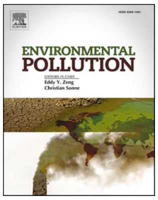
Environmental Pollution 342 (2024) 123127

Contents lists available at ScienceDirect

Environmental Pollution

journal homepage: www.elsevier.com/locate/envpol



Micro- and nanoplastics interact with conventional pollutants on microalgae: Synthesis through meta-analysis☆

Jingke Ge [a,1], Peng Jin [b,1], Shuyu Xie [a], John Beardall [a,c], Yuan Feng [a], Can Guo [a], Zengling Ma [d], Guang Gao [a,*]

[a] State Key Laboratory of Marine Environmental Science & College of Ocean and Earth Sciences, Xiamen University, Xiamen, 361005, China
[b] School of Environmental Science and Engineering, Guangzhou University, Guangzhou, 510006, China
[c] School of Biological Sciences, Monash University, Clayton, VI 3800, Australia
[d] Zhejiang Provincial Key Laboratory for Water Environment and Marine Biological Resources Protection, Wenzhou University, Wenzhou, 325035, China

ARTICLE INFO

Keywords:
Heavy metal
Microalgae
Microplastics
Nanoplastics
Pesticide
Physiological performance

ABSTRACT

Micro- and nanoplastics (MNPs) have been found to occur intensively in aquatic environments, along with other conventional pollutants (Po) such as heavy metals, pesticides, pharmaceuticals, etc. However, our understanding of how MNPs and Po interact on aquatic primary producers is fragmented. We performed a quantitative meta-analysis based on 933 published experimental assessments from 44 studies to examine the coupled effects of MNPs and Po on microalgae. Although the results based on interaction type frequency (the proportion of each interaction type in all results) revealed dominantly additive interactions (56%) for overall physiological performance, an overall antagonistic effect was observed based on the mean interaction effect sizes. A higher proportion of antagonistic interaction type frequency was found in marine species compared to fresh species. The antagonistic effects were particularly significant for growth, oxidative responses, and photosynthesis, which could be attributed to the adsorption effect of MNPs on Po and thus the decreasing concentrations of pollutants in the medium. Larger-sized, negatively charged or uncharged and aged MNPs had higher proportions of antagonistic effects compared to smaller-sized, positively charged and virgin MNPs, due to their stronger adsorption capacity to Po. This study provides a comprehensive insight into the interactive effects of MNPs and Po on microalgae.

1. Introduction

The development of plastics has brought great convenience to human society, and they have been increasingly used in all kinds of fields since the 1950s (Barnes et al. 2009). However, the constant input of plastics from land via rivers and rainfall run-off has allowed MNPs (micro- and nanoplastics) to distribute and accumulate in aquatic environments, both freshwater and marine (Shahul Hamid et al., 2018). Microplastics can exist in the water column for a long time due to their low-density and hard-to-degrade characteristics, although they can evolve into more forms, such as smaller sizes, different shapes, differently charged states, roughened surfaces etc., after undergoing a range of physical and chemical processes (Burrows et al., 2020; Davidson, 2012). Microplastics can pose a threat to aquatic organisms regardless of their existence in the water column or sediments and can be transferred to humans through, but not limited to, seafood (Alimi et al., 2018; Feng et al., 2019; Feng et al., 2020b). Most studies of the effects of plastics on the environment have focused on aquatic animals because they can ingest microplastics (Gao et al., 2021b). Meanwhile, there are increasing concerns about the effects on microalgae because they contribute about half of global primary productivity and O_2 production (Caroppo and Pagliara, 2022). Many existing laboratory studies have demonstrated the negative effects of microplastics on microalgae as well as neutral and positive effects (Bergami et al., 2017; Mao et al., 2018; Zhao et al., 2019b). For instance, polystyrene (PS) decreased the growth rate, chlorophyll content and lipid concentration of the marine microalgae *Tetraselmis suecica* and *Amphora subtropica* (Raju et al., 2021). Polyvinyl chloride (PVC) (5–100 mg L^{-1}) exerted adverse effects on *Karenia*

☆ This paper has been recommended for acceptance by Da Chen.

* Corresponding author.

E-mail address: guang.gao@xmu.edu.cn (G. Gao).

[1] These authors contributed equally to this work.

https://doi.org/10.1016/j.envpol.2023.123127
Received 11 August 2023; Received in revised form 10 November 2023; Accepted 7 December 2023

mikimotoi growth, chlorophyll content and photosynthetic efficiency (Zhao et al., 2019b). In contrast, it was also reported that PS enhanced the growth and maximum quantum yield of stationary phase cells of *Chlorella pyrenoidosa* (Mao et al., 2018). As for neutral results, those studies using environmental relevant concentrations were usually found to cause limited harm to microalgae (Niu et al., 2021; Phuong et al., 2016).

Before MNPs attracted a lot of attention, more conventional pollutants (Po, e.g., heavy metals, pharmaceuticals, nanoparticles, pesticides) had attracted much research attention because of their wide distribution in aquatic environments and potential harm to aquatic ecosystems (Pal et al., 2010; Petrie et al., 2015). Conventional pollutants mainly come from domestic sewage or industrial wastewater, and thus there are relatively high concentrations in estuaries or along the coast (Batool et al., 2016). Both freshwater and marine organisms, e.g., phytoplankton, zooplankton, fish, shrimp and shellfish, are threatened by these pollutants (Zhang et al., 2016). Conventional pollutants usually negatively influence the physiological parameters of microalgae, such as growth, oxidative responses, photosynthesis and pigment content (Miguez et al., 2021). For example, nonylphenol and octylphenol exerted an inhibitory effect on growth, photosynthesis, and PSII activity of *C. pyrenoidosa* and *Scenedesmus obliquus* (Yang et al., 2021) and sulfonamides also reduced the growth of *C. vulgaris* (Chen et al., 2020).

In the natural environment, MNPs and Po usually occur together. Therefore, there has been increasing research interest in investigating the interactive effects of MNPs and Po on microalgae. The most investigated physiological indicators include growth, photosynthesis, oxidative response and pigment content. The combined effects exhibited a range of responses, including synergistic (Tunali et al., 2020; Yi et al., 2019), antagonistic (Fu et al., 2019; Gunasekaran et al., 2020) and additive effects (Bellingeri et al., 2019; Gonzalez-Pleiter et al., 2021). Due to the variability in reported responses, a quantitative meta-analysis of the interactions between these two stressors on microalgae is needed. Meta-analysis allows us to exclude the interference of details like inconsistent experimental conditions and provides us with a more comprehensive understanding. Herein, we gathered and integrated published literature through a quantitative meta-analysis to facilitate bridging the knowledge gap involving the combined effects of MNPs and conventional pollutants on microalgae. The findings presented here supply new insights into the combined impacts of MNPs and Po exposure on microalgae, which can support decision-making processes for environmental sustainability and ecosystem well-being.

2. Materials and methods

2.1. Literature search

We conducted a systematic review of the literatures sourced from ISI Web of Science (v.5.35) and Google Scholar, focusing on studies that explore experimental responses of microalgae to both micro-/nanoplastic and conventional pollutants. Our search was driven by the following keywords: microplastic, nanoplastic, pollutant, contaminant, chemical, and microalgae. These investigations were carried out until 23 April 2023. Initially, we found 419 papers, which were then trimmed down to 933 published assessments from 44 published reports upon further scrutiny (refer to Supplementary Table S1 for more details).

2.2. Study selection criteria

We evaluated each piece of literature for its relevance, retaining only those studies that focused on the responses of microalgae to both MNPs and conventional pollutants through an exhaustive factorial experiment. This particular approach involved testing the MNPs and conventional pollutants individually and in synergy. This experimental design resulted in four independent treatments: control (C), micro-/nanoplastics (MNPs), conventional pollutants (Po), and a blend of micro-/nanoplastics and conventional pollutants (MNPs + Po). Only researches satisfying this criterion proceeded to the next level of analysis. Any study that failed to provide or made it impossible to figure out data variation (like standard deviation, standard error, confidence intervals or variance) or sample size (absence or pseudo-replication) was not taken into consideration, as per the guidelines outlined by the Preferred Reporting Items for Systematic Reviews and Meta-analyses guidelines (PRISMA) (O'Dea et al., 2021). We followed the PRISMA checklist for meta-analyses and reviewed papers/experiments to maintain high standards in reporting meta-analyses.

2.3. Data collection

We procured means, error estimates (standard deviation or standard error of mean), and sample sizes from published figures and tables. Data exhibited in tables was straightforward to extract. For graphical data, we asked for the primary data from the corresponding authors. For those data that could not be obtained through the above approaches, we utilized GetData Graph Digitizer (version 2.24) for data acquisition. For meta-analysis purposes, all harvested error estimates were converted into standard errors. To comply with the statistical principle of independence among observations in the meta-analysis (Hedges et al., 1999), we gathered data at the endpoint stages (e.g., cell numbers) or derived them via time trend, a rate (e.g., growth) reported at multiple time intervals throughout the study. If an experiment reported identical biological responses through various metrics at several time points, only the most comprehensive metric for that response variable was considered to preclude pseudo-replication (Kroeker et al., 2013). Data normalization was carried out as needed to maintain internal consistency within each response.

Assessments of literature on different biological responses to stressors encompassed: growth, cell abundance, pigment content, photosynthesis, concentrations of biochemical compounds, and enzymatic rates. These responses were segregated into five categories as outlined by Jin et al. (2019) with slight modifications, including (1) growth (based on cell abundance or chlorophyll level per volume water); (2) motility (based on swimming speed); (3) oxidative response (e.g., antioxidant enzymes activities, superoxide dismutase, SOD; oxidative stress biomarkers, malondialdehyde levels, MDA and reactive oxygen species content, ROS); (4) photosynthesis (e.g., maximum photochemical efficiency, F_v/F_m); (5) pigment (e.g., the content of carotenoids and chlorophylls *a, b, c* per cell). In addition to biological responses, data were subdivided into subcategories based on (1) taxonomical classifications (Bacillariophyta, Chlorophyta, Cryptophyta, Cyanophyta, Euglenozoa and Haptophyta), as per AlgaeBase (www.algaebase.org); (2) habitats (freshwater and marine); (3) exposure time (24 h, 48 h, 72 h, 96 h and >96 h); (4) the polymer type of MNPs (polyamide-PA, polyacrylonitrile polymer-PAN, polyethylene-PE, polyethylene terephthalate-PET, polylactic acid-PLA, polyoxymethylene-POM, polystyrene-PS, polyurethane-PU and polyvinyl chloride-PVC); (5) the characteristic of MNPs, including size (microplastic and nanoplastic), charge conditions (negative, neutral and positive), and aging conditions ('virgin' and aged); (6) and type of conventional pollutants (heavy metal, nanoparticle, natural organic matter, pesticide, pharmaceutical and plastic additive). Comprehensive details on the extracted data can be found in Table S2.

2.4. Effect size calculation

We determined individual, main, and interactive effect sizes for each test employing Hedge's *d* (Hedges, 1984) in accordance with methodologies stated by Gurevitch et al. (2000). The choice of Hedge's *d* over other available measures of effect size (e.g., natural logarithm of response ratio (lnRR)) was based on the following reasons (1) it aligns with the ANOVA model utilized in numerous publications where a significant interaction effect size indicates deviation from the null model of additive effects (Gurevitch et al., 2000); (2) it remains unaffected by unequal sampling variances in the associated groups and includes a correction factor (*J(m)*, see below) for small sample sizes; and (3) it is frequently used as a metric for meta-analyses, thus facilitating comparisons with findings reported elsewhere (Jin et al., 2021, 2022).

