Supporting Information

Enhancement of extracellular Cr(VI) reduction for anammox recovery using hydrazine: Performance, pathways, and mechanism

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Text S1: Synthetic wastewater compositions

Ammonium and nitrite were supplemented to mineral medium as required in the form of $(NH_4)_2SO_4$ and $NaNO_2$, respectively. $KHCO_3$ and $MgSO_4 \cdot 7H_2O$ were added at 1.25 g/L and 0.30 g/L, respectively. Additionally, three trace element solutions were also added into the synthetic wastewater, whose compositions were as follows. Trace element I (adding 1.25 mL/L): 5.00 g/L EDTA and 9.14 g/L $FeSO_4 \cdot 7H_2O$. Trace element II (adding 1.25 mL/L): 15.00 $g \cdot L^{-1}$ EDTA, 0.43 g/L $ZnSO_2 \cdot 7H_2O$; 0.24 g/L $CoCl \cdot 6H_2O$, 0.99 g/L $MnCl_2 \cdot 4H_2O$, 0.25 g/L $CuSO_2 \cdot 5H_2O$, 0.22 g/L $Na_2MoO_4 \cdot 2H_2O$, 0.21 g/L $NiCl_2 \cdot 6H_2O$ and 0.014 g/L H_3BO_4. Trace element III (adding 1.00 mL/L): 0.0056 g/L $CaCl_2 \cdot 2H_2O$ and 0.01 g/L $NaH_2PO_4 \cdot 2H_2O$.

Text S2: Optimization tests for N_2H_4 concentration

To explore the simultaneous alleviation effect of N_2H_4 on anammox activity under Cr(VI) exposure, N_2H_4 and Cr(VI) were added concurrently. N_2H_4 was supplied with different concentrations at 0, 1.75, 7.00, 13.13, and 26.25 mg/L and 7.5 mg/L Cr(VI) was added at a concentration of 7.5 mg/L in the batch-test I. Additionally, in order to clarify the recovery effect of N_2H_4 on anammox activity after Cr(VI) inhibition, N_2H_4 was added after Cr(VI) (7.5 mg/L) inhibition for 10 h. The inhibited sludge was taken out and washed with deionized water to remove Cr(VI) adsorbed on sludge surface. Then, the sludge was divided equally into 6 parts mixed with N_2H_4 at 0, 3.67, 7.34, 11.00, 14.67 and 18.34 mg/L, respectively, to conduct the batch-test II.

Supernatant samples were taken every 2 h for nitrogen detection and the specific anammox activity (SAA) calculation. The Cr(VI) inhibition was characterized by inhibition percentage (IP), and the recovery percentage (RP) of anammox activity was used as the indicator to evaluate the

alleviation and recovery effects of N_2H_4 on Cr(VI) inhibition. The optimal N_2H_4 concentrations for maximum SAA were selected to be applied in the subsequent Cr(VI) exposure and activity recovery experiments. The SAA, IP and RP were calculated as followed equations.

$$\text{SAA (kg N/(kg VSS·d))} = 24k/\text{VSS}/1000 \quad (1)$$

where k is the slope of the curve described by the decrease of substrate concentration during the time; VSS is the biomass concentration in the vials.

$$\text{RP (\%)} = \text{SAA}_t/\text{SAA}_c \times 100\% \quad (2)$$

where SAA_c is the SAA value calculated in the control group without N_2H_4 addition; SAA_t is the value measured in the experimental group with N_2H_4 addition.

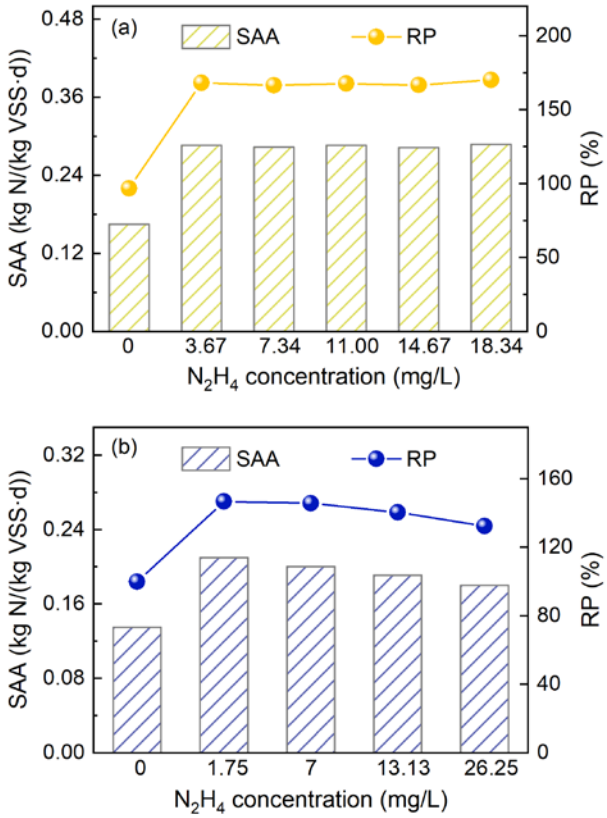

Fig. S1. Effects of the addition of N_2H_4 at different concentrations on the specific anammox activity (SAA) and recovery percentage (RP) in Cr(VI) exposure experiment (a) and activity recovery experiment (b).

