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# Amino acids and hexosamines as indicators of organic matter degradation state in North Sea sediments

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*Abstract*

Sediment cores from six stations in the eastern North Sea were analyzed for protein amino acids, the nonprotein amino acids β-alanine and γ-aminobutyric acid and the hexosamines galactosamine and glucosamine, and bulk parameters (organic carbon, nitrogen, total hydrolyzable amino acids and carbohydrates) in order to establish the degradation state of sedimentary organic matter. The study sites were selected on the basis of their different physical settings and macrofaunal communities so that a broad quality range in the organic matter would likely be covered. To test if the molecular parameters provide a robust matrix for quality determination, we integrated our results with complementary literature data ranging from marine source organisms to deep-sea environments. A principal component analysis based on the mole percent contribution of amino acids showed that there are systematic variations in the amino acid spectra as a consequence of degradation of organic matter. Comparison with more established quality parameters such as hexosamines confirmed that amino acids reflect the degradation state of the organic matter. The amino acids glycine, serine, and threonine were enriched in the more degraded material, and others, such as phenylalanine, glutamic acid, tyrosine, leucine, and isoleucine, became depleted with increasing degradation state. Selective preservation of structural compounds (diatom cell walls, chitinous organic matter) vs. preferential breakdown of cell plasma material appears to be the reason for the contrasting behavior of these molecular compounds. Some of the essential amino acids for macrofauna nutrition (arginine, methionine, and histidine) occurred in lower concentrations in the North Sea sediments compared to organism tissue and therefore may be limiting to growth of deposit-feeders.

Coastal marine environments are sites of intensive organic carbon production, turnover, and burial. A substantial fraction of the organic material produced in the euphotic zone reaches the sediment, where it forms the major food input for the benthos. During passage through the water column and burial into the sediment, organic matter is degraded with the result that the more labile components are preferentially lost. The amount and degradability of organic material finally arriving at the sediment has been found to be one of the major structuring factors in the trophic composition of benthic communities (e.g. Pearson and Rosenberg 1978; Tenore and Rice 1980).

A variety of biomarkers that undergo either selective preservation or loss during diagenesis have been used as source and maturity indicators of organic matter in suspended material and in sediments. Compounds such as lignin (preferential preservation) and pigments (preferential loss) were usually used as maturity indicators (e.g. Cowie et al. 1992; Hedges and Prahl 1993; Boon and Duineveld 1996). However, degradation state indicators based on specific trace compounds are problematic to use in cross-system comparison studies because of the nonuniform initial distribution and compound-specific degradation pathways. Furthermore, the sources of organic matter input are often multiple and may vary strongly between stations. Therefore it is often difficult to distinguish between preferential breakdown of a certain compound and a variable input. Moreover, biomarkers occur in trace quantities and consequently are not important as energy or nitrogen sources for heterotroph organisms.

The limitations of biomarkers can be avoided by the use of ubiquitous biopolymers such as proteins, hexosamines, and carbohydrates that account for a major part of the organic material both in marine and terrestrial organisms, as well as in particulate organic matter (Cowie and Hedges 1994). Nitrogen-containing compounds (e.g. proteins and hexosamines) are suggested to play a regulatory role in benthic ecology because organic nitrogen usually limits heterotrophic growth (Tenore 1983).

Until now few comprehensive studies of these major compounds have been carried out in the context of organic matter quality. Depth profiles of amino acids and hexosamines in sinking suspended matter (e.g. Lee and Cronin 1984; Haake et al. 1992, 1993) and sediments (e.g. Haugen and Lichtentaler 1991; Cowie and Hedges 1992; Liebezeit 1993) indicate that some amino acids and hexosamines become relatively enriched during degradation, whereas others are preferentially utilized. This results in compositional changes that can be used to determine the decomposition state of the organic material. The enzymatic formation of nonprotein amino acids such as β-alanine and γ-aminobutyric acid from protein amino precursors such as glutamic acid and aspartic

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acid is a well-known criterion of organic matter quality (e.g. Whelan 1977; Lee and Cronin 1982; Ittekkot et al. 1984*a*; Cowie and Hedges 1994). The ratios of the precursors to their respective degradation products can be interpreted as a measure of the decompositional state of sedimentary organic matter.

In coastal areas where the upper layer of the sediments (<20–30 cm) is inhabited and extensively bioturbated by macrofauna, vertical concentration profiles cannot be used to study the time evolution of organic matter (Boudreau 1994). As an alternative, degradation states can be assessed on a horizontal plane by comparing sedimentary inventories between stations. In this study, we compare six stations in the North Sea that differ in organic matter degradation state as suggested by macrofaunal characteristics and physical setting along the major current pattern of the North Sea. However, plankton is the ultimate origin of organic matter at all stations.

For these six stations we report complementary data on bulk and molecular parameters such as total organic carbon, total nitrogen, amino acids (14 protein and 2 nonprotein), hexosamines (glucosamine and galactosamine), and total carbohydrates. These data allow a comparison of the degradation state and the potential food value to deposit-feeders. We integrated our data with those published by Cowie and Hedges (1994), who have conducted a cross-system study with material varying from coastal trap material to highly decomposed deep-sea sediments.

## Materials and methods

*General features of sampling stations*—Six stations in the eastern part of the North Sea have been selected so that a range of degradation states would be expected (Fig. 1). The most southern stations are the very shallow Brouwershavensche-gat A and B (BG-A, BG-B), then the 20–40-m deep Broad Fourteens (BF), Frisian Front (FF), and German Bight (GB), and finally the 280-m-deep Skagerrak (SK). Details of the physical and chemical characteristics of the study stations are summarized in Table 1. The water columns at the shallow coastal stations were completely mixed, but the 280-m-deep Skagerrak was temperature stratified, with a minimum water temperature of 6°C at the bottom. The GB, BG-A, and BG-B B have somewhat lower salinities compared to the other stations due to the influence of river runoff.

Sediment granulometry reflects the hydrodynamic regime of the North Sea to a large extent (Otto et al. 1990; Wiesner et al. 1990). The southern part, with the highest current velocities, is largely dominated by sandy sediments (BG-A, BG-B, BF) mixed with varying amounts of mud. Temporary deposition areas (FF, GB), where slow currents allow particles to stay a few weeks or longer, consist of fine sands mixed with clay (Eisma and Kalf 1987a,b). Silty sediments are found at station GB and the deep final deposition area SK (Table 1).

The major source of organic material in the North Sea is local primary production as indicated by the stable carbon isotope composition ($\delta^{13}C$) of about −21‰ to −22‰ at all stations (Table 1), matching the value typically found in

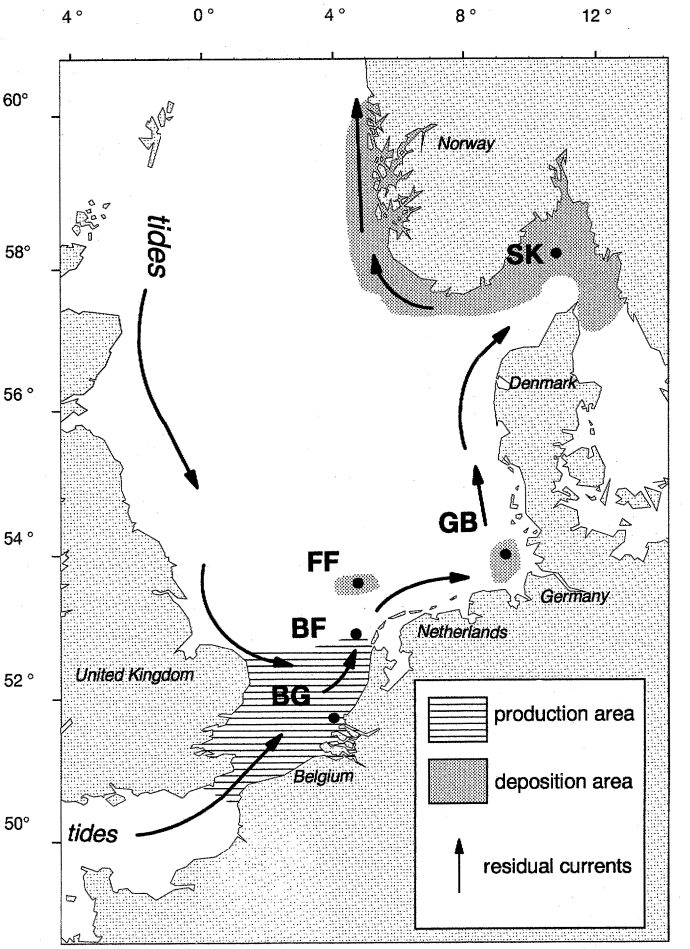


Fig. 1. Sampling stations in the North Sea: Brouwerhavenschegat A and B (BG-A and BG-B), Broad Fourteens (BF), Frisian Front (FF), German Bight (GB), and Skagerrak (SK). The main transport route of water and suspended matter is indicated by arrows.

North Sea marine plankton (e.g. Dauby et al. 1994). Most of the primary production takes place in the shallow, unstratified southern part of the North Sea (BG-A, BG-B, BF, FF). After the spring bloom, the fresh material sinks to the bottom, ages, and is transported by bed-load transport successively to the north, with SK as the final deposition area (Eisma and Kalf 1987*a,b*; van Weering et al. 1993; Lohse et al. 1995).

Factors such as primary productivity, water depth, sediment accumulation rate, and macrofaunal and microbial biomass are responsible for the amount and nutritional quality of organic matter in sediments. The quality of sedimentary organic matter as revealed by bacterial turnover rates is highest at station BF, which receives fresh material, and is lowest at stations with more refractory organic matter (e.g. SK, FF). Macrofauna biomass is lowest at the sandy stations (BG-B, BF) (Table 1). The stations have been chosen on the basis of these contrasting features to make sure that a broad quality range would be covered.

