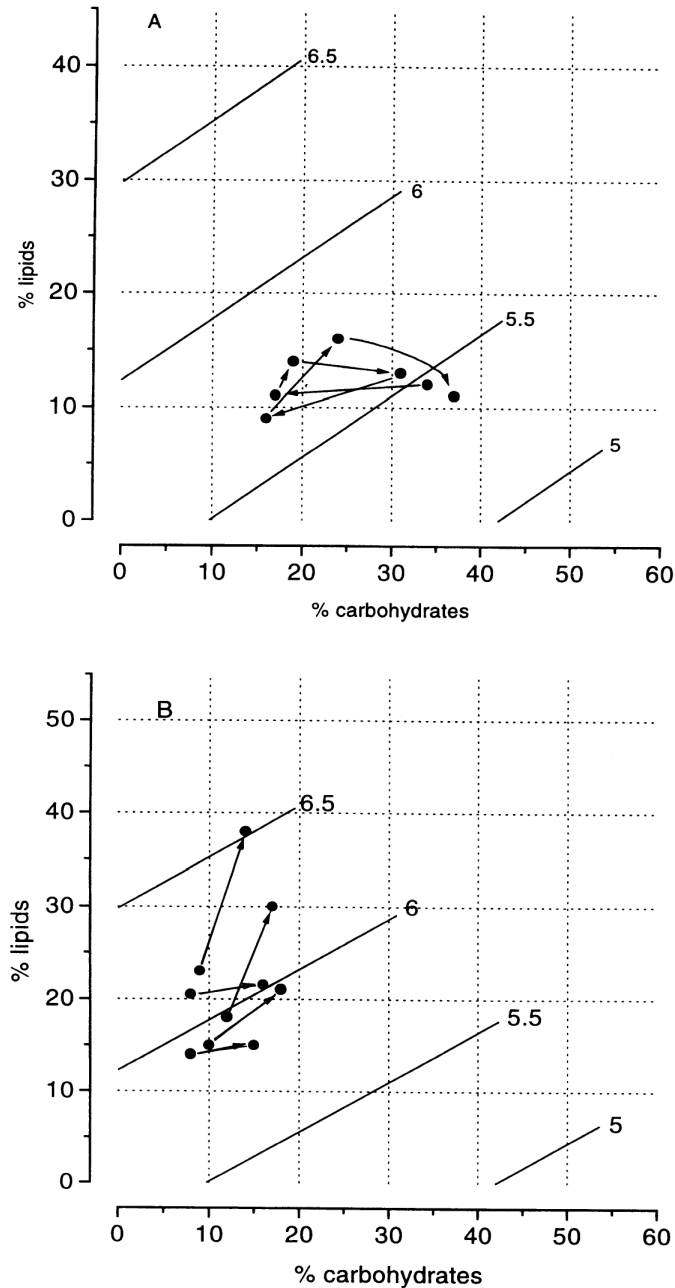


Figure 3 Seasonal changes in the chemical composition of the AFDW of the bivalve *Macoma balthica* in two areas : (A) at one station in the westernmost part of the Wadden Sea, The Netherlands, during two years (data from Beukema & De Bruin 1977) and (B) at five stations in the Gdansk Bay, Poland (data from Wenne & Styczynska-Jurewicz 1985). Arrows indicate seasonal changes: (in A) from summer (three rightmost points) via winter (two leftmost points) to spring (top two points) and (in B) from winter (leftmost points of each pair) to summer for five stations separately (lower values from inner part of the Bay, higher values from deeper outer parts). Caloric values are indicated by isoenergetic lines as in Figure 1 (viz. the punctuated lines of Fig. 1, showing caloric values at realistic conversion factors).

amounting to 6.5 cal mg^{-1} AFDW can be expected at this biochemical composition. This high value is clearly beyond the limit for the great majority of bivalves shown in Fig. 2.

