EXPERIMENTAL ARTICLES =

The Effect of Mineral Particulate Matter on the Productive Characteristics of Bacterioplankton and the Degradation of Labile Organic Material

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Abstract—The effect of mineral particulate matter on the population of bacterioplankton, its aggregation, and productive characteristics was studied in model experiments with different concentrations of particulate kaolin and the same concentration of organic substance (sodium humate). It was found that the presence of mineral particulate matter stimulated the aggregation of bacterioplankton, improved bacterial production, and extended the productive period of bacterioplankton. The integral specific production of aggregated bacterioplankton was higher than that of free-swimming bacterioplankton. The energy metabolic coefficient K_2 of bacterioplankton in the presence of mineral particulate matter was higher than in its absence.

Key words: bacterioplankton, bacterial production, mineral particulate matter, organic matter.

It is well known that organic particulate matter is essential in aqueous ecosystems. Pertinent information on the role of inert mineral particulate matter in these ecosystems is scarce [1–3]. The effect of solid interfaces on biological recycling was reviewed by Zvyagintsev [4]. Inert inorganic particles comprise the major portion of solid interfaces in bodies of water. Dissolved organic substances can adsorb on suspended mineral particles to form organomineral detritus, which promotes the transformation of biogenic materials in bodies of water [5–8].

The aim of this work was to evaluate the effect of mineral particulate matter on the functional characteristics of bacterioplankton and the dynamics of biochemical oxygen demand.

MATERIALS AND METHODS

Model experiments were carried out with Yenisey river water sampled in November, when the content of bacterio- and phytoplankton was minimal. The water was passed through paper filters to remove coarse mineral particles. One batch of the filtered water was used throughout the experiments. The water dispensed into sterile flasks was supplemented with humic acids (as an organic matter) and particulate kaolin, which is the major component of terrigenic mineral particulate matter occurring in bodies of water. The initial concentration of humic acids in all the experiments was the same (10 mg/l), whereas the concentration of kaolin particles in different experiments was different (5, 15, 45, or 90 mg/l). Particulate kaolin was suspended in distilled water, and the suspension was elutriated to give a mean size of kaolin particles of $1.64-1.86 \mu m$. The experiments were performed in 3-5 replicates and lasted 13 days.

Biochemical oxygen demand (BOD) was determined by the standard procedure [9]. Bacterial cells were enumerated by epifluorescence microscopy [10]. Daily bacterial production (P_t), specific bacterial production (P_t/B_t), and energy metabolic coefficient (K_2) were calculated by the formulas $P_t = B_t - B_{t-1}$; $P_t/B_{ts} =$ $\ln B_t - \ln B_{t-1}$; and $K_2 = P_t/(P_t + R)$, respectively, where $R = R_t - R_{t-1}$ is the average daily consumption of oxygen dissolved in water. The mean biomass was calculated by the formula $(B_t - B_{t-1})/(\ln B_t - \ln B_{t-1})$. The parameters B, P, and R were expressed in the same units [9]. The net results of production, respiratory, and mortal processes by the end of the *t*th day of incubation were calculated by the formulas $P = (B_t - B_0)/t$; $P/B = (\ln B_t - \ln B_0)/t$; $B = (B_t - B_0)/(\ln B_t - \ln B_0)$; $K_2 = P/(P + R)$, where $R = R_t/t$.

RESULTS AND DISCUSSION

The total number and biomass of bacteria during incubation changed by more than one order of magnitude reaching maximum values on the 3rd to 4th day of incubation in the experimental flasks with the riverine water supplemented with particulate kaolin, and on the 2nd day in the control flask with the riverine water not supplemented with particulate kaolin (Fig. 1). After 1 day of incubation, the masses of bacterioplankton in all the flasks were close. Throughout the rest of the incu-

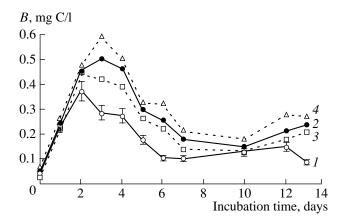


Fig. 1. The dynamics of the bacterioplankton biomass in the absence (I) and presence (2-4) of mineral particulate matter (kaolin). Curves 2, 3, 4 correspond to mean, minimum, and maximum biomass values in experimental flasks.

bation period, the mass of bacterioplankton in the control flask was lower than in the experimental flasks. On the 10th day of incubation, the masses of bacterioplankton in the control and experimental flasks became close, but then the biomass of bacterioplankton in the experimental flasks tended to increase, whereas the biomass in the control flask tended to decrease (Fig. 1).

