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Interrelationships Between Microbiological and Chemical Parameters of Sandy Beach Sediments, a Summer Aspect†

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At 12 stations located in sandy beach sediments of the brackish water Kiel Fjord and Kiel Bight (Baltic Sea, FRG), variations and interrelationships of microbiological, chemical, and physicochemical parameters were monitored. Depending upon location, wide variations of a number of parameters reflecting dissolved organic and inorganic nutrients, chlorophyll *a*, microbial number, and uptake activity of glucose were measured. Whereas most of the parameters generally showed the tendency to decrease from the inner to the outer Kiel Fjord, individual parameters (oxygen, particulate nitrogen, ribose, chlorophyll *a*, glucose/fructose ratio) increased with increasing distance from the inner Kiel Fjord. Similarities in the local variation pattern demonstrated various relationships between individual parameters. Among those, dissolved organic nutrients on the one hand and inorganic nutrients on the other hand were closely linked together. Variations of organic and inorganic nutrients corresponded to variations of microbial activity and physicochemical parameters. By comparing standing stock carbon with carbon production, a microbial biomass turnover time of about 100 h was calculated. Approximately 50% of the microphytobenthos primary production was fixed by microorganisms. Daily microbial carbon production (43 mg of C per m^2) was in the range of meiofauna carbon (35 mg of C per m^2).

The cycle of matter in sediments is only poorly understood, a discrepancy arising from the sparsity of information concerning microbial activity and variations in organic and inorganic nutrients. In most of the microbiological studies of sediments (2, 6, 13; L. W. Wood, Ph.D. thesis, North Carolina State University, Raleigh, 1970), only very few parameters could be included, thus restricting the understanding of the various interrelationships between microbiological and chemical parameters.

The study presented is part of a comprehensive joint research program carried out at different seasons of the year at 12 stations located in sandy beach sediments of the brackish water Kiel Fjord and Kiel Bight (Baltic Sea, FRG). General characteristics of the research area have been described by Rheinheimer and co-workers (14). The aim of this study is to obtain more information on interrelationships and variations of microbiological, chemical, and physicochemical parameters. The data presented were used to estimate microbial carbon production, which is compared to microphytobenthos primary production and meiofauna carbon.

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MATERIALS AND METHODS

Sampling was carried out at 12 stations of sandy wave-washed beaches of the brackish water Kiel Fjord and Kiel Bight during the period from 4 to 13 July 1977 (see Fig. 1). Stations A to F are located at the west side; the corresponding stations G to M are located at the east side of the Kiel Fjord and the Kiel Bight, respectively. Stations A/G are located in the inner part, B/H in the center part, and C/J in the outer part of the Kiel Fjord. Stations D/K, E/L, and F/M are located on the Kiel Bight. Water samples overlying the sediments from the same stations have already been analyzed during a previous autoradiography study (11).

Undisturbed sediment samples were collected by pushing plastic syringes (1.6 cm in diameter, 2-cm^2 surface area; top cut off) into the surface of sandy sediments (0 to 0.7 cm depth; approximately 1.9 g of dry-weight sediment), which at those locations was subjected to brief periods of exposure to air and brackish water, respectively. At this part of the Western Baltic Sea, the influence of the tide is negligible. For the analysis of organic and inorganic nutrients, interstitial water was squeezed out of the sediment cores by applying low vacuum (0.2 kPa per cm^2), using a filtration apparatus equipped with cellulose membranes (Schleicher & Schüll Co.; medium pore size, 4.4 μ; boiled in double-distilled water before use). Unless otherwise stated, duplicate samples were analyzed.

The techniques applied to determine the individual microbiological and chemical parameters followed procedures published in previous reports. Salinity, tem-