*Sampling procedure*—Stations BF, FF, GB, and SK were visited with the RV *Pelagia* in August 1994, and the south-

Table 1. Characteristics of the sediment at all sites.

| | | Bhgat-B | Broad fourteens | Frisian front | Bhgat-A | German bight | | Skagerra | Source* | Sed. depth (cm) |
|---|---|---|---|---|---|---|---|---|---|---|
| Latitude | | 51°46′N | 53°00′N | 53°42′N | 51°45′N | 54°05′N | | 58°12′N | | |
| Longitude | | 3°46′E | 3°52′E | 4°30′E | 3°48′E | 8°09′E | | 10°15′E | | |
| Water depth | m | 2.7 | 28 | 39 | 4.9 | 20 | | 270 | | |
| Temperature of bottom water | °C | 11.6 | 17.7 | 17 | 11.2 | 18.8 | | 6.5 | | |
| Salinity | | 31.6 | 34.6 | 34.3 | 31.5 | 31.2 | | 35.1 | | |
| Near-surface flow velocity | cm $s^{-1}$ | — | 30–40 | 10–35 | — | 15–40 | | ? | 1 | |
| Sedimentation rate | cm 100 yr | — | 0 | 10 | — | 25–60 (30) | | 35 | 2 | |
| Median grain size | μm | 285 | 233 | 77 | 143 | 38 | | 12 | | 0–15 |
| Percentage silt+clay (particles <50 μm) | % | 0.4 | 1 | 48 | 33 | 64 | | 98 | | 0–15 |
| Porosity | vol/vol | 0.39 | 0.37 | 0.53 | 0.45 | 0.68 | | 0.85 | | 0–15 |
| Chlorophyll a | μg $g^{-1}$ | — | 0.05 | 0.5 | — | 10.7,‡ 2.4§ | | 1.4 | | 0–1 |
| $\delta^{13}C$ | ‰ | −21.5 | −21.9 | −21.7 | −21.5 | −22.2 | | −22.2 | | 0–0.5 |
| Macrofauna biomass | g ADW $m^{-2}$ | 1.0 | 1.9 | 13.5 | 201† | 38.1 | | 11.4 | | 0–15 |
| Bacterial biomass | g C $m^{-2}$ | — | 2.7 | 5.0 | — | 11.4 | | 4.6 | 3 | 0–6.2 |
| Bacterial production | g C $m^{-2}$ $d^{-1}$ | — | 2.44 | 1.48 | — | 2.41 | | 0.38 | 3 | 0–6.2 |
| Bacterial turnover | *P/B* | — | 0.9 | 0.3 | — | 0.2 | | 0.08 | 3 | 0–6.2 |
| | | | | | | GB-1 | GB-2 | | | |
| TOC | mg $g^{-1}$ | 0.23 | 0.45 | 5.1 | 1.57 | 23.4 | 10.3 | 24.95 | | 0–15 |
| TN | mg $g^{-1}$ | 0.06 | 0.09 | 0.4 | 0.23 | 2.62 | 0.77 | 2.48 | | 0–15 |
| THAA | mg $g^{-1}$ | 0.11 | 0.26 | 1.12 | 1.06 | 8.9 | 2.72 | 6.11 | | 0–15 |
| Carbohydrates | mg $g^{-1}$ | 0.13 | 0.11 | 1.15 | 1.04 | 7.74 | 3.14 | 7.17 | | 0–15 |
| Hexosamines | mg $g^{-1}$ | 0.01 | 0.02 | 0.14 | 0.09 | 0.84 | 0.41 | 1.17 | | 0–15 |
| C:N | Molar ratio | 4.6 | 5.7 | 13.5 | 7.9 | 10.4 | 15.6 | 11.7 | | 0–15 |
| THAA:hexosamines | Molar ratio | 13 | 4 | 2 | 245‖ | 32 | 8 | 16 | | 0–15 |
| Asp:β-Ala | Molar ratio | 37 | 71 | 11 | 16 | 40 | 7 | 5 | | 0–15 |
| Glu:γ-Aba | Molar ratio | 44 | 448‖ | 13 | 16 | 21 | 12 | 8 | | 0–15 |

*1, Boon and Duineveld 1996; 2, de Haas and van Weering [1997] and pers. comm.; 3, van Duyl and Kop (1994), who visited the same stations during a cruise in August 1992.
† High biomass at BG-A because of some large animals.
‡ Surface layer (0–1 cm).
§ Deep layer (1–15 cm).
‖ Exceptionally high ratios due to low values of the denominator.

ernmost stations, BG-A and BG-B, were visited with the RV *Luctor* in May 1996 (Fig. 1). At each station four box cores were recovered and each was subsampled immediately with plastic liners having a diameter of 3.3 cm and a length of 50 cm. These subcores were successively sliced in vertical sections and samples from each horizon were pooled after sectioning. The cores were sliced at 0.25-cm resolution in the top centimeter, at 0.5-cm resolution down to 3 cm, at 1-cm resolution down to 7 cm, and at 1.5-cm thick slices down to 15-cm depth. The sediment was immediately frozen onboard and then freeze-dried.

*Chemical analysis*—Total organic carbon (TOC) and total nitrogen (TN) were determined on freeze-dried samples that had been finely powdered and homogenized. A 20–50-mg split was combusted at 1,010°C in a Carlo Erba elemental analyzer NA-1500 after removal of carbonate by in situ acidification with 25% HCl within silver sample cups (Nieuwenhuize et al. 1994). Reproducibility is ~2% for both TOC and TN.

Total hydrolyzable amino acids (THAA) and hexosamines were quantified by reverse-phase HPLC of their fluorescent derivatives (Roth 1971; Benson and Haare 1975) and comparison with known amounts of authentic standards (Sigma) following a modified procedure after Hill et al. (1979) and Lindroth and Mopper (1979) with precolumn *o*-phthaldaldehyde (OPA) derivatization. The OPA solution was prepared by dissolving 134 mg of OPA (Pierce) in 5 ml of methanol (gradient grade) and adding 100 μl of 2-mercaptoethanol (Merck). This solution was brought to 25-ml volume with borate buffer. The borate buffer was prepared by adjusting 0.4 M boric acid to pH 9.5 with 6 N NaOH. The OPA solution was prepared the day before use, stored in refrigeration, and protected from light.

About 0.1–0.25-g homogenized, freeze-dried samples were weighed into ampoules, hydrolyzed under nitrogen in 5 ml of 6 N HCl at 100°C for 24 h, centrifuged, and the supernatant was stored frozen at −40°C until analysis. A 250-μl aliquot of the hydrolysate was neutralized with 1 N NaOH and brought to volume with ultrapure, distilled Milli-Q water, resulting in 25–100 times dilution depending on expected concentrations.

Fluorescent OPA derivatives were formed online exactly 2 min prior to injection by adding 10 μl OPA solution to 100 μl of the neutralized sample. An autoinjector (WISP 717 plus) was used to standardize the derivatisation time. The HPLC system consisted of two WATERS pumps (model 510 and 6000), a guard column (Alltech), a WATERS fluorescence detector (model 420), and a Nova-Pak $C_{18}$ column (4 μm, 150 × 3.9 mm i.d.). Chromatographic peaks were recorded and integrated using Millennium software (WATERS). Sample concentrations were determined from integrated peak areas of the standards. An excitation wavelength of 328 nm and emission wavelengths of ≥450 nm were used for fluorometric detection. Eluant A was 0.04 M Na-acetate buffer at pH of 7 with 2% tetrahydrofuran and 10% methanol (HPLC grade). Eluant B was a mixture of 90% methanol (HPLC grade) and 10% 0.04 M Na-acetate buffer. Flow rate was 1 ml $min^{-1}$ and the gradient used was 100% A and 0% B to 0% A and 100% B in 21 min. The analytical precision expressed as standard deviation from multiple standard injections was <5% for most of the amino acids and hexosamines and 10–15% for methionine and the nonprotein amino acids. All glassware used in this procedure was cleaned with chromic acid solution to remove trace organic contamination. Method blanks showed negligible concentrations of amino acids and hexosamines from handling and reagent contamination compared to the sample concentrations reported in this paper. Our method does not allow measurement of tryptophan (Trp), cysteine (Cys), lysine (Lys), and proline (Pro), nor is it possible to identify the neutral forms of glutamic acid (Glu) and aspartic acid (Asp) named glutamine (Gln) and asparagine (Asn), respectively.

Total carbohydrate measurements were carried out using the phenol-sulphuric acid method (Liu et al. 1973). Approximately 10 mg of freeze-dried material was reacted with 10% phenol solution and concentrated sulphuric acid. The absorbence was read in a Perkin-Elmer Lambda 3B UV/VIS spectrophotometer at 485 nm against a reagent blank with a sediment–sulphuric acid interaction correction, and starch was used as a standard. The average reproducibility of the determinations was <5%.

Carbon isotopes have been determined using a Fisons elemental analyzer coupled online (via a continuous-flow interface) with a Finnigan delta S mass spectrometer. Results of the carbon isotope analyses are reported in the $\delta$ notation relative to Vienna PDB. Reproducibility based on replicate measurements was >0.1‰.

Chloroyphyll *a* was determined using reverse-phase HPLC according to Mantoura and Llewellyn (1983) after extraction in 90% acetone.

Macrofauna biomass was determined from three box cores of 50-cm diameter and >15-cm penetration depth at each station. Whole cores were sieved on 0.5-mm mesh width, stored in 4% buffered formaldehyde, stained with Rose Bengal, and sorted under a stereo microscope. Ash-free dry weight (AFDW) was measured by weight loss after combustion at 500°C. Grain-size spectra were measured with a Malvern particle sizer 3600 EC.

## Results

*Concentration of major organic components*—Organic carbon concentrations showed little variation with depth in the sediment (Fig. 2), except at station GB, where the surface layer is enriched in organic carbon compared to the deeper layers. The organic matter in the surface layer was also of higher quality compared to the deeper stratum as indicated by the higher Chl *a* content (Table 1). These results suggest that this layer was recently deposited and was not yet completely mixed by bioturbation, as at the other much more homogeneous stations.

The other major components (total nitrogen, carbohydrates, amino acids, and hexosamines) neatly followed the trends observed in the TOC profiles at all stations. Downcore compositional changes in the molecular THAA fraction were minor and are therefore not presented here. Because of the lack of trends in downcore profiles we used depth averaged values over a 0–15-cm horizon to compare among stations.

