**Supplementary materials**

**Interactive effects of microplastics and tetracycline on bioaccumulation and biochemical status in the Jian carp (*Cyprinus carpio* var. Jian)**

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**Supplementary text 1.** **Details of experimental methods of RNA extraction and cDNA synthesis**

Total RNA was extracted using AG RNAex Pro Rreagent (Accurate Biotechnology Co., Ltd, Hunan, China) following the manufacturer’s protocol. RNA quality was assessed by electrophoresis on a 1.0% agarose gel and its concentration was tested by mySPEC (VWR, Radnor, PA, USA). 1 μg total RNA was purified and the first-strand cDNA was synthesized using Evo M-ML RT Kit with gDNA Clean for qPCR (Accurate Biotechnology Co., Ltd, Hunan, China) according to the manufacturer’s instructions.

**Supplementary text 2. Details of the RT-PCR materials and program**

The real-time PCR assay was carried out using CFX Connect Real-Time System (Bio-Rad, CA, USA) and AG™ SYBR Green Premix Pro Taq HS qPCR Kit (Accurate Biotechnology Co., Ltd, Hunan, China) following the manufacturer’s recommendations. The real-time PCR program was set at 30 s for 95 ℃, followed by 40 cycles of 95℃ for 5 s, 60 ℃ for 30 s. Melting curves were obtained by increasing the temperature from 60 to 95℃ (0.5℃/s) to denature the double-stranded DNA. Each amplification reaction was run in triplicate. After finishing the program, the threshold cycle (Ct) values were obtained from each sample. Relative gene expression levels were evaluated using 2−ΔΔCT method. Bio-Rad CFX Maestro 1.0 version 4.0.2325.0418 was used for data collection.

**Supplementary text 3. Details of the specific method of IBR calculation**

The basis of the calculation is described here briefly. For each biomarker: (1) Calculation of mean and SD for each station. (2) Standardisation of data for each station: χ' i = (χi – mean χ) / s, where χ'i = standardized value of the biomarker, χi = mean value of a biomarker from each station, mean χ is the mean of the biomarker calculated for all the stations, and s = standard deviation calculated for the station-specific values of each biomarker. Result: variance = 1, mean = 0. (3) Using standardised data, addition of the value obtained for each station to the absolute (=non-negative) value of the minimum value in the data set: B = χ' i +│χmin│. Result: adjusts the lowest value in the set to zero. For all the biomarkers treated this way: (4) calculation of Star Plot areas by multiplication of the obtained value of each biomarker (Bi) with the value of the next biomarker, arranged as a set, dividing each calculation by 2 and (5) summing-up of all values: {[(B1· B2)/2] + [(B2· B3)/2] + … [(Bn-1· Bn)/2]}. Result: IBR (average of different arrangements of biomarkers in the set).

**Supplementary figure 1.** Bar graph presentation of TC concentration variation in liver of Jian carp treated with various exposure treatments of TC group and TC+MPs group during 48 h and 96 h exposure.

