Electronic Supplementary Material (ESI) for Environmental Science: Nano.

Does polystyrene nanoplastics aggravate the toxicity of single contaminants (okadaic acid)? Using AGS cells as a biological model

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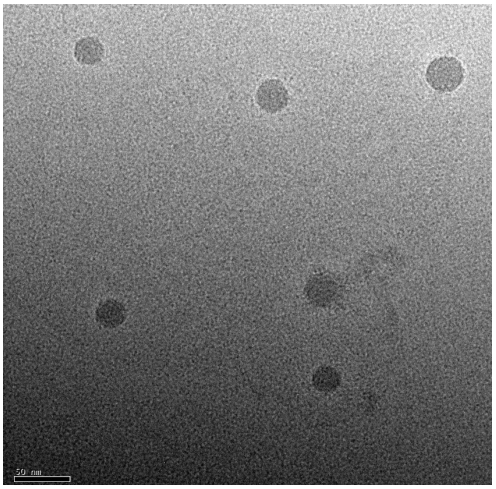

Fig. S1 The TEM images of PS.

Table S1 Characterization of PS NPs.

Characterization	Medium	Plain PS	Fluorescent PS
Size	H_2O	21.4±11.0	42.23±0.49
(nm)	F12	23.76±0.42	40.69±0.72
ζ-potentials	H_2O	-27.71±4.61	-35.93±3.13
(mV)	F12	-25.1±3.99	-8.06±0.51
PDI	H_2O	0.176±0.015	0.179±0.016
	F12	0.264±0.019	0.229±0.012

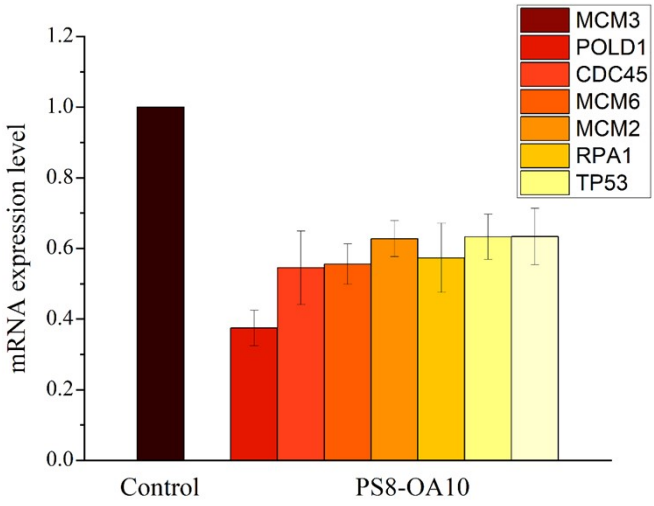

Fig. S2 Relative of expressions of mRNA by q-PCR.