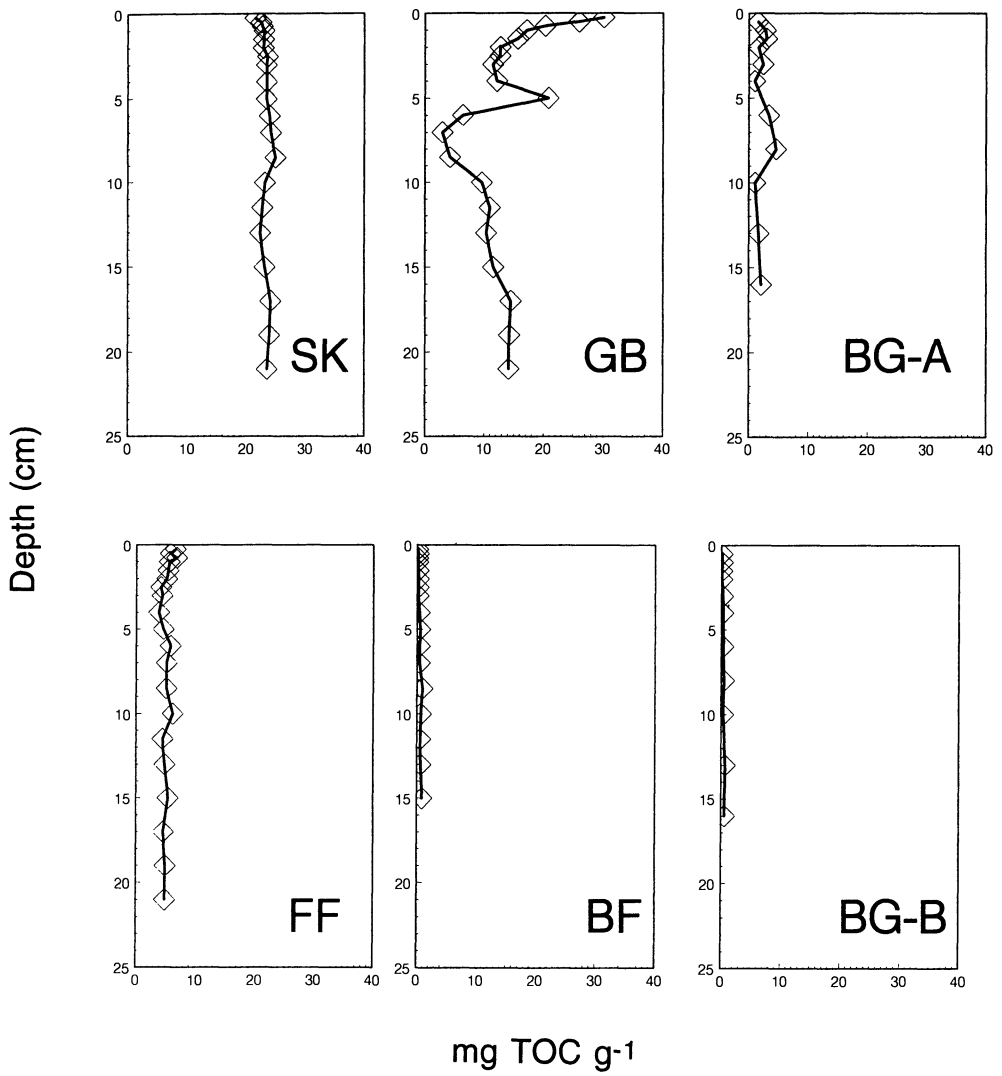


Fig. 2. Vertical profiles of total organic carbon (TOC) within the sediment at all stations. Stations are presented in the order proposed by the principal component analysis (*see discussion*).

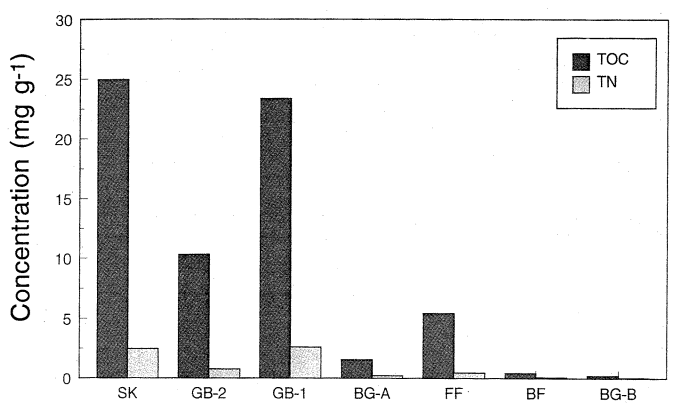


Fig. 3. Depth-averaged total organic carbon (TOC) and total nitrogen (TN). GB-1 (0–1 cm), GB-2 (1–15cm), and SK, FF, BG-A, BG-B, and BF (0–15 cm).

At station GB a distinction was made between the recently deposited organic material in the surface horizon (0–1 cm; GB-1) and the remaining 1–15 cm (GB-2).

Depth-averaged organic carbon contents (Fig. 3) varied from <0.5 mg TOC $g^{-1}$ in sandy sediments (BF and BG-B) stations to up to 24 mg TOC $g^{-1}$ in the silty to muddy sediments (SK and GB-1). Sediments at stations GB-2, FF, and BG-A showed intermediate organic carbon contents of ±2–11 mg TOC $g^{-1}$. The total amounts of the other components followed the same general trend as has been observed for TOC, with TN ranging from ±0.06 to 2.6 mg $g^{-1}$, THAA between ±0.1 and 9 mg $g^{-1}$, carbohydrates between ±0.1 and 8 mg $g^{-1}$, and hexosamines between ±0.01 and 1.2 mg (g DW)$^{-1}$ (Table 1). The molar C:N ratios of bulk organic matter varied between 5 and 16 (Table 1).

*Relative contribution of major compound groups*—At most stations, between ~20 and 60% of the TOC was identified as carbon derived from amino acids, carbohydrates,

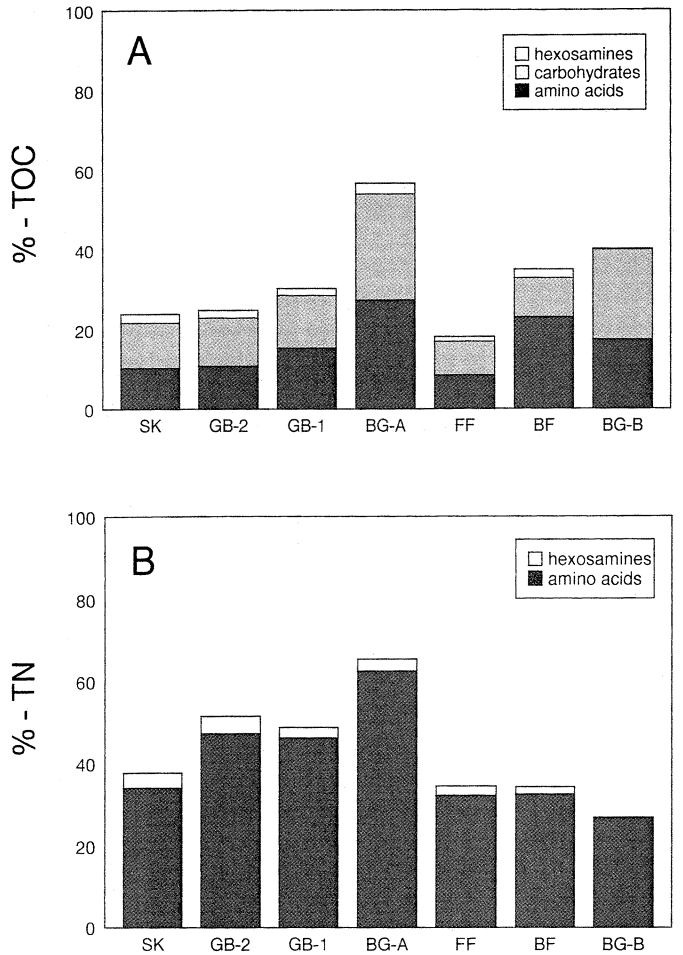


Fig. 4. Relative contributions of the major compounds to the total organic carbon pool (A) and to the total nitrogen pool (B).

and hexosamines (Fig. 4A), and the major fraction of the TOC remained unidentified. Amino acids and carbohydrates each composed between 8 and 28% of the TOC, and hexosamines accounted for a small fraction, with only ~2% TOC in most sediments. Compared to TOC, organic nitrogen could be identified to a larger extent (up to 66% of TN), with THAA and hexosamines contributing 25–60% and 0–4% of the TN in the sediment samples, respectively (Fig. 4B).

The identified fraction of TOC generally increased from the silty to muddy SK and GB sediments to the coarse grained BF and BG-B, suggesting a decreasing contribution of humified organic matter or other resistant biopolymers. The size of the identified TN fraction does not follow a clear trend.

*Relative contribution of molecular compounds—Amino acids*: These coastal sediments appeared to be rather similar in terms of relative concentrations of the individual amino acids (Fig. 5, Table 2). At all stations glycine dominated, with a contribution up to 22 mol%, followed by aspartic acid and alanine, which contributed 10–15 mol%. Glutamic acid, serine, valine, leucine, and threonine accounted for 5–10 mol%, and a minor group of arginine, isoleucine, phenylalanine, tyrosine, methionine, and histidine contributed <5 mol%. The molar C:N ratio of the THAA pool was ~3.5 at all stations.

Despite overall similarity between stations, some amino acids showed distinct molar concentration patterns (Fig. 5). Mole percent of glycine and methionine increased from stations BF and BG-B to SK and GB-2, whereas mole percent of leucine, isoleucine, and phenylalanine decreased. The contribution of the remaining protein amino acids varied less consistently between the stations.

The division of the protein amino acids into charge classes (Fig. 6) showed that the neutral amino acids (glycine + alanine + valine + leucine + isoleucine) dominate (45–50 mol%), and that acidic (glutamic acid and aspartic acid),

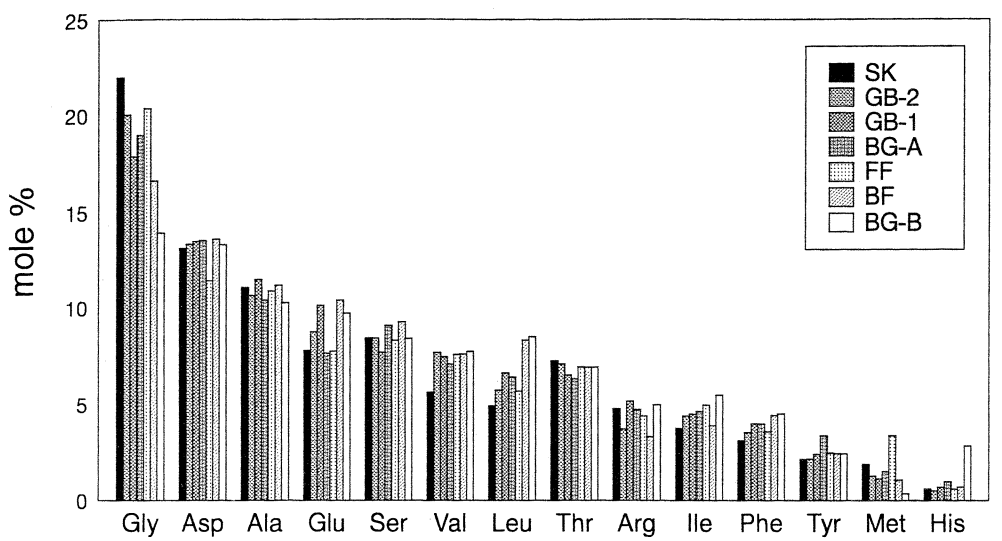


Fig. 5. Mole percent contributions of the individual protein amino acids to total hydrolyzable amino acid pool (THAA).