Direct calorimetry

The most commonly used instruments for the determination of the caloric value of dried marine animals appear to be the Parr semi-microbomb (Paine 1971) and particularly the Phillipson microbomb (Phillipson 1964). Both use benzoic acid ($6.318 \text{ cal mg}^{-1}$) as a standard for calibration. The accuracy of these calorimeters at standard operation is roughly equal, with a coefficient of variation (of a number of estimates on the same material) amounting to maximally a few percentage points (Paine 1971). However, several corrections should be applied and precautions taken to reach maximal precision and avoid systematic errors. Recommended procedures are discussed in some detail by Paine (1971) and Fraschetti et al. (1994).

Corrections

The main corrections are for acid production (from N-, S-, or Cl-containing compounds) and carbonate breakdown (formation of CaO by an endothermic reaction). Strictly speaking, these corrections should be applied, but in practice their magnitude is often negligible. Moreover, they partly counterbalance each other.

In marine animal material, the acid correction generally amounts to about 1% or 2%, to be subtracted from the estimated caloric value (Paine 1971). This is not usually done, although there are favourable exceptions (Thayer et al. 1973, Wacasey & Atkinson 1987). The overestimates caused by omission of this correction will be slight, usually less than 0.1 cal mg^{-1} .

The heat loss by carbonate breakdown is more variable, depending of course on the carbonate content of the material. It can lead to gross underestimates of caloric values in such materials as ground corals, echinoderms, sponges and opisthobranchs (Paine 1964, 1971). In bivalves and other molluscs it is highly recommended that the soft parts be separated from the shells. This separation not only reduces the correction for carbonate breakdown to negligible values ($<1\%$, to be added to the estimates), it also promotes the homogeneity and inflammability of the material to be combusted. Ash content of the dried soft parts of bivalves usually amounts to about 10%, up to occasional values of around 20% (references can be found in several publications mentioned in Table 2, namely, where the percentages are printed in italics).

Sources of serious bias

The ash content of the combusted material should be estimated separately at an adjustable temperature, close to 500°C . At higher temperatures, several salts would decompose, resulting in an underestimate of ash weight and consequently an overestimate of ash-free dry weight ($\text{AFDW} = \text{DW} - \text{ash}$) and thus an underestimate of the caloric value on an AFDW base. For the same reason, the ash content cannot be estimated by weighing the ash remaining in the bomb after combustion, as temperatures at combustion under the high oxygen pressure within the bomb will exceed 600°C by far. At this temperature, weight losses of carbonate can already be significant (Paine 1964, 1971). Therefore, such determinations are bound to produce underestimates, both in ash content and consequently also in calories per unit of AFDW. Sisula & Virtanen (1977) found the underestimate of ash content determined by oxygen-bomb combustion to be substantial (about one-third in mollusc tissue). In plant material, Reiners & Reiners (1972) found an underestimate amounting to nearly a quarter. Thus, particularly at ash contents of more than a few per cent, ash weights

should be determined independently. Even then, endothermic reactions within the bomb can lead to serious underestimates of caloric values of material with high ash content.

Other sources of serious bias include incomplete combustion, incomplete drying (see above) and prolonged storage. Incomplete drying will lead to underestimates of the caloric value. The effect of prolonged storage appears to be less uniform. Various organic materials tend to oxidize on standing, even when well dried or frozen. In particular, the oxidation of lipids would cause a decline in the caloric value of the material. However, any further loss of water during storage would have the opposite effect. Apparently, the latter effect prevailed in the experiments reported by Sisula & Virtanen (1977).

The error caused by the presence of tightly bound water is inevitable. Usually, it will amount to a few percentage points. At high contents of inorganic material, it can be more significant and will then generally be accompanied by a similar error caused by endothermic reactions of carbonate breakdown. Such errors and inadequate measurements of ash contents will explain the unrealistically low values so often associated with organisms characterized by a high ash content. In particular, this will be the case if errors accumulate: at high ash contents errors originating from improper ash determination and from endothermic reactions will both be significant. Paine (1964) presents actual examples.

