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INGESTION OF MICROPLASTICS BY THE HETEROTROPHIC DINOFLAGELLATE *OXYRRHIS MARINA*

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Incorporation of microplastics (MP) into the microbial food web and its further transport to higher trophic levels have been hitherto poorly studied. In this work, the patterns of MP ingestion by the unicellular heterotrophic dinoflagellate *Oxyrrhis marina* (OXY) were analyzed. The prymnesiophycean *Isochrysis galbana* (ISO), 5.6- μm polystyrene microspheres (MS), and their mixture (ISO-MS) were used as food objects for *O. marina*. Dynamics of the abundance of microorganisms and microspheres was investigated using a flow cytometer. As shown, the heterotroph *O. marina* ingested MP even in the presence of its natural prey (microalgae), and feeding on MP did not result in a decrease in the dinoflagellate abundance. The grazing rates of “preys” in the OXY-ISO-MS mixture were $(0.21 \pm 0.01) \text{ MS} \cdot \text{cell}^{-1} \cdot \text{h}^{-1}$ (\pm standard deviation) and $(0.38 \pm 0.01) \text{ ISO} \cdot \text{cell}^{-1} \cdot \text{h}^{-1}$. These rates were significantly lower than in the mono-diet experiments – with OXY-ISO [$(1.93 \pm 0.68) \text{ ISO} \cdot \text{cell}^{-1} \cdot \text{h}^{-1}$] and OXY-MS [$(0.45 \pm 0.04) \text{ MS} \cdot \text{cell}^{-1} \cdot \text{h}^{-1}$]. Thus, the expansion of the range of food objects led to a decrease in the grazing rate. In the mono-diet experiments, the clearance rates were (0.12 ± 0.04) and $(0.19 \pm 0.06) \mu\text{L} \cdot \text{cell}^{-1} \cdot \text{h}^{-1}$ for OXY-ISO and OXY-MS, respectively; thereby, *O. marina* spent less time on capturing ISO cells than on capturing MS. The same pattern was observed in the experiments with the OXY-ISO-MS mixture: the clearance rate for microalgae [$(0.17 \pm 0.02) \mu\text{L} \cdot \text{cell}^{-1} \cdot \text{h}^{-1}$] was slightly lower than that for MS [$(0.19 \pm 0.003) \mu\text{L} \cdot \text{cell}^{-1} \cdot \text{h}^{-1}$]. Since *O. marina* re-consumed MS even in the presence of its natural food object (*I. galbana*), no trophic adaptation of the dinoflagellate to MS occurred. No selective grazing of *O. marina* for any “prey” was revealed, either ISO or MS. The obtained results indicate the possibility (and high probability) of the incorporation of MP into the microbial food web and the significant role of unicellular organisms in the transport of MP to higher trophic levels.

Keywords: microplastics, ingestion, microspheres, microalgae, persistent organic pollutants, trophic transport, *Oxyrrhis marina*, *Isochrysis galbana*

The rapid development of plastics production in recent decades has led to the problem of accumulation of related waste. Marine debris is at least 60% plastic [Kozlovskii, Blinovskaia, 2015]. In the marine environment, its polymer base undergoes hydrolysis, photolysis, and microbiological redox reactions; this results in the degradation of plastic fragments [Ateia et al., 2020; Auta et al., 2017] and the formation of particles of various size, including microscopic ones (less than 5 mm), which many researchers [Barnes et al., 2009; Betts, 2008; Fendall, Sewell, 2009; Moore, 2008; Støttrup et al., 1986] consider microplastics (hereinafter MP). One of the main environmental risks associated with MP is their bioavailability to marine hydrobionts [Desforges et al., 2015; Egbeocha et al., 2018]. Having a positive or neutral buoyancy [Van Cauwenberghe et al., 2015], MP are incorporated into food webs of aquatic