Table 2. Mole percent composition of amino acids in North Sea sediments and material used in the comprehensive PCA (Phy-B, algae (Brown 1991); Phy-C, phytoplankton (Cowie and Hedges 1992); Bac, bacteria; Zoo, zooplankton; Saan, Saanich Inlet; Dab, Dabob Bay; T, trap material; S1, 0–1-cm surface sediment; S2, 1–15-cm-deep sediment; Tur-ox, oxidized turbidite; Tur-red, unoxidized turbidite.

| | Gly | Asp | Ala | Glu | Ser | Val | Leu | Thr | Arg | Ile | Phe | Tyr | Met | His | β-Ala | γ-Aba |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | mol% | | | | | | | | |
| SK | 21.2 | 13.1 | 11.1 | 7.8 | 8.4 | 5.6 | 4.9 | 7.2 | 4.8 | 3.7 | 3.1 | 2.1 | 1.9 | 0.6 | 2.51 | 1.01 |
| GB-2 | 20.4 | 13.4 | 10.7 | 8.8 | 8.4 | 7.7 | 5.8 | 7.1 | 3.7 | 4.4 | 3.6 | 2.2 | 1.3 | 0.5 | 1.92 | 0.71 |
| GB-1 | 20.0 | 13.5 | 11.5 | 10.1 | 7.7 | 7.5 | 6.6 | 6.5 | 5.2 | 4.5 | 4.0 | 2.4 | 1.1 | 0.7 | 0.34 | 0.49 |
| BG-A | 19.0 | 13.5 | 10.4 | 7.6 | 9.1 | 7.1 | 6.4 | 6.3 | 4.7 | 4.6 | 4.0 | 3.4 | 1.5 | 1.0 | 0.82 | 0.47 |
| FF | 17.9 | 11.4 | 10.9 | 7.7 | 8.3 | 7.6 | 5.7 | 6.9 | 4.4 | 5.0 | 3.6 | 2.5 | 3.4 | 0.6 | 1.05 | 0.58 |
| BF | 16.6 | 13.6 | 11.2 | 10.4 | 9.3 | 7.6 | 8.3 | 6.9 | 3.3 | 3.9 | 4.4 | 2.4 | 1.1 | 0.7 | 0.19 | 0.02 |
| BG-B | 13.9 | 13.3 | 10.3 | 9.7 | 8.4 | 7.7 | 8.5 | 6.9 | 5.0 | 5.5 | 4.5 | 2.4 | 0.3 | 2.8 | 0.36 | 0.22 |
| Phy-B | 10.1 | 11.4 | 11.3 | 12.5 | 8.2 | 7.0 | 8.2 | 6.7 | 7.3 | 4.6 | 4.5 | 3.5 | 2.0 | 1.6 | 0.00* | 1.00 |
| Phy-C | 12.4 | 11.9 | 11.5 | 13.7 | 6.3 | 6.8 | 8.9 | 5.9 | 4.5 | 5.2 | 4.6 | 3.0 | 2.7 | 1.9 | 0.09 | 0.74 |
| Bac | 11.2 | 11.7 | 13.7 | 16.1 | 4.6 | 7.4 | 8.4 | 5.1 | 4.5 | 5.1 | 3.6 | 2.4 | 2.7 | 1.9 | 0.35 | 1.42 |
| Zoo | 14.5 | 11.9 | 13.1 | 14.9 | 5.4 | 5.3 | 7.4 | 5.9 | 5.6 | 4.0 | 3.1 | 4.0 | 2.3 | 2.2 | 0.00* | 0.38 |
| Saan-T | 16.0 | 9.3 | 12.3 | 8.8 | 8.2 | 7.6 | 8.0 | 7.3 | 3.6 | 5.3 | 4.7 | 3.6 | 2.9 | 1.8 | 0.16 | 0.40 |
| Dab-T | 13.7 | 11.2 | 11.1 | 9.8 | 8.3 | 7.8 | 7.5 | 7.6 | 5.5 | 5.3 | 4.4 | 3.6 | 1.9 | 2.0 | 0.43 | 0.03 |
| Saan-S1 | 19.5 | 13.3 | 11.0 | 9.7 | 9.8 | 6.4 | 5.8 | 7.0 | 3.4 | 4.6 | 3.7 | 2.5 | 0.7 | 1.4 | 0.53 | 0.60 |
| Dab-S1 | 20.3 | 12.4 | 10.9 | 8.8 | 8.4 | 6.6 | 5.6 | 6.8 | 4.2 | 4.2 | 3.4 | 2.4 | 0.8 | 1.6 | 3.15 | 0.35 |
| Saan-S2 | 25.4 | 9.3 | 11.5 | 6.2 | 10.4 | 6.2 | 5.8 | 6.4 | 3.1 | 3.9 | 3.7 | 2.6 | 1.7 | 1.5 | 1.14 | 1.15 |
| Dab-S2 | 24.2 | 10.9 | 10.9 | 7.7 | 8.9 | 6.2 | 5.2 | 6.4 | 3.5 | 4.0 | 3.0 | 1.9 | 1.8 | 1.7 | 3.28 | 0.30 |
| Tur-red | 15.4 | 14.1 | 11.0 | 8.5 | 3.9 | 8.6 | 6.5 | 4.5 | 7.6 | 4.5 | 3.8 | 1.5 | 1.0 | 0.8 | 4.26 | 3.95 |
| Tur-ox | 13.9 | 15.5 | 8.5 | 8.2 | 4.2 | 5.6 | 2.6 | 3.9 | 5.8 | 1.6 | 1.4 | 0.6 | 0.6 | 0.4 | 12.47 | 14.73 |

* Values below the detection limit.

hydroxylic (serine + threonine), aromatic (tyrosine + phenylalanine), basic (arginine + histidine), and sulfuric (methionine) amino acids account for 24, 15, <7, <7, and <3 mol%, respectively. This lumping of amino acids in terms of functional groups generally masked the differences between the sediments compared to the trends observed in the individual amino acids. Only neutral amino acids decreased when going from SK and GB-2 (~51 mol%) to BG-B and BF (~46 mol%). The other charge classes often contain amino acids with little or contrasting individual concentration

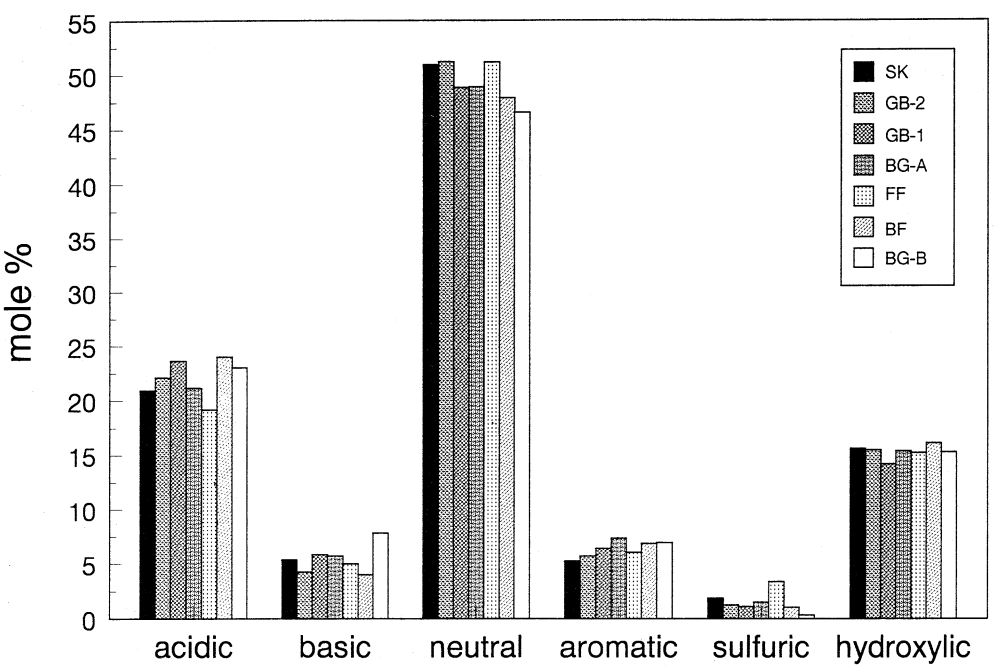


Fig. 6. Functional groups of protein amino acids. Basic: His (histidine), Arg (arginine). Acidic: Asp (aspartic acid), Glu (glutamic acid). Hydroxylic: Ser (serine), Thr (threonine). Neutral: Gly (glycine), Ala (alanine), Val (valine), Ile (isoleucine), Leu (leucine). Sulfuric: Met (methionine). Aromatic: Tyr (tyrosine), Phe (phenylalanine).

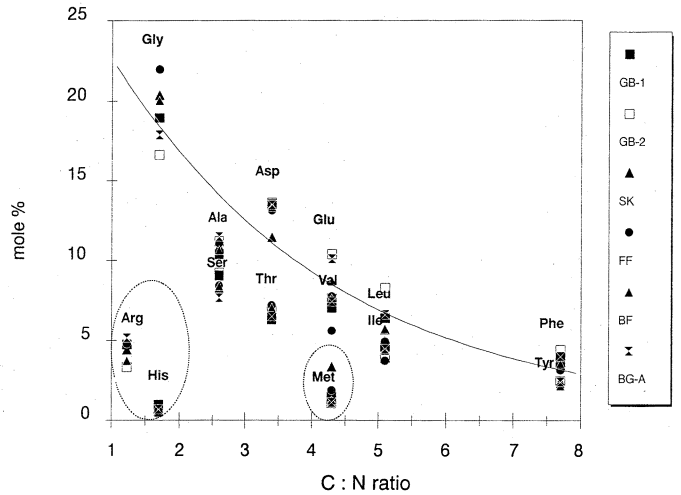


Fig. 7. C:N ratio of the individual amino acids vs. their relative mole percent contribution to THAA in all investigated sediments. The regression characteristics of the solid line are mol% = 22.63 $\times e^{-0.263 \times C:N\ ratio}$ ($r^2$ = 0.78).

patterns, resulting in small and irregular variation among the sediments.

The C:N ratio of individual amino acids correlates with the relative contribution of these compounds to the total protein pool in the sediment organic matter (Fig. 7). Short-chain, nitrogen-rich amino acids, such as glycine, occur in higher concentrations in sediment material than do nitrogen-poor amino acids such as the aromatic phenylalanine and tyrosine. The basic amino acids arginine and histidine, as well as the sulfur-containing methionine, are present in lower concentrations than would be expected by the general increase of mole percent with decreasing C:N ratio.