Individual effects denote the response when a singular stressor is present relative to the control, while main effects contrast the net effect of a stressor in the presence and absence of another stressor. The individual effects of MNPs (d_{mp}) and conventional pollutants (d_{po}) were calculated with respect to the control (d_c) using this equation (Crain et al., 2008; Gurevitch et al., 1992):

$$d_{mp} = \frac{Y_{mp} - Y_c}{s} J(m)$$

$$d_{po} = \frac{Y_{po} - Y_c}{s} J(m)$$

where Y_{mp}, Y_{po}, and Y_c are means of a variable in the treatment groups of MNPs, conventional pollutants, and the control correspondingly; *s* and *J (m)* signify pooled standard deviation and correction term for small samples, which were calculated using the equations below:

$$s = \sqrt{\frac{(n_c - 1)s_c^2 + (n_{mp} - 1)s_{mp}^2 + (n_{po} - 1)s_{po}^2 + (n_{mp+po} - 1)s_{mp+po}^2}{n_c + n_{mp} + n_{po} + n_{mp+po} - 4}}$$

$$J(m) = 1 - \frac{3}{4m - 1}$$

where n_c, n_{mp}, n_{po}, and n_{mp+po} represent the sample sizes, and s_c, s_{mp}, s_{po}, and s_{mp+po} are the standard deviations in the control and experimental groups of MNPs, Po and their combination (MNPs + Po), respectively; *m* denotes the degree of freedom ($m = n_c + n_{mp} + n_{po} + n_{mp+po} - 4$). The main effects of MNPs (d_{MP}), Po (d_{PO}), and their interaction (d_{MP+PO}) were deduced as follows:

$$d_{MP} = \frac{(Y_{MP} + Y_{MP+PO}) - (Y_{PO} + Y_C)}{2s} J(m)$$

$$d_{PO} = \frac{(Y_{PO} + Y_{MP+PO}) - (Y_{MP} + Y_C)}{2s} J(m)$$

$$d_{MP+PO} = \frac{(Y_{MP+PO} - Y_{PO}) - (Y_{MP} - Y_C)}{2s} J(m)$$

For an individual effect d_z (where z is *mp* or *po*), the sampling variance is (Gurevitch et al., 2000):

$$v_z = \frac{n_z + n_c}{n_z n_c} + \frac{d_z^2}{2(n_z + n_c)}$$

and for a main effect d_Z (where Z is MP or PO), the sampling variance is (Gurevitch et al., 2000):

$$v_Z = \frac{1}{4}\left[\frac{1}{n_{MP}} + \frac{1}{n_{PO}} + \frac{1}{n_{MP+PO}} + \frac{1}{n_C} + \frac{d_Z^2}{2(n_{MP} + n_{PO} + n_{MP+PO} + n_C)}\right]$$

The variance v_Z (where Z is MP + PO) for interaction was computed as:

$$v_Z = \frac{1}{n_{MP}} + \frac{1}{n_{PO}} + \frac{1}{n_{MP+PO}} + \frac{1}{n_C} + \frac{d_Z^2}{2(n_{MP} + n_{PO} + n_{MP+PO} + n_C)}$$

If the response traits indicate stress (e.g., activities of superoxide dismutase (SOD), contents of malondialdehyde (MDA) and reactive oxygen species (ROS)), the above-calculated effect sizes were transformed using the formula: 0-*d*, where *d* represents the main or individual effect of MNPs, conventional pollutants, or their interaction.

Consistent with Crain et al. (2008), we utilized individual effect sizes to categorize the interactions of MNPs and Po into one of three types, i.e., additive, synergistic or antagonistic. If the 95% CI of the interaction term overlapped with zero, it was interpreted as an additive interactive effect. Where individual effects were either both negative or one positive and one negative, interaction effects <0 were seen as synergistic while effects >0 were deemed antagonistic. If both stressors had a positive individual effect then interaction types were interpreted the opposite way, i.e., interaction effects <0 were antagonistic and >0 were synergistic (Piggott et al., 2015).

2.5. *Statistical analyses*

All analyses were performed with the statistical software R (R version 4.1.3), utilizing the function rma. mv (meta-analysis via multivariate/multilevel linear mixed-effects models) available in the *metafor* package (Viechtbauer, 2010). Initially, the *escalc* function was used to perform a weighted meta-analyse for estimating the mean interaction effect sizes and variances across diverse studies. In each analysis, we considered “Observation ID” as a random-effect to account for the random component of effect size variation among observations (Gurevitch and Hedges, 1993) (refer to Supplementary Table S3 for equations and model details). Additionally, with the application of random-effects meta-analyses, we then applied a series of mixed effects meta-analyses where selected categorical moderators (e.g., taxon, habitat, response trait, exposure time, characteristic of MNPs and type of conventional pollutants) were considered fixed effects to assess mean interactions at each category level (refer to Table S3 for model terms). To ascertain the frequency of interaction types, we calculated the proportion of each interaction type within all results, compared these using a chi-square test across different groups within the same category. For maintaining the robustness of the analysis, categories with fewer than four samples ($n < 4$) were not included in the analysis. Within these analyses, heterogeneity between (Q_M) and within (Q_E) moderator levels (e.g., taxon, response trait, habitat) was compared using mixed models to ascertain the significance of each categorical moderator (Borenstein et al., 2011). A notable Q_T (total heterogeneity) suggests that the variance of effect sizes across different studies surpasses what would be expected from sampling error, hinting a possible inherent data structure. Q_M describes the amount of heterogeneity, which can be attributed to the chosen categories (refer to Table S3 for model structure). A significant Q_M implies that the categorical moderator exerts a substantial effect. Q_E describes the amount of heterogeneity, which remains unexplained when the model is considered. Thus, a significant value of Q_E signifies the presence of additional variance within effect sizes that requires explanation.

Methodological factors such as MNPs concentration, Po concentration, MNPs size and experimental duration could potentially influence the effect size. Therefore, we determined a range of continuous variables above for each data point where viable. Subsequently, a continuous random-effects meta-analysis was conducted to examine the influences of these factors on the effect size, treating MNPs concentration, Po concentration, MNPs size and duration of experiment as distinct continuous variables (Kroeker et al., 2013). Linear or quadratic regressions were utilized to quantify the relationship. Moreover, since the ranges of MNPs/Po concentration and MNPs size varied largely among different studies, we calculated the log base 10 of these variables as x axis. Additionally, separate random-effects meta-analyses were conducted for each response trait to analyse these data more detailed.

We tested for publication bias through Rosenthal’s fail-safe numbers method to assess the reliability of observed effects (Rosenthal, 1979). Rosenthal’s fail-safe number denotes the number of non-significant effect sizes required to alter the significance level (*p*-value) as reported by the model. We calculated a fail-safe number of 294,600 noticeably exceeding the minimum threshold suggested based on our sample size (294,600 > 5n+10, where n representing the total number of observations). Additionally, the trim and fill method (Duval and Tweedie, 2000) was employed to estimate any missing studies required to restore symmetry in the funnel plot for the overall dataset, which failed to identify ultimately (missing studies = 0). After passing these two tests, we concluded that our data set showed no signs of publication bias.

3. Results and discussion

3.1. Overview of the datasets

Forty-four articles reporting 933 assessments of microalgal responses to MNPs and Po were analysed in this study, and the data sources can be found in Supplementary Table S1. The majority of observations were from Chlorophyta (n = 549, 59% of total observations) and Cyanophyta (n = 278, 30% of total observations), with approximately 1~6% of the assessments recorded for Bacillariophyta, Cryptophyta, Euglenozoa and Haptophyta (Table S3). Most of the data points stem from studies with microalgae from freshwater habitats (n = 672), with 261 observations from marine habitats (Fig. 1a). Based on the selection criteria, our analysis included five different response traits, which were growth (n = 382), motility (n = 10), oxidative response (n = 341), photosynthesis (n = 84) and pigments (n = 90) (Fig. 1b). Most studies (475) used polystyrene (PS), and accounted for 78% of all polymer materials (Fig. 2a). In addition, the data with smaller-sized, uncharged and virgin MNPs had higher proportions (55%, 69% and 93%, respectively) compared to smaller-sized, positive or negative charged and aged MNPs (Fig. 2b). Short-term experiments (≤96 h) dominated the assessments (89%) (Fig. 1b), with a maximum study duration of 28 days (S'anchez-Fortún et al., 2022). Finally, our analysis also included six different conventional pollutants according to specific type, which were heavy metals (n = 237), nanoparticles (n = 167), natural organic matter (n = 30), pesticides (n = 248), pharmaceuticals (n = 206), and plastic additives (n = 35) (Fig. 3).

Globally, our results showed a significant (i.e., the 95% CIs did not overlap with zero) negative effect of both MNPs, d_{MP} (Hedge's $d = -2.875$, 95% CI = [−3.328, −2.423], $Z = -12.460$, $p < 0.001$) and conventional pollutants, d_{PO} (Hedge's $d = -4.146$, 95% CI = [−4.669, −3.624], $Z = -15.546$, $p < 0.001$) alone on microalgae, which is consistent with most previous studies (Chen et al., 2020; Yang et al., 2021). However, the 95% CI of the overall interaction term, $d_{MP + PO}$ showed a positive effect (Hedge's $d = 1.918$, 95% CI = [1.570, 2.266], $Z = 10.812$, $p < 0.001$) (Fig. 1a & Table S3). These results suggested that the negative effects of both MNPs and Po on microalgae were reversed by their combination, leading to a positive effect. Although adverse effects of conventional pollutants have been widely found (Chen et al., 2020; Yang et al., 2021), some studies demonstrate that conventional pollutants at low concentrations could enhance microalgal growth (Gu et al., 2020; Lozano et al., 2014). This could explain how the combination of MNPs and Po stimulated the physiological performance of microalgae as the adsorption of Po by MNPs can lead to decreases in the concentrations of Po to stimulatory levels.

Regarding the interaction type frequency, our results showed that additive effects dominated (56%), followed by over a quarter of antagonistic effects (37%) and a small fraction of synergistic effects (7%) (Fig. 1a). The relatively smaller proportion of antagonistic interactions outweighed additive and synergistic interactions, which led to opposite responses when MNPs and Po acted together (Fig. 1a). This antagonistic interaction has also been reported in previous studies. For instance, the strong adsorption capacity of MNPs for glyphosate resulted in antagonistic effects when combined, reducing their ability to inhibit the growth of *Microcystis aeruginosa* (Zhang et al., 2018). Furthermore, growth of *C. vulgaris* was inhibited by Cu^{2+}, but was stimulated by the combination of Cu^{2+} and MNPs (Fu et al., 2019). The adsorption of Cu^{2+} by MNPs may explain the stimulating effect since some studies show that MNPs can adsorb other environmental pollutants due to their hydrophobicity and large specific surface area (Holmes et al., 2014; Liu et al., 2022).

3.2. Response of different microalgae phyla and habitats to MNPs and Po

Overall, significant differences among microalgal taxonomic groups were found when assessing their responses to MNPs ($Q_M = 53.804$, $p < 0.001$) and Po ($Q_M = 72.834$, $p < 0.001$) (Table S3 and Fig. 1a). MNPs showed negative effects on Bacillariophyta (Hedge's $d = -2.158$, 95% CI = [−4.033, −0.282], $Z = -2.255$, $p = 0.024$), Chlorophyta (Hedge's $d = -1.936$, 95% CI = [−2.509, −1.364], $Z = -6.633$, $p < 0.001$) and Cyanophyta (Hedge's $d = -5.339$, 95% CI = [−6.149, −4.529], $Z = -12.916$, $p < 0.001$), while Po exerted negative effects on Chlorophyta (Hedge's $d = -2.906$, 95% CI = [−3.560, −2.252], $Z = -8.713$, $p < 0.001$), Cyanophyta (Hedge's $d = -7.428$, 95% CI = [−8.352, −6.504], $Z = -15.757$, $p < 0.001$) and Euglenozoa (Hedge's $d = -2.933$, 95% CI = [−5.763, −0.102], $Z = -2.030$, $p = 0.042$), suggesting that Chlorophyta and Cyanophyta are sensitive to both MNPs and Po (Fig. 1a). No significant effects of either MNPs or Po were found for Cryptophyta or Haptophyta (all $p > 0.05$) (Fig. 1a). This could be attributed to motility of algae in these two phyla because algal motility can help them escape from environmental stress (Margalef, 1978). In addition, the limited number of data sets (n = 3 for Cryptophyta and n = 22 for Haptophyta) may also contribute to the large variance, leading to statistical insignificance. Under combined MNPs and Po exposure (Fig. 1a),

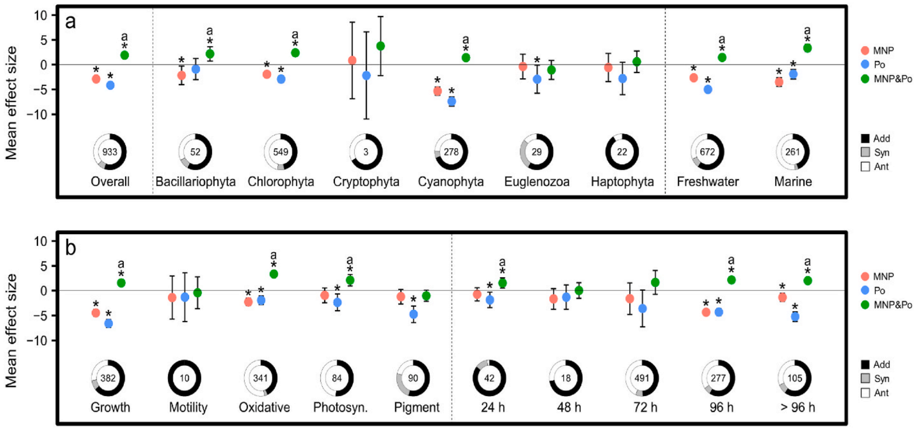


Fig. 1. Mean effect sizes (Hedge's *d*) of micro- and nanoplastics (MNPs) (red), pollutants (Po) (blue) and their interaction (green) observed for the overall dataset, different phylum, habitats (a), and microalgal response trait and exposure time (b). Error bars indicate 95% confidence intervals. Significant effects are denoted by asterisks. For interactions, confidence intervals overlapping 0 indicate additive effects; those >0 or <0 indicate a significant antagonistic (highlighted with the letter "a") or synergistic interaction, respectively. Pie charts indicate the frequencies (%) of additive (black), synergistic (gray) and antagonistic (white) interaction types. Numbers inside pie charts indicate the number of observations. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