biota since MP are comparable in size to phytoplankton – the initial link of a food web. By entering the gastrointestinal tract of zooplankton, MP can form aggregates; this, in turn, leads to a reduced excretion rate [Egbeocha et al., 2018; Ogonowski et al., 2016] and, consequently, to an increased probability of MP transport to higher trophic levels. In addition to physiological damage to an organism (movement disorder, false satiety, etc.), the negative effect of this process is related to MP vector function: MP are involved in the transport of toxic substances included in their composition (plasticizers, dyes, styrene, antimicrobials, and so on) via the food web [Egbeocha et al., 2018; Kwon et al., 2014; Wright et al., 2013]. During grazing, these substances can leach and accumulate in animal tissues [Batel et al., 2016; Kozlovskii, Blinovskaia, 2015]. Moreover, polymer particles adsorb hydrophobic persistent organic pollutants which are present in seawater [Ogata et al., 2009], and this increases their bioavailability for MP uptake by marine biota [Batel et al., 2016]. Concentrations of persistent organic pollutants on MP surface can be several times higher than background levels [Avio et al., 2015]. This carries a risk of the transport of toxins from animal intestinal tract to tissues [Avio et al., 2015; Koelmans, 2015; Rehse et al., 2016; Watts et al., 2014], and hydrobionts inhabiting coastal waters are most vulnerable to this adverse effect [McCormick et al., 2016].

The relationship between MP pollution and trophic processes in microorganisms that form the basis of the food web is poorly studied [Rehse et al., 2016]. In the present work, we analyzed the incorporation into the diet of a heterotrophic dinoflagellate *Oxyrrhis marina* Dujardin, 1841 – an inhabitant of the Black Sea coasts – of MP that correspond in size to cells of microalgae (the main food item of this species under natural conditions). *O. marina* was chosen as an object of study due to the fact that this dinoflagellate inhabits the coastal zone most exposed to MP pollution and, therefore, is at risk. Moreover, it is one of the main phyto- and bacterioplankton consumers in coastal marine ecosystems and the species actively involved in carbon recycling [Hansen, 1991; Roberts et al., 2010]. *O. marina* is successfully used in aquaculture production to feed copepods [Støttrup et al., 1986] – the main and highest quality feed for fish larvae [Khanaichenko, Bitjukova, 1999]. Interestingly, 90% of aquaculture equipment is made of various types of plastics, and this, due to the factors described above, inevitably leads to MP pollution of the environment where hydrobionts are cultured. The possibility of MP incorporation into food webs significantly reduces the quality of final aquaculture products [Wu et al., 2020]. Thus, the heterotrophic dinoflagellate *O. marina* can serve as a model object to study plastic consumption by unicellular organisms.

The aim of this work was to study the presence/absence in *O. marina* of grazing selectivity towards MP and cells of the haptophyte microalga *Isochrysis galbana* Parke, 1949 in their food mixture and to quantify several trophic indicators of each of these food items, including the medium clearance rate (F) and grazing rate (G).

MATERIAL AND METHODS

We carried out a comparative analysis of the main indicators of cell consumption of the haptophyte *I. galbana* and plastic microspheres by the dinoflagellate *O. marina*: the clearance rate (captured volume per cell per time unit) (F); grazing rate (cell abundance per cell per time unit) (G); and selective grazing of one or another “prey” in the mixture.

In the experiment, we used *O. marina* and *I. galbana* cultures from the working collection of IBSS aquaculture and marine pharmacology department, as well as Polychromatic Red polystyrene dyed microspheres, 5.6 µm in diameter (excitation, 491 nm; manufacturer, Polysciences, Inc., the USA).

Microalgae were cultured on Walne medium [Coutteau, 1996] at (24 ± 1) °C, constant irradiance of 5,000 lux, without aeration. Aliquots of cultures were used in the exponential growth phase. *O. marina* culture was pre-adapted to the experimental conditions and maintained for a day without feed.

Experimental scheme. *I. galbana*, microspheres, and their mixture (hereinafter ISO, MS, and ISO-MS, respectively) were introduced as food items into prepared *O. marina* culture (hereinafter OXY) so that the total volume of the medium was 18 mL. The incubation in conical glass vessels lasted for 3 h under constant stirring.

To account for MS loss due to their settling and adhering to vessel walls, an additional glass vessel was placed – the one containing only nutrient medium and MS. Thus, the abundance dynamics of microorganisms and MS was studied in four types of vessels (each in three replicates): 1) OXY-ISO; 2) OXY-MS; 3) OXY-ISO-MS; and 4) MS. The initial abundance of *O. marina*, *I. galbana*, and MS in the experimental vessels was as follows: from 25×10^3 to 50×10^3 cells·mL⁻¹ (OXY); from 10×10^3 to 50×10^3 cells·mL⁻¹ (ISO); and from 5×10^3 to 10×10^3 MS·mL⁻¹.