Table 3. Mole percent composition of essential amino acids (except lysine and tryptophane) in a composite marine organism (crab, abalone, euphausiid, tunicate, coral, sea urchin) as given by Phillips (1984).

| | Mean | Std | % deviation |
|---|---|---|---|
| Thr | 5.15 | 0.46 | 8.9 |
| Val | 5.35 | 0.72 | 13.4 |
| Met | 2.50 | 0.39 | 15.6 |
| Ileu | 4.55 | 0.56 | 12.3 |
| Leu | 7.32 | 0.91 | 12.4 |
| Phe | 5.10 | 1.44 | 28.2 |
| His | 2.38 | 0.24 | 10.1 |
| Arg | 6.97 | 1.64 | 23.6 |

The mole percent contributions of essential amino acid in the North Sea sediments were compared to a "composite" marine organism tissue (Table 3) to investigate whether some of these essential compounds were deficient and consequently limiting in deposit-feeder diets (Fig. 8). The sulfur-containing methionine and the basic histidine and arginine seemed to be the most deficient essential amino acids in sedimentary organic matter relative to heterotroph organism tissue, with depletions $\leq -90\%$ for methionine, $\leq -80\%$ for histidine, and $\leq -50\%$ for arginine.

*Nonprotein amino acids*: Nonprotein amino acids occurred in minor concentrations in the sediments, with $\beta$-alanine (up to 2.5 mol%) being more abundant than $\gamma$-aminobutyric acid (<1 mol%). These molecular compounds

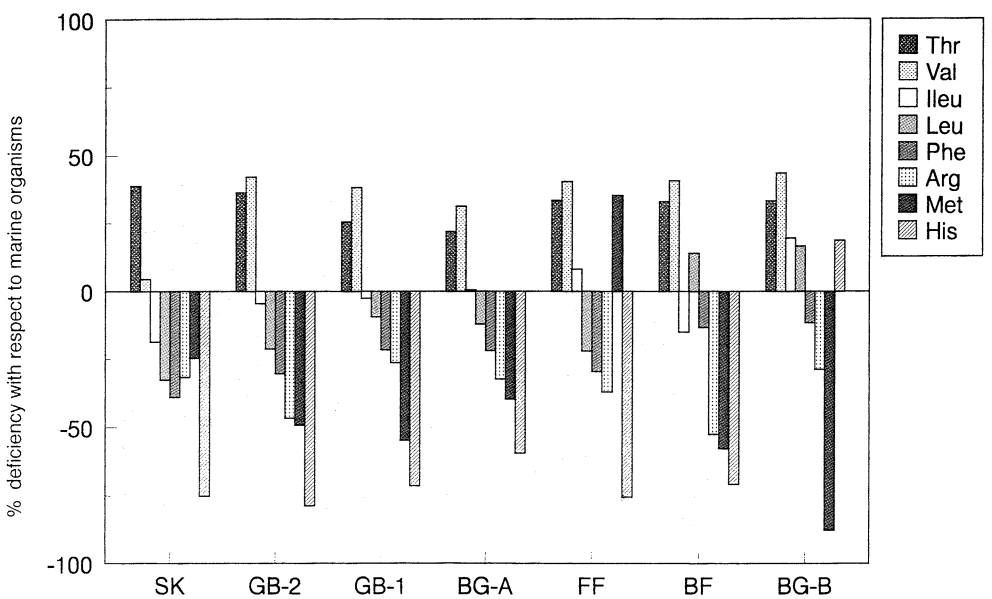


Fig. 8. Relative deficiencies or excesses of essential amino acids (essAA) in all investigated sedimentary organic matter in comparison to tissue essAA profile of a "composite" marine invertebrate (Table 3) (Phillips 1984). Percent deficiency was calculated as $[(C_{sed} - C_{ani})/C_{ani}]100$, where $C_{sed}$ is the percentage of the particular essAA in the sediment as percent of total amino acids and $C_{ani}$ is the same for animal tissue (Fong and Mann 1980).

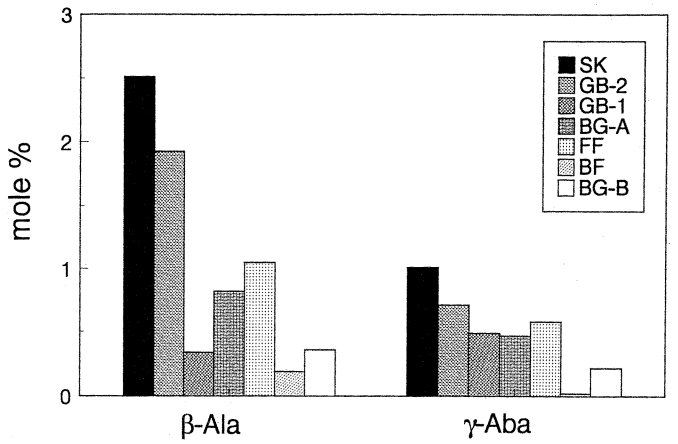


Fig. 9. Mole percent contributions of the nonprotein amino acids γ-aminobutyric acid and β-alanine.

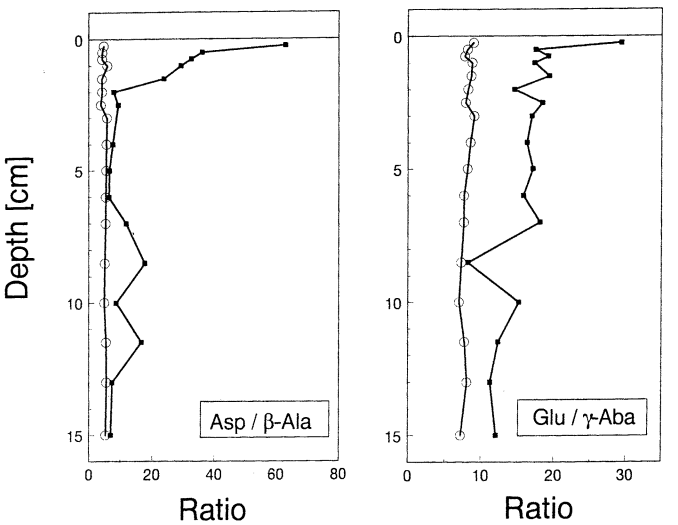


Fig. 10. Profiles within the sediment of the ratios of the protein amino acids glutamic acid and aspartic acid to their nonprotein derivates γ-aminobutyric acid and β-alanine, respectively. ■, station GB; ○ station SK .

increased in relative abundance when going from BF and BG-B to SK (Fig. 9). Moreover, Glu : γ-Aba and Asp : β-Ala ratios decreased when going from station BF and BG-B to GB-2 and SK (Table 1), indicating that degradation products accumulated along this array of stations. An exceptionally high Glu : γ-Aba ratio was found at BF, since only traces of degradation products were detected. Decreasing Glu : γ-Aba and Asp : β-Ala ratios were also found with depth in sediment at station GB, but not at the other stations, which are rather homogeneous with depth (Fig. 10).

*Hexosamines*: Total hexosamines reached concentrations up to 1.2 mg $g^{-1}$ dry sediment (Table 1), with their contribution to the total amino acid pool decreasing from the Skagerrak (glucosamine, 7.7 mol%; galactosamine, 3.3 mol%) to sta. BG-B (glucosamine and galactosamine <0.1 mol%) (Fig. 11).

## Discussion

Molar concentrations of amino acids usually appear to be rather constant with depth in the bioturbated zone, but may change over large depth scales (in terms of meters) and, hence, longer time scales (Henrichs and Farrington 1987; Cowie and Hedges 1992). These vertical changes have been attributed to differences in the reactivity of the individual amino acids (e.g. Haugen and Lichtentaler 1991; Cowie et al. 1992) depending on their nitrogen content, chain length, and functional groups. Our results support these observations, since amino acids are distributed rather homogeneously with depth in the bioturbated zones (Figs. 2, 10), but there are systematic differences of molar concentrations of THAA between North Sea stations on a horizontal scale (Fig. 5).

To test whether the intersite trends observed in mole percent contribution of the amino acids can be used to order the stations in terms of organic matter quality, a principal component analysis (PCA) was carried out. The North Sea sediments were analyzed together with literature data, including end-members such as labile organic matter present in source organisms (bacteria, phytoplankton, zooplankton), sediment trap material, and more degraded organic matter present in coastal sediments and refractory deep-sea deposits (Fig. 12). Hexosamines were not integrated into the PCA analysis because there is no comprehensive literature dataset available.

PCA is a variance-oriented method in which standardized variables are used to represent each site in a multidimensional space, the orthogonal axes of which are expressed in standard deviations of the original variables. Our basic variables (mole percent of the different amino acids) are standardized by subtracting the mean and dividing by their standard deviation. Means and standard deviations of the variables in our dataset are given in Table 4. The derived principal components or orthogonal axes are linear combinations of the original ones, but have the property that the maximum variance is found along the first axis, the maximum of the remainder variance along the second axis, and so on. In our analysis we only use the first axis and estimate the position of a station on this axis (i.e. the site score),

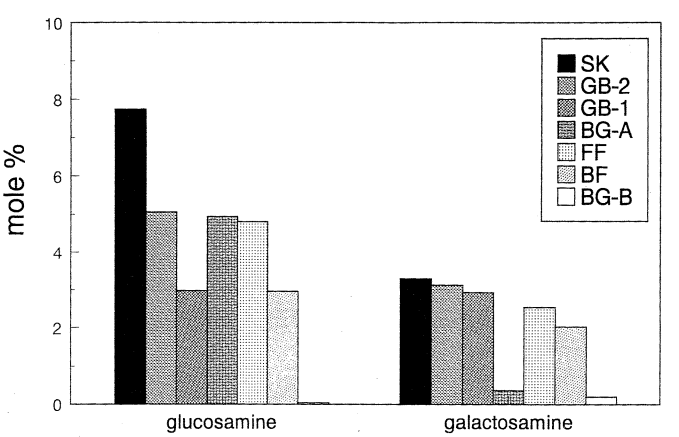


Fig. 11. Relative contributions of galactosamine and glucosamine to the sum of THAA and hexosamine.