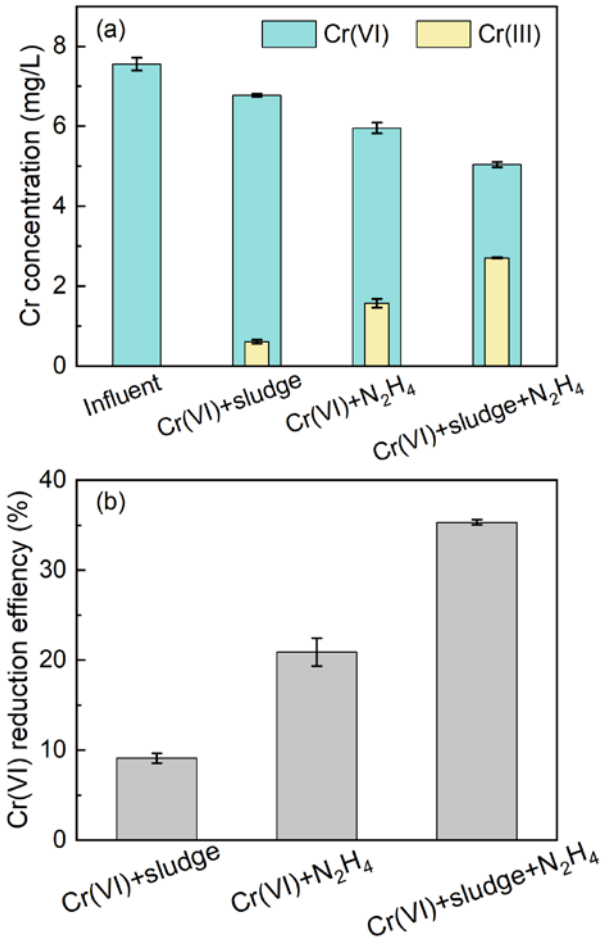

Fig. S2. Variation of Cr(VI) and Cr(III) concentrations in the influent and effluents collected at the end of the batch experiments. The batch experiments included the biotic test (Cr(VI)+N_2H_4+sludge), abiotic test (Cr(VI)+N_2H_4) and biotic control (Cr(VI)+sludge).

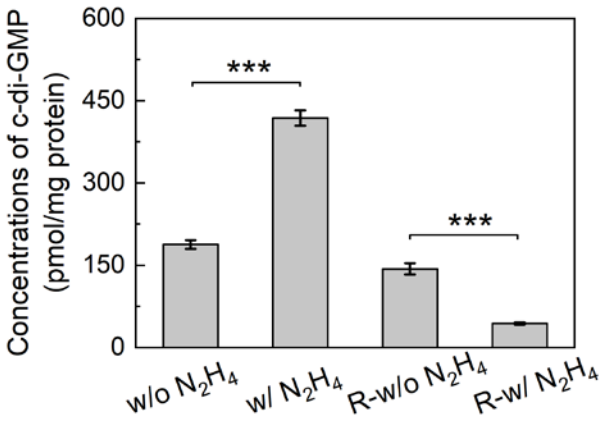

Fig. S3. Effect of N_2H_4 on c-di-GMP concentration of anammox consortia under Cr(VI) exposure and after Cr(VI) inhibition.

Table S1

The operating parameters of HPLC-ICP-MS.

ICP-MS parameters	
Plasma gas flow	15.0 L/min
Carrier gas flow	0.90 L/min
RF power	1550 W
Auxiliary gas flow	0.90 L/min
Collision cell gas and flow	He, 3 mL/min
Selected ion (*m/z*)	52
Dwell time	0.5 s
HPLC parameters	
Guard column	Dionex IonPac AG19-4μm (4×50 mm)
Analytical column	Dionex IonPac AG19-4μm (4×250 mm)
Column temperature	Room temperature
Mobile phase	60 mmol/L NH_4NO_3
(optimum condition)	pH=7.0
Injection volume	10 μL
Flow rate ($mL \cdot min^{-1}$)	0.4
Gradient program	Isocratic elution