

## Supplemental Material

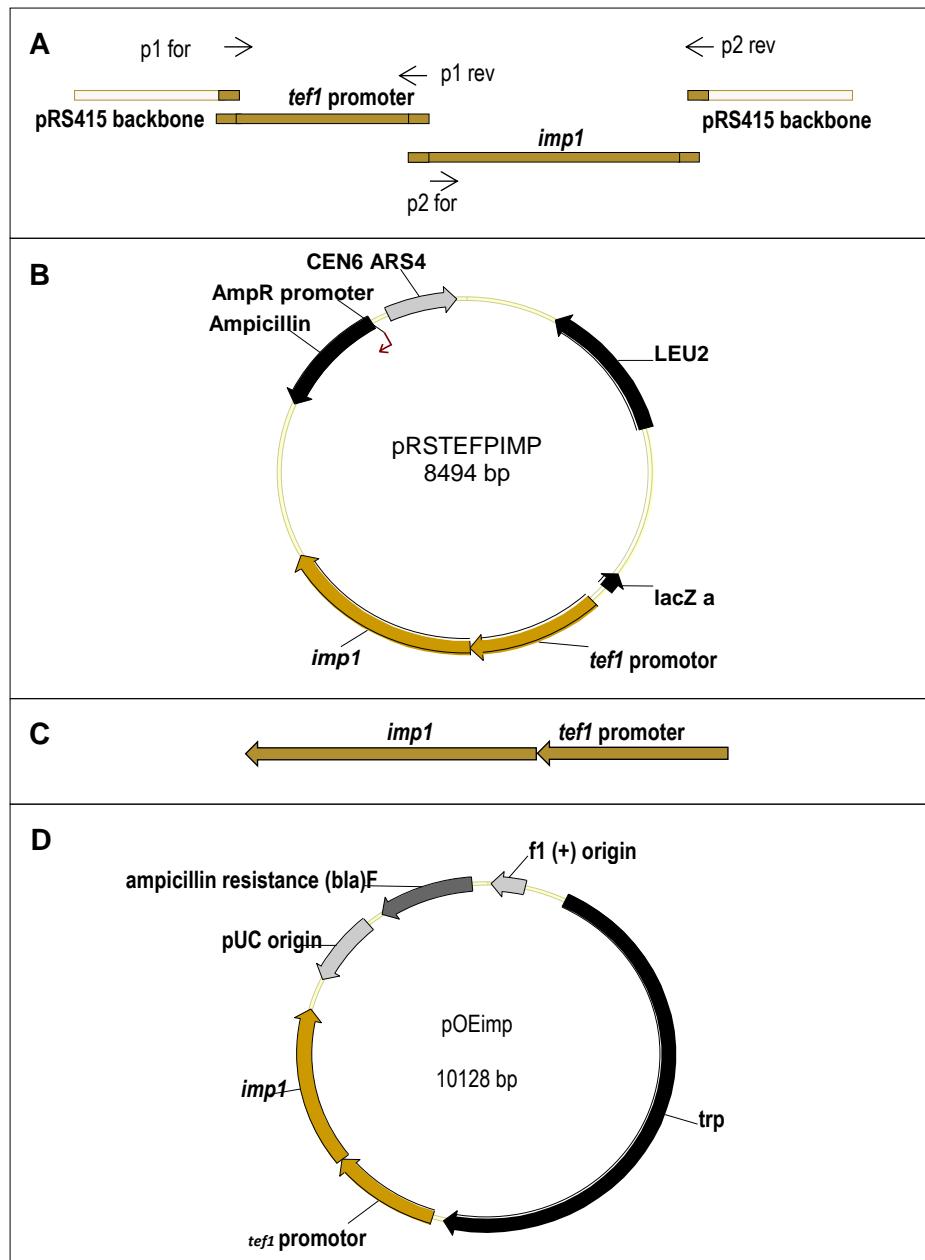
**Suppl. Tab. S1:** *S. commune* strains used in this study.

