Title: Validation of reference genes for transcriptional analyses in *Pleurotus ostreatus* using RT-qPCR.

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SUPPLMENTAL INFORMATION

Materials and methods

Using Blastp (cutoff e^{-15}) *vs* the filtered model protein databases of *Phanerochaete chrysosporium, Coprinopsis cinerea* and *Postia placenta* (deposited on the JGI webpage) we identified the orthologous models of *sar, pep* and *phos* genes. Gene expression datasets corresponding to the fungal models were downloaded from the GEO portal of NCBI (http://www.ncbi.nlm.nih.gov/gds). The individual expression of the orthologous of our three best reference genes was recorded and displayed in a heatmap in $\log_2$ scale. The mean expression of the whole genome was used as a proxy of expression stability.

SUPPLEMENTARY TABLES

Table S1. Intra-group variation of SD_N.

Ranking	SmF		s-SSF		SSF	
	Gene	SD_N	Gene	SD_{NORM}	Gene	SD_N
1	sar1	0.0552	actin1	0.0398	lip	0.0399
2	lip	0.0754	sar1	0.0473	gapdh1	0.0413
3	pas	0.1869	pep	0.2538	sar1	0.0414
4	actin1	0.2264	phos	0.3502	pep	0.0507
5	phos	0.2495	chs	0.3827	vsn	0.0819
6	vsn	0.3295	cyph	0.5765	tub	0.0828
7	tub	0.3591	cyt	0.6335	cyph	0.1058
8	cyph	0.5161	gapdh1	0.6768	phos	0.192
9	pep	0.5324	pas	1.2489	cyc	0.2263
10	gapdh1	0.7029	vsn	1.4482	cyt	0.3129
11	chs	0.9616	lip	1.4949	chs	0.4413
12	cyt	1.0499	tub	1.9659	actin1	0.5344
13	cyc	1.4165	cyc	3.568	pas	0.587

Table S2. Inter-group variation of SD_N. Genes removed from the analysis are shown in red.

Gene	SD_N		
	SmF	sSSF	SSF
pep	0.1025	0.1457	-0.2482
cyc	0.1637	-0.587	0.4233
phos	-0.1111	0.1639	-0.0528
tub	0.4202	0.2527	**-0.6729**
lip	-0.2783	0.153	0.1252
vsn	**0.6768**	-0.6287	-0.0481
cyph	0.3934	0.1326	-0.526
actin1	-0.5544	-0.3202	**0.8747**
cyt	-0.1292	0.3158	-0.1867
gapdh1	-0.1876	-0.4507	0.6383
pas	**-0.782**	0.3109	0.4711
sar1	0.2558	-0.0409	-0.2148
chs	0.0301	0.5529	-0.5831

Table S3. Spearman correlation of *mnp3* expression levels obtained using five
normalization strategies (in parenthesis).

	mnp3 (I)	mnp3 (II)	mnp3 (III)	mnp3 (IV)	mnp3 (V)
mnp3 (I)	1	0.98	0.96	0.62	0.49
mnp3 (II)		1	0.96	0.64	0.51
mnp3 (III)			1	0.65	0.55
mnp3 (IV)				1	0.67
mnp3 (V)					1

Table S4. Spearman correlation of *lacc10* expression levels obtained using five
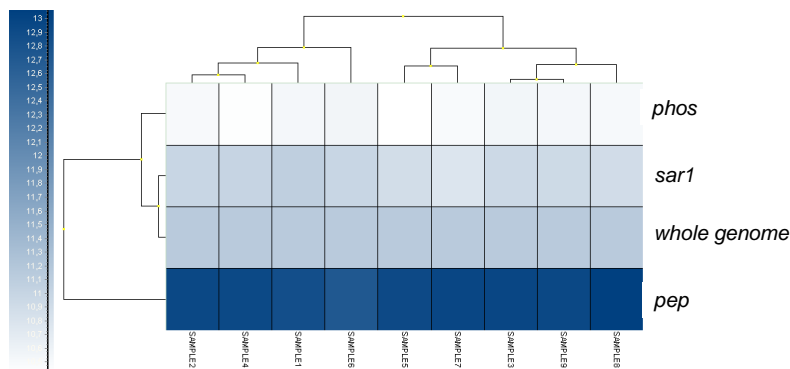normalization strategies (in parenthesis).