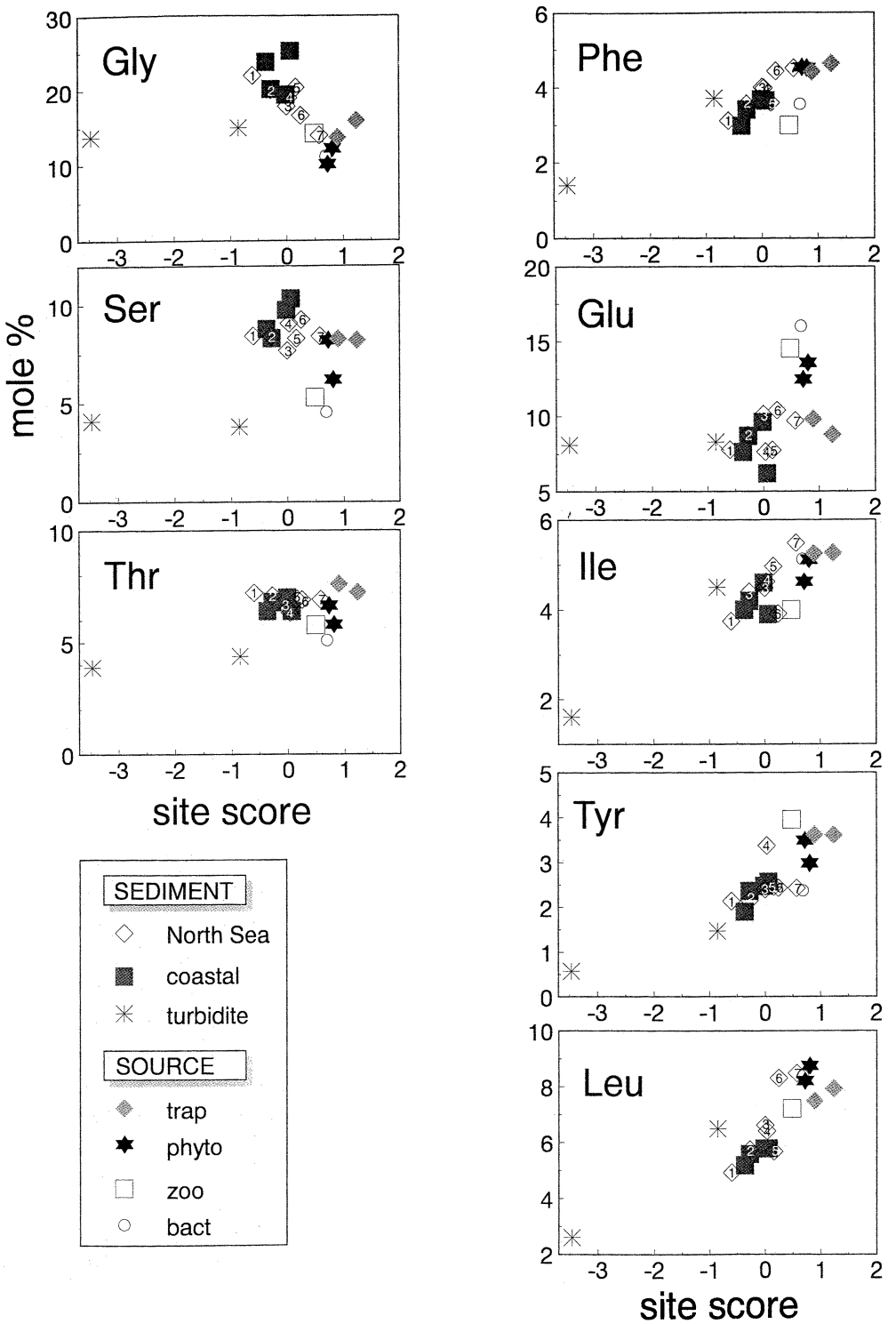


Fig. 12. PCA site scores on the first component vs. the mole percentages of individual protein amino acids in different substrates and North Sea sediments. Data labels of the North Sea sites: 1, SK; 2, GB-2; 3, GB-1; 4, BG-A; 5, FF; 6, BF; 7, BG-A. One of the datasets for phytoplankton was derived from Brown (1991). Data for fresh phytoplankton, bacteria, and zooplankton were derived from Cowie and Hedges (1992), data of sediment and 30-m trap material from Dabob Bay were extracted from Cowie and Hedges (1992, 1993), and Saanich Inlet data were derived from Cowie et al. (1992) and Cowie and Hedges (1993). Data of turbidites were extracted from tables in De Lange et al. (1994) and Cowie et al. (1995). The characteristics of the sample stations Dabob Bay and Saanich Inlet are described by Hedges et al. (1988) and Cowie et al. (1992), respectively.

Table 4. Parameters of the PCA based on the comprehensive analysis (A) and on the North Sea stations (B) (for coding, *see* Table 2).

A. Comprehensive analysis*

| Site score | | Factor coefficient | | Avg | Std |
|---|---|---|---|---|---|
| Tur-ox | −3.467 | β-Ala | −0.136 | 1.74 | 2.81 |
| Tur-red | −0.856 | γ-Aba | −0.125 | 1.50 | 3.22 |
| SK | −0.603 | Asp | −0.093 | 12.36 | 1.55 |
| Dab-S2 | −0.367 | Arg | −0.028 | 4.73 | 1.22 |
| GB-2 | −0.278 | Gly | −0.026 | 17.17 | 4.17 |
| Dab-S1 | −0.277 | Ser | 0.053 | 7.69 | 1.84 |
| Saan-S1 | −0.018 | Val | 0.054 | 6.95 | 0.85 |
| GB-1 | −0.003 | Glu | 0.058 | 9.84 | 2.57 |
| BG-A | 0.029 | Met | 0.071 | 1.66 | 0.82 |
| Saan-S2 | 0.062 | His | 0.084 | 1.35 | 0.66 |
| FF | 0.155 | Thr | 0.088 | 6.39 | 0.93 |
| BF | 0.247 | Ala | 0.100 | 11.20 | 1.05 |
| Zoo | 0.481 | Tyr | 0.118 | 2.583 | 0.798 |
| BG-B | 0.568 | Leu | 0.122 | 6.632 | 1.548 |
| Bac | 0.682 | Phe | 0.122 | 3.742 | 0.752 |
| Phy-B | 0.718 | Ile | 0.126 | 4.419 | 0.836 |
| Phy-C | 0.805 | | | | |
| Dab-T | 0.894 | | | | |
| Saan-T | 1.235 | | | | |

B. North Sea stations†

| Site score | | Factor coefficient | |
|---|---|---|---|
| SK | −1.561 | β-Ala | −0.122 |
| FF | −0.689 | γ-Aba | −0.127 |
| GB-2 | −0.502 | ASP | 0.058 |
| BG-A | 0.208 | ARG | −0.004 |
| GB-1 | 0.276 | GLY | −0.134 |
| BF | 0.873 | SER | 0.031 |
| BG-B | 1.397 | VAL | 0.095 |
| | | GLU | 0.099 |
| | | MET | −0.094 |
| | | HIS | 0.094 |
| | | THR | −0.060 |
| | | ALA | −0.036 |
| | | TYR | 0.043 |
| | | LEU | 0.133 |
| | | PHE | 0.137 |
| | | ILE | 0.070 |

*Forty-four percent of variation explained.
†Forty-five percent of variation explained.

which provides the best projection of its position in multidimensional space onto the axis. Hence, the site scores quantify the relative positions of the stations, and they are calculated from the values of the original variables with the aid of the factor coefficients according to the formula Site score $= \Sigma_i(\text{var}_i - \text{avg var}_i/\text{std var}_i)\text{fac.coeff}_i$, where $\text{var}_i$ is the original (nonstandardized) mole percent of amino acid *i*, avg $\text{var}_i$ and std $\text{var}_i$ are its mean and standard deviation in our dataset, and $\text{fac.coeff}_i$ is the factor coefficient for amino acid *i* (Table 4). The part of the formula within parentheses performs the standardization. This equation allows for inclusion of data external to the dataset used here, and other sediments can be compared with our sediments by calculating their scores on the first axis described here.

The first component of the comprehensive analysis of North Sea and literature sites explains 44% of the variance (Table 4A). The first axis in a PCA based on protein and nonprotein amino acids of only the North Sea stations explains 45% of the variance (Table 4B). These two PCA analyses basically reveal the same sequence for the North Sea stations, with the exception that FF and GB-1 were swapped in position. This indicates that amino acid composition provides a robust, yet powerful, measure to reveal differences between sedimentary organic matter having a variable quality and degradation history. In the following discussion the amino acid-based station scores from the comprehensive analysis of North Sea and literature data are used to order the organic matter (Table 4A).

The highest scores on the first axis were found in the sandy sediments of the Southern Bight (BG-B and BF), with station scores of 0.568 and 0.247, respectively, resembling those of the labile source materials phytoplankton, bacteria, and trap material (>0.6). The lowest score in the North Sea (−0.603) was found in the deep Skagerrak, where organic matter finally becomes buried. The refractory organic matter in the deep sea turbidites has even lower scores, namely −0.856 for the initial and −3.467 for the oxidized turbidite. Also, the deeper layers of the deposition area GB-2 score relatively low (−0.278) compared to the surface layer (−0.003).

The comprehensive PCA analysis reveals that the quality range of organic matter in North Sea sediments is relatively wide, varying between sourcelike material and material resembling deep-sea deposits. Scores on the first axis of the PCA can therefore be interpreted as an index of organic matter degradation. This degradation state index is supported by independent quality parameters such as the ratio of precursors to nonprotein amino acids and hexosamine concentration (*see below*). The quality of organic matter in North Sea sediments appears to be SK < GB-2 < GB-1 ≈ BG-A ≈ FF < BF < BG-B.

The degradation state of the organic matter at the intermediate sites is less conclusive than those of the end-members. First, the intermediate stations (FF and GB-1) swapped position in the PCA based only on the North Sea sites. Second, parameters such as ratio of precursors to nonprotein amino acids (Fig. 10) and glucosamine content (Fig. 11) indicate that GB-1 contains mainly labile organic matter, whereas the comprehensive PCA suggest that it resembles the deeper sediment (GB-2) at this site.

These differences in degradation state are not only the result of in situ degradation, but also of the degradation history before deposition. Degradation in situ occurs at all stations but is difficult to trace because bioturbation has resulted in extensive mixing of the sediments, with a result that compositional changes with depth are nearly absent except for the German Bight. At this station there is clear evidence of in situ degradation (Figs. 2, 10).

In marine sediments >95% of the organic matter is sorbed to the sediment surface, and there is a rather constant relationship between organic carbon content, sediment surface area, and grain size (Mayer 1994; Keil et al. 1998). The organic matter on the surface is probably in some kind of

dynamic equilibrium with the local environment (Keil et al. 1997).

In the North Sea, particles are sorted according to hydrodynamic conditions. The more sandy material is deposited in the southern North Sea, whereas the silty material is transported by the residual currents to the final depositional area in the Skagerrak (Fig. 1) (e.g. Anton et al. 1993; van Weering et al. 1993; Lohse et al. 1995). During this transport from the southern North Sea to the Skagerrak these silty particles are involved in a number of deposition–resuspension cycles, and there is extensive time for degradation. This interplay between grain-size sorting due to hydrodynamic conditions and organic matter surface area (grain-size) relationships determines the predepositional degradation history.

Skagerrak sediments are silty and are consequently relatively rich in organic matter but of low average quality because they have been degraded extensively before deposition. Sediments in the southern North Sea (BF, BG-B) are sandy and hence relatively poor in organic matter, but the organic matter is rather fresh because little decomposition has occurred before deposition.