Caloric values observed in bivalves

From the literature, 95 values (from 73 species) could be compiled (Table 3) of caloric values from bomb calorimetry and that were stated to be on an AFDW base for the entire soft parts of marine bivalves (additional values on a wet- or dry-weight base and for separate parts were not included). The average of these 95 estimates amounts to 5.21 cal mg^{-1} . Note that Table 1b presents a slightly different mean value for bivalves (namely, 5.30 as an average of 115 estimates). This difference will have arisen from $115 - 95 = 20$ times of multiple use of individual values in the calculation of the average given in Table 1b.

Three out of these 95 values will be excluded from further discussion because they are far beyond the range of probable values (namely, the three figures 3.5, 4.05, and 6.95).

In 32 out of the remaining 92 cases, the ash content had not been determined separately (or statements on this point were lacking). Most (22) of these 32 determinations stem from one publication (Dauvin & Joncourt 1989). The average value of these 32 imperfect determinations of caloric value amounts to 4.86 cal mg^{-1} AFDW. Because of the inadequate method of ash determination (see above) this average must be an underestimate of true caloric values of bivalves. It is indeed below the range of probable values shown in Fig. 2.

In 60 cases (marked by at least one * in Table 3), ash contents were assessed by separate ashing at a controlled temperature. The average of the caloric values thus determined amounted to 5.43 cal mg^{-1} AFDW, with a standard error of 0.05. As expected, it is significantly higher than the above 32-values mean of 4.86 for estimates without separate ash determination ($p < 0.001$, both in a Wilcoxon and a t-test). The difference between the two averages does not necessarily reflect the error in ash determination, as there are other differences. Some of the higher estimates included an acid correction (marked by ** in Table 3) and these tend to be slightly higher (** values average 5.55 compared with 5.30 for * values). This is contrary to expectation, as an acid correction should be subtracted from the estimate (see above). The 32 acidcorrected values were taken from only two publications (Thayer et al. 1973, Wacasey & Atkinson 1987).

Table 3 Published records of caloric values (cal mg^{-1} AFDW) in various species of bivalves as obtained by bomb calorimetry (micro: Phillipson micro-bomb; semi: Parr semi-micro bomb; bomb: Gallenkamp bomb). Applied precautions and corrections are indicated: * separate ash determination, ** also acid corrections, *** separate ash

determinations and correction for endothermic carbonate breakdown (if shells were not removed). The three odd values printed in italics were not included in calculations of mean values.