Strain	Genetic background	Source
12-43	<i>ura</i> <sup>-</sup>	Jena Microbial Resource Collection (JMRC), Germany
T33	<i>matA3,1; matB3,2; trp; ura</i> <sup>-</sup>	JMRC
EVC1	<i>matA3,1; matB3,2</i>	JMRC
OEIMP4	<i>matA3,1; matB3,2; tef1p::imp1</i>	JMRC
OEIMP6	<i>matA3,1; matB3,2; tefp::imp1</i>	JMRC

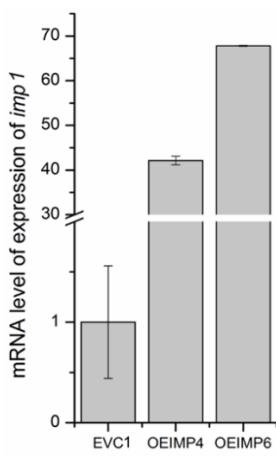
**Suppl. Tab. S2:** Primers used in this study.

Name	Gene	Sequence (5'-3')	Length (bp)	Efficiency
p1for	pRS415::ptef1	CTAGTTCTAGAGCGGCCGCCACCGCCGGAAAAGAACAAAGACGTGT*		
p1rev	<i>tef1::imp</i>	TAGTCGGCGATGGTAAGGTCGGTGGCATTGAGTGTGTTCTAAGTGAG*		
p2for	<i>tef1::imp</i>	TCACCTAGAAAACACTCAAATGCCACCACCTTACCAT*		
p2rev	<i>Imp::pRS415</i>	CTAAAGGGAACAAAGCTGGACACACGAGGATGACGGTT*		
mio3	Myo-inositol oxygenase	For: GGATCTACAAGCCGCACTGT Rev: CACGGTGCCAGGGATAGAAG	149	92.80%
imp	Inositol monophosphatase	For: GAAGCCC GTGCTCGGTG Rev: TGGTTGTTGATGCCCTCCGT		
ipp1	Hypothetical inositol polyphosphate phosphatase	For: GACCGCATAGCGGACTACAAC Rev: TCCATGAGGGAATAGCCCGA	188	109.95%
act	Actin-1	For: CTGCTCTTGTATTGACAATGGTTCC Rev: AGGATACCACGCTTGGACTGAGC	178	96.31%
tef	Translation elongation factor 1a	For: AGCTCGGCAAGGGTTCCTTCA Rev: AACTTCCAGAGGGCGATATCA	97	97%
ubi	Ubiquitin-conjugating-protein	For: GAAGGAGTACGATGCGAAGG Rev: TCCTCCTCTGCCTTCTTGC	93	89.5%