This interplay between organic matter grain-size relationships and predepositional history related to grain size may account for the degradation history of bulk organic matter, but local inputs may modify this picture. The organic carbon-rich surface layer at station GB is clearly related to the recent deposition of labile organic matter. Chl *a* concentrations and benthic biomass data indicate that there is perhaps more labile organic matter at stations FF, GB, BG-A, and SK than at the sandy stations BG-B and BF (Table 1). At each site there is a distribution of organic matter quality classes (Middelburg 1989), and Boudreau and Ruddick (1991) have shown that the overall lability of organic matter is dominated by the preponderance of refractory organic matter. Similarly, our degradation index based on amino acid composition provides a measure for the bulk organic matter quality and is not very sensitive to minute amounts of very labile organic matter that are reflected in Chl *a* and benthic biomass data.

*Protein amino acids*—The overall amino acid spectra in North Sea sediments with high abundances of glycine, followed by aspartic acid, alanine, and glutamic acid (Fig. 5), resemble those found in coastal sediments dominated by planktonic inputs, e.g. the Gulf of Maine (Mayer et al. 1988), Oslo Fjord (Haugen and Lichtentaler 1991), Saanich Inlet (Cowie et al. 1992), and Potomac estuary (Sigleo and Shultz 1993). However, it is also clear that the neutral leucine and isoleucine, the aromatic phenylalanine and tyrosine, and the acidic glutamic acid increase in mole percent contribution with increasing lability of the organic material. The neutral amino acid glycine and the hydroxylic serine and threonine tend to decrease with increasing lability (Figs. 5, 12).

Some authors divided the amino acids into functional groupings (e.g. Sigleo et al. 1983; Sigleo and Shultz 1993) and found a decrease for acidic amino acids and an increase in basic and neutral amino acids during degradation over large time scales in continental slope sediments (Steinberg et al. 1987) and in anoxic fjord sediments (Brown et al. 1972). In North Sea sediments there is a systematic trend within the neutral group, but there is little variation within the acidic (19–23 mol% acidic amino acids) or the basic groups (5–8 mol% basic amino acids). There are two problems regarding biochemical groupings. First, they may obscure the behavior of individual amino acids with contrasting accumulation patterns, such as glutamic and aspartic acids, and consequently give an inconsistent picture. Second, abundant amino acids, such as glycine, determine the overall trend of the neutral group, whereas less abundant amino acids are of little influence.

Variation of individual amino acid mole percentages depends on their association with cell wall and cytoplasma, their embedding into structural matrices, such as silicate tests of diatoms or calcite tests of coccolithophorids (Cowie and Hedges 1996), and sorption to mineral surfaces (Henrichs and Sugai 1993) that may physically limit microbial degradation (Mayer 1994), rather than on individual differences in lability based on functional groups or molecule size.

Cowie et al. (1992) calculated relative reactivities of individual amino acids by their ratios of average annual fluxes to corresponding accumulation rates in the surface sediment in the 50-m-deep Saanich Inlet. They found intracellular amino acids (glycine and serine) in diatoms having higher relative reactivities than cell wall-associated amino acids (tyrosine and methionine), suggesting selective preservation of cell wall protein relative to intracellular protein. This concept has been supported by kinetic studies that revealed amino acids associated with these structural cell wall elements to be less susceptible to enzymatic degradation than those in cell plasma (Laursen et al. 1996). This has also been found for intracellular sugars that are digested by zooplankton with higher efficiencies than diatom cell wall sugars (Cowie and Hedges 1996). Glycine and the hydroxyl amino acids serine and threonine are enriched in diatom cell walls (Hecky et al. 1973) and are preferentially accumulated in diatomaceous material during sinking and decomposition in the water column (e.g. Siezen and Mague 1978; Lee and Cronin 1984; Müller et al. 1986). In North Sea sediments glycine increases from 14 mol% in the most labile organic matter compared to 22 mol% in the most refractory organic matter (Figs. 5, 12). The indicative value of glycine for the refractory nature of organic matter has been pointed out earlier for particulate material in the Potomac estuary by Sigleo and Shultz (1993) and for sediments underlying a sewer outlet by Compiano and Romano (1988). Moreover, glycine often increases with sediment depth (Haugen and Lichtentaler 1991; Cowie et al. 1992). Sediment trap data also suggest an accumulation of glycine as degradation progresses (Lee and Cronin 1984). This relatively conservative behavior of glycine may not only be related to its presence in the structural matrices of diatoms and bacteria, but possibly also to its comparatively minor food value to micro- and macroconsumers because of its short chain length and because it can be synthesized from many other amino acids in heterotrophic metabolism.

Glutamic acid and the aromatic species tyrosine and phenylalanine are generally concentrated in cell plasma (Hecky et al. 1973) and show strong depletion with increasing state of decomposition over the entire range, as well as in the more restricted range in the North Sea (Fig. 12). The mole percent of leucine, isoleucine, and phenylalanine correlate

well with the degradation index, suggesting that these components are good indicators for the lability of organic matter. Glutamic acid and tyrosine exhibit less linear trends in the North Sea system, increasing only ~25%, and therefore are less valuable indicators. Strongly decreasing vertical profiles of glutamic acid in sediment cores with increasing depth (Haugen and Lichtentaler 1991) and depletion relative to phytoplankton (e.g. Cowie and Hedges 1992) also indicate that it is easily degraded. Moreover, sediment trap data show that tyrosine, phenylalanine, and glutamic acid are the most labile species (Cowie and Hedges 1992; Cowie et al. 1992).

The essential amino acids arginine, histidine, and methionine do not follow the general trend of increasing mole percent concentration with increasing nitrogen content of amino acids in the bioturbated North Sea sediments (Fig. 7). Moreover, these very same amino acids are also the most deficient in sediment organic matter relative to marine organism tissue (Fig. 8), which means that they have to be preferentially taken up in order to fulfill the animal nutritional needs and therefore may be limiting to animal growth (Phillips 1984). Because the deficiencies generally increase from the more labile organic matter at BF and BG-A to the refractory organic matter at SK (Fig. 8), we propose that sediment-ingesting animals may be more nitrogen limited in sediment containing highly degraded organic matter. Phillips (1984) also found deficiencies for arginine and methionine in particulate organic matter, although to a smaller extent ($<-15\%$) than found in the North Sea sediments, indicating that the deficiency trends may be inherited from the source and become even stronger in the sediments. However, it is difficult to clearly demonstrate the effects of essential amino acid deficiencies in marine invertebrates, since gut microbes could also partially supply some of the essential amino acids lacking in food (Fong and Mann 1980). Moreover, the amino acid distribution in the bio-available fraction of the sedimentary may vary from the total hydrolyzable fraction measured in this study (Mayer et al. 1995), since animals use an array of enzymes to solubilize amino acids from food rather than strong acids.

*Nonprotein amino acids*—Nonprotein amino acids are generally much less reactive than protein amino acids (Cowie et al. 1992) and are not incorporated into biota, resulting in net accumulation during mineralization. Increasing mole percentages of nonprotein amino acids points to bacterial breakdown of particulate organic matter, with values >10 mol% indicating substantially degraded material, which are typically encountered in deep-sea sediments (Wakeham et al. 1993; de Lange et al. 1994). In abyssal environments the sum of $\beta$-alanine and $\gamma$-aminobutyric acid can exceed that of all protein amino acids, with values up to 70% in highly degraded pelagic clays (Whelan 1977).

The sum of $\beta$-alanine plus $\gamma$-aminobutyric acid in the North Sea sediments (0.2–3.5 mol%) fits well into the range generally encountered in coastal sediments and source organisms (Cowie and Hedges 1994) . Concentrations in the most labile North Sea sediments BG-B and BF (Fig. 9) are as low as found in the deepest trap of the Scotia Sea (Müller et al. 1986), with values up to 0.36 mol% $\beta$-alanine and 0.19 mol% $\gamma$-aminobutyric acid. This is consistent with the high quality score derived from the amino acid spectra and also with the high bacterial turnover rates measured at BF (Table 1). Nonprotein amino acids are most abundant in the most refractory North Sea sediments (SK). The mole percent range of $\beta$-alanine and $\gamma$-aminobutyric acid increases systematically in the station sequence based on the amino acid spectra (Fig. 9).

The diagenetic origin of nonprotein amino acid has been demonstrated by their reverse downcore behavior in absolute amounts with respect to the parent amino acids (e.g. Cowie and Hedges 1994). Consequently, the ratios of the precursors (aspartic acid, glutamic acid) to their decompositional products ($\beta$-alanine and $\gamma$-aminobutyric acid) have been widely used to verify variations in the intensity of organic matter decomposition in the water column (Lee and Cronin 1982; Ittekkot et al. 1984*a*; Haake et al. 1993) and to a smaller extent in sediment profiles (Whelan 1977; Cowie and Hedges 1994). Our results show that Asp : $\beta$-Ala and Glu : $\gamma$-Aba ratios correlate strongly with the degradation state of organic matter based on protein amino acids (Fig. 13A,B). The most refractory North Sea sediment (SK) reaches ratios nearly as low as the turbidites and anoxic Dabob Bay sediments ($\pm 5$ for Asp : $\beta$-Ala and $\pm 8$ for Glu : $\gamma$-Aba). However, the most labile organic matter in BG-B has ratios $\pm 37$ for Asp : $\beta$-Ala and $\pm 44$ for Glu : $\gamma$-Aba, which are comparable with labile source compounds as bacteria, phytoplankton, zooplankton, and suspended matter. The oxidized turbidites do not fit into the nearly linear relation displayed by the other sediments due to a 3–5 times elevation of their nonprotein amino acid mole concentrations (11–12 mol% $\beta$-alanine and 12–15 mol% $\gamma$-aminobutyric acid) compared to the coastal sediments and the reduced turbidites. This increase in nonprotein amino acids compared to reducing turbidites has been attributed to in situ oxidation and subsequent association with noncarbonate phases (like aluminosilicates) (Cowie et al. 1995).

