

Captions for supplemental Table and figures

Table S1. qPCR, genome walking, and PCR primers.

Figure S1. Construction of the overexpression plasmid.

Figure S2. The sensitivity of wild-type *P. eryngii* var. *ferulae* in CYM medium with two different antifungal compounds (a) *cbx* resistance; (b) *hygB* resistance.

Figure S3. Effect of different osmotic pressure stabilizers on protoplast preparation. The eyepiece and objective of the optical microscope (starMICRO, WuXi, China) were magnified at 10x and 40x, respectively (a) mannitol; (b) sorbitol; (c) MgSO₄; (d) Sucrose; (e) KCl.

Figure S4. Verification of the insertion of exogenous plasmids into the four transformant genomes (*P. eryngii* var. *ferulae*::*pfgpd1-egfp*, *P. eryngii* var. *ferulae*::*pfgpd1-egfp*, *P. eryngii* var. *ferulae*::*pfgpd1-egfp*, and *P. eryngii* var. *ferulae*::*pfsar1-egfp*) (a) Amplify Fragment1 (*egfp*, 717 bp) in transformants; (b) Amplify Fragment2 (promoter + *egfp*) in transformants, which are 1821, 1527, 1324, and 923 bp, respectively. M: marker. NC: negative control.

Supplemental Table

Table S1 qPCR, genome walking, and PCR primers

qPCR primes				
Gene	ID	Prime name	Primer sequence (5′—3′)	Amplicon length (bp)
<i>egfp</i>		Fw	TACGTGCAGGAGAGGACCAT	148
		Rv	ACTTGTGGCCGAGGATGTTT	
<i>sar1</i> ^a	1052294	Fw	GGATAGTCTTCCTCGTCGATAG	133
		Rv	GGGTGCGTCAATCTTGTTAC	
Genome walking primes				
Gene	ID	Prime name	Primer sequence (5′—3′)	
<i>pfsdlhb</i>	KDQ23485.1	S-SP1	TCCAGAAAGAATTCAAGATCTACCG	
		S-SP2	CTCGATCCGCGCTTTCTCCACCTCG	
		S-SP3	AACGTCTGCGCCCGACAGAACTATC	
<i>pfgpd1</i>	KDQ24107.1	G1-SP1	TGGGCCTTGGAATAAACAA	
		G1-SP2	GACTTCGAGCGCGTTACGGA	
		G1-SP3	CCAGATGTATTTCGAGTTAG	
<i>pfgpd2</i>	KDQ24081.1	G2-SP1	CACCATATAATCGAGATCAA	
		G2-SP2	AGGGCGTTACGAAAGACGAT	
		G2-SP3	TCCCACTTCCTGATGGACGC	
<i>pfsar1</i>	KDQ33428.1	SA-SP1	ACTTGACGTTTCCGATGGCA	
		SA-SP2	ATCATTTTTCAACATATGAA	
		SA-SP3	TGGCGTTCTTGTGGAGCAGG	
<i>pfras</i>	KDQ24534.1	R-SP1	TGATGATAGATGGTCGTATG	
		R-SP2	CACCCATGGTGAAAATAGAG	
		R-SP3	GGTCAAGGCTGATTTTCCGA	
PCR primes				
Gene	ID	Prime name	Primer sequence (5′—3′)	
Mutant- <i>pfsdlhb</i>	KDQ23485.1	MS1-Fw	CTAGCTAGCTAGATCGAAGTTATTAGGATCCAGCATAT GTTTTGGGCTGGA	
		MS1-Rv	GACGTACAATTGAAGATGGTGAGGCAACGGAACAGA CTCA	
		MS2-Fw	TGAGTCTGTTCCGTTGCCTCACCATCTTCAATTGTACGT C	
		MS3-Rv	ATTGCCCCGAGCCGTTAGATGTCTCA	
<i>P_{pfgpd1}</i>		Fw	GCCCCAAAACATATGCTGGATCCAGCTTTCTCGCTTCGT CAGAGG	
		Rv	GTCGACTAATTTGAGCTCGATTCTAGAGACGTTGACCT ATAACAAG	
<i>P_{pfgpd2}</i>		Fw	GCCCCAAAACATATGCTGGATCCCGGCTGTATGGCTTG AGGC	
		Rv	GTCGACTAATTTGAGCTCGATTCTAGAGACGTTGACCT	