Species	Calmg ⁻¹ AFDW	Type	Corr	Reference
<i>Abra alba</i>	5.23	micro	*	Brunswick (1973)
<i>Abra alba</i>	4.61	micro		Dauvin & Joncourt (1989)
<i>Abra prismatica</i>	4.49	micro		Dauvin & Joncourt (1989)
<i>Anadara transversa</i>	5.44	micro	**	Thayer et al. (1973)
<i>Anomia simplex</i>	5.66	micro	**	Thayer et al. (1973)
<i>Arctica islandica</i>	5.07	micro	*	Arntz & Brunswick (1975)
<i>Arctica islandica</i>	4.71	semi	*	Steimle & Terranova (1985)
<i>Astarte borealis</i>	5.37	semi	**	Wacasey & Atkinson (1987)
<i>Astarte montagui</i>	5.25	semi	**	Wacasey & Atkinson (1987)
<i>Astarte sulcata</i>	5.69	micro		Norrbin & Båmstedt (1984)
<i>Astarte undata</i>	5.09	semi	*	Steimle & Terranova (1985)
<i>Aulacomya ater</i>	5.85	semi	*	Duarte et al. (1980)
<i>Callista chione</i>	4.67	micro		Dauvin & Joncourt (1989)
<i>Cardium echinatum</i>	4.54	micro		Dauvin & Joncourt (1989)
<i>Cardium edule</i>	4.92	bomb		Chambers & Milne (1979)
<i>Cardium edule</i>	5.18	micro		Swennen (1976)
<i>Cerastoderma edule</i>	5.29	semi	*	Zwarts & Wanink (1993)
<i>Cerastoderma glaucum</i>	5.07	micro	*	Ivell (1979b)
<i>C. glaucum</i> (oven-dried)	5.11	micro	*	Ivell (1983)
<i>C. glaucum</i> (freeze-dried)	5.29	micro	*	Ivell (1983)
<i>Chione cancellata</i>	5.61	micro	**	Thayer et al. (1973)
<i>Chlamys islandica</i>	5.27	semi	**	Wacasey & Atkinson (1987)
<i>Chlamys islandicus</i>	5.54	semi	*	Steimle & Terranova (1985)
<i>Choromytilus chorus</i>	5.62	semi	*	Duarte et al. (1980)
<i>Clinocardium ciliatum</i>	5.43	semi	**	Wacasey & Atkinson (1987)
<i>Corbula gibba</i>	4.58	micro		Dauvin & Joncourt (1989)
<i>Crassostrea gigas</i>	4.85	micro		Héral & Deslous-Paoli (1983)
<i>Crassostrea virginica</i>	5.07	micro		Dame (1972)
<i>Crassostrea virginica</i>	6.03	micro	**	Thayer et al. (1973)
<i>Cultellus pellucidus</i>	5.02	micro		Dauvin & Joncourt (1989)
<i>Cyclocardia borealis</i>	4.59	semi	*	Steimle & Terranova (1985)
<i>Dosinia exoleta</i>	4.47	micro		Dauvin & Joncourt (1989)
<i>Dosinia lupinus</i>	4.43	micro		Dauvin & Joncourt (1989)
<i>Ensis minor</i>	3.5			Slobodkin (1962)
<i>Glycymeris glycymeris</i>	4.96	micro		Dauvin & Joncourt (1989)
<i>Hiatella arctica</i>	5.28	semi	**	Wacasey & Atkinson (1987)
<i>Macoma balthica</i>	4.05	micro		Gilbert (1973)
<i>Macoma balthica</i>	5.08	micro	**	Thayer et al. (1973)

Species	Cal mg ⁻¹ AFDW	Type	Corr	Reference
<i>Macoma balthica</i>	5.33	micro	*	Beukema & De Bruin (1979)
<i>Macoma balthica</i>	4.77	bomb		Chambers & Milne (1979)
<i>Macoma balthica</i>	5.25	semi	*	Zwarts & Wanink (1993)
<i>Macoma calcarea</i>	5.45	semi	**	Wacasey & Atkinson (1987)
<i>Macoma moesta</i>	5.41	semi	**	Wacasey & Atkinson (1987)
<i>Macoma tenta</i>	6.27	micro	**	Thayer et al. (1973)
<i>Mercenaria mercenaria</i>	6.15	micro	**	Thayer et al. (1973)
<i>Modiolus demissus</i>	5.76	micro	**	Thayer et al. (1973)
<i>Modiolus modiolus</i>	5.40	semi	*	Steimle & Terranova (1985)
<i>Musculus discors</i>	5.62	semi	**	Wacasey & Atkinson (1987)