Cell condition monitoring and quantification of microorganisms and MS were performed by microscopy techniques and flow cytometry. Specifically, 1 mL of a sub-sample was taken from every experimental vessel at the very beginning and then every 20 min.

The clearance rate (F) and grazing rate (G) were calculated according to [Frost, 1972], but a feeding-independent process – MS adhering to vessel walls – was taken into account as well. To study the grazing selectivity of *O. marina* with a mixture of *I. galbana* and MS (the OXY-ISO-MS experiment), the selectivity index was used [Ivlev, 1961]. It was calculated as follows: $(R_i - P_i) / (R_i + P_i)$, where R_i is the proportion of the i -th food item in the predator diet; P_i is the proportion of the i -th food item in the medium. The values of the selectivity index varied from -1 (complete avoidance) to +1 (maximum preference).

Microscopy. MS and microorganisms were microphotographed under a Nikon Eclipse TS100-F microscope equipped with a digital camera, in epifluorescence mode (a set of light filters for excitation in the blue area of the spectrum). Due to bright green fluorescence, MS were clearly visible in the nutrient medium and in *O. marina* digestive vacuoles (see Fig. 2).

Cytometric analysis. A Cytomics™ FC500 flow cytometer (Beckman Coulter, the USA), equipped with a 488-nm single-phase argon laser, and CXP software were used to study abundance dynamics of MS and abundance and size of *O. marina* and *I. galbana* cells in the experimental vessels. Total microalgae abundance was determined in unstained samples by gating a cell population on 2-parameter cytograms – forward scatter (FS) and autofluorescence in the red (FL4, 675 nm) and green (FL1, 525 nm) areas of the spectrum on dimensionless logarithmic scales (Fig. 1).

MS and microalgae concentrations were calculated from sample flow rates (15 and 60 µL·min⁻¹, respectively), time of counting (100–360 s), and the abundance of cells (or MS) recorded during this time interval (in microalgae samples, a minimum of 3,000 cells for each replicate). Measurement quality was controlled using Flow-Check™ calibration fluorospheres (Beckman Coulter) with a known concentration in the sample.

RESULTS

A rapid decline in the abundance of MS indicated high rates of their grazing by the dinoflagellate. Specifically, within the first hour of the experiment, MS abundance dropped to extremely low values ($< 10^2$ MS·mL⁻¹) in all vessels (see Fig. 3A, C). *O. marina* cells containing MS in their digestive

vacuoles acquired green fluorescence. As a consequence, a subcluster of points with high FL1 values was formed on the cytograms (Fig. 1).

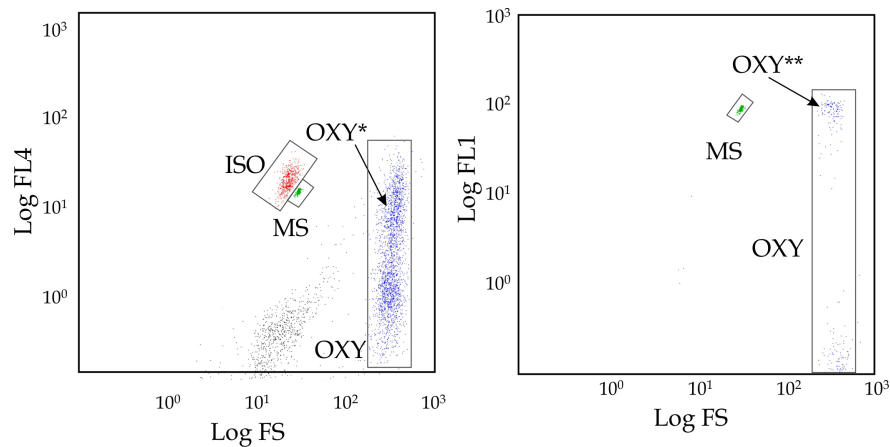


Fig. 1. Gating of the heterotrophic dinoflagellate *Oxyrrhis marina* (OXY) and its food objects [polystyrene microspheres (MS) and the haptophyte *IsochrYSIS galbana* (ISO)] on two-parameter cytograms – of forward scatter (FS) and of red (FL4, 675 nm) and green (FL1, 525 nm) fluorescence. OXY* denotes dinoflagellate cells grazing both *I. galbana* and microspheres; OXY**, dinoflagellate cells with microspheres in their digestive vacuoles

MS grazing by *Oxyrrhis marina* cells was recorded under a microscope as well. In some cases, up to 5–6 brightly fluorescent microspheres could be identified in digestive vacuoles of one cell (Fig. 2).