		CCGTACGGGT
<i>P_{pfsar1}</i>	Fw	GCCCCAAAACATATGCTGGATCCCGCGAGTGGATCTGT
		AAGGT
	Rv	GTCGACTAATTTGAGCTCGATTCTAGAGCAGGCCTGC
		AGACCGAATA
<i>P_{pfras}</i>	Fw	GCCCCAAAACATATGCTGGATCCTGCAATCAACGAGCA
		ACAGC
	Rv	GTCGACTAATTTGAGCTCGATTCTAGAGGTGGCCTATA
		TGGCCATCT
<i>T_{pfsdhh}</i>	Fw	TCGAGCTCAAATTAGTCGACACCCTAGTCAACAGCCA
		CGG
	Rv	CGTTCGACGATCTAGCTAGCATTGCCCCGAGCCGTTAG
		ATG
<i>egfp</i>	Fw	CGTCTCTAGAATGGGTAAGGGAGAAGAAGT
	Rv	GGTGTGCGACTTATTTGTATAGTTCATCCA

Supplemental figures

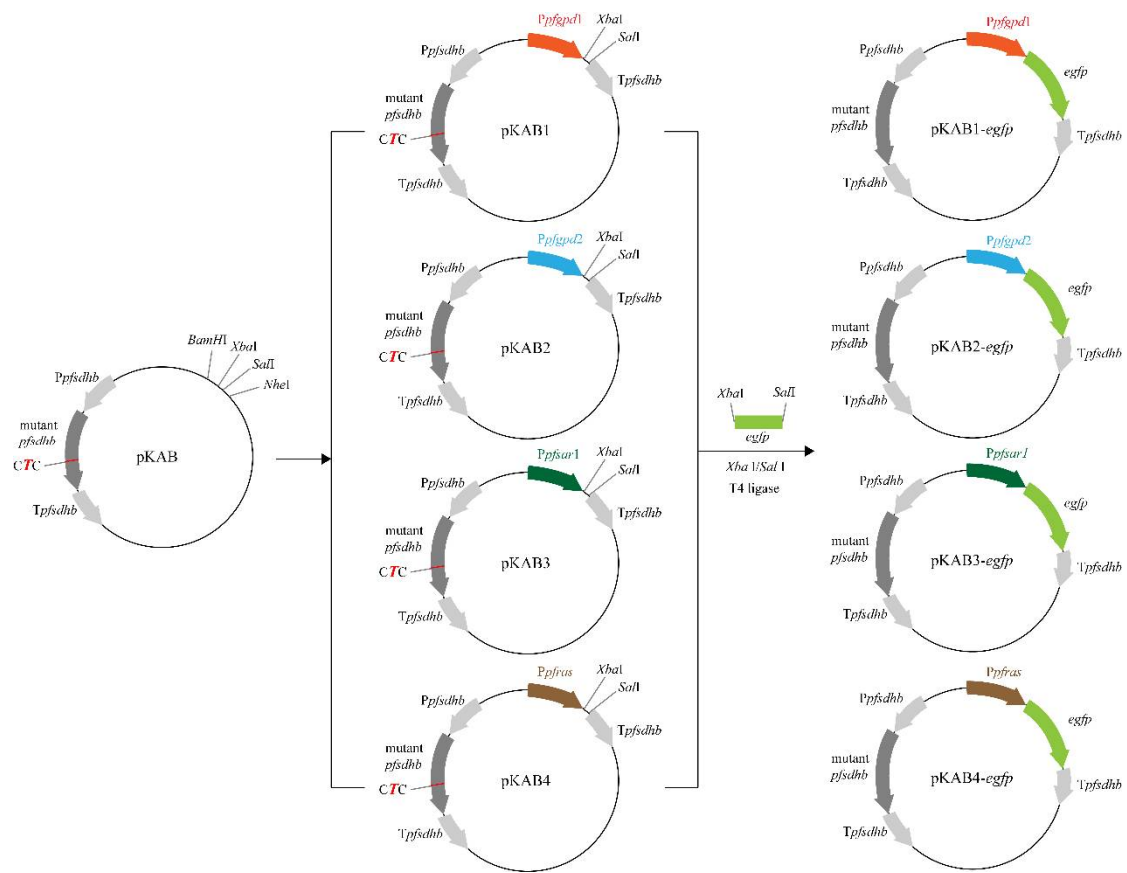


Figure S1. Construction of the overexpression plasmid.