Table 3 Continued

Species	Cal mg ⁻¹ AFDW	Type	Corr	Reference
<i>Musculus niger</i>	5.54	semi	**	Wacasey & Atkinson (1987)
<i>Mya arenaria</i>	4.98	semi		Edwards & Huebner (1977)
<i>Mya arenaria</i>	5.18	micro		Winther & Gray (1985)
<i>Mya arenaria</i>	5.18	semi	*	Zwarts & Wanink (1993)
<i>Mya truncata</i>	5.16	semi	**	Wacasey & Atkinson (1987)
<i>Mytilus chilensis</i>	5.72	semi	*	Duarte et al. (1980)
<i>Mytilus edulis</i>	5.40	bomb		Heppleston (1971)
<i>Mytilus edulis</i>	5.36	bomb	*	Hansson (1976)
<i>Mytilus edulis</i>	5.30	bomb		Chambers & Milne (1979)
<i>Mytilus edulis</i>	5.66	semi	**	Wacasey & Atkinson (1987)
<i>Mytilus edulis</i> (1–2 cm)	5.55	micro	*	Barkati & Ahmed (1990)
<i>Mytilus edulis</i> (4–7 cm)	5.47	micro	*	Barkati & Ahmed (1990)
<i>Mytilus edulis</i>	5.55	semi	*	Zwarts & Wanink (1993)
<i>Nucula belloti</i>	5.88	semi	**	Wacasey & Atkinson (1987)
<i>Nucula hanleyi</i>	5.00	micro		Dauvin & Joncourt (1989)
<i>Nucula proxima</i>	5.73	semi	*	Steimle & Terranova (1985)
<i>Nucula proxima</i>	5.52	semi	***	Steimle & Terranova (1985)
<i>Nucula turgida</i>	4.65	micro		Dauvin & Joncourt (1989)
<i>Nuculana minuta</i>	5.48	semi	**	Wacasey & Atkinson (1987)
<i>Nuculana pernula</i>	5.49	semi	**	Wacasey & Atkinson (1987)
<i>Ostrea edulis</i>	5.21	micro	*	Rodhouse (1978)
<i>Pandora glacialis</i>	5.12	semi	**	Wacasey & Atkinson (1987)
<i>Pecten groenlandicus</i>	5.50	semi	**	Wacasey & Atkinson (1987)
<i>Pecten irradians</i>	6.95	micro	**	Thayer et al. (1973)

Species	Cal mg ⁻¹ AFDW	Type	Corr	Reference
<i>Peripoloma abyssorum</i>	5.28	semi	**	Wacasey & Atkinson (1987)
<i>Pinna serrata</i>	6.32	micro	**	Thayer et al. (1973)
<i>Placopecten magellanicus</i>	5.21	semi	*	Steimle & Terranova (1985)
<i>Portlandia intermedia</i>	6.22	semi	**	Wacasey & Atkinson (1987)
<i>Scrobicularia plana</i>	5.10	bomb	*	Hughes (1970)
<i>Scrobicularia plana</i>	5.21	semi	*	Zwarts & Wanink (1993)
<i>Serripes groenlandicus</i>	5.56	semi	**	Wacasey & Atkinson (1987)
<i>Solecurtus scopula</i>	4.59	micro		Dauvin & Joncourt (1989)
<i>Spisula elliptica</i>	4.64	micro		Dauvin & Joncourt (1989)
<i>Spisula solidissima</i>	4.83	semi	*	Steimle & Terranova (1985)
<i>Tagelus divisus</i>	5.03	micro	**	Thayer et al. (1973)
<i>Tapes rhomboïdes</i>	4.45	micro		Dauvin & Joncourt (1989)
<i>Tellina crassa</i>	4.85	micro		Dauvin & Joncourt (1989)
<i>Tellina fabula</i>	4.64	micro		Dauvin & Joncourt (1989)
<i>Tellina squalida</i>	5.16	micro		Dauvin & Joncourt (1989)
<i>Teredo navalis</i>	5.67	micro	**	Thayer et al. (1973)
<i>Thracia phaseolina</i>	4.90	micro		Dauvin & Joncourt (1989)
<i>Thyasira flexuosa</i>	4.65	micro		Dauvin & Joncourt (1989)
<i>Thyasira gouldi</i>	5.32	semi	**	Wacasey & Atkinson (1987)
<i>Venus casina</i>	4.73	micro		Dauvin & Joncourt (1989)
<i>Venus fasciata</i>	4.73	micro		Dauvin & Joncourt (1989)
<i>Venus ovata</i>	5.17	micro		Dauvin & Joncourt (1989)
<i>Yoldia hyperborea</i>	5.40	semi	**	Wacasey & Atkinson (1987)