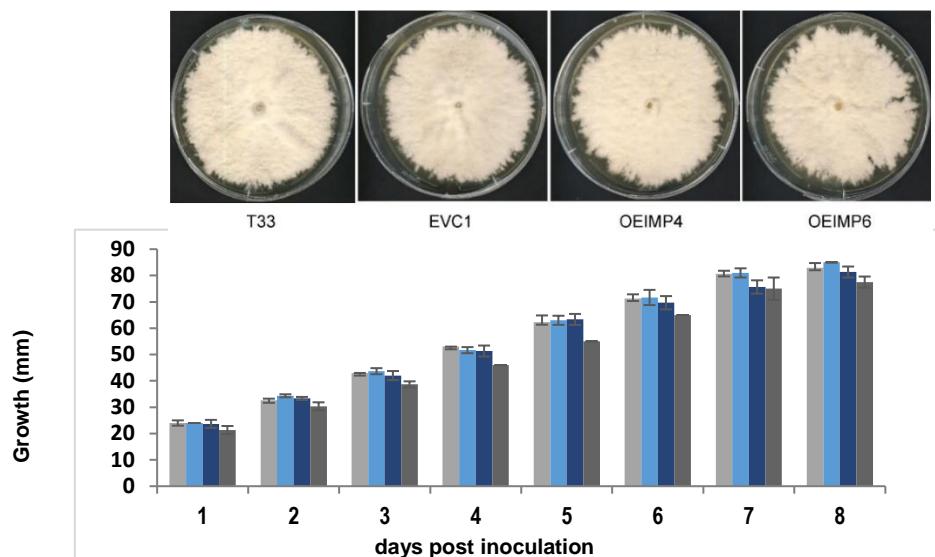
\* Overhang for annealing in italics



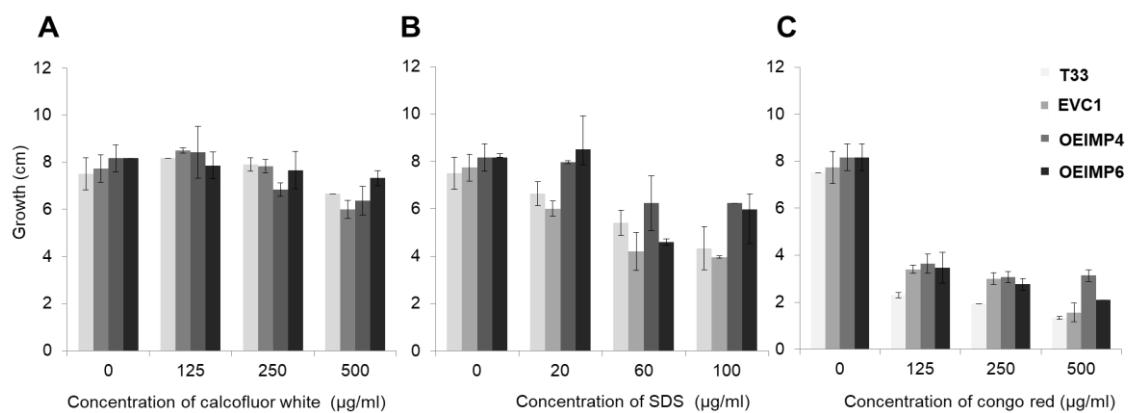
**Suppl. Fig. S1:** Plasmid construction for generation of *imp1* overexpression strains. (A) Recombination in *S. cerevisiae* was carried out using pRS415 to generate the product fusing the PCR products for the *tef1* promoter (*tef1p*) and the *imp1* gene. (B) Overlapping sequences between two adjacent fragments *tef1p* and *imp1* were used (p1rev and p2for). (C) Fusion product *tef1p::imp1* was cloned in pRS415 to generate pRSTEFPIMP. (D) The *tef1p::imp1* region was excised and cloned into pSK containing the selection marker *trp1* to arrive at pOEIMP.



**Suppl. Fig. S2:** Verification of *imp1* overexpression by RT-qPCR. Comparison is given for the empty vector control EVC1 and overexpression transformants OEIMP4 and OEIMP6.



**Suppl. Fig. S3:** Effect of *imp1* overexpression on colony morphology (top) and growth (below) of *S. commune* T33 (grey bars), empty vector control EVC1 (blue), and overexpression transformants OEIMP4 (dark blue) and OEIMP6 (dark grey); n=3.



**Suppl. Fig. S4:** Susceptibility towards calcofluor white (A), SDS (B), and Congo red (C). Comparison is given in triplicates for the wildtype parental *S. commune* T33, the empty vector control EVC1, and overexpression transformants OEIMP4 and OEIMP6.