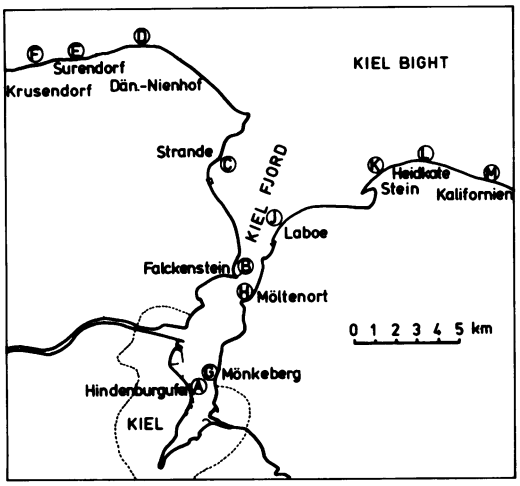

FIG. 1. *Map of sampling stations located at the Kiel Fjord and the Kiel Bight.*

perature, oxygen, and pH were recorded in situ with portable probes (Yellow Springs Instruments Co., Inc.). Dissolved inorganic matter (phosphate, ammonia, nitrite, nitrate), as well as dissolved organic carbon (DOC) and particulate organic carbon (POC) and nitrogen (PON), were measured by procedures cited by Grasshoff (3). Determinations of DOC were carried out after filtration of interstitial water through 0.2-μ cellulose ester membranes. POC and PON were determined in portions of the homogenized sediment cores (4). Generally, this determination composes both the particulate and dissolved fraction; however, the overwhelming portions of carbon are present in the form of particulate material (Table 1). Because of this, the fraction determined after homogenization of the sediment cores is considered to be mainly POC, which reflects, on an average, 99% of the total organic carbon in the sediments. For determination of chlorophyll *a*, we followed procedures recommended by UNESCO (16). For the analysis of natural free dissolved monosaccharides (glucose, fructose, ribose, total monosaccharides) and physicochemical properties of the sediment (water content, organic matter content, sand grain size, and shape), compare a previous report (13) and literature cited therein.

The number of saprophytic bacteria was determined by the spread plate technique after homogenization (4) and subsequent dilution of the sediment. The culture medium was a yeast-extract peptone agar (ZoBell 2216 E, modified) prepared with filter-sterilized natural seawater overlying the sediment at the individual stations. The same water was used for preparing dilutions. The plates were incubated at in situ temperature for 14 days. Total number and biomass of microorganisms were analyzed by epifluorescence microscopy (13) from the homogenized and diluted sediment samples. Biomass determinations were based on a size fractionation of the individual cells (9).

For the determination of microbial uptake of glucose in undisturbed sediment cores incubated close to in situ conditions, we followed procedures published

TABLE 1. *Range and mean of the parameters examined at 12 stations of sandy beach sediments of the Kiel Fjord and the Kiel Bight*

Parameter	Range	Mean
Physicochemical parameters		
Salinity (‰)	10.7–14.5	13.4
Temp (°C)	18.5–26.0	21.0
pH	7.5–8.0	7.8
Dissolved inorganic matter		
Phosphate (μg at P liter^{-1})	1.2–4.5	2.5
Ammonia (μg at N liter^{-1})	1.4–6.6	3.2
Nitrite (μg at N liter^{-1})	0.2–1.7	0.5
Nitrate (μg at N liter^{-1})	0–6.4	1.6
Oxygen (ppm)	7.2–8.9	8.2
Particulate organic matter		
Carbon (mg g^{-1})	0.43–2.40	1.41
Nitrogen (mg g^{-1})	0.03–0.43	0.12
Carbon/nitrogen ratio	1.21–95.9	28.2
DOC		
Glucose (μg liter^{-1})	21.6–317.6	106.4
Fructose (μg liter^{-1})	3.9–1,144	153.0
Glucose/fructose ratio	0.3–9.7	2.1
Ribose (μg liter^{-1})	2.1–53.9	19.8
Total monosaccharides (μg liter^{-1})	42.1–1,483	284.7
DOC (mg liter^{-1})	6.0–23.7	13.5
Chlorophyll *a* (μg g^{-1})	0.10–2.94	1.08
Sediment properties		
Water content (%)	10.8–22.5	17.6
Organic matter content (mg g^{-1})	3.3–9.6	5.9
Grain size (mm)	0.208–0.489	0.293
Grain shape (% grains with rounded edges)	62–86	76
Microbial no.		
Saprophytic bacteria (10^6 g^{-1})	4.51–34.90	16.82
Pigmented saprophytic bacteria (%)	3.9–50	25.6
Total microbial no. (10^8 g^{-1})	5.65–20.20	10.26
Total microbial biomass (10^{-3} mg C g^{-1})	11.04–36.71	20.44
Microbial activity		
Uptake rate of [^{14}C]-glucose (10^{-3} μg g^{-1} h^{-1})		
Net uptake	1.84–11.91	7.65
Respiration	0.19–1.04	0.66
Gross uptake	2.02–12.61	8.35
Actual uptake rate of glucose (10^{-1} μg g^{-1} h^{-1})	0.72–4.11	2.54
Turnover time of glucose (h)	0.28–1.69	0.53