Even when ash determinations were performed in the correct way, unrealistically low caloric values (between 4.5 and 5.0) resulted in a few cases (3 out of 56), whereas one value was unrealistically high (the already mentioned one of 6.95). If these four outliers are excluded, the average and median values of the remaining 57 acceptable observations with correct ashing amount to 5.47 cal (22.9 J) and 5.43 cal (22.7 J) mg⁻¹ AFDW, respectively. This mean is only 1% lower than the average of 5.55 for the determinations including an acid correction (this group did not contain clear outliers). Thus a representative value for directly determined caloric values for the soft parts of marine bivalves would be close to 5.5 cal (23.0 J) mg⁻¹ AFDW. Because of inevitable imperfect drying of the material used, a likely “true” (but in practice not obtainable) value will be between 5.6 and 5.7 cal mg⁻¹ AFDW (dry in the sense of completely waterfree organic matter). The latter value is not too far from the centre of the window shown in Fig. 2. Note that Fig. 2 refers to completely water-and ash-free material, composed exclusively of carbohydrates, proteins and lipids.

Comparisons between estimates from two methods

Wet oxidation versus other methods

Craig et al. (1978) and Henken et al. (1986) showed that the wet-oxidation method consistently underestimates caloric values as compared to both direct bomb calorimetry and indirect calculation from the proximate chemical composition. Wet oxidation appears to be a less appropriate method and will not be further discussed.

Bomb calorimetry versus calculations from chemical composition

Relatively few investigators determined caloric values of bivalves by the two methods from the same material. Some of them found higher values by direct calorimetry (Héral & Deslous-Paoli 1983), others by the indirect method (Beukema & De Bruin 1979, Shafee 1981, Barkati & Ahmed 1990). In most cases, causes of the observed differences can easily be identified.

In their analysis of oysters *Crassostrea gigas*, Héral & Deslous-Paoli (1983) will have underestimated at least one of the main biochemical components, because the sums of their estimates account for only 64–84% of the total AFDW. Using a mean deficit of 28.8%, their estimate of 4.3 cal mg⁻¹ can be recalculated to 6.05 cal mg⁻¹ (assuming that the deficit had the same caloric value as the determined fraction). A further correction of this estimate can be made by application of a more realistic conversion value for the lipid fraction (8.5 instead of 9.45), resulting in a twice-corrected estimate of 5.9 cal mg⁻¹. The difference with their direct estimate of 4.85 (or 4.92 cal mg⁻¹ after application of a separately determined ash content) is still large. The chemical composition of the material (>12% lipids and >6% carbohydrates) suggests (Fig. 1) a value closer to 5.9 than to 4.9 cal mg⁻¹ AFDW. Therefore, their directly obtained value will have been a serious underestimate as well. Incomplete drying of the material may have been the common cause of both the too low direct estimate and the large unexplained deficits in the proximate biochemical composition.

Estimates by the two methods in soft parts of mussels *Mytilus edulis* by Barkati & Ahmed (1990) were similar, though on average slightly higher by the indirect method (which involved the attribution of a caloric value of 4.2 cal mg⁻¹ to all material not accounted for in the performed analyses, and which did not include one for carbohydrates). A reduced conversion factor for lipids (8.5 instead of the 9.45 cal mg⁻¹ applied by the authors) would result in nearly identical estimates for the two methods. Any more detailed comparison is meaningless because the authors used mussels of different size groups in the two kinds of estimates.

The analysis of soft parts of the scallop *Chlamys varia* by Shafee (1981) showed higher values by the indirect method than by bomb calorimetry. The differences were particularly large in gonads and remain large even if a lower conversion factor (of 8.5) for lipids is applied. As the author states, the difference is inexplicable.