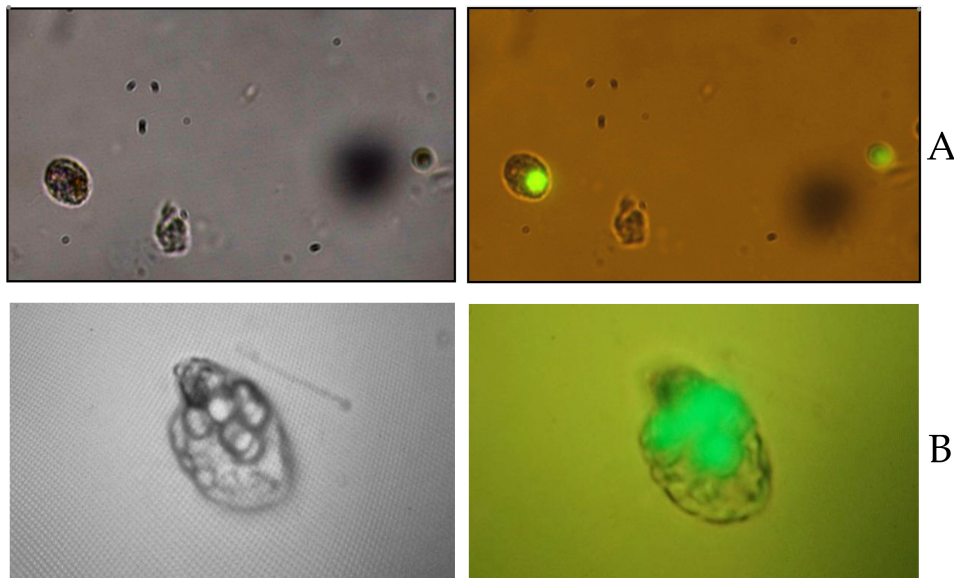


Fig. 2. Cells of the dinoflagellate *Oxyrrhis marina* with one (A) and several (B) microspheres in its digestive vacuoles in bright field (images on the left) and in epifluorescence mode (images on the right)

During the first 2 hours of the experiment, the dinoflagellate almost completely grazed *I. galbana* (Fig. 3B). MS abundance in the medium was restored with time and reached about 10^3 MS·mL⁻¹ by the end of the experiment (Fig. 3A, C). Such an unusual dynamics occurs since MP cannot be digested in digestive vacuoles. An increase in MS abundance meant that the dinoflagellate excreted them back into the medium.