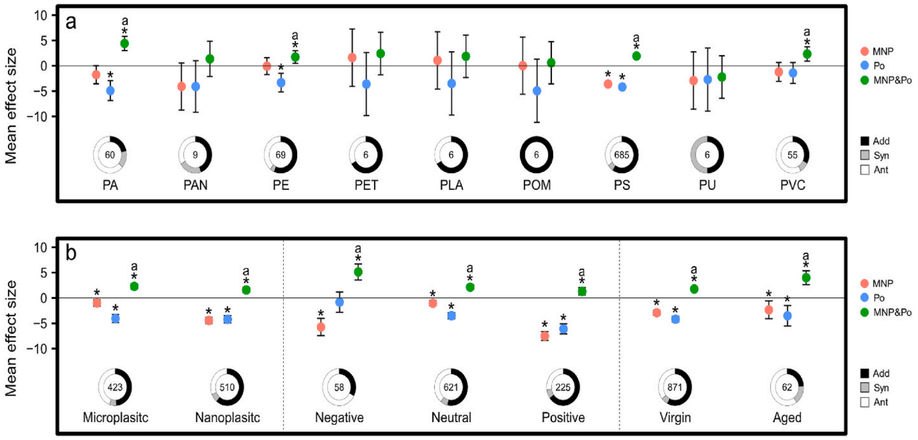


Fig. 2. Mean effect sizes (Hedge's *d*) of micro- and nanoplastics (MNPs) (red), pollutants (Po) (blue) and their interaction (green) observed for the polymer type of MNPs (a), the size rank, charge and aging condition of MNPs (b). Error bars indicate 95% confidence intervals. Significant effects are denoted by asterisks. For interactions, confidence intervals overlapping 0 indicate additive effects; those >0 or <0 indicate a significant antagonistic (highlighted with the letter "a") or synergistic interaction, respectively. Descriptions of pie charts, numbers and color coding see Fig. 1. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

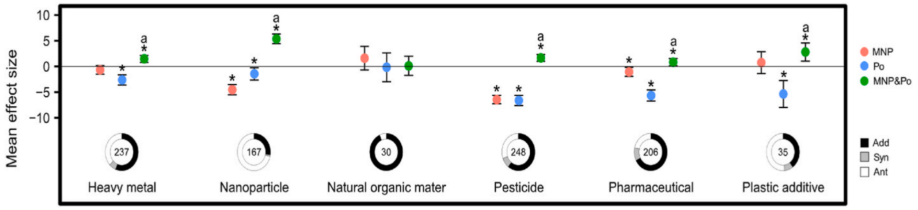


Fig. 3. Mean effect sizes (Hedge's *d*) of micro- and nanoplastics (MNPs) (red), pollutants (Po) (blue) and their interaction (green) observed for the type of pollutants. Error bars indicate 95% confidence intervals. Significant effects are denoted by asterisks. For interactions, confidence intervals overlapping 0 indicate additive effects; those >0 or <0 indicate a significant antagonistic (highlighted with the letter "a") or synergistic interaction, respectively. Descriptions of the error bars, pie charts, numbers and color coding see Fig. 1. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

Bacillariophyta (Hedge's $d = 2.180$, 95% CI = [0.727, 3.632], $Z = 2.941$, $p = 0.003$), Chlorophyta (Hedge's $d = 2.390$, 95% CI = [1.930, 2.851], $Z = 10.168$, $p < 0.001$) and Cyanophyta (Hedge's $d = 1.415$, 95% CI = [0.799, 2.032], $Z = 4.499$, $p < 0.001$) all showed significant positive responses, while the remaining taxonomic groups were tolerance (all $p > 0.05$). In terms of interaction type frequency, additive interactions were prevalent in all taxonomic groups (47–91%), followed by antagonistic interactions (9–47%) ($\chi^2 = 77.481$, $p < 0.001$, df = 10, n = 933, Fig. 1a & Table S4). However, there were more synergistic interactions than antagonistic interactions for Euglenozoa (Fig. 1a). Algal species in Euglenozoa can ingest MNPs like zooplankton and thus the antagonistic interactions between MNPs and Po are reduced but the synergistic interactions were enhanced (Sun et al., 2021).

Microalgae from either freshwater or marine habitat showed significant negative responses to MNPs (both $p < 0.05$) ($Q_M = 2.796$, $p = 0.095$) (Fig. 1a & Table S3). Exposure to conventional pollutants also exerted negative effects on microalgae both from freshwater (Hedge's $d = -4.998$, 95% CI = [−5.602, −4.394], $Z = -16.216$, $p < 0.001$) and marine habitats (Hedge's $d = -1.912$, 95% CI = [−2.883, −0.941], $Z = -3.859$, $p < 0.001$). Effect size was different for freshwater and marine microalgae, with freshwater microalgae being more sensitive to the stress of Po ($Q_M = 27.981$, $p < 0.001$) (Fig. 1a & Table S3). When MNPs and Po acted together, however, both freshwater and marine microalgae were positively impacted (both $p < 0.001$), with a larger effect size for marine species ($Q_M = 21.157$, $p < 0.001$). There were also significant differences in the frequency of interaction type between two habitats ($\chi^2 = 36.631$, $p < 0.001$, df = 2, n = 933), and the antagonisms showed a higher proportion in marine (52%) than freshwater (32%) habitats (Table S4). Our results could be well explained by Liu et al. (2020), who argued that the salinity of water could influence the aggregation of MNPs and Po and thus affect their toxicity. As solid evidence, Vockenberg et al. (2020) had proved that the sorption capacity of MNPs to amine micropollutants increased with increasing salinity. Additionally, pH (Holmes et al., 2014) and temperature (Fonte et al., 2016) are also critical environmental factors that could influence the interaction between MNPs and Po, which should be considered in further research. To sum up, the present study demonstrates that microalgae from different phyla respond differentially to individual effects of MNPs or Po and their combined effects, which suggests that the responses of microalgae to the combination of MNPs and Po are species and habitat dependent.

3.3. *Response of different physiological parameters to MNPs and Po*

There was high variability of effect sizes among different response traits when exposed to MNPs ($Q_M = 33.620$, $p < 0.001$) or conventional pollutants ($Q_M = 68.358$, $p < 0.001$) (Fig. 1b & Table S3). Growth parameters showed a significant negative response to MNPs or Po alone (MNPs: Hedge's $d = -4.464$, 95% CI = [−5.173, −3.755], $Z = -12.342$, $p < 0.001$; Po: Hedge's $d = -6.598$, 95% CI = [−7.399, −5.797], $Z = -16.142$, $p < 0.001$). Oxidative parameters responded similarly but to a relatively smaller extent (MNPs: Hedge's $d = -2.264$, 95% CI = [−3.011, −1.516], $Z = -5.937$, $p < 0.001$; Po: Hedge's $d = -1.935$, 95% CI = [−2.780, −1.091], $Z = -4.493$, $p < 0.001$) (Fig. 1b). It is not surprising that MNPs or Po alone exert negative effects on growth and oxidative response of microalgae as many previous studies have shown these phenomena (Chen et al., 2020; Gao et al., 2017). It has been documented that growth and oxidative response may be the parameters most sensitive to environmental pollutants (Fu et al., 2019; Liao et al., 2020). Our results confirmed this conclusion by analysing 933 sets of data. In addition, exposure to conventional pollutants rather than MNPs dramatically decreased algal photosynthesis (Hedge's $d = -2.356$, 95% CI = [−4.050, −0.662], $Z = -2.726$, $p = 0.006$) and pigment content (Hedge's $d = -4.736$, 95% CI = [−6.391, −3.080], $Z = -5.607$, $p < 0.001$) (Fig. 1b). However, when MNPs and Po acted together, growth (Hedge's $d = 1.558$, 95% CI = [1.028, 2.089], $Z = 5.758$, $p < 0.001$), oxidative parameters (Hedge's $d = 3.344$, 95% CI = [2.749, 3.938], $Z = 11.021$, $p < 0.001$) and photosynthesis (Hedge's $d = 2.104$, 95% CI = [0.961, 3.247], $Z = 3.608$, $p < 0.001$) had significant positive responses, indicating that MNPs and Po had an antagonistic effect (Fig. 1b). This may also be related to the adsorption of conventional pollutants to MNPs, which leads to the decreased availability of Po in waters. It has been reported that Po at low concentrations could stimulate the oxidative response (e.g., enhanced SOD) of microalgae (Gunasekaran et al., 2020). The enhanced oxidative response can reduce the harm caused by ROS, alleviating the negative effects on growth. Elevated enzyme activity could indirectly promote the performance of photosynthesis and growth in the cellular level. Additionally, Po at low concentrations could stimulate the growth of microalgae directly through providing necessary elements (Gu et al., 2020; Lozano et al., 2014), e.g., Cu and Fe ions were important components for relative photosynthetic proteins; the leaching out of few MNPs additive is also confirmed to slightly stimulate algal growth, known as "Hormesis" phenomenon (Chae et al., 2019). Po also reduced pigment content of microalgae while MNPs or MNPs + Po did not show significant effects (Fig. 1b). Neither MNPs nor Po showed influences on microalgal motility (Fig. 1b), indicating microalgal motility is tolerant to these two environmental stressors. Meanwhile, more studies are needed to justify this conclusion because there are only 10 data sets available for motility.

There were significant differences in the frequencies of interaction types among various physiological traits ($\chi^2 = 118.569$, $p < 0.001$, df = 8, n = 907) (Fig. 1b). Although additive interactions were prevalent in all physiological traits, their proportion varied: 43% for oxidative response and 100% for motility. Growth had a middle antagonistic interaction type frequency (27%) with oxidative response most (53%) and motility least (0%), which suggests that growth is the integrated result of other parameters after regulation and trade-off between them. Synergistic interactions were the least frequent type in all physiological traits (0–24%) (Fig. 1b).

3.4. The effects of MNPs with different characteristics on effect size

Regarding the polymer type of MNPs (Fig. 2a), only PS showed negative effects on physiological performance of microalgae (PS: Hedge's $d = -3.612$, 95% CI = [−4.144, −3.080], $Z = -13.302$, $p < 0.001$). PS is recognized as more toxic than other types of MNPs (Podbielska and Szpyrka, 2023). There were also significant differences among MNPs polymer types when evaluating the interaction of MNPs and Po ($Q_M = 16.358$, $p = 0.038$). The combination of PA, PE, PS, or PVC with Po had a positive effect on microalgae while the combination of other polymer types with Po did not show significant effects. Different frequencies of interaction type were also found among MNPs polymer types, with PA (63%), PVC (58%) and PE (39%) having higher proportion of antagonistic interaction ($\chi^2 = 72.936$, $p < 0.001$, df = 16, n = 902) (Fig. 2a & Table S4). Different polymer materials of MNPs possess different abilities to adsorb smaller-size matters or molecular-form pollutants. It was reported that PE possesses the highest adsorption ability, followed by PP and PVC (Alimi et al., 2018; Teuten et al., 2009), which could partially explain the higher proportions of antagonism for PVC or PE with Po.

As for MNPs size (Fig. 2b), both microplastics and nanoplastics exerted negative effects on microalgae (MP: Hedge's $d = -0.990$, 95% CI = [−1.636, −0.344], $Z = -3.005$, $p = 0.003$; NP: Hedge's $d = -4.436$, 95% CI = [−5.030, −3.842], $Z = -14.642$, $p < 0.001$). Meanwhile their combined effects with Po appeared to be positive (both $p < 0.05$) and microplastics (Hedge's $d = 2.276$) showed larger positive effect than nanoplastics (Hedge's $d = 1.578$) in combination with Po. There were significant differences in the frequencies of interaction type between the two scales of MNPs ($\chi^2 = 16.515$, $p < 0.001$, df = 2, n = 933), and the proportion of antagonism in microplastics (44%) was higher than in nanoplastics (32%). The results above suggest that the size of MNPs also plays a decisive role when MNPs act with Po. Previous work based on transcriptomic analysis has also revealed that Cd + PS-NPs is more toxic than Cd + PS-MPs to *Euglena gracilis* (Liao et al., 2020). As for a lower proportion of antagonistic interactions between nanoplastics and Po, the main reason may be that nanoparticles could destroy the cytoplasmic membrane of microalgae by contact and enhance its permeability so that more free-form Po can enter cells (Cao et al., 2022; You et al., 2021). It has been documented that higher intercellular Cu content in *Platymonas helgolandica* could be found while cultured with nano PS compared to micro PS (Gao et al., 2022).