On the 1st day of incubation, the average bacterial production in the control and experimental flasks was the same within the experimental error (Table 1). On the 2nd day, bacterial production in the control was higher than in the experimental flasks. On the 3rd day, the loss of bacterial cells in the control and in the experimental flask with a minimal content of particulate kaolin exceeded their production; the loss of bacterial cells in the experimental flask with 15 mg/l particulate kaolin was equal to their production; and the loss of bacterial cells in the experimental flasks containing 45 or 90 mg/l particulate kaolin was lower than their production.

Over the entire incubation period (13 days), the total bacterial production (the sum of the average daily productions) in the experimental flasks with 5, 15, 45, and 90 mg/l particulate kaolin exceeded the total production

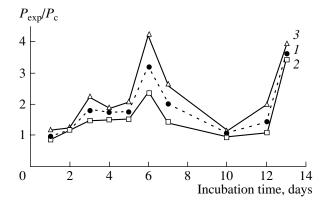


Fig. 2. The dynamics of the ratio of the current bacterial productions in the experimental and control flasks (P_{exp}/P_c) . Curves 1, 2,3 correspond to mean, minimum, and maximum ratio values in experimental flasks.

in the control flask by 1.4, 1.5, 1.6, and 1.7 times, respectively. By the end of the incubation period, the biomass of bacterioplankton in the experimental flasks exceeded that in the control flask on the average by 3.6 times. Consequently, the presence of mineral particulate matter in water augments the production of bacteria and allows them to occur longer in a productive state (or to survive unfavorable conditions, such as nutritional starvation).

The curves showing the current integral bacterial production, which is equal to the difference between the final and original biomasses in the flasks, were similar to the curves presented in Fig. 1. The ratio of the current bacterial productions in the experimental and control flasks (P_{exp}/P_c) were at maxima on the 6th and 13th days of incubation (Fig. 2). In addition, the experimental flasks with 45 and 90 mg/l particulate kaolin showed a statistically significant peak in this ratio on the 3rd day of incubation (Fig. 2). The dependence of bacterial production on the presence of particulate kaolin was most profound on the 3rd and 4th days of incubation, bacterial production in the flasks with 45 and 90 mg/l particulate kaolin of production in the flasks with 45 and 90 mg/l particulate kaolin of the 3rd and 4th days of incubation. Between the 5th and 12th days of incubation, bacterial production in the flasks with 45 and 90 mg/l particulate kaolin was slightly suppressed. On

Particulate kaolin, mg/l	Days of incubation										
	1	2	3	4	5	6	7	10	12	13	
0	0.20	0.14	-0.09	-0.01	-0.10	-0.08	0.00	0.01	0.01	-0.06	
5	0.17	0.23	-0.03	0.04	-0.19	-0.01	-0.12	0.01	0.06	-0.04	
15	0.18	0.22	0.00	0.05	-0.17	-0.01	-0.11	-0.03	0.02	0.05	
45	0.19	0.23	0.08	-0.05	-0.22	-0.07	-0.05	0.00	0.01	0.08	
90	0.23	0.19	0.14	-0.20	-0.07	-0.10	-0.04	-0.02	0.03	0.01	

Table 1. The average daily production of bacterioplankton (mg C/l day) in flasks with different concentrations of particulate kaolin

MICROBIOLOGY Vol. 73 No. 1 2004

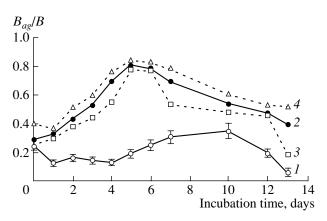


Fig. 3. The dynamics of the fraction of aggregated bacterioplankton in the total bacterial mass. Curve designation as in Fig. 1.

the 13th day of incubation, the ratio P_{exp}/P_c was maximum for all the kaolin concentrations.