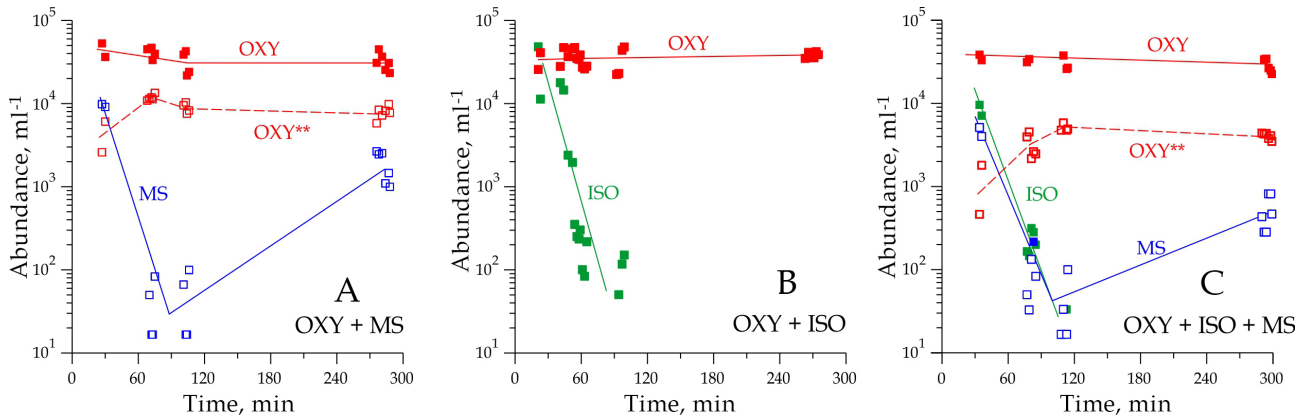


Fig. 3. Dynamics of the abundance of microspheres (MS), the haptophyte *Isochrysis galbana* (ISO), and the heterotrophic dinoflagellate *Oxyrrhis marina* (OXY) in the experimental vessels with different diets: A, microspheres only (OXY-MS); B, haptophytes only (OXY-ISO); C, the mixture (OXY-ISO-MS). Dynamics of *O. marina* cell abundance with MS in their digestive vacuoles (OXY**) is shown separately in plots A and C

Thus, a sharp drop in MS concentration in the medium at the initial stage of the experiment resulted from MS rapid grazing and their accumulation in digestive vacuoles. Then, MS were excreted back into the medium, and this led to a “compensation” of their abundance, which, however, did not reach the initial values. In accordance with this scheme of the processes, the abundance of *O. marina* cells containing MS in their vacuoles first increased and then reached a certain plateau (Fig. 3A, C).

The values of *O. marina* abundance in all series of the vessels remained almost the same throughout the experiments (Fig. 3A, B, C). This could indicate the lack of negative effect of MS on the dinoflagellate division rate. However, the duration of the experiments may have been insufficient to reveal such an effect.

Calculations of the clearance rates (F) by the dinoflagellate showed as follows. In the OXY-ISO series, the F value was $(0.12 \pm 0.04) \mu\text{L}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}$; this was lower than in the OXY-MS series $[(0.19 \pm 0.06) \mu\text{L}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}]$. So, in the first case, *O. marina* cells captured a smaller volume *per* time unit than in the second case (Fig. 4). Moreover, *O. marina* cells spent less time on capturing *I. galbana* cells than on capturing MS.

The same pattern, though to a lesser extent, was maintained in the series with the mixture of microalgae and microspheres as the feed (OXY-ISO-MS). The clearance rate calculated taking into account the haptophyte microalgae $[(0.17 \pm 0.02) \mu\text{L}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}]$ was slightly lower than the value obtained for MS $[(0.19 \pm 0.003) \mu\text{L}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}]$ (Fig. 4). The grazing rates (G) of these two “preys” in the OXY-ISO-MS mixture were $(0.21 \pm 0.01) \text{MS}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}$ (\pm standard deviation) and $(0.38 \pm 0.01) \text{ISO}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}$. These values were significantly lower than in the OXY-ISO and OXY-MS mono-diet experiments $[(1.93 \pm 0.68) \text{ISO}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}$ and $(0.45 \pm 0.04) \text{MS}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}$, respectively], *i. e.*, the expansion of the range of food items resulted in a decrease in the grazing rate (Fig. 4). The maximum of G was observed in the OXY-ISO experiment $[(1.93 \pm 0.68) \text{ISO}\cdot\text{cells}^{-1}\cdot\text{h}^{-1}]$ (Fig. 4), which means that *O. marina* consumed *I. galbana* with the highest efficiency.

However, differences in the grazing rates and clearance rates did not affect *O. marina* grazing selectivity. The values of the Ivlev selectivity index obtained for *I. galbana* and MS were close to zero (-0.03 and 0.05 , respectively), which reflected the lack of grazing selectivity: the dinoflagellate ingested MP on a par with live food. This result indicated that MP could be incorporated into the food web of marine ecosystems at the lowest trophic levels.