in previous reports (10, 13). Briefly, undisturbed sediment cores were incubated with 1 ml of filter-sterilized pore water spiked with 10 μl of D-[*U*-^{14}C]glucose (New England Nuclear Corp.; specific activity, >150 mCi/mmol; final concentration, 0.0029 μg of [^{14}C]glucose per ml of pore water). Compared to the natural concentration, [^{14}C]glucose was present in tracer concentrations. Triplicate samples and a blank (fixed with 10 μl of concentrated Formalin per ml of pore water) were run at one substrate concentration for different incubation times (1, 2, 3, 5, and 7 min) at a temperature within 1 to 2°C of the in situ temperature. Respiration and incorporation of [^{14}C]glucose (the latter defined as retention of label within cells, determined after oxidation of the sediment cores) were determined as previously described (10, 13). Radioactivity was measured in a liquid scintillation counter (Betazint 5000; Berthold and Frieseke). Counts were corrected for apparatus background, blank activity, counting and oxidation efficiency. The radioactivity that was abiotically fixed by the sediment (blank activity) normally was very low (maximally 100 cpm per g of dry-weight sediment). Fixation of $^{14}CO_2$ by the minerals in the sediment or the sediment itself was negligible.

Respiration and net uptake rates (micrograms of [^{14}C]glucose per gram of dry-weight sediment per hour) were calculated by linear regression from the slope of the curves obtained by plotting incubation time versus the amount of substrate respired and incorporated, respectively (10). Gross uptake rates were determined from the slope of the curves calculated for the sum of the amount of [^{14}C]glucose respired and taken up. Actual uptake rates (flux; micrograms of glucose per gram of dry-weight sediment per hour) follow from multiplying gross uptake rates by the quotient of the total amount of glucose present in the sample (^{12}C and ^{14}C) and the amount of [^{14}C]-glucose added. Turnover times of glucose (T) were calculated by the equation $T = S \times v^{-1}$ (S = substrate concentration; v = actual uptake rate).

Conversions from dry weight (in grams) to wet volume (in milliliters) were carried out on the basis of the corresponding dry-weight content of the sediment (average value, 82%). This enabled us to do the extrapolation from wet volume (in milliliters) to surface area (in square meters) of sediment (down to a depth of 1 cm).

RESULTS AND DISCUSSION

The range and the mean for each of the 31 parameters monitored are shown in Table 1. The hydrographical conditions were relatively constant during the sampling period (4 to 13 July 1977). With the exception of two stations (C and G; Fig. 1), only slight variations of temperature (between 19 and 22°C) and salinity (between 13 and 14‰) were observed. The weather was sunny with light winds from north to west directions.

Despite the relatively constant hydrographical conditions, wide variations in most of the chemical and microbiological parameters were observed depending upon location. Dissolved inorganic matter in the pore water varied by a factor of 4 (phosphate) and 9 (nitrite), respectively; particulate organic matter varied by a factor of 6 (carbon) and 17 (nitrogen), respectively. Whereas variations in the concentration of DOC were relatively small (factor of 4), great variations were measured for individual components of DOC (e.g., total free dissolved monosaccharides varied by a factor of 35). Distinct variations were also noticed in the concentrations of chlorophyll *a* (factor of 29) and the number of saprophytic bacteria and uptake activity of glucose. However, variations in total number of microorganisms and biomass were relatively small (factor of 3).