Another problem with the inclusion of turbidite and deep-sea samples into the comparison with coastal stations is the difference in input source and adsorptive properties. In contrast to the siliceous diatom-dominated temperate coastal sites, open-ocean areas may be dominated by input from calcareous organisms such as coccolithophorids (e.g. Ittekkot et al. 1984*b*). Carbonate-rich sediments may change the whole amino acid spectrum because of the enhanced adsorption capacities for acidic groups (Carter and Mitterer 1978; Ittekkot et al. 1984*a*; Wakeham et al. 1993; de Lange et al. 1994; Cowie et al. 1995). Consequently, calcareous deposits are generally characterized by a strong predominance of aspartic acid (up to 25 mol%) and a high Asp : Gly ratio (up to 2), whereas coastal sediments with diatomaceous organic matter sources are characterized by a predominance of glycine (up to 23 mol%) and a low Asp : Gly ratio (0.6–0.8) (Ittekkot et al. 1984*b*; Müller et al. 1986; Wakeham et al. 1993). This may explain why the carbonate-rich turbidite sediments do not fit in the linear relationship exhibited by the remaining noncarbonate stations for glycine, serine, and threonine concentrations (Fig. 12). These results make it clear that amino acid spectra, and especially the parameters directly related to mole percent of the acidic amino acids such as the Asp : $\beta$-Ala and Glu : $\gamma$-Aba ratios, can be mis-

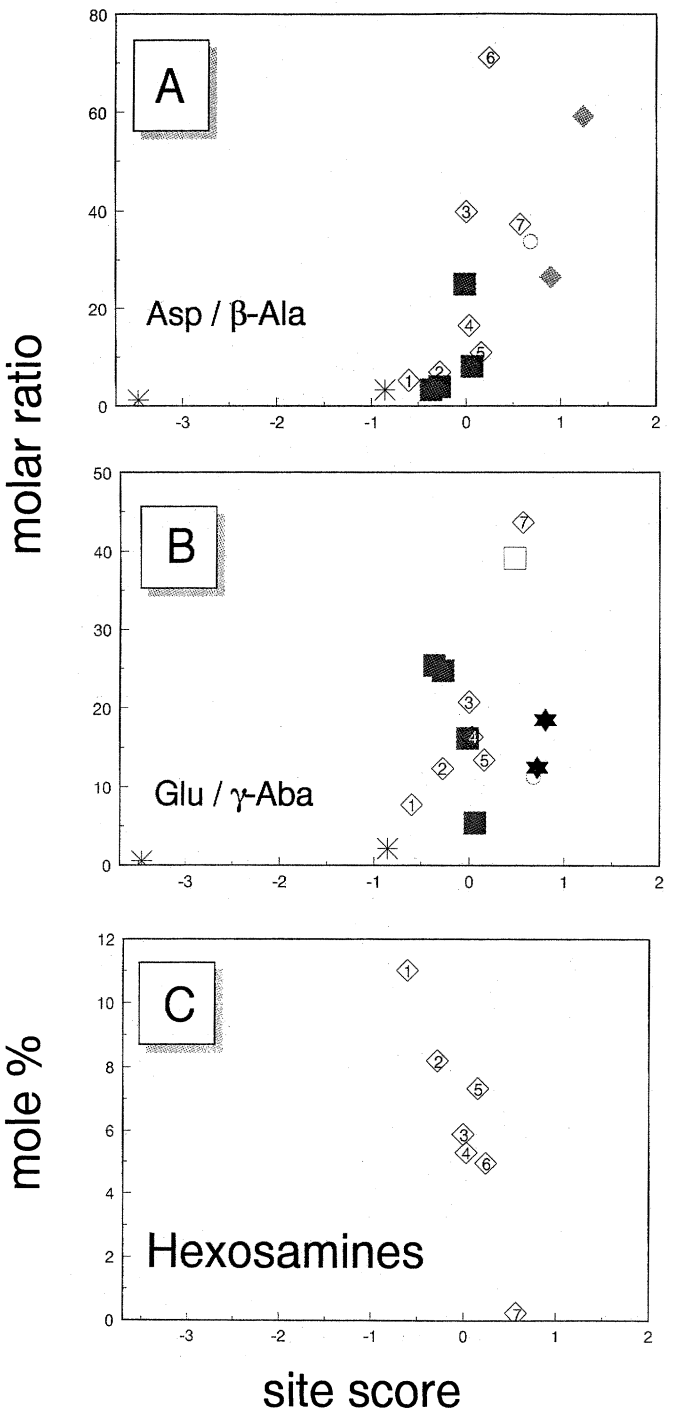


Fig. 13. (A, B) PCA site scores on the first component vs. molar ratios of precursors/degradation products and (C) total hexosamines at the North Sea stations vs. sites score (=degradation index). Plankton samples and station BF data were not included in panels A and B since extremely low concentrations of nonprotein amino acids resulted in exceptional high ratios (Table 1).

leading indices for the quality of organic matter in comparison of sites with obviously different sources of organic matter.

With regard to vertical sediment profiles, a uniform source is much more certain. The downcore decrease in the Asp: β-Ala ratio in the German Bight (GB) from 63 in the upper layer to 12.9 in the deeper layers (Fig. 10) confirms the conclusion made above that the surface layer consists of freshly deposited material not yet mixed significantly with refractory material by bioturbation. This German Bight station is the most eutrophic of all investigated North Sea stations (Hickel et al. 1993), and a high concentration of organic matter coincides with a high reactivity (Fig. 3).

In summary, it appears that Asp: β-Ala and Glu: γ-Aba ratios are widely applicable, even though these ratios are potentially ambiguous due to combined effects of cumulative error, compositional variability of source organisms, and the fact that the degradation of aspartic and glutamic acid does not always result in the production of β-alanine and γ-aminobutyric acid (Cowie and Hedges 1994).

*Hexosamines*—The hexosamines glucosamine and galactosamine can be used to trace the origin of the organic material as well as its degradation state (e.g. Ittekkot et al. 1984b; Liebezeit 1993). Although in marine sediments hexosamines are present in minor quantities compared to amino acids, they contribute considerably to sedimentary nitrogen in the more degraded North Sea sediments (Fig. 4), which makes them valuable as food source for heterotroph organisms as well. Owing to their general incorporation into structural biopolymer matrices, such as bacterial cell walls and chitinous material (e.g. Ternay 1976), hexosamines are relatively resistant to decomposition and can therefore be used as an independent parameter to verify the order of the stations proposed by the amino acid spectra. Both hexosamines are well correlated with the amino acid-based degradation index and become more concentrated in refractory sediments (Figs. 11, 13C). This is consistent with an increase of hexosamines relative to THAA during decomposition in the water column (Ittekkot et al. 1984*b*; Müller et al. 1986; Haake et al. 1992). Hexosamine concentrations in the more labile North Sea sediments (<1 mol%) are consistent with sedimenting (Haake et al. 1993) and suspended matter (Müller et al. 1986) and recent sediments (Mayer et al. 1985). Moreover, the high THAA: hexosamine ratios (Table 1) resemble those measured in sediment trap samples in the deep Arabian Sea of 8–22 (Haake et al. 1992). Extremely high values up to >80 have been reported for phytoplankton since hexosamines are present only in trace amounts. The ratio we found for most refractory organic carbon in SK sediments in this study is higher than the lowest value reported for the deepest trap (3,800 m) in the Arabian Sea (~3–5), but still indicates that SK sediments contain the most degraded organic matter in the North Sea.

*Major biochemical groups*—The order based on the protein amino acid spectra is consistent with the results of more established independent parameters, such as ratio of precursors to nonprotein amino acids and hexosamine concentrations (Fig. 13), and also the general expectations based on the physical setting of the investigated stations. Amino acid composition therefore gives a good indication of organic matter degradation state. The contributions of total carbohydrate and THAA to TOC have been used as a measure of

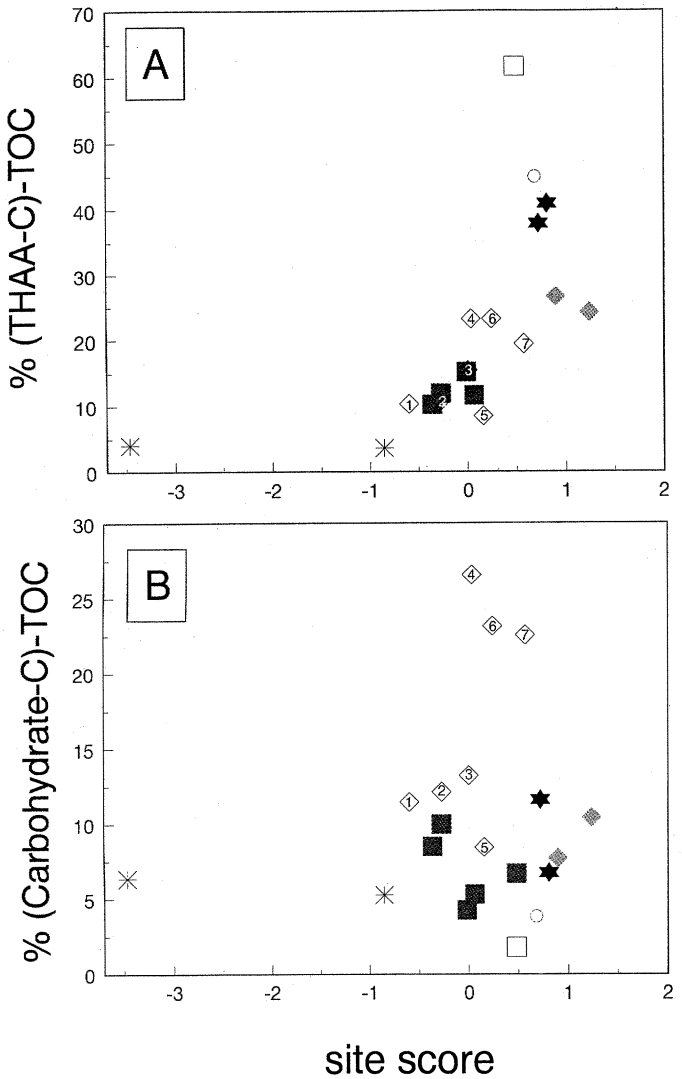


Fig. 14. PCA site scores on the first component vs. the contributions of THAA (A) and carbohydrates (B) to total organic carbon. THAA data were extracted from the literature cited in Fig. 12. The contribution of total amino acids to TOC was calculated on basis of the individual spectra. Carbohydrate data were derived from Brown (1991), Cowie et al. (1992, 1995), and Cowie and Hedges (1984, 1993). A conversion factor of 0.4 from total aldose yield to TCHO to mg carbon was used.

organic matter quality (Ittekkot 1988; Cowie and Hedges 1994). The relative contribution of THAA to TOC confirms the array of the sites based on the PCA (Fig. 14A), with the less degraded material having the highest THAA : TOC ratio. However, the contribution of carbohydrate to TOC reveals a rather scattered picture (Fig. 14B), indicating either a nonselective behavior during decomposition because members of this group are simultaneously part of structural elements, humic substances, and cell storage products and therefore display variable reactivity patterns (de Leeuw and Largeau 1993), or that differences in analytical methods and windows (GC and sulfuric acid method) may mask any trend. Accordingly, the decrease of the sum of these compounds with increasing degradation found by Cowie and Hedges (1994) are mainly determined by a selective mineralization of proteinaceous material rather than by carbohydrates. Previous studies already demonstrated that the proteinaceous fraction of the organic matter generally is more labile than the carbohydrate pool (Cowie et al. 1992; Colombo et al. 1996). Cowie and Hedges (1994) also reported a decrease of organic carbon concentration with increasing degradation state. Our data of the North Sea confirm this trend (Fig. 3), with the exception of the high concentration at the surface of station GB-1 that is due to recent deposition.

In conclusion, the integrated PCA analysis shows that the measurement of amino acid composition gives valuable and consistent information about the degradation state in North Sea sediments and also on a broader scale if the source of the organic matter is the same.

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