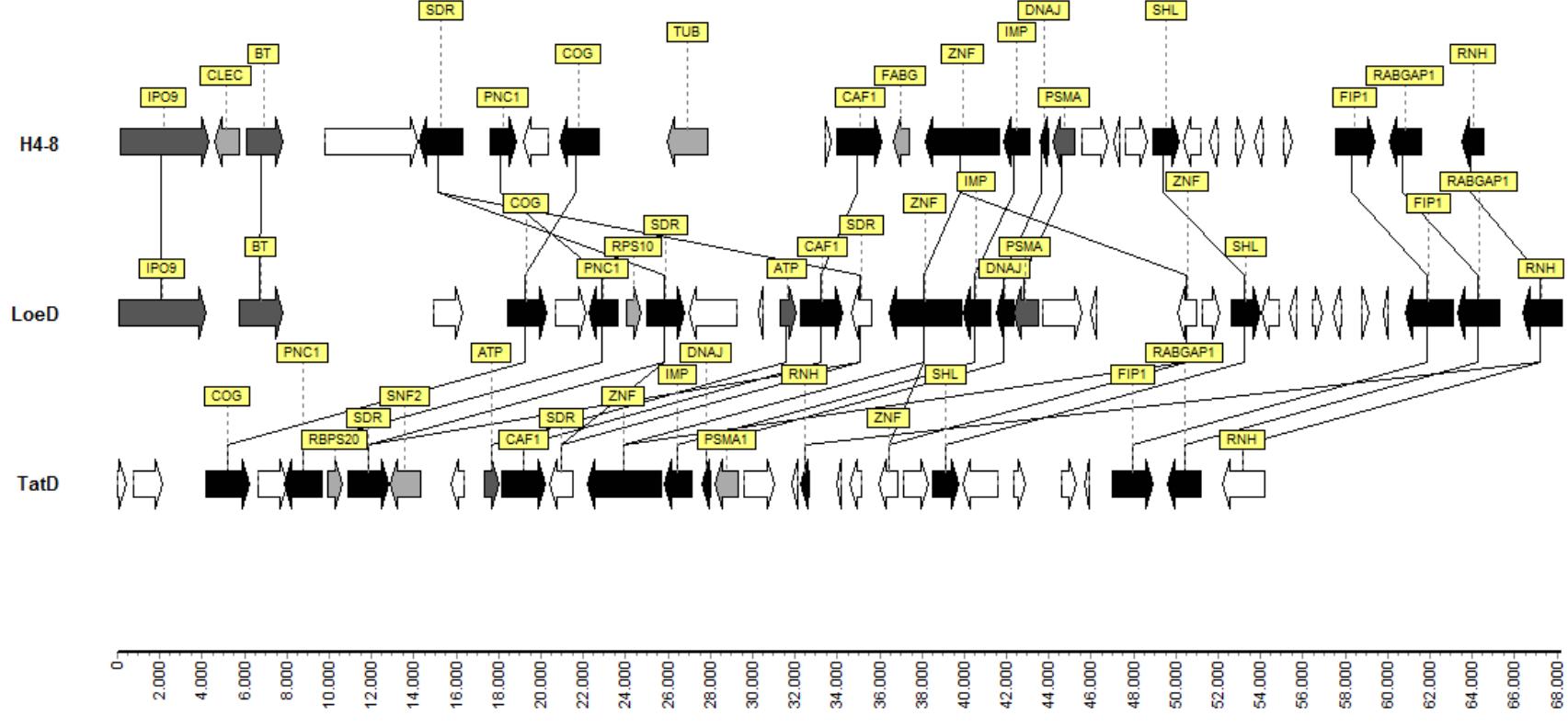
**Suppl. Tab. S3:** Regulation of genes involved in the inositol signalling cycle. The microarray analysis (Erdmann et al., 2012) is showing comparisons between *S. commune* 12-43 grown on CYM versus CYM with seepage water from a former mining site (HSW) and co-isogenic *S. commune* W22 grown on CYM with 0.01 mM cadmium nitrate.