For MNPs charge conditions (Fig. 2b), negatively charged (Hedge's $d = -5.742$, 95% CI = [−7.437, −4.047], $Z = -6.640$, $p < 0.001$), neutrally (Hedge's $d = -1.032$, 95% CI = [−1.540, −0.523], $Z = -3.976$, $p < 0.001$) and positively charged (Hedge's $d = -7.504$, 95% CI = [−8.360, −6.648], $Z = -17.181$, $p < 0.001$) MNPs all presented negative effects on microalgae. Also, there was a consistent result when MNPs acted with Po, revealing a positive effect on microalgae (all $p < 0.05$). The frequencies of interaction type among these three categories showed significant differences ($\chi^2 = 33.453$, $p < 0.001$, df = 4, n = 904), where negatively charged MNPs had the highest proportion of antagonistic interactions with Po (67%), followed by neutral MNPs (39%), and positive charged MNPs being the lowest (28%). Previous studies showed that carboxylated (negatively charged) polystyrene microplastics decreased the TiO_2 nanoparticles toxicity to *Chlorella* sp. (Thiagarajan et al., 2019; Thiagarajan et al., 2021). This finding is consistent with our results, which could be attributed to the negatively charged cell membrane and cell wall of microalgae, so it is harder for negatively charged MNPs that trap pollutants to make contact with microalgae because the same charges repel each other. There were also some exceptions; for example, PS-SO_3H combined with tetracycline enhanced the toxicity of single negatively charged PS-SO_3H or tetracycline alone on *S. costatum* (Feng et al., 2020a). As such, special exceptions such as this should also be taken into consideration and need further exploration.

For MNPs of different aged conditions (Fig. 2b), both virgin and aged MNPs exposure showed significant negative effects on microalgae (Virgin: Hedge's $d = -2.913$, 95% CI = [−3.382, −2.445], $Z = -12.185$, $p < 0.001$; Aged: Hedge's $d = -2.356$, 95% CI = [−4.105, −0.606], $Z = -2.639$, $p = 0.008$). And the combination of MNPs and Po had positive effects on microalgae for both virgin and aged MNPs (both $p < 0.001$). However, there existed significant differences in frequency of interaction type ($\chi^2 = 28.243$, $p < 0.001$, df = 2, n = 933), and the proportion of antagonism in aged MNPs (60%) was largely higher than that in virgin MNPs (36%). Aged MNPs have more rough surface areas and more micropores, which could adsorb more other pollutants and thus may reduce their toxicity (Fu et al., 2019; Wang et al., 2021).

3.5. The effects of different conventional pollutants on effect size

Although the adsorption of Po to MNPs has been widely reported (Li et al., 2022; Santana-Viera et al., 2021; Wang et al., 2020c; Wang et al., 2020a), there are few comparisons of different pollutant types when acting together with MNPs on microalgae. Chemical bonding could be found between MNPs and the antibiotic sulfamethoxazole while electrostatic force interaction was reported to be the main adsorption mechanism between heavy metals and microplastics (Liu et al., 2022; Zhang et al., 2022). According to the various properties of different conventional pollutants, we hypothesized that they may have differential effects on microalgae when interacting with MNPs. Our results showed that except for natural organic matter (e.g., humic acids), the physiological performances of microalgae were negatively impacted when exposed to other conventional pollution, especially pesticides (Hedge's $d = -6.628$, 95% CI = [−7.165, −5.641], $Z = -13.168$, $p < 0.001$), pharmaceuticals (Hedge's $d = -5.664$, 95% CI = [−6.749,

−4.579], $Z = -10.236$, $p < 0.001$) and plastic additive (Hedge's $d = -5.358$, 95% CI = [−7.966, −2.750], $Z = -4.027$, $p < 0.001$) that showed more significantly negative effects (Fig. 3). After combining with MNPs, high variability among pollution types was found ($Q_M = 67.255$, $p < 0.001$) (Table S3); MNPs positively impacted the physiological performances of microalgae with heavy metals, nanoparticles, pesticides, pharmaceuticals and plastic additives (all $p < 0.05$) but had neutral effects for natural organic matter (Fig. 3). Similarly, we found that the frequencies of interaction types differed significantly among various conventional pollutants ($\chi^2 = 133.018$, $p < 0.001$, df = 10, n = 923) (Table S4). Additive interaction accounted for 56–93% of observations for most pollutant types. In terms of antagonistic interaction, highest proportion was found in nanoparticles (71%) followed by plastic additives (48%), heavy metals (37%) and pesticides (31%), with the least in pharmaceuticals (21%). The highest proportion of antagonistic interaction in nanoparticles may be attributed to the strong aggregation between MNPs and nanoparticles (Li et al., 2021; Thiagarajan et al., 2021). Po that has lower proportions of antagonistic interaction usually has weaker adsorption ability to MNPs. For instance, the adsorption amounts of pharmaceuticals (34−111 ng g^{-1}) to MPs are lower than that (61−963 ng g^{-1}) for pesticides (Santana-Viera et al., 2021; Wang et al., 2020a). Synergistic interaction was the least frequent (0−11% of observations) for all pollutant types (Fig. 3).

3.6. *The effects of continuous variables on the effect size of microalgal physiology*

To assess whether MNPs and Po concentration, MNPs size and exposure time influence effect size, we mined data on these factors with different magnitudes, and tested their relationship with effect size using general linear models or nonlinear models.

For MNPs concentration, it has been reported that the negative effects of MNPs on microalgae increased with increasing MNPs concentration in several physiological responses (e.g., growth, F_v/F_m and

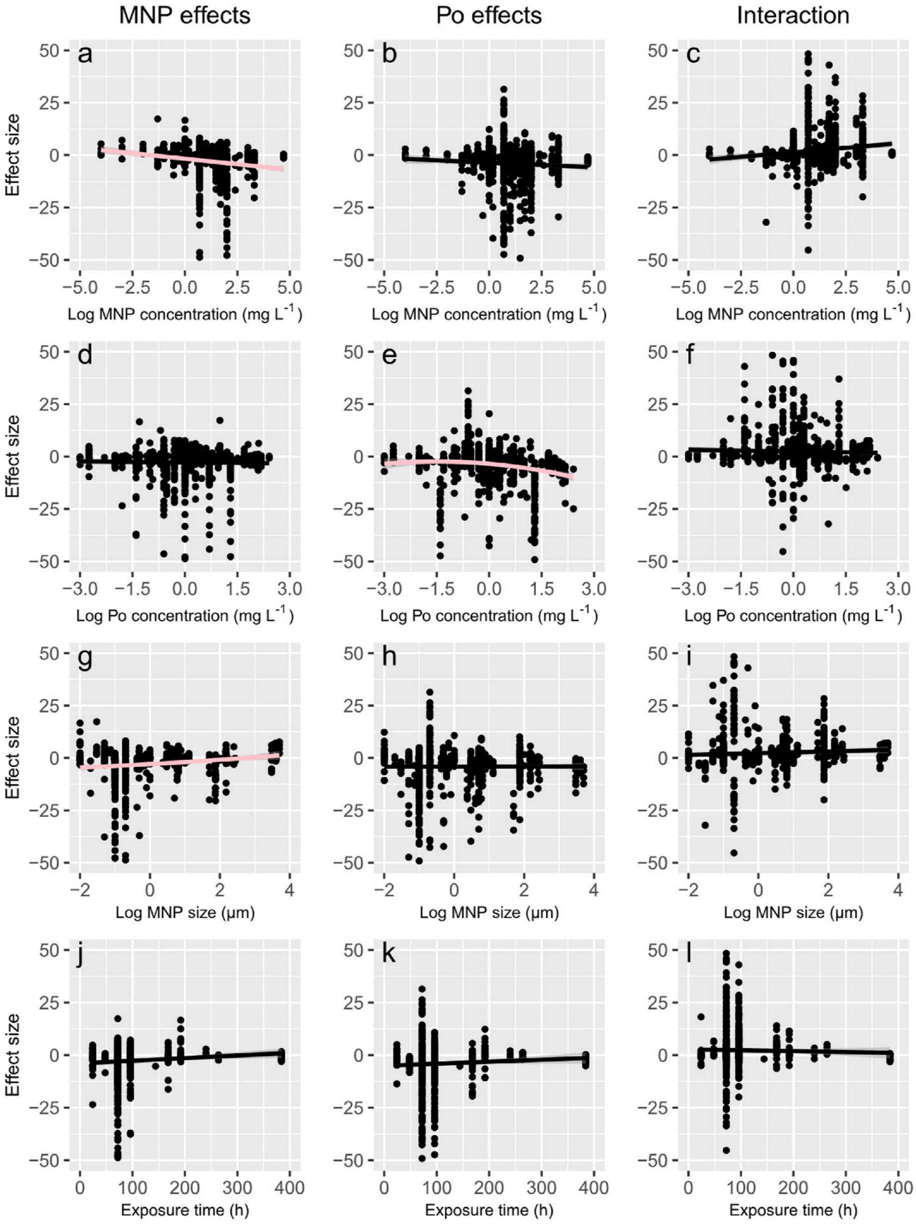


Fig. 4. The effect of MNP concentration (mg L^{-1}) (a–c), pollutant concentration (mg L^{-1}) (d–f), MNP size (μm) (g–i) and exposure time (hour) (j–l) on the overall effect size (Hedge's *d*) of micro- and nano-plastic (MNP), pollutant (Po) and their interaction. The data were fitted with linear or binomial regressions. Pink represents these fitted curves accord with $p < 0.05$. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

pigment) (Gao et al., 2021b). The present study showed a similar trend, where the negative effects became larger with the increase of MNPs concentration (Fig. 4a & Table S5). In contrast, the relationship tended to be positive from neutral when MNPs acted with Po, although there was insignificant in statistical analysis (Fig. 4c & Table S5), indicating that their combined effects were likely to be buffered or antagonistic. Enhanced aggregation could reduce the concentration of MNPs and Po and thus the positive effects of MNPs combined with Po became more apparent with the increase of MNP concentration. For Po concentration, negative effects firstly decreased with enhanced Po concentration in the low concentration scale ($<\sim$4.395 mg/L) and then increased with Po concentration (quadratic regression, $y = -32.16x^2 - 41.36x - 5.08$, $p = 0.031$) (Fig. 4e). Nevertheless, there was a flat trend when MNPs acted with Po, and the value of Hedge's *d* close to zero all the time (Fig. 4f & Table S5), suggesting MNPs alleviate the negative effects no matter at low or high concentrations of Po.

It was not surprising that the physiological performances of microalgae to MNPs alone were positively correlated, from minus tend to zero, with increasing MNPs size (Fig. 4g & Table S5), suggesting that negative effects of MNPs on microalgae were buffered with increasing MNPs size. Meanwhile, persistent negative values under Po treatment were adjusted to neutral in the nearly all MNPs size (Fig. 4h, I & Table S5). Exposure time did not show significant effects on physiological performances of microalgae cultured under any conditions (Fig. 4 j-l & Table S5).

