

## **Supplementary Material 1**

### **The direct and indirect effects of fungicides and nontarget pesticides on deoxynivalenol accumulation**

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## **1. The extraction and detection method of H<sub>2</sub>O<sub>2</sub>**

### **(1) Preparation of standard curve**

Reaction solutions including 10 mM phosphate buffer (PH 7.0), 1 M KI, 10 mM (NH)MoO were prepared in advance. Then take seven 5-mL centrifuge tubes, add the reaction solutions according to the **Table S5**, shake gently, and let stand for 15 minutes. The absorbance value of the final reaction solution was measured at a wavelength of 352 nm, and a standard curve was made.

### **(2) Extraction and determination of H<sub>2</sub>O<sub>2</sub> from the TBI medium**

The liquid medium was collected and centrifuged at 4000 rpm for 5 min. Then, 1.5 mL of the upper layer was filtered through 0.45- $\mu$ m nylon syringe filters and transferred 0.5 mL filtrate to a 5-mL centrifuge tube. Then add 0.5 mL of 10 mM H<sub>3</sub>PO<sub>4</sub>, 1 mL of 1 M KI, 0.1 mL of 10 mM (NH)MoO, shake gently, and let stand for 15 minutes. The absorbance value of the final reaction solution was measured at a wavelength of 352 nm. All the sample concentration was quantified by the standard curve.

### **(3) Extraction and determination of H<sub>2</sub>O<sub>2</sub> from wheat leaves**

Two grams of wheat leaves were cut off and ground into powder in a mortar with liquid nitrogen. Transferred the powder into a 50-mL centrifuge tube. Then add 10 mL of precooled 5% TCA solution, mix well, and centrifuged at 12000 rpm for 15 minutes. Then, 5 mL of the upper layer was transferred to a 10-mL centrifuge tube and purified with 200 mg of GCB to reduce the interference of pigments. The resulting mixture centrifuged at 4000 rpm for 1 min, and the resulting supernatant was

diluted 5 times with phosphate buffer. Then transferred 0.5 mL diluted solution to a 5-mL centrifuge tube. Add 0.5 mL of 10 mM  $\text{H}_3\text{PO}_4$ , 1 mL of 1 M KI, 0.1 mL of 10 mM  $(\text{NH})\text{MoO}_4$ , shake gently, and let stand for 15 minutes. The absorbance value of the final reaction solution was measured at a wavelength of 352 nm. All the sample concentration was quantified by the standard curve.

## **2. The extraction and detection method of DON**

### **(1) Extraction from TBI medium**

The liquid medium was collected and centrifuged at 4000 rpm for 5 min. Then, 1.5 mL of the upper layer was transferred to a 2-mL centrifuge tube and purified with 15 mg of  $\text{C}_{18}$  and 5 mg of graphitized carbon black (GCB). The resulting mixture was vortexed and centrifuged again, and the resulting supernatant was filtered through 0.22- $\mu\text{m}$  nylon syringe filters for UPLC–MS/MS analysis.

### **(2) Extraction from wheat leaves and ears**

The leaves or ears of wheat were completely ground into powder in a mortar with liquid nitrogen, then homogenized in 8 mL of water for 2 min and then shaken vigorously for 10 min using a shaker. The mixture was centrifuged at 4000 rpm for 5 min. Then, 1.5 mL of the upper layer was transferred to a 2-mL centrifuge tube and purified with 25 mg of  $\text{C}_{18}$  and 10 mg of GCB. The resulting mixture was vortexed and centrifuged again, and the resulting supernatant was filtered through 0.22- $\mu\text{m}$  nylon syringe filters for UPLC–MS/MS analysis. The absolute mass of DON was calculated from its concentration and the volume of water.

### (3) Detection method of DON

All the sample extracts of DON were separated using a Waters Acquity UPLC system. This system included a Waters Acquity UPLC binary solvent manager, Acquity UPLC manager, and Acquity column heater equipped with a Waters Acquity UPLC BEH C<sub>18</sub> column (2.1×100 mm, 1.7μm particle size; Milford, MA, USA). A gradient UPLC elution using methanol as mobile phase A and 0.1% formic acid aqueous solutions as mobile phase B was performed. The analytes were separated at a flow rate of 0.4 mL min<sup>-1</sup> with an elution gradient of 0.00 min/95% B, 2.00 min/95% B, 6.40 min/5% B, 8.40 min/5% B, 8.41 min/95% B, and 10.00 min/95% B. The column was held at 40 °C to decrease the viscosity, and the sample manager temperature was set to 5 °C. The injection volume was 1 μL. Separation and stabilization occurred within 10.00 min.