	KOG Annotation	Gene Location Scho1	12-43 vs. 12-43 HSW			12-43 vs. W22 Cd		
			x-fold	p-value	Q	x-fold	p-value	Q
PIS	KOG3240: PI synthase	estExt_fgenesh1_pm.C_20467	-1.45	1.79E-01	1	2.11	1.16E-02	1
	KOG0693: Myo-inositol-1-phosphate synthase	estExt_fgenesh1_pm.C_70303	1.25	4.55E-01	1	2.45	6.76E-03	1
		estExt_fgenesh1_pm.C_70303	2.00	2.81E-03	1	-2.50	2.52E-04	1
		estExt_fgenesh1_pm.C_70303	1.08	6.86E-01	1	-1.50	3.75E-02	1
PI4K	KOG2381: PI4-kinase	augustus-scaffold_8.g641	2.24	1.21E-01	1	4.61	6.59E-03	1
		e_gw1.2.405.1	1.25	4.86E-01	1	2.81	3.84E-03	1
PI5K	KOG0903: PI4-kinase, involved in intracellular trafficking and secretion,	estExt_Genewise1Plus.C_21667	3.82	9.87E-02	0	1.73	4.86E-01	0
PI5K	KOG0230: PI-4-phosphate 5-kinase and related FYVE finger-containing proteins	estExt_fgenesh1_kg.C_60138	-1.75	6.41E-02	1	1.07	8.11E-01	1
		estExt_Genewise1.C_12180	1.04	8.60E-01	1	3.04	1.67E-04	1
	KOG0229: PI-4-phosphate 5-kinase	estExt_fgenesh1_kg.C_110031	2.41	1.38E-02	1	1.75	9.89E-02	1
PI3K	KOG0906: PI3-kinase VPS34, involved in signal transduction,	fgenesh1_pm.C_scaffold_5000272	-1.08	8.16E-01	1	1.34	3.56E-01	1
PLC	KOG0169: PI-specific phospholipase C	estExt_Genewise1.C_170034	1.30	2.09E-01	1	-1.06	7.71E-01	1
		estExt_fgenesh1_pm.C_60287	1.18	4.20E-01	1	1.83	9.07E-03	1
		augustus-scaffold_6.g78	-1.81	2.19E-01	1	-1.00	1.00E+00	1
		augustus-scaffold_3.g897	-4.17	5.43E-02	1	2.59	1.87E-01	0
PKC	KOG2397: Protein kinase C substrate, 80 kD protein, heavy chain	estExt_Genewise1Plus.C_11897	-1.11	7.10E-01	1	2.56	4.35E-03	1
IMK	KOG1620: Inositol polyphosphate multikinase, component of the ARGR transcription regulatory complex, KOG4749: Inositol polyphosphate kinase	e_gw1.1.1005.1	1.79	5.90E-01	1	6.94	8.66E-02	1
		estExt_Genewise1Plus.C_100427	-15.82	5.24E-03	1	-2.71	2.63E-01	1
		e_gw1.2.893.1	-13.45	2.05E-03	1	-6.98	1.46E-02	1
IP <sub>3</sub> P	KOG1089: Myotubularin-related PI3-phosphate 3-phosphatase MTM6	estExt_fgenesh1_pm.C_20581	3.88	2.81E-02	1	2.11	2.04E-01	1
IP <sub>4</sub> P								
IP <sub>5</sub> P	KOG0565: Inositol polyphosphate 5-phosphatase and related proteins	estExt_fgenesh1_pm.C_20547	1.35	7.50E-01	1	7.38	4.55E-02	1
		estExt_Genewise1.C_170073	-1.20	4.45E-01	1	1.13	6.01E-01	1
		augustus-scaffold_5.g421	-9.43	1.26E-01	1	-5.25	2.51E-01	1
		estExt_fgenesh2_pg.C_100071	-8.56	1.58E-04	1	-12.9	2.38E-05	1
IPP	KOG1382: Multiple inositol polyphosphate phosphatase	gw1.1.3820.1	1.86	1.58E-01	1	1.96	1.29E-01	1
		gw1.8.732.1	1.55	2.01E-01	1	2.12	3.59E-02	1
		estExt_Genewise1Plus.C_30697	1.35	4.05E-01	1	-2.21	3.69E-02	1
		e_gw1.7.1022.1	-1.62	5.16E-01	1	-1.10	9.02E-01	1