3.6.1. *The effects of continuous variables on the effect size for growth*

After analysing the dataset of growth, nearly the same patterns as the overall parameter were found (Fig. 5), although the linear or nonlinear trends are not statistically significant (all $p > 0.05$, Table S6). The negative effects of MNPs or Po on growth showed a rising trend with increasing concentration of MNPs or Po (Fig. 5a and d), while an increasing positive effect and a flat trend were found respectively, when MNPs and Po combined (Fig. 5c and f). The positive effect turned to become a negative effect on microalgal growth with the increase of MNPs concentration (Fig. 5a). This positive effect is consistent with that of Song et al. (2020) who showed that the activity of microalgae could

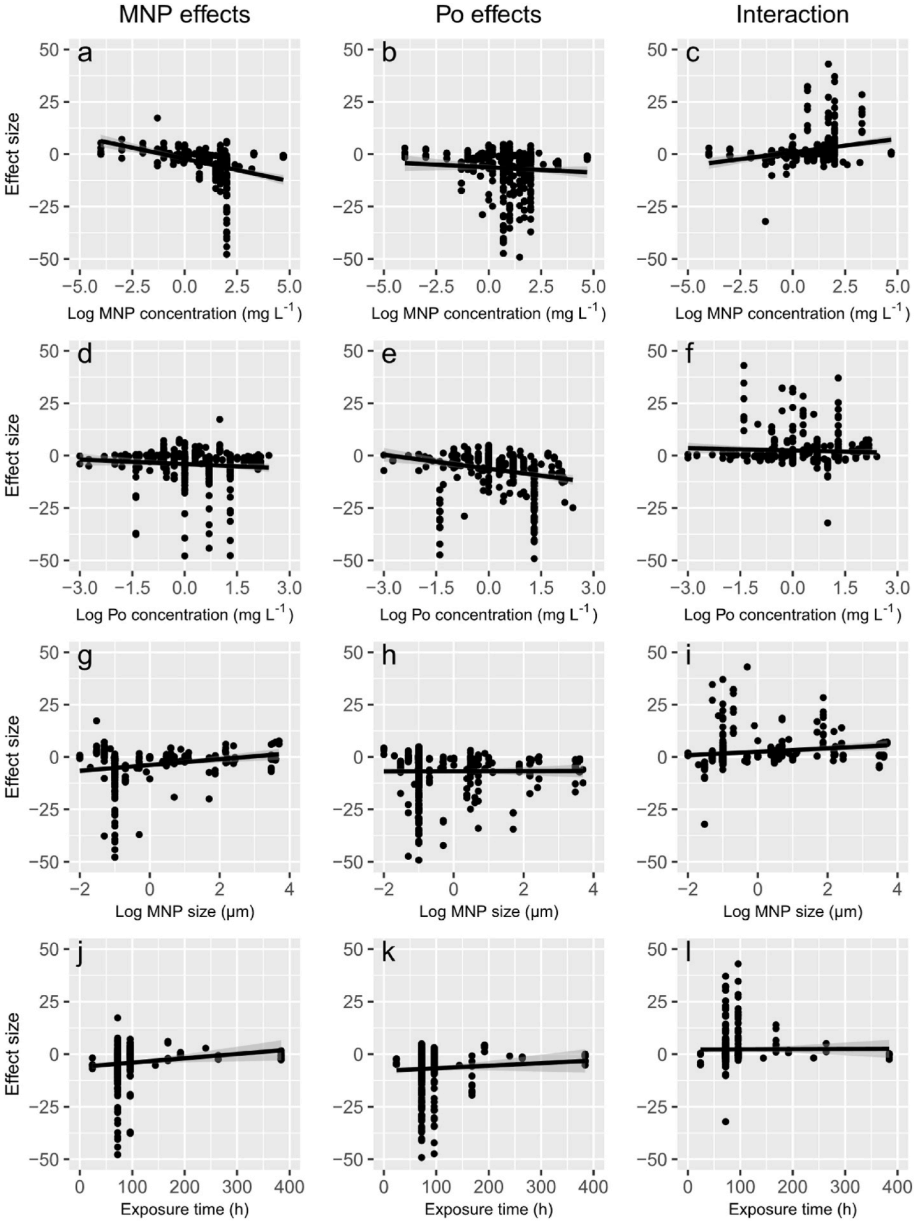


Fig. 5. The effect of MNP concentration (mg L^{-1}) (a–c), pollutant concentration (mg L^{-1}) (d–f), MNP size (μm) (g–i) and exposure time (hour) (j–l) on the effect size (Hedge's *d*) of micro- and nanoplastics (MNPs), pollutants (Po) and their interaction in growth. The data were fitted with linear regressions or binomial regressions.

be stimulated by low levels of additives leaching from MNPs. Meanwhile, harms of MPs was generally observed with further increasing MNPs concentration (Zhao et al., 2019a). However, an opposite trend is found under the treatment of MNPs + Po (Fig. 5c). The adsorption of Po to MNPs was attributed to this phenomenon.

3.6.2. The effects of continuous variables on the effect size for oxidative response

Microalgal oxidative response increased firstly with increasing MNPs concentration for MNPs treatment and then decreased after the MNPs concentration reached approximately 1 mg L^{-1} (Fig. 6a & Table S7) (quadratic regression, $y = 24.47x^2 - 0.02x - 2.63$, $p = 0.014$); however, it showed an opposite trend for MNPs + Po treatment (Fig. 6c & Table S7) (quadratic regression, $y = -78.43x^2 - 3.19x + 9.32$, $p = 0.012$), suggesting the negative effects of MNPs or Po alone on the oxidative response of microalgae would turn to be positive when they acted together with increasing MNPs concentration. In the beginning, increasing positive results under MNPs + Po treatment could be explained with effective adsorption of MNPs to Po; however, the toxicity of MNPs tended to appear with further increasing MNPs concentration. Similar trends were also found with increasing Po concentration (Fig. 6d, f & Table S7) (quadratic regression, $y = -99.47x^2 + 0.50x + 9.56$, $p = 0.001$), suggesting that low concentration of Po benefits the positive feedback of oxidative response firstly, and then reduced.

For MNPs size, the negative effects of MNPs on microalgal oxidative response declined with increased MNPs size (Fig. 6g) while an opposite trend was found for Po treatment (Fig. 6f). For MNPs + Po treatment, the positive effects on oxidative response increased with MNPs size (Fig. 6i & Table S7) (quadratic regression, $y = 29.86x^2 - 88.50x + 9.32$, $p = 0.012$). NPs (600 nm), approaching to MPs, could reduce the oxidative stress induced by ibuprofen by accelerated the degradation of ibuprofen in *Chlorella pyrenoidosa* (Wang et al., 2020b). Liao et al. (2020) also

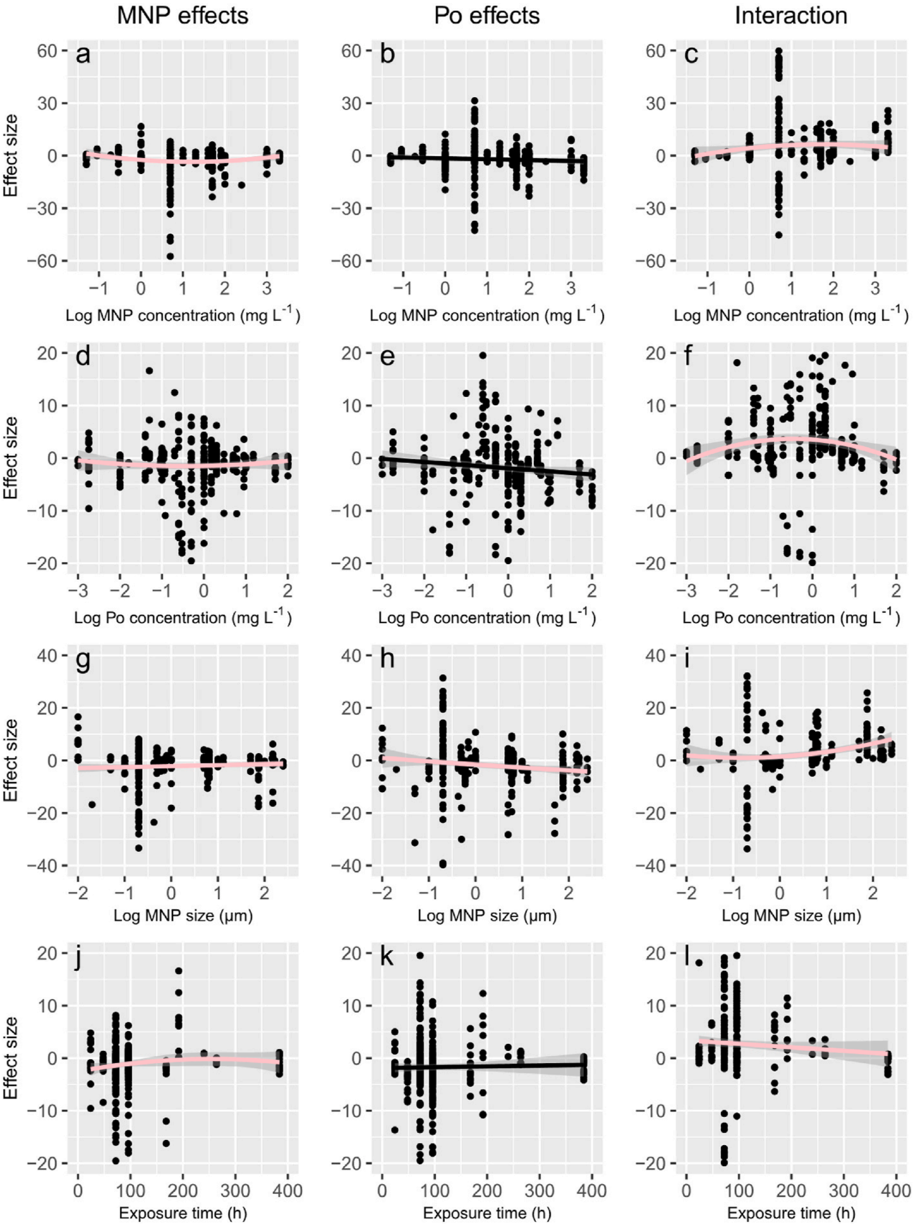


Fig. 6. The effect of MNP concentration (mg L^{-1}) (a–c), pollutant concentration (mg L^{-1}) (d–f), MNP size (μm) (g–i) and exposure time (hour) (j–l) on the effect size (Hedge's *d*) of micro- and nanoplastics (MNPs), pollutants (Po) and their interaction in oxidative response. The data were fitted with linear regressions or binomial regressions. Pink represents these fitted curves accord with $p < 0.05$. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

reported that the combination of MPs (5 μm) with Cd imposed lower inhibition on growth of *Euglena gracilis* compared to the combination of NPs (100 nm) and Cd, which could be attributed to smaller harm from MPs to photosynthesis-related pathways. In addition, negative effects under MNPs alone and positive effects under MNPs + Po treatment after short-term culture both tended to be neutral as culture time extended (Fig. 6j and i), indicating algal recovery and acclimation to MPNs and Po.

3.6.3. *The effects of continuous variables on the effect size for photosynthesis*

Photosynthesis is an important index to evaluate the state of algal cells (Hartmann et al., 2014). The photosynthetic response to either MNPs or Po alone did not significantly change with increasing MNPs concentration, as well as MNPs + Po treatment (Fig. 7a–c & Table S8). The negative effect of Po alone on photosynthesis increased obviously with increasing Po concentration (Fig. 7e & Table S8), suggesting the apparent harm from Po to photosynthesis. Our results showed that the negative effect of Po was alleviated in the presence of MNPs, and their joint effect with Po was not significantly affected by Po concentration (Fig. 7e, f & Table S8).

The effect size of MNPs on microalgal photosynthesis changed from negative to zero with increasing MNPs size (Fig. 7g). However, there was no significant trend for MNPs + Po treatment (Fig. 7i & Table S8). The individual effect of Po on photosynthesis was significantly affected by exposure time; the negative effect increased first and then decreased with extended exposure time (Fig. 7 & Table S8) (quadratic regression, $y = 4.48x^2 + 7.51x - 2.37, p = 0.028$), which could be attributed to rapid toxicity of Po in short term and recovery after a longer period of acclimation. No significant relationship between the effects of MNPs alone or MNPs + Po treatment on photosynthesis and exposure time was found (Fig. 7j, l & Table S8).

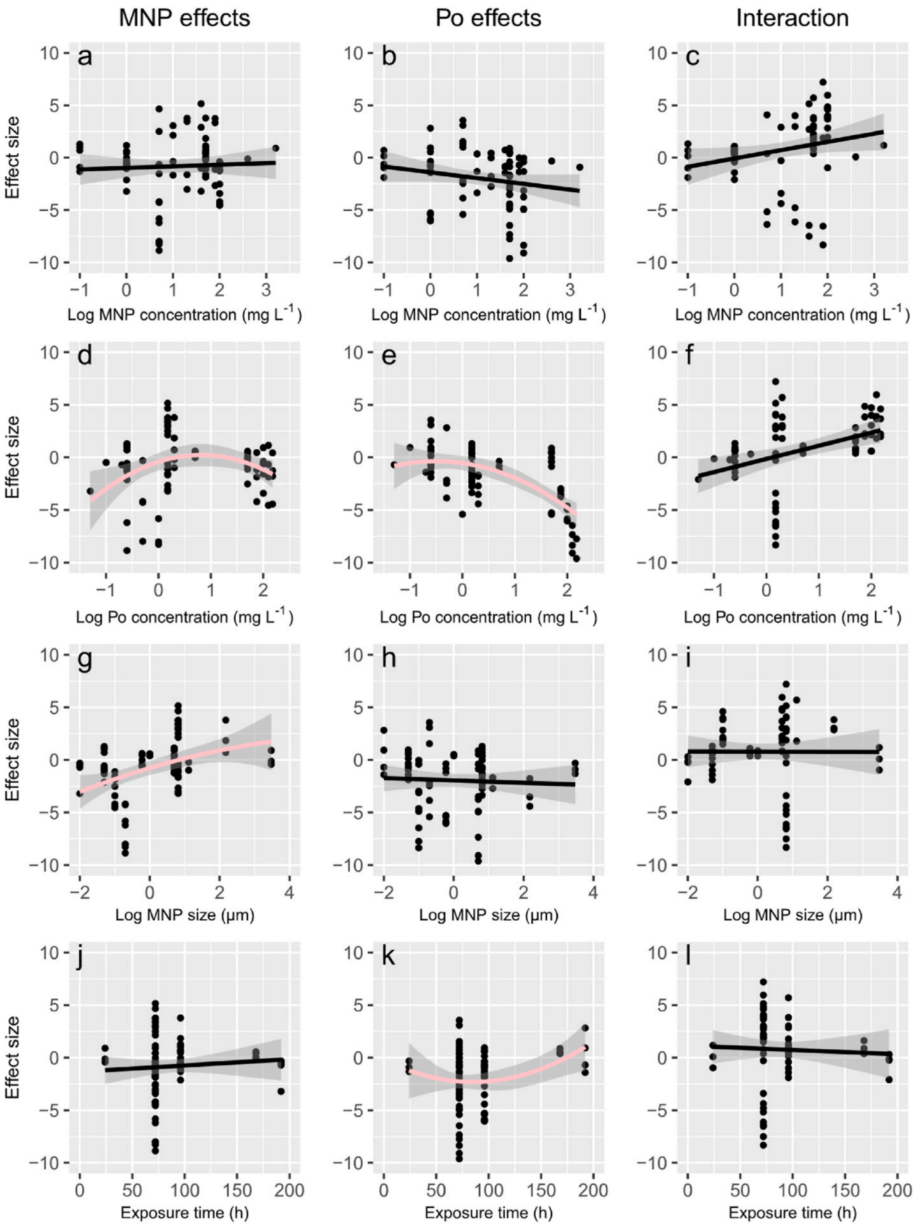


Fig. 7. The effect of MNP concentration (mg L^{-1}) (a–c), pollutant concentration (mg L^{-1}) (d–f), MNP size (μm) (g–i) and exposure time (hour) (j–l) on the effect size (Hedge's *d*) of micro- and nanoplastics (MNPs), pollutants (Po) and their interaction in photosynthesis. The data were fitted with linear regressions or binomial regressions. Pink represents these fitted curves accord with $p < 0.05$. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