Concentration of the DON was determined by a triple-quadrupole mass spectrometer (TQD, Waters Crop., Milford, MA, USA) using the multiple reaction monitoring (MRM) mode and positive electrospray ionization (ESI+) mode. The following conditions were typical: the capillary voltage was set to 3.0 kV, and the source and desolvation temperatures were held at 120 and 350°C, respectively. The desolvation gas and cone gas (nitrogen, 99.95% purity) used flow rates of 600 and 50 L h<sup>-1</sup>, respectively. The collision gas, which was high-purity argon, was maintained at 0.15 mL min<sup>-1</sup> with a pressure of 2×10<sup>-3</sup> mbar in the T-wave cell. The concrete MS-MS parameter for DON is listed in **Table S6**. The sample concentration was quantified by the external standard method with matrix-matched calibration curves.

The calibration curves showed good linearity for DON in TBI medium or in wheat leaves or ears with all  $R^2$  values greater than 0.9956.

### **3. The extraction and detection method of pesticides**

#### **(1) Extraction of pesticides from wheat tissues**

The wheat leaves powder (0.5 g) or ears powder (0.5 g) were weighed and homogenized in 2 mL of water and 8 mL of acetonitrile for 2 min and then shaken vigorously for 10 min using a shaker. Subsequently, 4 g of anhydrous  $\text{MgSO}_4$  and 1 g of NaCl were added and vortexed for 1 min at maximum speed. The mixture was centrifuged at 4000 rpm for 5 min to acquire the supernatant. Then, 1.5 mL of the upper layer was transferred to a 2-mL centrifuge tube and purified with 25 mg of PSA, 25 mg of  $\text{C}_{18}$ , 150 mg of anhydrous  $\text{MgSO}_4$  and 10 mg of GCB. The resulting mixture was vortexed and centrifuged again, and the resulting supernatant was filtered through 0.22- $\mu\text{m}$  nylon syringe filters for UPLC–MS/MS analysis.

#### **(2) Detection method of pesticides**

All the sample extracts of pesticides were separated using a Waters Acquity UPLC system. This system included a Waters Acquity UPLC binary solvent manager, Acquity UPLC manager, and Acquity column heater equipped with a Waters Acquity UPLC BEH  $\text{C}_{18}$  column (2.1 $\times$ 100 mm, 1.7 $\mu\text{m}$  particle size; Milford, MA, USA). A gradient UPLC elution using methanol as mobile phase A and 0.1% formic acid aqueous solutions as mobile phase B was performed. The analytes were separated at a flow rate of 0.4 mL min<sup>-1</sup> with an elution gradient of 0.00 min/95% B, 2.00 min/95%

B, 6.40 min/5% B, 8.40 min/5% B, 8.41 min/95% B, and 10.00 min/95% B. The column was held at 40 °C to decrease the viscosity, and the sample manager temperature was set to 5 °C. The injection volume was 1 µL. Separation and stabilization occurred within 10.00 min.

Concentration of the pesticides was determined by a triple-quadrupole mass spectrometer (TQD, Waters Crop., Milford, MA, USA) using the multiple reaction monitoring (MRM) mode and positive electrospray ionization (ESI+) mode. The following conditions were typical: the capillary voltage was set to 3.0 kV, and the source and desolvation temperatures were held at 120 and 350°C, respectively. The desolvation gas and cone gas (nitrogen, 99.95% purity) used flow rates of 600 and 50 L h<sup>-1</sup>, respectively. The collision gas, which was high-purity argon, was maintained at 0.15 mL min<sup>-1</sup> with a pressure of  $2 \times 10^{-3}$  mbar in the T-wave cell. The concrete MS-MS parameter for these pesticides is listed in **Table S6**. The sample concentration was quantified by the external standard method with matrix-matched calibration curves. The calibration curves showed good linearity for pesticides in wheat leaves or wheat ears with all  $R^2$  values greater than 0.9992.