	KOG Annotation	Gene Location Scho1	12-43 vs. 12-43 HSW			12-43 vs. W22 Cd		
			x-fold	p-value	Q	x-fold	p-value	Q
ITP	KOG0566: Inositol-1,4,5-triphosphate 5-phosphatase (synaptotjanin), INP51/INP52/INP53 family	estExt_Genewise1.C_11249	-9.00	4.61E-02	1	-14.53	1.79E-02	1
IMP	KOG2951: Inositol monophosphatase	e_gw1.1.586.1	1.33	4.63E-01	1	1.12	7.67E-01	1
		e_gw1.1.586.1	-1.05	8.93E-01	1	-1.48	2.59E-01	1
		e_gw1.1.586.1	-1.30	2.04E-01	1	-1.20	3.79E-01	1

**Suppl. Tab. S4:** Intracellular trafficking associated proteins regulated in *imp1* overexpressing transformant pOEIMP4 vs. EVC

ID	x-fold	KOG class and description	BLAST hit, organism, % identity
2598708	2.9	cyto-skeleton	Drebrins and related actin binding proteins Actin-binding related protein, <i>Laccaria bicolor</i> , 70%
2541047	2.7		Kinesin-like protein Kinesin-like protein, <i>Armillaria solidipes</i> , 78%
2674409	2.3		Predicted actin-bundling protein Protein FRG1, <i>Hypsizygus marmoreus</i> , 68%
2681330	2.3		Predicted actin-bundling protein Ubiquitin-activating enzyme E1-like, <i>Trametes pubescens</i> , 63%
2704529	23.8		Signal recognition particle, subunit Srp19 Signal recognition particle, SRP19 subunit, <i>Armillaria solidipes</i> , 61%
2262522	23.7		Protein involved in glucose derepression and pre-vacuolar endosome protein sorting Snf7-domain-containing protein, <i>Armillaria solidipes</i> , 74%
2603193	21.7		Mitochondrial Fe/S cluster exporter, ABC superfamily Iron-sulfur clusters transporter ATM1, <i>Coprinopsis cinerea okayama</i> , 75%
2597192	21.7		Golgi transport complex subunit Dor1-domain-containing protein, <i>Armillaria gallica</i> , 48%
2678633	4.3		Vesicle coat complex AP-3, beta subunit acyltransferase ChoActase/COT/CP, <i>Fistulina hepatica</i> , 60%
2693248	3.5		Signal recognition particle, subunit Srp72 Signal recognition particle subunit SRP72, <i>Trametes pubescens</i> , 47%
2460397	3.0	intracellular trafficking, secretion, and vesicular transport	Mitochondrial import inner membrane translocase, subunit TIM8 Mitochondrial import inner membrane translocase <i>Heterobasidion irregularare</i> , 68%
2700443	3.0		Guanine nucleotide exchange factor Sec7 guanine nucleotide exchange factor, <i>Moniliophthora roreri</i> , 72%
1185145	2.4		Vacuolar assembly/sorting protein DID2 Vacuolar protein sorting protein 46, <i>Moniliophthora roreri</i> , 90%
2698910	2.2		Membrane coat complex Retromer, subunit VPS5/SNX1, Sorting nexins, and related PX domain-containing proteins PX-domain-containing protein, <i>Armillaria gallica</i> , 75% Sorting nexin-41, <i>Hypsizygus marmoreus</i> , 76%
2678145	2.1		Cysteine protease required for autophagy - Apg4p/Aut2p Thiamine pyrophosphokinase, <i>Coprinopsis cinerea okayama</i> , 49%
2007874	2.1		Nuclear transport factor 2 Nuclear transport factor 2, <i>Neolentinus lepideus</i> , 75%
2595664	2.1		SNARE protein Syntaxin 1 and related proteins t-SNARE, <i>Armillaria solidipes</i> , 80%
2573839	2.2	poorly characterized	Vacuolar sorting protein VPS1, dynamin, and related proteins Dynamin protein dnm1, <i>Moniliophthora roreri MCA 2997</i> , 75%



**Suppl. Fig. S5:** Chromosomal map of *imp1* surrounding genes in three different strains *S. commune*, H4-8, LoeD, and TatD. White arrows indicate unknown genes, black arrows show that the corresponding genes are present in all three genomes.