Throughout the incubation period, the aggregation of bacterial cells in the control flask was lower than in the experimental flasks (Fig. 3). In the control flask, the number of aggregated cells increased between the 4th and 10th days of incubation, but then tended to decrease reaching almost zero on the 13th day. In the experimental flasks, the degree of bacterial aggregation or adsorption on mineral particles was maximum (about 80%) between the 5th and 6th day of incubation (Fig. 3).

It is known that aggregated bacterial cells are functionally more active [11, 12]. The proportions between the current productivities of aggregated bacteria and the total production of bacterioplankton are shown in Table 2. In the control flask, the production of aggregated bacterioplankton was notable only between the 7th and 10th day of incubation. In the experimental flasks, the relative production of aggregated bacteria reached a maximum between the 5th and 6th day of incubation and then tended to decrease.

The specific bacterial production P/B, which is equal to the growth rate constant K [13], of aggregated

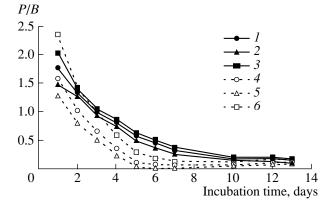


Fig. 4. The dynamics of the average daily production of (1-3) aggregated and (4-6) free-swimming bacterioplankton. Curves 1, 2, 3 and 4, 5, 6 correspond to mean, minimum, and maximum production values in experimental flasks.

and unaggregated bacterioplankton was comparable only on the 1st day of incubation (Fig. 4). Then the average daily specific production of aggregated bacterioplankton was higher than that of unaggregated bacterioplankton.

The kinetics of the biochemical oxygen demand of model media during their incubation did not follow an exponential curve either in the control or in the experimental flasks (Fig. 5), which is in agreement with the data of other researchers [14, 15]. In the absence of oxygen consumption by other organisms, the rate of oxygen consumption calculated from BOD corresponded to the daily respiration of bacterioplankton. The consumption of oxygen in the flasks had three stages. At the first stage (three days of incubation), oxygen consumption in all the flasks was intermediate between autocatalytic and exponential. The second stage ended on the 7th day for the experimental flask with 90 mg/l particulate kaolin, while on the 6th day for all the other flasks. The third stage was not so distinct as the first two stages, especially in the control flask and in the flasks with 5 and 15 mg/l particulate kaolin. At the same time, the third stage was distinct when oxygen consumption was calculated per one cell (Fig. 6). In this

Particulate kaolin, mg/l	Days of incubation									
	1	2	3	4	5	6	7	10	12	13
0	0.10	0.15	0.13	0.11	0.17	0.24	0.33	0.38	0.17	0.00
5	0.31	0.48	0.65	0.76	0.98	0.96	0.69	0.59	0.49	0.16
15	0.40	0.39	0.53	0.78	0.92	0.85	0.83	0.58	0.61	0.52
45	0.36	0.55	0.62	0.83	1.04*	1.09*	1.04*	0.75	0.55	0.58
90	0.32	0.38	0.45	0.57	0.81	0.83	0.85	0.66	0.46	0.39
Average value	0.34	0.45	0.55	0.74	0.93	0.92	0.85	0.65	0.52	0.42

Table 2. The fraction of the current integral production of aggregated bacterioplankton in the total bacterial production (P_{ag}/P)

* Negative production of free-swimming bacteria on the 5th through 7th days of incubation.

case, the third stage in the flask with 90 mg/l particulate kaolin ended one day later than in the control flask and the flask with 45 mg/l particulate kaolin.

The presence of mineral particulate matter also influenced the energy metabolic coefficient K_2 , which was calculated from the integral bacterial production and oxygen requirements. As can be seen from Fig. 7, the energy metabolic coefficients for the control and experimental flasks were close only on the 1st and 10th days of incubation. On the other days, the energy metabolic coefficient for the experimental flasks was higher than for the control flask, suggesting that mineral particulate matter slowed down the process of bacterioplankton aging.