	lacc10 (I)	lacc10 (II)	lacc10 (III)	lacc10 (IV)	lacc10 (V)
lacc10 (I)	1	1.00	0.99	0.90	0.95
lacc10 (II)		1	0.98	0.93	0.97
lacc10 (III)			1	0.89	0.93
lacc10 (IV)				1	0.94
lacc10 (V)					1

Table S5. Results of the Mann–Whitney test comparing the expression rations of *mnp3* and *lacc10* between the three culture media.

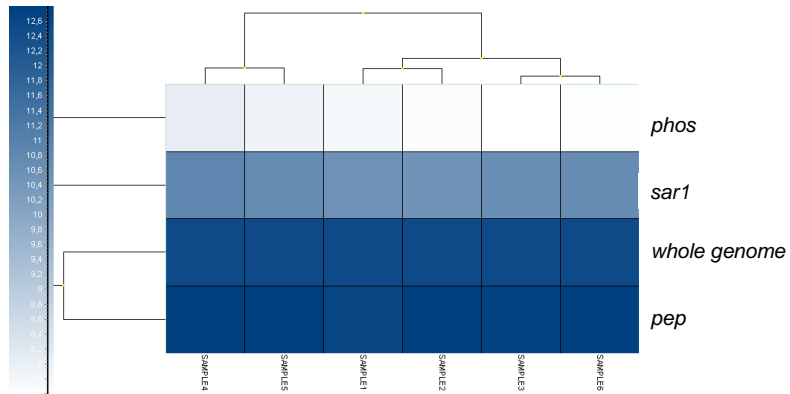|	Group 1	Group 2	*p* Value
	Expression (SmF)	Expression (s-SSF)	1,47E-06
mnp3	Expression (SmF)	Expression (SSF)	2,18E-07
	Expression (s-SSF)	Expression (SSF)	0,66829079
	Expression (SmF)	Expression (s-SSF)	1,26E-05
lacc10	Expression (SmF)	Expression (SSF)	3,74E-05
	Expression (s-SSF)	Expression (SSF)	0,00105003

SUPPLEMENTARY FIGURES

A) *Phanerochaete chrysosporium*
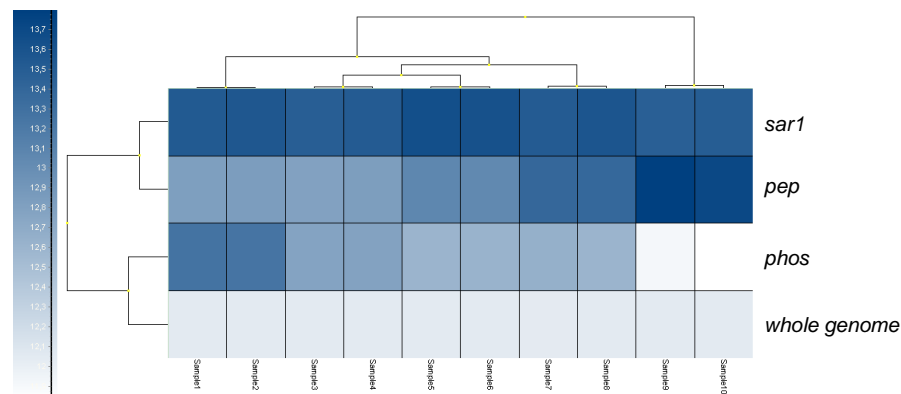

B) *Postia placenta*

C) *Coprinopsis cinerea*

Fig S1. Exploratory analysis of *sar1*, *pep* and *phos* expression stability in other fungal models