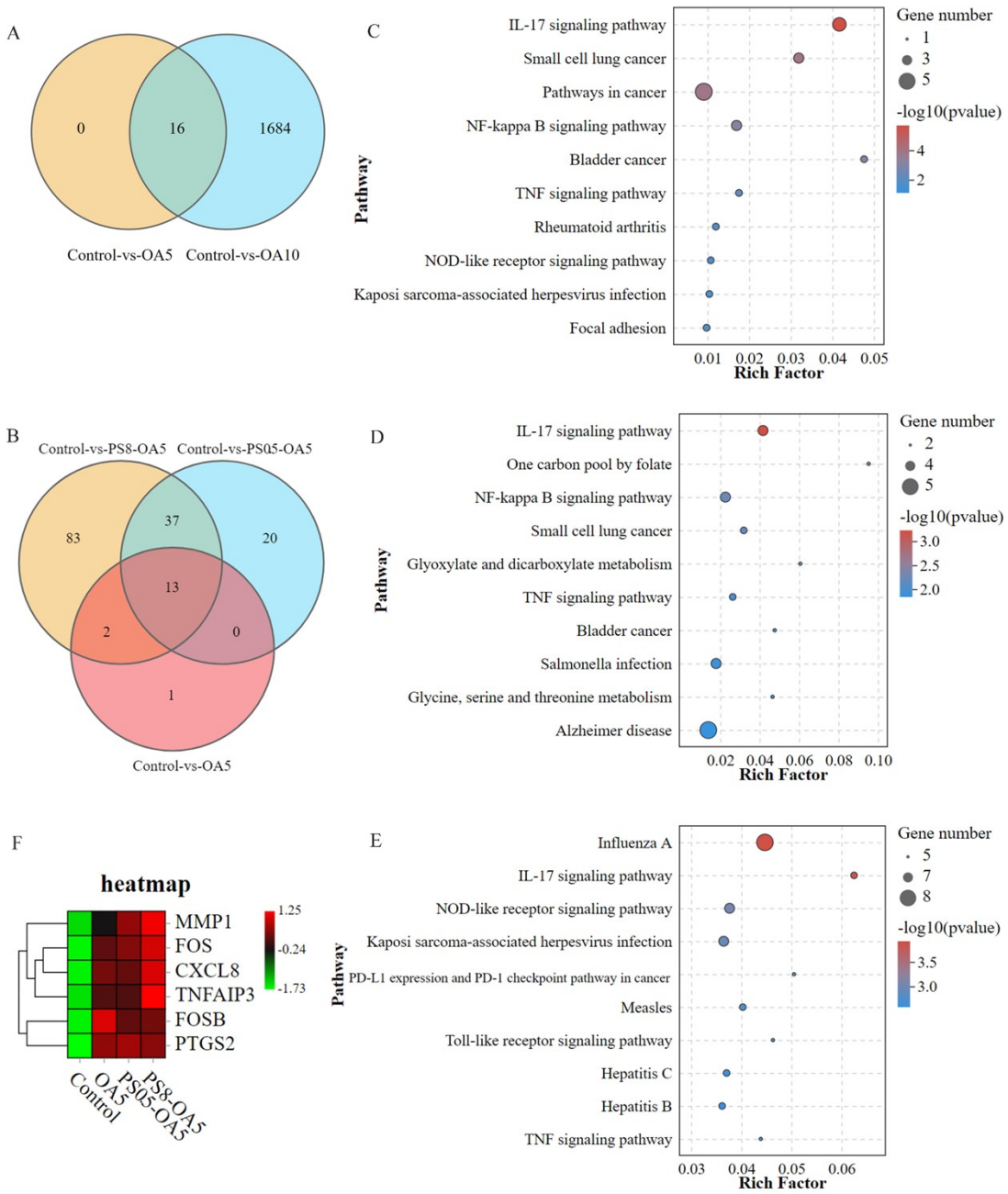

Fig. S3 Genes expression in AGS cells after exposures of OA5, OA10, PS0.5-OA5 and PS8-OA5. (A) Venn diagrams of DEGs about OA5 and OA10; (B) Venn diagrams of DEGs about OA5, PS0.5-OA5 and PS8-OA5; (C-E) Enrichment of KEGG pathways in OA5, PS0.5-OA5 and PS8-OA5; (F) Heat map of DEGs about IL-17 signaling pathway.

Table S2 Primer sequences used in this study.

Oligo Name	Sequence (5’ to 3’)
MCM3 forward	gaa ggc gag gaa tgt tgg
MCM3 reverse	gtg gga tgg gaa gta ggg
CDC45 forward	ggg agt ggt ata ata ggg
CDC45 reverse	agt gag ttc cag gac agc
MCM6 forward	tgt cag tgg tgt tga tgg ata tg

MCM6 reverse	gct gtc tgt tcc tca tct ctg
MCM2 forward	cgg gac tat gtg atc gaa gac
MCM2 reverse	acc tgc tct gcc act aac tgc t
FEN1 forward	gcg tct gga atg taa gga agt g
FEN1 reverse	gtg cag ggt ccg agg t
POLD1 forward	caa cct ggt cac tgc ctc ac
POLD1 reverse	gtc ccg ctt cct cat cct ct
RPA1 forward	aag tgg aga cct aca acg ac
RPA1 reverse	aca acc acc tga gcg tat
TP53 forward	cct cac cat cat cac act gg
TP53 reserve	tct gag tca ggc cct tct gt
βactin forward	agc gag cat ccc cca aag tt
βactin reverse	ggg cac gaa ggc tca tca tt