Inorganic nutrients, DOC, and microbial biomass generally showed the tendency to decrease from the inner to the outer part of the Kiel Fjord. For other parameters, a distinct trend could be demonstrated only at those stations located at the east side of the Kiel Fjord and the Kiel Bight, respectively (stations G to M; Fig. 1). Organic matter content, POC, carbon/nitrogen ratio, glucose, fructose, total monosaccharides, saprophytic bacteria, and respiration rate of glucose decreased with increasing distance from the inner part to the outer part of the Kiel Fjord. In contrast, the concentrations of oxygen, particulate nitrogen, ribose, chlorophyll *a*, and the glucose/fructose ratio increased. For the stations located at the west side of the Kiel Fjord and the Kiel Bight, respectively, a corresponding trend could be demonstrated only for concentrations of fructose and mean grain size, which increased with increasing distance from the inner part to the outer part of the Kiel Fjord (Fig. 2, 3).

Stations C and G differed from the other stations by extremely high or low values of individual parameters. Station C (west side of the outer Kiel Fjord) represented the situation of a calm protected bay characterized by high water temperature and high concentrations of inorganic and organic nutrients (organic matter content, POC, free dissolved monosaccharides). The number of saprophytic bacteria and actual uptake rate of glucose reflected high microbial activity. However, oxygen content, POC, carbon/nitrogen ratio, and concentration of chlorophyll *a* were extremely low. At station G (east side of the Kiel Fjord, strongly influenced by land and shipping) extremely low values of salinity, oxygen content, concentrations of dissolved inorganic matter, PON, and chlorophyll *a* were recorded. However, concentrations of DOC and free dissolved monosaccharides were relatively high. Low microbial activity was reflected by the low total number of cells and the high turnover time of glucose (Fig. 2, 3).

By comparing visually the local variations, similarities in the variation pattern of a number of parameters are obvious. Among the physicochemical parameters of the interstitial water, salinity and temperature reveal indirect relationships (Fig. 2). Phosphate, ammonia, nitrite, and nitrate show similar variations, indicating that the dissolved inorganic nutrients are closely linked together and governed by biological and chemical processes (Fig. 2; see variations of ammonia and nitrite). Among the parameters of DOC, local variations of glucose, fructose, ribose, and total free dissolved monosaccharides are similar (Fig. 3; see variations of glucose and

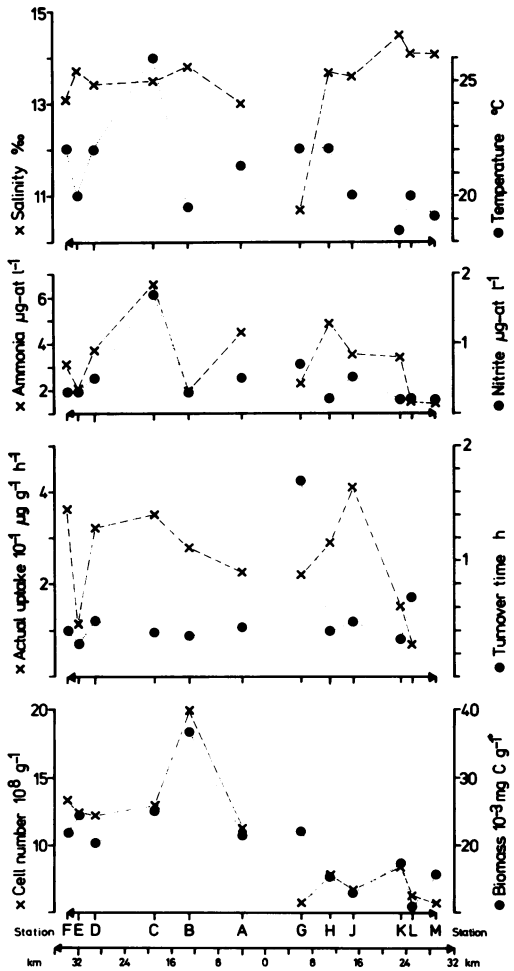

FIG. 2. *Local variations of microbiological parameters (total number of microorganisms and biomass; actual uptake rate and turnover time of glucose), dissolved inorganic matter (ammonia and nitrite), and physicochemical parameters (salinity and temperature) monitored at 12 stations of sandy beach sediments at the Kiel Fjord and the Kiel Bight.*