3.6.4. The effects of continuous variables on the effect size for pigments

The individual effect size of MNPs on pigment content of microalgae turned to negative values and then levelled off gradually with increasing MNPs concentration (quadratic regression, $y = 6.92x^2 - 14.48x - 1.59$, $p = 0.001$). However, Po alone or their joint effect was not obviously affected by MNPs concentration (Fig. 8a–c & Table S9). Similarly, the response of pigments to Po alone (quadratic regression, $y = -31.02x^2 - 48.55x - 6.13$, $p < 0.001$) was negatively correlated with increasing Po concentration, but this phenomenon disappeared in the combined interaction (Fig. 8e & Table S9), suggesting joint action greatly reduced the toxicity of MNPs or Po to pigment despite at their respective higher concentrations. However, it has also been observed that microplastics-pharmaceutical mixtures led to a reduction in pigment content of *Tetraselmis chuii* to a larger degree than the pharmaceuticals alone, which was explained by the acceleration of uptake of the pharmaceuticals induced by microplastics (Prata et al., 2018). Our results indicate that antagonistic interaction due to the adsorption of Po to MNPs dominates their effects on pigment content of microalgae although there are also some exceptions.

For MNPs size, no significant relation between the effect size of MNPs or Po on pigment content and MNPs size was found (Fig. 8g, h and Table S9). Nevertheless, the effect size of MNPs + Po tended to negative firstly and then turned to neutral with increasing MNPs size (quadratic regression, $y = 18.09x^2 - 4.23x - 1.16$, $p < 0.001$, Fig. 8i), suggesting MNPs may have the most destructive power to cell membrane so that more Po can enter cells (Thiagarajan et al., 2019) when the cell size is around 1.3 μm. MNPs can influence the cell membrane protein function and generate a gap between the cell wall and cell membrane, which induces the damage of Po to the internal biochemical composition, including protein and pigment content (Huang et al., 2019; Wang et al., 2023). The specific reasons for this tipping point remain unknown and more studies are needed to confirm this result and explore potential

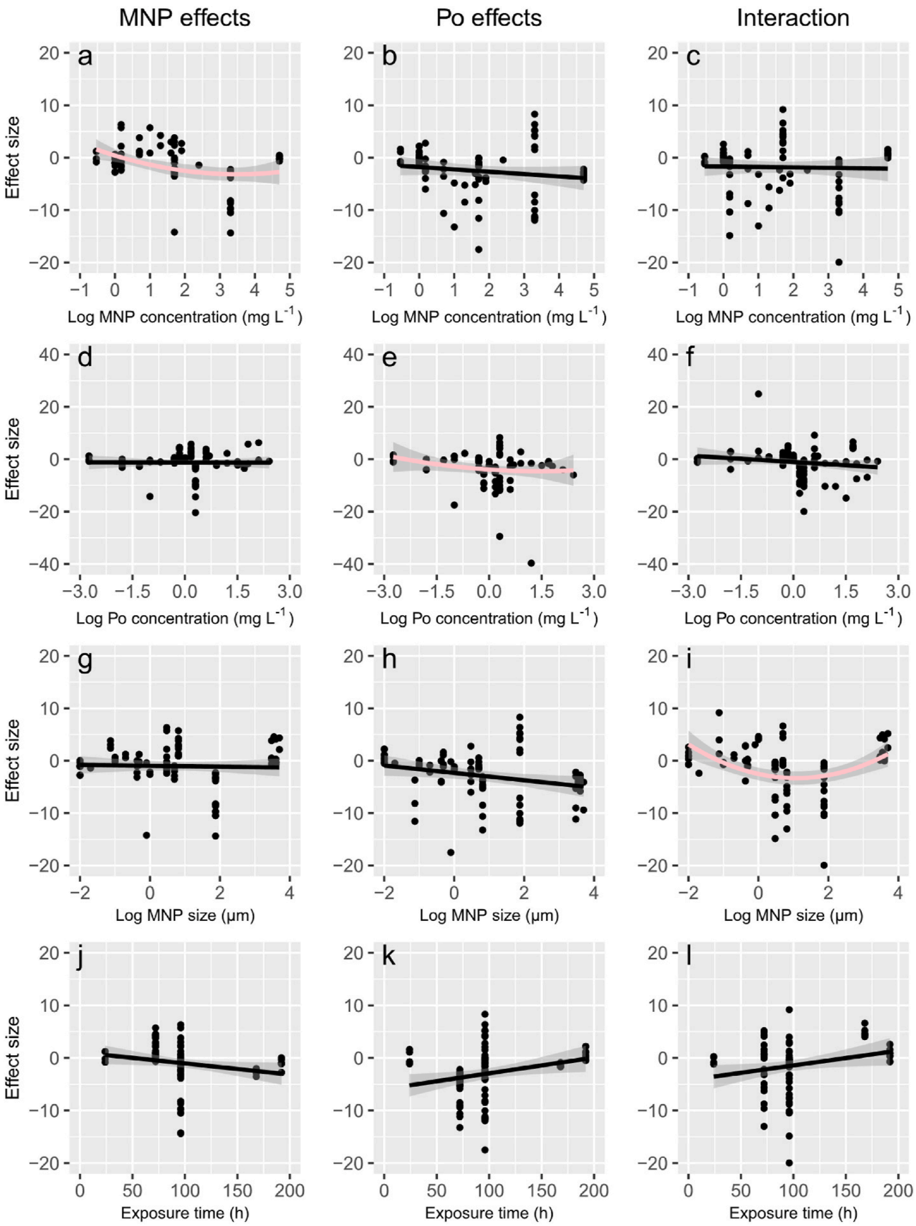


Fig. 8. The effect of MNP concentration (mg L^{-1}) (a–c), pollutant concentration (mg L^{-1}) (d–f), MNP size (μm) (g–i) and exposure time (hour) (j–l) on the effect size (Hedge's *d*) of micro- and nanoplastics (MNPs), pollutants (Po) and their interaction in pigment content. The data were fitted with linear regressions or binomial regressions. Pink represents these fitted curves accord with $p < 0.05$. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

mechanisms. In addition, we did not observe a changeable trend for exposure time in pigment content when exposure to MNPs, Po alone or their combined effect (Fig. 8j-l & Table S9).

4. Conclusions and future research needs

This study is the first to systematically analyse the interaction between MNPs and Po on physiological performance of microalgae using meta-analysis. The findings demonstrate that the overall negative effects of MNPs or Po alone disappear when they act together, and even turn to positive effects, suggesting antagonistic effects between MNPs and Po when acting on microalgae. This may be caused, to a large extent, by the adsorption of Po to MNPs. Therefore, if there are techniques that can capture MNPs effectively from waters, they can also remove a large number of Po adsorbed by MNPs at the same time. It is worth noting that the overall antagonistic effect between MNPs and Po does not mean that the input of MNPs and Po into aquatic environments can be ignored because there are still many studies showing substantial additive, and a fraction of synergistic, effects of MNPs and Po on microalgae. There are actually more studies showing additive effects although the smaller proportion of antagonistic interactions outweigh additive actions based on the mean interaction effect sizes (Hedge's *d*) and variances. Furthermore, MNPs could act as vectors or carriers to concentrate and transport biological and chemical pollutants, which can be ingested by microalgae in small MNPs size and more easily for aquatic animals, aggravating the toxicity of Po. In terms of future research needs, we put forward some suggestions.

4.1. Experiment design and date presentation

Full factorial experimental design is recommended. The data of Control, micro-/nanoplastics alone (MNPs), conventional pollutants alone (Po), and a mixture of micro-/nanoplastics and conventional pollutants (MNPs + Po) are needed for satisfying the requirement of meta-analysis. Raw data is encouraged to be deposited in a publicly accessible repository so that they can be reused easily.

4.2. Environmental reality

It is suggested that the setting of concentrations and characteristics of MNPs and Po should be taken into account in an environmentally real manner. This means that research should consider the real-world levels and behaviours of these substances within natural environments, mirroring their actual occurrences and impacts on ecosystems. By doing so, the results derived from such experiments will have higher ecological validity and applicability, providing a more accurate understanding of the ongoing environmental challenges related to MNPs and Po.

4.3. Noteworthy pollutants

Other common conventional pollutants, including mercury, chromium, phthalates, perfluorochemicals, etc., should be adopted and applied to coupling researches with MNPs. Currently, these pollutants are significantly underrepresented in the previous studies. Mercury and chromium, two toxic heavy metals known for its harmful effects on the environment and human health, require further examination with MNPs. Additionally, phthalates and perfluorochemicals, widely used in various industries and known for their potential endocrine-disrupting properties, must be included in future studies. Phthalates, commonly found in plastics, and perfluorochemicals, often used in non-stick cookware and stain-resistant fabrics, are ubiquitous in our daily lives. These pollutants' interaction with MNPs could provide valuable insights into the combined impacts of emerging pollutants on aquatic ecosystems.

4.4. Noteworthy marine algae

There was a scarcity of studies involving dinoflagellates and especially macroalgae, which deserves our attention. For instance, dinoflagellates are known for their ability to form harmful algal blooms (HABs), which can lead to significant economic losses and potentially devastating impacts on human health (Zhang et al., 2023). Likewise, macroalgae play an important in carbon sequestration, nutrient cycling, providing food and shelter for aquatic life (Gao et al., 2021a). Therefore, studies on these algae exposed to MNPs and Po are particularly necessary.

4.5. The mechanism of combined effects of MNPs and Po

The mechanisms involved in the combined effects of MNPs and Po present an intricate area of investigation that warrants further exploration. This refers to the interaction among MNPs, Po and microalgae and the acclimation of microalgae with exposure time. Another critical aspect of this exploration should be understanding the molecular responses of microalgae to the combination of MNPs and Po. These could include any potential damage or alteration to cell structures and functions, as well as the examination of cellular responses such as apoptosis.

Author contributions

GG conceptualized the study. JG, YF, CG and SX acquired the data. JG, PJ and YF performed the statistical analysis. JG and PJ generated the figures. JG, GG, SX and JB drafted the manuscript, and all authors discussed and edited the manuscript.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Acknowledgements

This work was supported by the National Natural Science Foundation of China (42076154), the Natural Science Foundation of Fujian Province of China (2022J01026, 2021J01026), the Marine Economic Development Special Fund Project of Fujian Province of China (FJHJF-L-2022-11), and the Zhejiang Provincial Natural Science Foundation of China (No. LZ21C030001).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2023.123127.

References

Alimi, O.S., Farner Budarz, J., Hernandez, L.M., Tufenkji, N., 2018. Microplastics and nanoplastics in aquatic environments: aggregation, deposition, and enhanced contaminant transport. Environ. Sci. Technol. 52 (4), 1704–1724. https://doi.org/10.1021/acs.est.7b05559.

Barnes, D.K., Galgani, F., Thompson, R.C., Barlaz, M., 2009. Accumulation and fragmentation of plastic debris in global environments. Phil. Trans. R. Soc. B 364 (1526), 1985–1998. https://doi.org/10.1098/rstb.2008.0205.

