

**Figure S1.** Relative expression of the *atrB* gene in three fludioxonil-resistant isolates and one representative sensitive isolate. The transcript levels of the *atrB* gene in fludioxonil-resistant isolates are shown relative to the sensitive one (SH-S172), which was assigned a value of 1.0. Values represent the mean  $\pm$  SD of two biological replicates.



**Figure S2.** Alignment of Bos1 protein sequences from different *Botrytis cinerea* isolates. Each type of protein sequence is indicated by one representative isolate. SH-94: without resistance related mutations, SH-458: with I365S mutation, SH-473: with I365N mutation, SH-714: with N373S mutation, SH-517: with Q369P/N373S mutations.

Fungicide	Active ingredient (a.i.)	Provider		
fludioxonil	95%	Syngenta Biotechnology Co., Ltd., Shanghai, China		
azoxystrobin	98%	Syngenta Biotechnology Co., Ltd., Shanghai, China		
pyrimethanil	95.2%	Syngenta Biotechnology Co., Ltd., Shanghai, China		
carbendazim	98%	Guangxin Agricultural Chemicals Co., Ltd, Anhui, China.		
boscalid	95%	Guangxin Agricultural Chemicals Co., Ltd., Anhui, China.		
fluopyram	98.8%	Guangxin Agricultural Chemicals Co., Ltd., Anhui, China.		
difenoconazole	95%	Yulong Chemical Industrial Co., Ltd., Hangzhou, China.		
cyprodinil	98.5%	Beijing Mindleader Agroscience Co.,Ltd., Beijing, China		
iprodione	95.5%	Bayer CropScience Co., Ltd., Beijing, China.		

Table S1. Fungicides used in this study.

Fungicide	Medium	Discrimination method <sup>a</sup>	Discriminatory concentration (µg/mL)	Reference
fludioxonil	PDA	MIC	10, 100	22
carbendazim	YG	MIC	100	23
azoxystrobin	YBA	MIC	100	24
boscalid	YBA	MIC	10	25
fluopyram	YBA	MIC	10	26
difenoconazole	YG	EC <sub>50</sub>	2.68	Unpublished data
pyrimethanil	GKMN	MIC	1	25, 27
cyprodinil	GKMN	$EC_{50}$	10	28
iprodione	PDA	$EC_{50}$	1	28

## Table S2. Sensitivity test of different fungicides.

<sup>a</sup> MIC means the minimum inhibitory concentration, and  $EC_{50}$  indicates the effective concentration inhibiting 50% of colony growth.