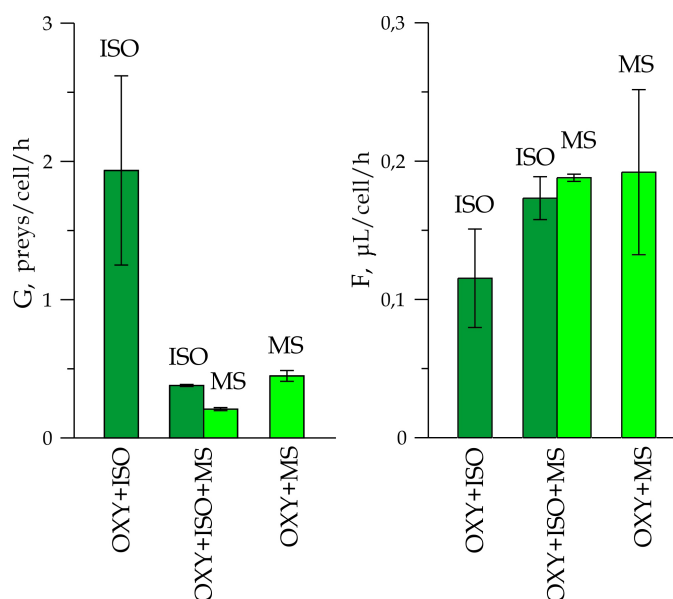


Fig. 4. Grazing rate (G) for the heterotroph *Oxyrrhis marina* (OXY) [the “preys” are the haptophyte *Isochrysis galbana* (ISO) and polystyrene microspheres (MS)] and clearance rate (F) for *O. marina* cells in the experimental vessels with different diets: OXY-MS, microspheres only; OXY-ISO, microalgae only; OXY-ISO-MS, the mixture of microalgae and microspheres

DISCUSSION

To date, reports on MP consumption by multicellular hydrobionts (detritophages, *Artemia*, copepods, mussels, crabs, *etc.*) are quite numerous [Batel et al., 2016; Egbeocha et al., 2018; Procter et al., 2019; Watts et al., 2014; Wu et al., 2020], whereas the incorporation of artificial particles into the diet of unicellular organisms has been studied poorly [Christaki et al., 1998; Rillig, Bonkowski, 2018]. Our results fill this gap to some extent by demonstrating the ability of marine protists to actively consume MP. However, we had to question the ability of the heterotrophic dinoflagellate *O. marina* to distinguish artificial particles from its normal food items – unicellular microorganisms.

The ideas of chemotactic or other warning signals in heterotrophic protists, which enable them to recognize plastic particles and prevent their phagocytosis, remain rather controversial. Specifically, *O. marina* is known for size-selective grazing [Hansen et al., 1996]. It has also been noted in several studies that *O. marina* may reject certain types of food items after capturing prey, at the stage of grazing [Flynn et al., 1996; Hansen et al., 1996; Wolfe et al., 1997]. Moreover, it was established that *O. marina* has receptors on the cell surface, with which it recognizes a prey [Martel, 2009]. According to preliminary experimental data [Hartz, 2010], *O. marina* plasma membrane contains rhodopsin, which makes this heterotroph capable of recognizing photoautotroph cells by red chlorophyll autofluorescence. The presence of such a prey recognition mechanism indicates *O. marina* ability to distinguish microalgae from other, non-pigmented food objects, *inter alia* MP.

Only three experimental studies are known in which artificial particles were incorporated into *O. marina* diet. Specifically, E. Wootton *et al.* [2007] established a new biochemical mechanism for prey recognition in unicellular protists, including *O. marina*. The core of this mechanism is in the presence of a special receptor – mannose-binding lectin – on the predator cell surface. As found, blocking this receptor significantly inhibited *O. marina* feeding on the microalga *I. galbana*, and application of mannose to the surface of plastic microspheres doubled the grazing rate. Pre-incubation of the dinoflagellate with mannose

solution completely deprived it of the ability to identify any plastic particles coated with a polysaccharide layer. The characteristic feature of this study was that plastic microspheres were used in experiments only after a special treatment – application of sugars on their surface [Wootton et al., 2007]. Thus, the results presented by the authors did not allow making a confident conclusion on the ability of *O. marina* to consume MP.

In the second investigation [Hammer et al., 1999], artificial particles (silicon and plastic microspheres, albumin microbeads, etc.) were offered to the heterotrophic dinoflagellate as prey along with its natural food items (live phytoplankton). *O. marina* was found to confidently consume artificial particles, but the rate was significantly lower than when feeding on regular food. The authors explained this selectivity by the magnitude of a charge on the surface of a prey: a more negative potential of an artificial particle prevented its capture by the predator. Differences between surface charges for *O. marina* and its prey increased the probability of collision and duration of their contact; consequently, the grazing rate rose [Hammer et al., 1999]. The initiation of the capture of the prey with a more negative surface charge occurred at a shorter distance from the prey (only 0.5 to 2 μm), whereas that with a higher potential occurred at significantly greater distance (6 to 10 μm), which increased the capture probability [Hammer et al., 1999]. In any case, this study established the incorporation of artificial particles into *O. marina* diet. However, the ability of the dinoflagellate to identify these food items was shown as well, which contradicts our results.