Batool, S., Ab Rashid, S., Maah, M.J., Sarfraz, M., Ashraf, M.A., 2016. Geographical distribution of persistent organic pollutants in the environment: a review. J. Environ. Biol. 37 (5), 1125–1134.

Bellingeri, A., Bergami, E., Grassi, G., Faleri, C., Redondo-Hasselerharm, P., Koelmans, A. A., Corsi, I., 2019. Combined effects of nanoplastics and copper on the freshwater alga *Raphidocelis subcapitata*. Aquat. Toxicol. 210, 179–187. https://doi.org/10.1016/j.aquatox.2019.02.022.

Bergami, E., Pugnalini, S., Vannuccini, M.L., Manfra, L., Faleri, C., Savorelli, F., Dawson, K.A., Corsi, I., 2017. Long-term toxicity of surface-charged polystyrene nanoplastics to marine planktonic species *Dunaliella tertiolecta* and *Artemia franciscan*. Aquat. Toxicol. 189, 159–169. https://doi.org/10.1016/j.aquatox.2017.06.008.

Borenstein, M., Hedges, L.V., Higgins, J.P.T., Rothstein, H.R., 2011. Introduction to Meta-Analysis. John Wiley & Sons, Chichester.

Burrows, S.D., Frustaci, S., Thomas, K.V., Galloway, T., 2020. Expanding exploration of dynamic microplastic surface characteristics and interactions. TrAC, Trends Anal. Chem. 130 https://doi.org/10.1016/j.trac.2020.115993.

Cao, J., Liao, Y., Yang, W., Jiang, X., Li, M., 2022. Enhanced microalgal toxicity due to polystyrene nanoplastics and cadmium co-exposure: from the perspective of physiological and metabolomic profiles. J. Hazard Mater. 427, 127937 https://doi.org/10.1016/j.jhazmat.2021.127937.

Caroppo, C., Pagliara, P., 2022. Microalgae: a promising future. Microorganisms 10 (8). https://doi.org/10.3390/microorganisms10081488.

Chae, Y., Kim, D., An, Y.-J., 2019. Effects of micro-sized polyethylene spheres on the marine microalga Dunaliella salina: focusing on the algal cell to plastic particle size ratio. Aquat. Toxicol. 216 https://doi.org/10.1016/j.aquatox.2019.105296.

Chen, S., Wang, L., Feng, W., Yuan, M., Li, J., Xu, H., Zheng, X., Zhang, W., 2020. Sulfonamides-induced oxidative stress in freshwater microalga *Chlorella vulgaris*: evaluation of growth, photosynthesis, antioxidants, ultrastructure, and nucleic acids. Sci. Rep. 10 (1), 8243. https://doi.org/10.1038/s41598-020-65219-2.

Crain, C.M., Kroeker, K., Halpern, B.S., 2008. Interactive and cumulative effects of multiple human stressors in marine systems. Ecol. Lett. 11 (12), 1304–1315.

Davidson, T.M., 2012. Boring crustaceans damage polystyrene floats under docks polluting marine waters with microplastic. Mar. Pollut. Bull. 64 (9), 1821–1828. https://doi.org/10.1016/j.marpolbul.2012.06.005.

Duval, S., Tweedie, R., 2000. Trim and fill: a simple funnelplot-based method of testing and adjusting for publication bias in meta-analysis. Biometrics 56 (2), 455–463.

Feng, L.J., Shi, Y., Li, X.Y., Sun, X.D., Xiao, F., Sun, J.W., Wang, Y., Liu, X.Y., Wang, S.G., Yuan, X.Z., 2020a. Behavior of tetracycline and polystyrene nanoparticles in estuaries and their joint toxicity on marine microalgae *Skeletonema costatum*. Environ. Pollut. 263 (Pt A), 114453 https://doi.org/10.1016/j.envpol.2020.114453.

Feng, Z., Zhang, T., Li, Y., He, X., Wang, R., Xu, J., Gao, G., 2019. The accumulation of microplastics in fish from an important fish farm and mariculture area, Haizhou Bay, China. Sci. Total Environ. 696 https://doi.org/10.1016/j.scitotenv.2019.133948.

Feng, Z., Zhang, T., Wang, J., Huang, W., Wang, R., Xu, J., Fu, G., Gao, G., 2020b. Spatio-temporal features of microplastics pollution in macroalgae growing in an important mariculture area, China. Sci. Total Environ. 719, 137490 https://doi.org/10.1016/j.scitotenv.2020.137490.

Fonte, E., Ferreira, P., Guilhermino, L., 2016. Temperature rise and microplastics interact with the toxicity of the antibiotic cefalexin to juveniles of the common goby (*Pomatoschistus microps*): post-exposure predatory behaviour, acetylcholinesterase activity and lipid peroxidation. Aquat. Toxicol. 180, 173–185. https://doi.org/10.1016/j.aquatox.2016.09.015.

Fu, D., Zhang, Q., Fan, Z., Qi, H., Wang, Z., Peng, L., 2019. Aged microplastics polyvinyl chloride interact with copper and cause oxidative stress towards microalgae *Chlorella vulgaris*. Aquat. Toxicol. 216, 105319 https://doi.org/10.1016/j.aquatox.2019.105319.

Gao, G., Gao, L., Jiang, M., Jian, A., He, L., 2021a. The potential of seaweed cultivation to achieve carbon neutrality and mitigate deoxygenation and eutrophication. Environ. Res. Lett. 17 (1) https://doi.org/10.1088/1748-9326/ac3fd9.

Gao, G., Zhao, X., Jin, P., Gao, K., Beardall, J., 2021b. Current understanding and challenges for aquatic primary producers in a world with rising micro- and nano-plastic levels. J. Hazard Mater. 406, 124685 https://doi.org/10.1016/j.jhazmat.2020.124685.

Gao, Q.T., Wong, Y.S., Tam, N.F.Y., 2017. Antioxidant responses of different microalgal species to nonylphenol-induced oxidative stress. J. Appl. Phycol. 29 (3), 1317–1329. https://doi.org/10.1007/s10811-017-1065-y.

Gao, Z.-y., Wang, S.-c., Zhang, Y.-x., Liu, F.-f., 2022. Single and combined toxicity of polystyrene nanoplastics and copper on Platymonas helgolandica var. tsingtaoensis: perspectives from growth inhibition, chlorophyll content and oxidative stress. Sci. Total Environ. 829 https://doi.org/10.1016/j.scitotenv.2022.154571.

Gonzalez-Pleiter, M., Pedrouzo-Rodriguez, A., Verdu, I., Leganes, F., Marco, E., Rosal, R., Fernandez-Pinas, F., 2021. Microplastics as vectors of the antibiotics azithromycin and clarithromycin: effects towards freshwater microalgae. Chemosphere 268, 128824. https://doi.org/10.1016/j.chemosphere.2020.128824.

Gu, P., Li, Q., Zhang, W., Zheng, Z., Luo, X., 2020. Effects of different metal ions (Ca, Cu, Pb, Cd) on formation of cyanobacterial blooms. Ecotoxicol. Environ. Saf. 189, 109976 https://doi.org/10.1016/j.ecoenv.2019.109976.

Gunasekaran, D., Chandrasekaran, N., Jenkins, D., Mukherjee, A., 2020. Plain polystyrene microplastics reduce the toxic effects of ZnO particles on marine microalgae *Dunaliella salina*. J. Environ. Chem. Eng. 8 (5) https://doi.org/10.1016/j.jece.2020.104250.

Gurevitch, J., Morrow, L.L., Wallace, A., Walsh, J.S., 1992. A meta-analysis of competition in field experiments. Am. Nat. 140 (4), 539–572.

Gurevitch, J., Hedges, L.V., 1993. Meta-analysis: combining the results of independent experiments. In: Scheiner, S., Gurevitch, J. (Eds.), Design and Analysis of Ecological Experiments. Chapman and Hall, New York, pp. 347–370.

Gurevitch, J., Morrison, J.A., Hedges, L.V., 2000. The interaction between competition and predation: a meta-analysis of field experiments. Am. Nat. 155 (4), 435–453. https://doi.org/10.1086/303337.

Hartmann, P., Bechet, Q., Bernard, O., 2014. The effect of photosynthesis time scales on microalgae productivity. Bioproc. Biosyst. Eng. 37 (1), 17–25. https://doi.org/10.1007/s00449-013-1031-2.

Hedges, L.V., 1984. Advances in statistical methods for meta-analysis. N. Dir. Progr. Eval. 1984 (24), 25–42. https://doi.org/10.1002/ev.1376.

Hedges, L.V., Gurevitch, J., Curtis, P.S., 1999. The meta-analysis of response ratios in experimental ecology. Ecology 80 (4), 1150–1156. https://doi.org/10.1890/0012-9658(1999)080[1150:Tmaorr]2.0.Co;2.

Holmes, L.A., Turner, A., Thompson, R.C., 2014. Interactions between trace metals and plastic production pellets under estuarine conditions. Mar. Chem. 167, 25–32. https://doi.org/10.1016/j.marchem.2014.06.001.

Huang, B., Wei, Z.B., Yang, L.Y., Pan, K., Miao, A.J., 2019. Combined toxicity of silver nanoparticles with hematite or plastic nanoparticles toward two freshwater algae. Environ. Sci. Technol. 53 (7), 3871–3879. https://doi.org/10.1021/acs.est.8b07001.

Jin, P., Overmans, S., Duarte, C.M., Agusti, S., 2019. Increasing temperature within thermal limits compensates negative ultraviolet-B radiation effects in terrestrial and aquatic organisms. Global Ecol. Biogeogr. 28 (11), 1695–1711. https://doi.org/10.1111/geb.12973.

Jin, P., Zhang, J., Wan, J., Overmans, S., Gao, G., Ye, M., Dai, X., Zhao, J., Xiao, M., Xia, J., 2021. The combined effects of ocean acidification and heavy metals on marine organisms: a Meta-Analysis. Front. Mar. Sci. 8 https://doi.org/10.3389/fmars.2021.801889.

Jin, P., Wan, J., Zhang, J., Overmans, S., Xiao, M., Ye, M., Dai, X., Zhao, J., Gao, K., Xia, J., 2022. Additive impacts of ocean acidification and ambient ultraviolet radiation threaten calcifying marine primary producers. Sci. Total Environ. 818, 151782 https://doi.org/10.1016/j.scitotenv.2021.151782.

Kroeker, K.J., Kordas, R.L., Crim, R., Hendriks, I.E., Ramajo, L., Singh, G.S., Duarte, C.M., Gattuso, J.P., 2013. Impacts of ocean acidification on marine organisms: quantifying sensitivities and interaction with warming. Global Change Biol. 19 (6), 1884–1896. https://doi.org/10.1111/gcb.12179.

Li, J., Mao, S., Ye, Y., Lu, J., Jing, F., Guo, Y., Liu, H., Wang, P., Ma, W., Qi, P., Zheng, J., Qu, C., 2021. Micro-polyethylene particles reduce the toxicity of nano zinc oxide in marine microalgae by adsorption. Environ. Pollut. 290, 118042 https://doi.org/10.1016/j.envpol.2021.118042.

Li, P., Liu, J., Zhang, H., 2022. Insights into the interaction of microplastic with silver nanoparticles in natural surface water. Sci. Total Environ. 805, 150315 https://doi.org/10.1016/j.scitotenv.2021.150315.

Liao, Y., Jiang, X., Xiao, Y., Li, M., 2020. Exposure of microalgae *Euglena gracilis* to polystyrene microbeads and cadmium: perspective from the physiological and transcriptional responses. Aquat. Toxicol. 228, 105650 https://doi.org/10.1016/j.aquatox.2020.105650.

Liu, P., Zhan, X., Wu, X., Li, J., Wang, H., Gao, S., 2020. Effect of weathering on environmental behavior of microplastics: properties, sorption and potential risks. Chemosphere 242, 125193. https://doi.org/10.1016/j.chemosphere.2019.125193.

Liu, Q., Wu, H., Chen, J., Guo, B., Zhao, X., Lin, H., Li, W., Zhao, X., Lv, S., Huang, C., 2022. Adsorption mechanism of trace heavy metals on microplastics and simulating their effect on microalgae in river. Environ. Res. 214 (Pt 1), 113777 https://doi.org/10.1016/j.envres.2022.113777.

Lozano, P., Trombini, C., Crespo, E., Blasco, J., Moreno-Garrido, I., 2014. ROI-scavenging enzyme activities as toxicity biomarkers in three species of marine microalgae exposed to model contaminants (copper, Irgarol and atrazine). Ecotoxicol. Environ. Saf. 104, 294–301. https://doi.org/10.1016/j.ecoenv.2014.03.021.