In their analysis of the soft parts of *Macoma balthica*, Beukema & De Bruin (1979) found almost equal values by the two methods: 5.33 cal mg⁻¹ AFDW by bomb calorimetry and slightly higher values for the indirect method. Their maximal calculated estimate (using an unrealistically high conversion factor of 9.45 cal mg⁻¹ for lipids and attributing a value of 4.1 cal mg⁻¹ for all ash-free material not accounted for in the chemical analyses) amounted to 5.68 cal mg⁻¹ AFDW. Application of their separately determined caloric value of 8.62 cal mg⁻¹ for the chloroform-extracted lipids reduces the indirect estimate to 5.59 cal mg⁻¹ AFDW. For a comparison with the direct estimate (in which material dried at 60 °C was used), this value should be further reduced, as drying at this temperature does not remove all tightly bound water. Part of the material not accounted for in the chemical analysis will have been water, not having any caloric value at

combustion. According to the results of drying experiments by the authors, this will apply to about 3% of the weight of the material dried at 60 °C. Therefore, a comparable indirect estimate further drops to 5.48 cal mg⁻¹ AFDW and this estimate comes close to the direct estimate of 5.33 cal mg⁻¹ AFDW. A difference of only about 3% remains unexplained. Maybe, the conversion factors for proteins (6.25 for N to protein and 5.65 for protein to calorie) were too high for the material used. A factor of 5.8 instead of 6.25 for the conversion of N to protein, as recommended by Gnaiger & Bitterlich (1984) and applied by Thompson & MacDonald (1990), might have been more realistic and would lead to almost complete correspondence between the directly and indirectly determined caloric values in *Macoma balthica*.

Note that the above values of 5.33 and 5.48 refer to a special definition of DW, namely, the weight obtained after prolonged drying at only 60 °C, which does not evaporate all water. A “true” caloric value of completely water-free material would amount to about 5.6 rather than to about 5.5 cal mg⁻¹ AFDW. The higher value is theoretically rather than practically obtained, because more thorough drying involves the risk of unnoticed disappearance of volatile material and oxidation of easily oxidized fractions. Freeze-drying appears to be preferable to minimize such losses and will result in slightly higher caloric values. Indeed Ivell (1983) found 2–4% higher caloric values after freeze-drying than after oven-drying in tissue of *Cerastoderma* and *Loligo*.

Conclusions

The grounds for this review were (a) curiosity as to the cause of unrealistically low records of caloric values reported in the literature for soft parts of various species of bivalves and (b) the observation that there is often an imperfect correspondence between estimates obtained by direct and indirect calorimetric methods. Estimates calculated from chemical composition should give at least an indication of the range of possible values. Directly observed values lying outside such a range should be considered with extreme caution.

An example of incautious use of unrealistic values is the inclusion of some low caloric values for big molluscs in Fig. 3 and Table 2 of Griffiths (1977), giving rise to a statistically highly significant negative relationship between body weights and cal g⁻¹ AFDW in a sample of 21 species of big (>10mgDw) molluscs. Needless to say this correlation is, in my opinion, spurious.

Observed deviations from a normal distribution of energy content values (Slobodkin 1962, Brey et al. 1988) may have arisen from the inclusion of too many unrealistically low values. If so, such deviations do not need a specific explanation. Norrbin & Båmstedt (1984), who found comparatively high caloric values in all groups (compare Tables 1a and 1e), observed a fair agreement with a normal distribution. Compare Slobodkin (1962) for a discussion of the possible significance of a skewed distribution of energy values.

Comparisons between material originating from different species can be made most suitably on the base of ash-free dry material. Unlike estimates of organic carbon, estimates of AFDW can be obtained easily and cheaply. Moreover, many more data are available on energy values based on AFDW. Caloric values on the base of DW are also numerous, but they are much more variable than those based on AFDW (Brey et al. 1988). Values based on organic carbon appear to be even less variable (Salonen et al. 1976).

Ideally, AFDW should exactly equal the total of organic material. However, in practice AFDW is defined operationally as weight loss at incineration (for a certain period at a certain temperature) after drying (again prescribed in terms of duration and temperature). Even drying to constant weight is not a guarantee for complete evaporation of all tightly bound water. If values (of different material or obtained by different methods) are compared, it should be sure that they are based on an identical definition of AFDW.