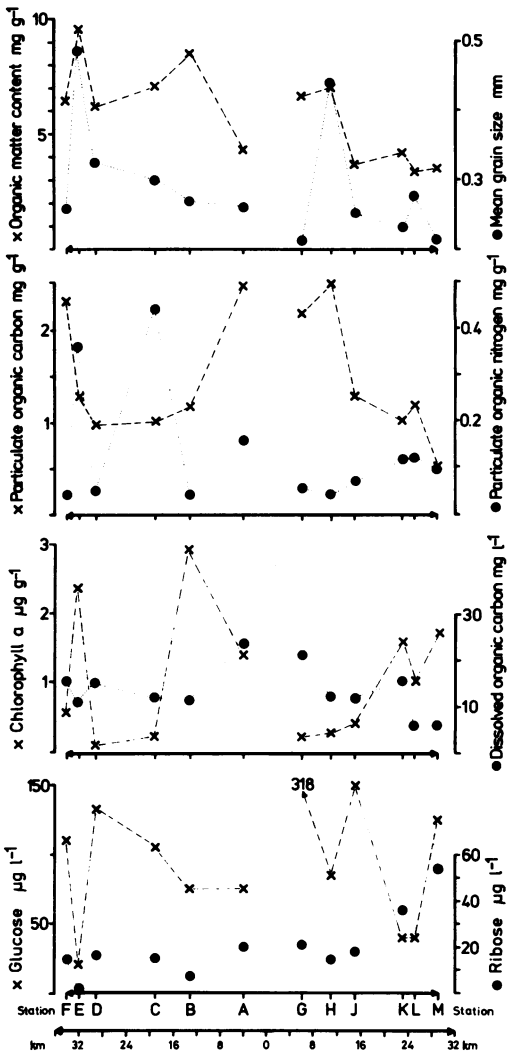

FIG. 3. *Local variations of glucose and ribose; chlorophyll a and dissolved organic carbon; POC and PON; organic matter content and mean grain size monitored at 12 stations of sandy beach sediments located at the Kiel Fjord and the Kiel Bight.*

ribose). This seems to indicate that liberation and consumption of monosaccharides follow mechanisms similar for different individual monosaccharides. The same was already suspected from a previous study (12). Solubility, adsorption, and exchange of dissolved organic and inorganic nutrients are governed by salinity, temperature, or both. This can be demonstrated by comparing the local variation pattern of salinity, temperature, or both on the one hand and, for example, glucose and nitrite, respectively, on the other hand (Fig. 2, 3). Between dissolved and

particulate organic matter, mutual exchange processes can be expected as indicated by the inverse variation pattern of glucose and organic matter content of the sediments (Fig. 3). Indirect relationships can also be demonstrated for variations in the concentration of chlorophyll *a* and both concentration and turnover time of glucose (Fig. 2, 3). These relationships documenting the close connection between primary and secondary production need further investigation. The close relationship between chlorophyll *a* and POC found by Steele and Baird (15) could not be confirmed in this study.

Relationships between microbiological and chemical parameters are of special interest. Variations in the total number of microbial cells correspond to organic nutrients (2) and microbial activity (L. W. Wood, Ph.D. thesis, North Carolina State University, Raleigh, 1970). High numbers of cells reflect high microbial biomass and organic matter content of the sediment. However, the higher the number of cells, the lower the concentration and the turnover time of glucose (Fig. 2, 3). Close indirect relationships between grain size and organic matter content and number of microorganisms, demonstrated by a number of authors (2, 5, 6, 17, 18; L. W. Wood, PhD. thesis, North Carolina State University, Raleigh, 1970) could not be documented from the data presented. This might be dependent on the narrow range of grain sizes studied and the limited number of samples. From the calculations of microbial uptake activity (see above), it is understandable that variations of both actual uptake rate and turnover time of glucose correspond to variations of glucose and total monosaccharides. Besides the relationships between microbial activity and dissolved and particulate organic matter (see above), activity relates to dissolved inorganic matter. Parallel to an increase in the actual uptake rates of glucose, the concentration of ammonia in the interstitial water increases (Fig. 2). Interrelationships between salinity, temperature, or both and microbial activity are not very pronounced. The physicochemical parameters obviously influence microbial activity indirectly by the various relationships existing between salinity, temperature, or both and parameters of DOC, which in turn relate to microbial activity (see above).