Mao, Y., Ai, H., Chen, Y., Zhang, Z., Zeng, P., Kang, L., Li, W., Gu, W., He, Q., Li, H., 2018. Phytoplankton response to polystyrene microplastics: perspective from an entire growth period. Chemosphere 208, 59–68. https://doi.org/10.1016/j.chemosphere.2018.05.170.

Margalef, R., 1978. Life-forms of phytoplankton as survival alternatives in an unstable environment. Oceanol. Acta 1 (4), 493–509.

Miguez, L., Esperanza, M., Seoane, M., Cid, A., 2021. Assessment of cytotoxicity biomarkers on the microalga *Chlamydomonas reinhardtii* exposed to emerging and priority pollutants. Ecotoxicol. Environ. Saf. 208, 111646 https://doi.org/10.1016/j.ecoenv.2020.111646.

Niu, Z., Vandegehuchte, M.B., Catarino, A.I., Everaert, G., 2021. Environmentally relevant concentrations and sizes of microplastic do not impede marine diatom growth. J. Hazard Mater. 409, 124460 https://doi.org/10.1016/j.jhazmat.2020.124460.

O'Dea, R.E., Lagisz, M., Jennions, M.D., Koricheva, J., Noble, D.W.A., Parker, T.H., Gurevitch, J., Page, M.J., Stewart, G., Moher, D., Nakagawa, S., 2021. Preferred reporting items for systematic reviews and meta-analyses in ecology and evolutionary biology: a PRISMA extension. Biol. Rev. Camb. Phil. Soc. 96 (5), 1695–1722. https://doi.org/10.1111/brv.12721.

Pal, A., Gin, K.Y., Lin, A.Y., Reinhard, M., 2010. Impacts of emerging organic contaminants on freshwater resources: review of recent occurrences, sources, fate and effects. Sci. Total Environ. 408 (24), 6062–6069. https://doi.org/10.1016/j.scitotenv.2010.09.026.

Petrie, B., Barden, R., Kasprzyk-Hordern, B., 2015. A review on emerging contaminants in wastewaters and the environment: current knowledge, understudied areas and recommendations for future monitoring. Water Res. 72, 3–27. https://doi.org/10.1016/j.watres.2014.08.053.

Phuong, N.N., Zalouk-Vergnoux, A., Poirier, L., Kamari, A., Chatel, A., Mouneyrac, C., Lagarde, F., 2016. Is there any consistency between the microplastics found in the field and those used in laboratory experiments? Environ. Pollut. 211, 111–123. https://doi.org/10.1016/j.envpol.2015.12.035.

Piggott, J.J., Townsend, C.R., Matthaei, C.D., 2015. Reconceptualizing synergism and antagonism among multiple stressors. Ecol. Evol. 5 (7), 1538–1547. https://doi.org/10.1002/ece3.1465.

Podbielska, M., Szpyrka, E., 2023. Microplastics - an emerging contaminants for algae. Critical review and perspectives. Sci. Total Environ. 885, 163842 https://doi.org/10.1016/j.scitotenv.2023.163842.

Prata, J.C., Lavorante, B., Mdc, B.S.M.M., Guilhermino, L., 2018. Influence of microplastics on the toxicity of the pharmaceuticals procainamide and doxycycline on the marine microalgae *Tetraselmis chuii*. Aquat. Toxicol. 197, 143–152. https://doi.org/10.1016/j.aquatox.2018.02.015.

Raju, P., Santhanam, P., Pandian, S.S., Divya, M., Arunkrishnan, A., Devi, K.N., Ananth, S., Roopavathy, J., Perumal, P., 2021. Impact of polystyrene microplastics on major marine primary (phytoplankton) and secondary producers (copepod). Arch. Microbiol. 204 (1), 84. https://doi.org/10.1007/s00203-021-02697-6.

Rosenthal, R., 1979. The file drawer problem and tolerance for null results. Psychol. Bull. 86 (3), 638–641. https://doi.org/10.1037//0033-2909.86.3.638.

S′anchez-Fortún, A., D'Ors, A., Fajardo, C., Martin, C., Nande, M., Mengs, G., Costa, G., Martin, M., Sanchez-Fortun, S., 2022. Influence of contaminant-spiked polyethylene-type microplastics on the growth and primary production of the freshwater phytoplankton species Scenedesmus armatus and Microcystis aeruginosa. Environ. Exp. Bot. 203 https://doi.org/10.1016/j.envexpbot.2022.105061.

Santana-Viera, S., Montesdeoca-Esponda, S., Torres-Padron, M.E., Sosa-Ferrera, Z., Santana-Rodriguez, J.J., 2021. An assessment of the concentration of pharmaceuticals adsorbed on microplastics. Chemosphere 266, 129007. https://doi.org/10.1016/j.chemosphere.2020.129007.

Shahul Hamid, F., Bhatti, M.S., Anuar, N., Anuar, N., Mohan, P., Periathamby, A., 2018. Worldwide distribution and abundance of microplastic: how dire is the situation? Waste Manag. Res. 36 (10), 873–897. https://doi.org/10.1177/0734242X18785730.

Song, C., Liu, Z., Wang, C., Li, S., Kitamura, Y., 2020. Different interaction performance between microplastics and microalgae: the bio-elimination potential of *Chlorella* sp. L38 and *Phaeodactylum tricornutum* MASCC-0025. Sci. Total Environ. 723, 138146 https://doi.org/10.1016/j.scitotenv.2020.138146.

Sun, L., Sun, S., Bai, M., Wang, Z., Zhao, Y., Huang, Q., Hu, C., Li, X., 2021. Internalization of polystyrene microplastics in Euglena gracilis and its effects on the protozoan photosynthesis and motility. Aquat. Toxicol. 236, 105840 https://doi.org/10.1016/j.aquatox.2021.105840.

Teuten, E.L., Saquing, J.M., Knappe, D.R., Barlaz, M.A., Jonsson, S., Bjorn, A., Rowland, S.J., Thompson, R.C., Galloway, T.S., Yamashita, R., Ochi, D., Watanuki, Y., Moore, C., Viet, P.H., Tana, T.S., Prudente, M., Boonyatumanond, R., Zakaria, M.P., Akkhavong, K., Ogata, Y., Hirai, H., Iwasa, S., Mizukawa, K., Hagino, Y., Imamura, A., Saha, M., Takada, H., 2009. Transport and release of chemicals from plastics to the environment and to wildlife. Phil. Trans. R. Soc. B 364 (1526), 2027–2045. https://doi.org/10.1098/rstb.2008.0284.

Thiagarajan, V., Iswarya, V., P Aj, Seenivasan, R., Chandrasekaran, N., Mukherjee, A., 2019. Influence of differently functionalized polystyrene microplastics on the toxic effects of P25 TiO2 NPs towards marine algae *Chlorella* sp. Aquat. Toxicol. 207, 208–216. https://doi.org/10.1016/j.aquatox.2018.12.014.

Thiagarajan, V., Alex, S.A., Seenivasan, R., Chandrasekaran, N., Mukherjee, A., 2021. Toxicity evaluation of nano-TiO2 in the presence of functionalized microplastics at two trophic levels: algae and crustaceans. Sci. Total Environ. 784, 147262 https://doi.org/10.1016/j.scitotenv.2021.147262.

Tunali, M., Uzoefuna, E.N., Tunali, M.M., Yenigun, O., 2020. Effect of microplastics and microplastic-metal combinations on growth and chlorophyll a concentration of *Chlorella vulgaris*. Sci. Total Environ. 743, 140479 https://doi.org/10.1016/j.scitotenv.2020.140479.

Viechtbauer, W., 2010. Conducting meta-analyses in R with the metafor package. J. Stat. Software 36 (3), 1–48. https://doi.org/10.18637/jss.v036.i03.

Vockenberg, T., Wichard, T., Ueberschaar, N., Franke, M., Stelter, M., Braeutigam, P., 2020. The sorption behaviour of amine micropollutants on polyethylene microplastics - impact of aging and interactions with green seaweed. Environ Sci Process Impacts 22 (8), 1678–1687. https://doi.org/10.1039/d0em00119h.

Wang, F., Gao, J., Zhai, W., Liu, D., Zhou, Z., Wang, P., 2020a. The influence of polyethylene microplastics on pesticide residue and degradation in the aquatic environment. J. Hazard Mater. 394, 122517 https://doi.org/10.1016/j.jhazmat.2020.122517.

Wang, F., Wang, B., Qu, H., Zhao, W., Duan, L., Zhang, Y., Zhou, Y., Yu, G., 2020b. The influence of nanoplastics on the toxic effects, bioaccumulation, biodegradation and enantioselectivity of ibuprofen in freshwater algae *Chlorella pyrenoidosa*. Environ. Pollut. 263 (Pt B), 114593 https://doi.org/10.1016/j.envpol.2020.114593.

Wang, Q., Zhang, Y., Wangjin, X., Wang, Y., Meng, G., Chen, Y., 2020c. The adsorption behavior of metals in aqueous solution by microplastics effected by UV radiation. J. Environ. Sci. 87, 272–280. https://doi.org/10.1016/j.jes.2019.07.006.

Wang, Q., Wang, J., Chen, H., Zhang, Y., 2023. Toxicity effects of microplastics and nanoplastics with cadmium on the alga Microcystis aeruginosa. Environ. Sci. Pollut. Res. Int. 30 (7), 17360–17373. https://doi.org/10.1007/s11356-022-23278-0.

Wang, Z., Fu, D., Gao, L., Qi, H., Su, Y., Peng, L., 2021. Aged microplastics decrease the bioavailability of coexisting heavy metals to microalga *Chlorella vulgaris*. Ecotoxicol. Environ. Saf. 217, 112199 https://doi.org/10.1016/j.ecoenv.2021.112199.

Yang, W., Gao, X., Wu, Y., Wan, L., Lu, C., Huang, J., Chen, H., Yang, Y., Ding, H., Zhang, W., 2021. Chemical- and species-specific toxicity of nonylphenol and octylphenol to microalgae *Chlorella pyrenoidosa* and *Scenedesmus obliquus*. Environ. Toxicol. Pharmacol. 81, 103517 https://doi.org/10.1016/j.etap.2020.103517.

Yi, X., Chi, T., Li, Z., Wang, J., Yu, M., Wu, M., Zhou, H., 2019. Combined effect of polystyrene plastics and triphenyltin chloride on the green algae *Chlorella pyrenoidosa*. Environ. Sci. Pollut. Res. 26 (15), 15011–15018. https://doi.org/10.1007/s11356-019-04865-0.

You, X., Cao, X., Zhang, X., Guo, J., Sun, W., 2021. Unraveling individual and combined toxicity of nano/microplastics and ciprofloxacin to *Synechocystis* sp. at the cellular and molecular levels. Environ. Int. 157, 106842 https://doi.org/10.1016/j.envint.2021.106842.

Zhang, Q., Qu, Q., Lu, T., Ke, M., Zhu, Y., Zhang, M., Zhang, Z., Du, B., Pan, X., Sun, L., Qian, H., 2018. The combined toxicity effect of nanoplastics and glyphosate on *Microcystis aeruginosa* growth. Environ. Pollut. 243 (Pt B), 1106–1112. https://doi.org/10.1016/j.envpol.2018.09.073.

Zhang, X., Liu, L., Chen, X., Li, J., Chen, J., Wang, H., 2022. The fate and risk of microplastic and antibiotic sulfamethoxazole coexisting in the environment. Environ. Geochem. Health. https://doi.org/10.1007/s10653-022-01385-8.

Zhang, Y., Lu, X., Wang, N., Xin, M., Geng, S., Jia, J., Meng, Q., 2016. Heavy metals in aquatic organisms of different trophic levels and their potential human health risk in Bohai Bay, China. Environ. Sci. Pollut. Res. 23 (17), 17801–17810. https://doi.org/10.1007/s11356-016-6948-y.

Zhang, Y., Whalen, J.K., Cai, C., Shan, K., Zhou, H., 2023. Harmful cyanobacteria-diatom/dinoflagellate blooms and their cyanotoxins in freshwaters: a nonnegligible chronic health and ecological hazard. Water Res. 233 https://doi.org/10.1016/j.watres.2023.119807.

Zhao, T., Tan, L., Huang, W., Wang, J., 2019a. The interactions between micro polyvinyl chloride (mPVC) and marine dinoflagellate *Karenia mikimotoi* the inhibition of growth, chlorophyll and photosynthetic efficiency. Environ. Pollut. 247, 883–889. https://doi.org/10.1016/j.envpol.2019.01.114.

Zhao, T., Tan, L., Huang, W., Wang, J., 2019b. The interactions between micro polyvinyl chloride (mPVC) and marine dinoflagellate *Karenia mikimotoi*: the inhibition of growth, chlorophyll and photosynthetic efficiency. Environ. Pollut. 247, 883–889. https://doi.org/10.1016/j.envpol.2019.01.114.