Finally, D. Lyakurwa [2017] demonstrated in his experiment that *O. marina* ingested plastic microspheres just as intensively as a natural prey – the cryptophyte alga *Rhodomonas baltica* Karsten, 1898. These observations are fully consistent with our results. In both cases, *O. marina* did not reject the plastic microspheres proposed. Equally high grazing rates by the heterotrophic dinoflagellate both of artificial and natural food items suggested a very limited ability of these protist to distinguish MP from its natural prey. We can only assume that selective grazing failed to be revealed both by us and [Lyakurwa, 2017] due to rapid adsorption of biopolymers (microalgae exometabolites and bacteria) on MP surface, which are abundant in the experimental medium. “Packaging” microspheres in an organic “film” disguised them as an “edible” prey and thus could hinder their identification by the predator.

The sorption properties of plastic particles have recently become the subject of much research attention as they determine their vector function – the ability to accumulate and transport organic pollutants [Ateia et al., 2020]. We believe that a thin organic film on MP surface, the same as a bacterial film or fouling community, increases MP bioavailability and facilitates their incorporation into trophic processes. At the same time, “taste quality” of particles may depend on the rate of formation of an organic film and its chemical composition. These characteristics are difficult to control even under experimental conditions since they can vary greatly in different nutrient media and cultured microorganisms. Accordingly, the phagocytosis rate of MP is an unpredictable value. This hypothesis well explains, on the one hand, the fact of MP consumption by predatory protists and, on the other hand, contradictory data on the ability of protists to distinguish MP from food items.

Interestingly, in the OXY-ISO series, where *O. marina* fed on *I. galbana* cells only, the grazing rate was higher than that for MS in the OXY-MS series (Fig. 4). This may be due to the fact that MP, having entered *O. marina* digestive vacuoles, stayed in the predator cell for a longer time as they could not be digested. Therefore, the grazing rate decreased in this case. *I. galbana* cells were rapidly digested, which allowed the predator to maintain a high rate of food consumption. The mobility of *I. galbana* cells could also increase their attractiveness to *O. marina*.

The abundance dynamics of *O. marina* cells with microspheres inside (Fig. 3A, C) indicated recaptures of MS after their excretion. This occurred both in the absence of an alternative food source (*I. galbana*) in the OXY-MS series and in its presence in the OXY-ISO-MS series. It can be assumed that *O. marina* did not identify MP upon re-encounter as a less suitable food object and captured them for the second time on a par with *I. galbana* cells. So, no trophic adaptation occurred. At the same time, MS consumption had no negative effect on the dinoflagellate: there were no significant differences in *O. marina* abundance in all experimental series within the time range studied (Fig. 3A, B, C). The same result was obtained in another experiments [Lyakurwa, 2017]. The negative effect of MS consumption could be manifested over longer exposure time; this assumption requires further experimental verification.

It should be noted that microspheres used in the experiment were microgranules of polystyrene, which is a major contributor to the chemical composition of microplastic pollution in marine coastal waters [Cordova et al., 2019]. This type of plastic is widely used in various human activities and is an unstable product [Cooper, Corcoran, 2010; Koelmans, 2015]. Thus, under solar radiation, its brittleness increases; at high temperatures, the polymer disintegrates to form a monomer [Cooper, Corcoran, 2010]. So, we can argue that polystyrene microgranules are capable of incorporating into the microbial food web, being food items even for unicellular organisms. The results obtained in this work highlight the need for further study of MP incorporation into trophic webs, MP effect on the physiological state of the smallest hydrobionts, and the possibility of biomagnification of persistent organic pollutants by planktonic organisms.