Interrelationships between saprophytic bacteria (colony-forming units) and other parameters are not very pronounced. Saprophytes do not correlate well with total number and biomass. The only pronounced relationship exists between saprophytes and respiration rate of glucose. Pigmented saprophytes (percentage of the total number of saprophytes) show interrelationships with the concentration of glucose and total monosaccharides and actual uptake rates of glucose. These relationships are difficult to interpret, and the number of samples may not be sufficient to draw any conclusions from these observations.

The data presented were used to calculate microbial biomass production (Fig. 4). The overwhelming portion of organic carbon in sediments is present in the form of POC (1.4 mg of carbon per g of dry-weight sediment). The average concentration of DOC is two orders of magnitude lower. Microbial carbon (2.0×10^{-2} mg of carbon per g of sediment) amounts to 1.5% of POC. Natural free dissolved monosaccharides account for less than 1% of DOC. The main components of the free dissolved monosaccharides are glucose and fructose which make up more than 90% of the total. Microbial uptake activity is in the order of 10^{-4} mg of carbon (glucose) per g per h. If we assume that microbial glucose uptake reflects one quarter of the total carbon uptake, the total microbial carbon uptake amounts to 4×10^{-4} mg of carbon per g per h. This assumption is derived from observations that approximately 2% of total DOC exists in the form of labile DOC (1% amino acids, 1% monosaccharides) from which glucose roughly represents one quarter (12). As an average value, 40% of the microbial uptake of labeled material is respired (7). Under the assumption that 10% of the net uptake is lost by excretion, microbial biomass production amounts to 2.2×10^{-4} mg of carbon per g per h, which is equivalent to 1.8 mg of carbon per m^2 per h (sediment depth, 0 to 1.0 cm; see above). From comparing microbial standing stock carbon with biomass production, a biomass turnover time or approximately 100 h was calculated. About 50% of the microphytobenthos primary production (8) is fixed by microbial secondary production. This corresponds to 50% of the phytoplankton production which is consumed by heterotrophic processes in the sea (1). Investigations of short-term variations of microbiological and chemical parameters in the water column have implied that despite seasonal and daily variations, microbial standing stock biomass and activity in one and the same body of water do not change drastically from one day to the other (12). If this holds true for the sediments investigated, most of the biomass production of microorganisms is available for organisms feeding on microorganisms. Meiofauna carbon amounts to 35 mg of carbon per m^2 (A. Faubel, personal communication), which is in the range of daily microbial biomass production (43 mg of carbon per m^2). This indicates that the microbial biomass production is high enough to provide

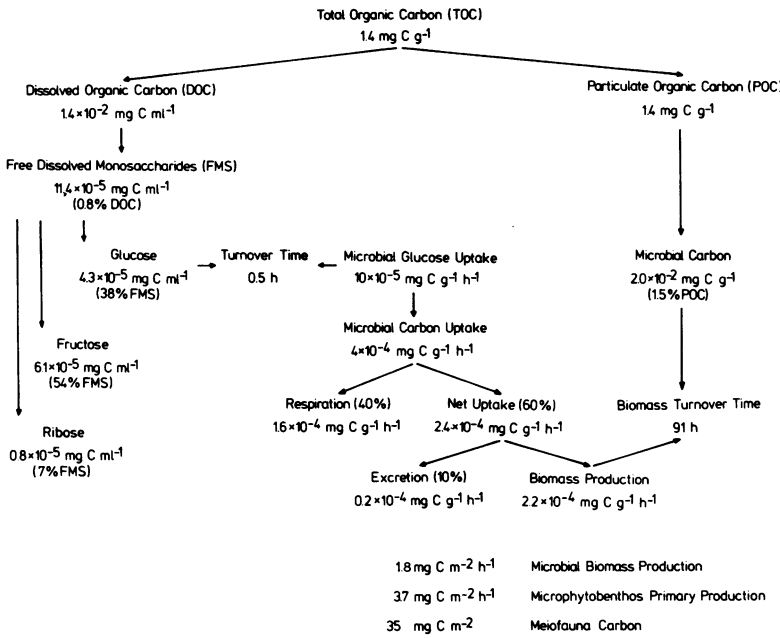

FIG. 4. *Diagram of carbon cycle as influenced by microbial activity in sandy beach sediments of the Kiel Fjord and the Kiel Bight.*

carbon for a daily turnover of the meiofauna carbon.

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