Brey et al. (1988) propose a general conversion factor of 23 J (i.e. 5.5 cal) mg⁻¹ AFDW (in the operational sense) for aquatic macrobenthic invertebrates. They base this value on 255 observations in 229

species, showing an average of 23.0 and a median value of 23.1 Jmg⁻¹ AFDW. The similarity with the proposed values stated above for bivalves (average 23.0 and median 22.8) is somewhat misleading, because the representative bivalve values I propose refer to a selected set of data, including only those obtained after correct ash determination and excluding some outliers. Brey et al. (1988) did not exclude any value and in fact included several highly unrealistic figures. About 40 out of their 255 values were <21 Jmg⁻¹ AFDW, so at least 15% will have been underestimates. Exclusion of such low values would raise their median and average values for aquatic macrobenthic invertebrates to a more realistic value of about 24 J (or 5.7 cal) mg⁻¹ AFDW.

Reliable energy values for bivalve tissue (with a mean of 23 Jmg⁻¹ AFDW) appear to be slightly lower than those for most taxa of benthic invertebrates (compare the above corrected average of 24 J mg⁻¹ AFDW for all aquatic invertebrates). A comparison of the mean and median values for the various taxonomic groups included in Table 1b and in Brey et al. (1988) corroborates this conclusion. The relatively low energy values in bivalves will be due to the generally high share of carbohydrates in their body composition (Giese 1966). Bivalves are nearly all benthic animals and the bottom appears to be inhabited by animals with low caloric values as compared to planktonic ones (Wissing et al. 1973, Griffiths 1977, Norrbin & Båmstedt 1984). This difference between benthic and pelagic animals appears to arise not only from a difference in major-taxa composition of these two groups, but is also present within some major taxa (Griffiths 1977, Norrbin & Båmstedt 1984). Similarly, species living at high latitudes generally show higher energy values than those at lower latitudes (Norrbin & Båmstedt 1984). The use of lipids as a prevalent energy storage will be the common cause of such higher values in planktonic and high-latitude organisms.

Application of a general value for bivalve tissue of 23 J (or 5.5 cal) mg⁻¹ AFDW appears to be acceptable if some margin is allowed. Only seven out of the 55 published estimates that were obtained by a satisfactory procedure deviate more than 10% from this value, whereas a majority (nearly two out of three) of these values deviate less than 5% (see Table 3).

Acceptance of the use for specific populations of well established mean values, such as 23Jmg⁻¹AFDW for bivalves or 24 J for animal taxa with generally lower carbohydrate and higher lipid contents, involves the introduction into the estimates of an uncertainty that will generally be less than 5% or 10%. This uncertainty remains if the energy value is expressed in Jmg⁻¹ organic carbon instead of AFDW. A general average will be close to 46 Jmg⁻¹ C (Salonen et al. 1976), but again such values are higher in lipid-rich than in carbohydrate rich material (as the higher proportions of C in lipid-rich material do not fully compensate for the much higher energy values of such material). Therefore, despite the higher accuracy claimed for determinations based on organic C (Salonen et al. 1976), an uncertainty of about 5% cannot be circumvented without further chemical analysis.

In many cases, such extra uncertainty will be acceptable. Other terms of an energy budget of a population will be even less accurate, e.g. estimates of numerical densities, of food intake, or of metabolic expenses of individuals at varying activity levels. In many cases of population studies, the decision not to perform any actual measurements of caloric values, therefore, appears to be warranted. This is even more true in studies of energy flow in multi-species natural communities.

If it is decided to perform actual measurements of caloric values, both of the two principal methods deserve serious consideration. Indirect estimates from an analysis of the chemical composition of the material are satisfactory only if the components add up to close to 100%. Moreover, a separately determined appropriate conversion factor for lipids should be applied. There is one significant and unique advantage of the indirect method: it also allows an estimate of caloric values at incomplete physiological oxidation.

Direct estimates by a bomb calorimeter are satisfactory only in well dried material with relatively low carbonate contents. Ash content should definitely be determined separately.

Of course, application of both methods is preferable to the use of only a single one. Mutual checks can be applied and differences in the results may indicate errors that would otherwise have escaped attention.

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