The energy metabolic coefficients calculated for each day of incubation are presented in Table 3. On the 1st day of incubation, bacterioplankton in the control flask spent more energy for constructive metabolism than bacterioplankton in the experimental flasks. On the 2nd day, the situation was quite the reverse. On the 3rd day, bacterioplankton in the flask with 90 mg/l particulate kaolin dissipated in vain only 30% of the energy, whereas cell loss in the control flask exceeded cell production.

As a results of intense cell lysis occurring on the 3rd and 4th days of incubation, the medium became enriched in readily metabolizable organic matter, which could be easily utilized by intact bacterial cells. For this reason, after the period of intense cell lysis, the energy metabolic coefficient reached values close to 1.0. In the presence of mineral particulate matter, intense cell lysis and hence the subsequent cell growth began later than in the control. In the presence of 45 and especially 90 mg/l particulate kaolin, bacterioplankton had an opportunity of utilizing part of the organic matter that is not available to bacterioplankton incubated without or with mineral particulate matter at a low concentration. This may account for the greater bacterial production in the flasks with a high content of mineral particulate matter.

Thus, the model experiments showed that the number of aggregated bacterial cells in the absence of mineral particulate matter was smaller than in its presence. The degree of bacterial aggregation or adsorption on mineral particles was maximum (about 80%) between the 5th and 6th day of incubation. The presence of mineral particulate matter augmented the production and extended the productive (or survival) period of bacterioplankton under starvation conditions. By the end of the incubation period, the biomass of bacterioplankton in the experimental flasks with mineral particulate matter exceeded that in the control flask on the average by 3.6 times. The integral specific production of aggregated bacterioplankton was higher than that of freeswimming bacterioplankton (except for the 1st day of incubation). The energy metabolic coefficient of bacterioplankton in the presence of mineral particulate matter was higher than in its absence throughout the incubation period.

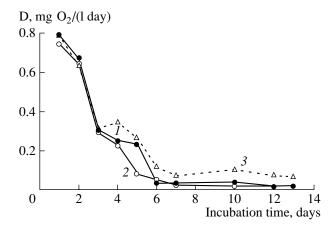


Fig. 5. The dynamics of the respiration of bacterioplankton in the absence (1) and presence of particulate kaolin at concentrations of (2) 5 and 15 mg/l and (3) 45 and 90 mg/l.

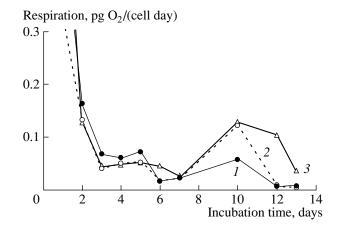


Fig. 6. The dynamics of the respiration of bacterioplankton as calculated per one cell in the absence (1) and presence of particulate kaolin at concentrations of (2) 45 and (3) 90 mg/l.

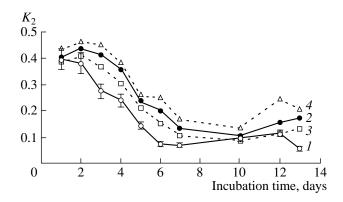


Fig. 7. The dynamics of the integral energy metabolic coefficient K_2 . Curve designation as in Fig. 1.

MICROBIOLOGY Vol. 73 No. 1 2004

Particulate kaolin, mg/l	Days of incubation										
	1	2	3	4	5	6	7	10	12	13	
0	0.54	0.24	_	_	_	_	0.49	0.49	0.83	_	
5	0.39	0.49	-	0.31	_	_	_	0.71	0.94	-	
15	0.39	0.38	0.33	0.26	_	_	_	_	0.92	0.94	
45	0.44	0.43	0.57	_	_	_	_	0.18	0.56	0.96	
90	0.39	0.39	0.70	_	_	_	_	_	0.44	0.20	

Table 3. Average daily values of the energy metabolic coefficient K_2

Note: The "-" signs correspond to negative values of daily bacterial production.

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