Table S3 DEGs of PS0.5 and PS8

Group	DEGs
PS0.5	*OAS2,XAF1,SPATA1,DDX60L,MX2,H2C12,AC116366.2,BST2,HEPHL1*
PS8	*LAMP3, RAB4B-EGLN2, HEPHL1*

Table S4 Enrichment of KEGG pathways in OA10, PS0.5-OA10 and PS8-OA10

	OA10		PS0.5-OA10		PS8-OA10	
	Pathway	P	Pathway	P	Pathway	P
1	DNA replication	0.000	DNA replication	0.000	DNA replication	0.000
2	Fanconi anemia pathway	0.000	Bladder cancer	0.000	MicroRNAs in cancer	0.000
3	TNF signaling pathway	0.000	MAPK signaling pathway	0.000	Bladder cancer	0.000
4	MicroRNAs in cancer	0.000	TNF signaling pathway	0.000	beta-Alanine metabolism	0.000
5	Cell cycle	0.000	Cell cycle	0.000	TNF signaling pathway	0.000
6	MAPK signaling pathway	0.001	Osteoclast differentiation	0.001	Cell cycle	0.000
7	Mismatch repair	0.001	Glycine, serine and threonine metabolism	0.001	Mismatch repair	0.000
8	Bladder cancer	0.001	Small cell lung cancer	0.001	ECM-receptor interaction	0.000
9	Focal adhesion	0.001	Nucleotide excision repair	0.001	Fanconi anemia pathway	0.000
10	Small cell lung cancer	0.001	beta-Alanine metabolism	0.002	Small cell lung cancer	0.000

11	beta-Alanine metabolism	0.001	Mismatch repair	0.002	Glycine, serine and threonine metabolism	0.000
12	Central carbon metabolism in cancer	0.002	MicroRNAs in cancer	0.003	Pathways in cancer	0.000
13	Nucleotide excision repair	0.002	FoxO signaling pathway	0.003	MAPK signaling pathway	0.000
14	Homologous recombination	0.002	Cellular senescence	0.003	Focal adhesion	0.000
15	FoxO signaling pathway	0.002	Hepatitis B	0.003	Nucleotide excision repair	0.001
16	Cellular senescence	0.002	Focal adhesion	0.004	Central carbon metabolism in cancer	0.001
17	Hepatitis B	0.002	Base excision repair	0.005	Osteoclast differentiation	0.002
18	Fructose and mannose metabolism	0.003	Complement and coagulation cascades	0.006	Fatty acid metabolism	0.002
19	Osteoclast differentiation	0.003	Gastric cancer	0.007	AGE-RAGE signaling pathway in diabetic complications	0.002
20	Glycine, serine and threonine metabolism	0.003	IL-17 signaling pathway	0.009	FoxO signaling pathway	0.004
21	Base excision repair	0.004	Central carbon metabolism in cancer	0.010	Histidine metabolism	0.005
22	Toxoplasmosis	0.004	Toxoplasmosis	0.013	Base excision repair	0.005
23	Pentose phosphate pathway	0.004	ECM-receptor interaction	0.015	Endocrine resistance	0.006
24	Pathways in cancer	0.005	Endocrine resistance	0.017	Human papillomavirus infection	0.006
25	AGE-RAGE signaling pathway in diabetic complications	0.008	Pathways in cancer	0.017	Homologous recombination	0.009
26	Sphingolipid metabolism	0.009	Phenylalanine metabolism	0.018	Fluid shear stress and atherosclerosis	0.010
27	Metabolic pathways	0.010	Non-small cell lung cancer	0.019	Parathyroid hormone synthesis, secretion and action	0.010
28	Valine, leucine	0.010	Fatty acid	0.019	Cushing syndrome	0.012

	and isoleucine degradation		metabolism			
29	Amino sugar and nucleotide sugar metabolism	0.012	Relaxin signaling pathway	0.023	IL-17 signaling pathway	0.013
30	ECM-receptor interaction	0.012	Parathyroid hormone synthesis, secretion and action	0.023	Valine, leucine and isoleucine degradation	0.014
31	Influenza A	0.013	Hepatitis C	0.025	Hepatitis B	0.016
32	Parathyroid hormone synthesis, secretion and action	0.013	HTLV-I infection	0.026	Relaxin signaling pathway	0.019
33	Gastric cancer	0.013	Human papillomavirus infection	0.028	Pentose phosphate pathway	0.020
34	Phenylalanine metabolism	0.017	Arginine and proline metabolism	0.029	Metabolic pathways	0.020
35	Galactose metabolism	0.022	One carbon pool by folate	0.030	Phenylalanine metabolism	0.020
36	Apoptosis	0.022	Melanoma	0.031	Tight junction	0.020
37	Human papillomavirus infection	0.027	Fanconi anemia pathway	0.035	Influenza A	0.021
38	Aldosterone-regulated sodium reabsorption	0.027	Breast cancer	0.036	Gastric cancer	0.022
39	Breast cancer	0.030	Non-homologous end-joining	0.043	Glycosylphosphatidylinositol (GPI)-anchor biosynthesis	0.022
40	Fatty acid metabolism	0.030	Pancreatic cancer	0.044	Breast cancer	0.027
41	Peroxisome	0.032	Histidine metabolism	0.047	Toxoplasmosis	0.028
42	Arginine and proline metabolism	0.035			Cellular senescence	0.029
43	Endocrine	0.036			Proteoglycans in cancer	0.030