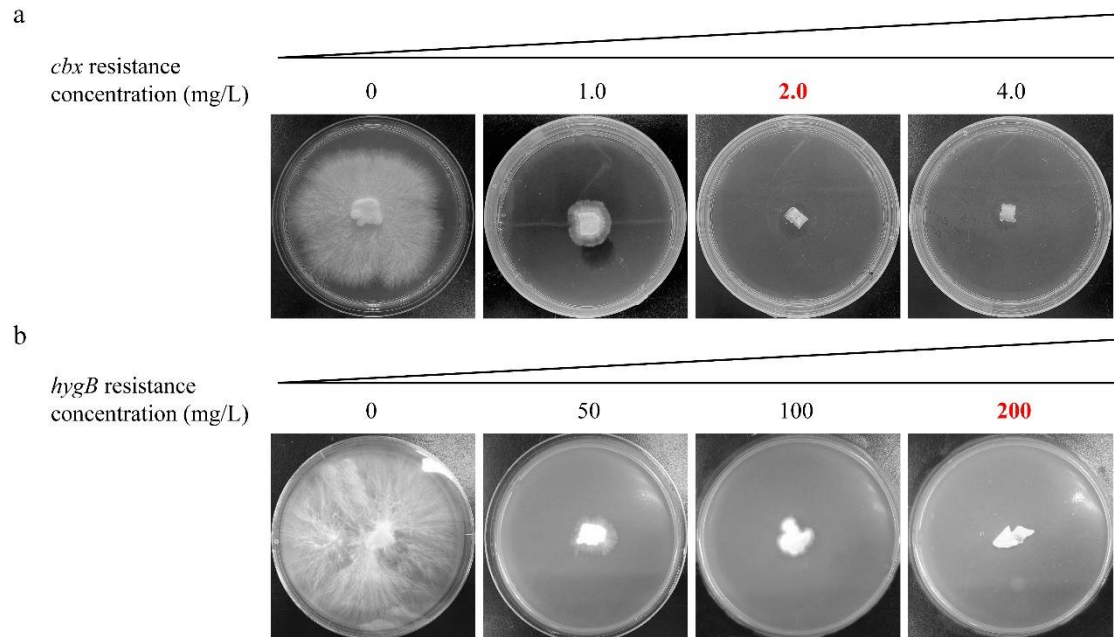


Figure S2. The sensitivity of wild-type *P. eryngii var. ferulae* in CYM medium with two different antifungal compounds (**a**) *cbx* resistance; (**b**) *hygB* resistance.