Conclusions:

1. The heterotrophic dinoflagellate *Oxyrrhis marina* ingests plastic microspheres even in the presence of alternative food objects from its natural diet.
2. Consumption rates of *Isochrysis galbana* cells and microspheres by the dinoflagellate in the OXY-ISO-MS mixture were significantly lower than in the OXY-ISO and OXY-MS mono-diet experiments, *i. e.*, the expansion of the range of food items led to a decrease in the grazing rate.
3. The high clearance rate when the dinoflagellate fed on live prey (*I. galbana*) indicated that it takes less time for the predator to capture microalga cells than to capture plastic microspheres.
4. The values of the Ivlev selectivity index obtained for *I. galbana* and microspheres indicated the lack of selective grazing in *O. marina*. This confirms the possibility of incorporation of microplastics into the microbial food web.
5. Trophic adaptation of *O. marina* cells to microplastics did not occur: cells consumed polystyrene microspheres for the second time even in the presence of an alternative food item.
6. No significant differences in the dynamics of *O. marina* abundance when feeding on live prey (*I. galbana*) and microplastics were revealed. Thus, there was no negative effect of microplastics on the predator, at least on the time scale of the experiment (hours).

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ПОТРЕБЛЕНИЕ ЧАСТИЦ МИКРОПЛАСТИКА ГЕТЕРОТРОФНОЙ ДИНОФЛАГЕЛЛЯТОЙ *OXYRRHIS MARINA*

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Включение частиц микропластика (МП) в микробную пищевую цепь и их дальнейшая передача на более высокие трофические уровни практически не исследованы. В данной работе закономерности поглощения МП одноклеточными организмами анализировали в культуре гетеротрофной динофлагелляты *Oxyrrhis marina* (ОХУ). В качестве пищевых объектов для *O. marina*

использовали гаптофитовую микроводоросль *Isochrysis galbana* (ISO), полистирольные микросферы (MS) размером 5,6 мкм, а также их смесь (ISO-MS). Динамику численности микроорганизмов и микросфер изучали с помощью проточного цитометра. Показано, что гетеротроф *O. marina* потреблял частицы МП даже в условиях наличия своих обычных жертв — микроводорослей; при этом МП не оказывал на него негативного влияния. Скорости выедания «жертв» в смеси OXY-ISO-MS составили $(0,21 \pm 0,01) \text{ MS} \cdot \text{кЛ}^{-1} \cdot \text{ч}^{-1}$ (\pm стандартное отклонение) и $(0,38 \pm 0,01) \text{ ISO} \cdot \text{кЛ}^{-1} \cdot \text{ч}^{-1}$ и были достоверно ниже, чем в экспериментах с монодиетами OXY-ISO [$(1,93 \pm 0,68) \text{ ISO} \cdot \text{кЛ}^{-1} \cdot \text{ч}^{-1}$] и OXY-MS [$(0,45 \pm 0,04) \text{ MS} \cdot \text{кЛ}^{-1} \cdot \text{ч}^{-1}$], то есть усложнение состава (расширение спектра) пищевых объектов вело к снижению скорости их потребления. Скорость осветления среды динофлагеллятой *O. marina* в экспериментах с монодиетами составила $(0,12 \pm 0,04)$ и $(0,19 \pm 0,06) \text{ мКЛ} \cdot \text{кЛ}^{-1} \cdot \text{ч}^{-1}$ для OXY-ISO и OXY-MS соответственно, а значит, на поимку клеток ISO динофлагеллята *O. marina* затрачивала меньше времени, чем на поимку MS. Эту же закономерность наблюдали и в экспериментах со смесью пищевых объектов (OXY-ISO-MS): скорость осветления среды в сосудах с ISO [$(0,17 \pm 0,02) \text{ мКЛ} \cdot \text{кЛ}^{-1} \cdot \text{ч}^{-1}$] была незначительно ниже, чем в сосудах с MS [$(0,19 \pm 0,003) \text{ мКЛ} \cdot \text{кЛ}^{-1} \cdot \text{ч}^{-1}$]. Трофической адаптации *O. marina* к MS не происходило, на что указывал факт их вторичного потребления даже в условиях наличия альтернативного кормового объекта — *I. galbana*. Не была выявлена и селективность питания *O. marina* ни к одной из «жертв», будь то *I. galbana* или пластиковые микросферы. Полученные результаты указывают на возможность (и высокую вероятность) включения МП в микробную пищевую цепь и на важную роль одноклеточных организмов в передаче МП на более высокие трофические уровни.

Ключевые слова: микропластик, поглощение, микросферы, микроводоросли, стойкие органические загрязнители, трофический перенос, *Oxyrrhis marina*, *Isochrysis galbana*