	resistance			
44	Pertussis	0.037	Carbon metabolism	0.031
45	IL-17 signaling pathway	0.039	Pancreatic cancer	0.032
46	RIG-I-like receptor signaling pathway	0.039	Sphingolipid metabolism	0.032
47	Non-small cell lung cancer	0.039	Amoebiasis	0.032
48	Hepatitis C	0.043	Apoptosis	0.034
49	Relaxin signaling pathway	0.043	Aldosterone-regulated sodium reabsorption	0.034
50	Growth hormone synthesis, secretion and action	0.044	Growth hormone synthesis, secretion and action	0.035
51	Endometrial cancer	0.045	Cortisol synthesis and secretion	0.036
52	Adherens junction	0.047	Fructose and mannose metabolism	0.036
53	Cytokine-cytokine receptor interaction	0.049	Prion diseases	0.036
54			Other glycan degradation	0.037
55			One carbon pool by folate	0.037
56			Cytokine-cytokine receptor interaction	0.038
57			Platinum drug resistance	0.039
58			Type II diabetes mellitus	0.040
59			Melanoma	0.042
60			Peroxisome	0.044
61			Arginine and proline metabolism	0.044
62			Colorectal cancer	0.046
63			Inflammatory mediator regulation of TRP channels	0.047
64			Tryptophan metabolism	0.049

Table S5 Enrichment of KEGG pathways in OA5, PS0.5-OA5 and PS8-OA5

OA5	PS0.5-OA5	PS8-OA5

	Pathway	P	Pathway	P	Pathway	P
1	IL-17 signaling pathway	0.000	IL-17 signaling pathway	0.001	Influenza A	0.000
2	Small cell lung cancer	0.000	One carbon pool by folate	0.003	IL-17 signaling pathway	0.000
3	Pathways in cancer	0.000	NF-kappa B signaling pathway	0.006	NOD-like receptor signaling pathway	0.001
4	NF-kappa B signaling pathway	0.001	Small cell lung cancer	0.007	Kaposi sarcoma-associated herpesvirus infection	0.001
5	Bladder cancer	0.001	Glyoxylate and dicarboxylate metabolism	0.008	PD-L1 expression and PD-1 checkpoint pathway in cancer	0.001
6	TNF signaling pathway	0.006	TNF signaling pathway	0.011	Measles	0.002
7	Rheumatoid arthritis	0.013	Bladder cancer	0.013	Toll-like receptor signaling pathway	0.002
8	NOD-like receptor signaling pathway	0.016	Salmonella infection	0.013	Hepatitis C	0.002
9	Kaposi sarcoma-associated herpesvirus infection	0.017	Glycine, serine and threonine metabolism	0.013	Hepatitis B	0.003
10	Focal adhesion	0.019	Alzheimer disease	0.014	TNF signaling pathway	0.003
11	One carbon pool by folate	0.023	Yersinia infection	0.015	Yersinia infection	0.004
12	Salmonella infection	0.023	Osteoclast differentiation	0.017	Osteoclast differentiation	0.005
13	Human cytomegalovirus infection	0.025	Pathways in cancer	0.023	Bladder cancer	0.005
14	Antifolate resistance	0.033	Hippo signaling pathway	0.027	Glycine, serine and threonine metabolism	0.006
15	Glyoxylate and dicarboxylate metabolism	0.035	Hepatitis B	0.030	Gap junction	0.008
16	Apoptosis -	0.035	Adherens	0.035	Human	0.008

	multiple species		junction		papillomavirus infection	
17	Glycine, serine and threonine metabolism	0.046	NOD-like receptor signaling pathway	0.040	C-type lectin receptor signaling pathway	0.013
18			Kaposi sarcoma-associated herpesvirus infection	0.043	Necroptosis	0.013
19					Parathyroid hormone synthesis, secretion and action	0.013
20					One carbon pool by folate	0.013
21					Toxoplasmosis	0.015
22					NF-kappa B signaling pathway	0.016
23					Amphetamine addiction	0.021
24					Adherens junction	0.023
25					Alcoholism	0.023
26					Relaxin signaling pathway	0.026
27					Pertussis	0.028
28					Epstein-Barr virus infection	0.030
29					Glyoxylate and dicarboxylate metabolism	0.031
30					Apoptosis	0.031
31					Leishmaniasis	0.037
32					Salmonella infection	0.040
33					Human immunodeficiency virus 1 infection	0.040
34					Pathways in cancer	0.041
35					Complement and coagulation cascades	0.041
36					Oxytocin signaling	0.044

	pathway	
37	Phagosome	0.044
38	Small cell lung cancer	0.044
39	HTLV-I infection	0.045
40	Human cytomegalovirus infection	0.046