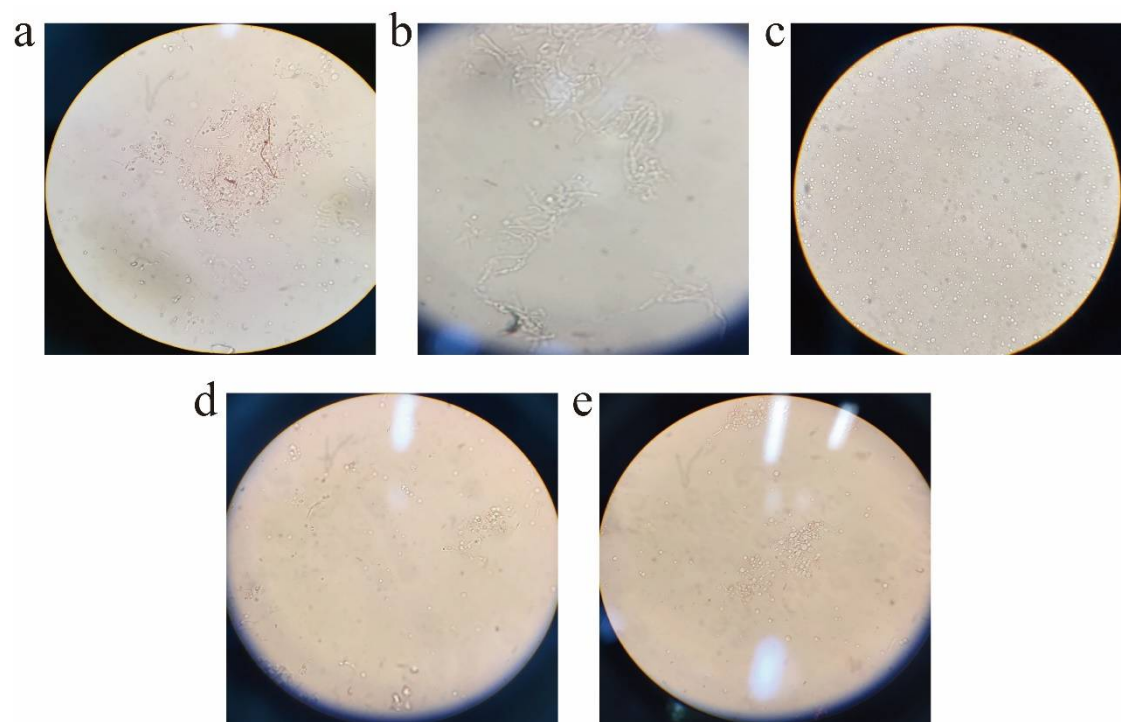


Figure S3. Effect of different osmotic pressure stabilizers on protoplast preparation. The eyepiece and objective of the optical microscope (starMICRO, WuXi, China) were magnified at 10x and 40x, respectively (a) mannitol; (b) sorbitol; (c) MgSO_4 ; (d) Sucrose; (e) KCl.

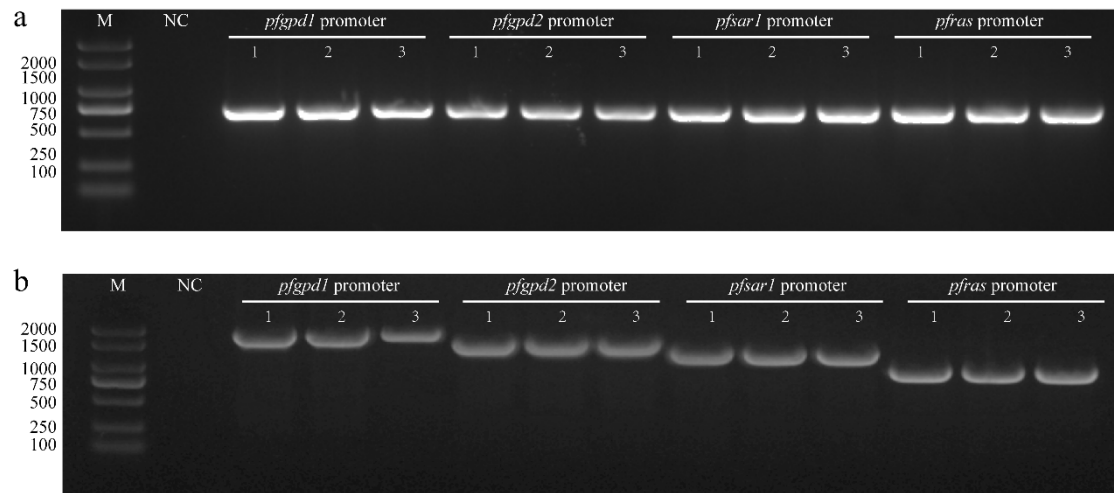


Figure S4. Verification of the insertion of exogenous plasmids into the four transformant genomes (*P. eryngii* var. *ferulae*::*pfgpd1-egfp*, *P. eryngii* var. *ferulae*::*pfgpd2-egfp*, *P. eryngii* var. *ferulae*::*pfgpd1-egfp*, and *P. eryngii* var. *ferulae*::*pfsar1-egfp*) (a) Amplify Fragment1 (*egfp*, 717 bp) in transformants; (b) Amplify Fragment2 (promoter + *egfp*) in transformants, which are 1821, 1527, 1324, and 923 bp, respectively. M: marker. NC: negative control.