1 Supplementary materials

2 Supplementary Figure 1.

3 Phylogenetic analysis of Pmt proteins in filamentous fungi and yeasts. Genetyx 4 software with an UPGMA method was used for constructing the phylogenetic tree. $\mathbf{5}$ UniProt database accession numbers for the proteins are as follows. CnPmt1,Q5KIZ1; 6 *Cn*Pmt2,Q5KAF1; *Cn*Pmt4,A3E242; *Sp*Oma1/Ogm1,O13898; 7SpOma2/Ogm2,Q9C100; SpOma4/Ogm4,O42933; ScPmt1,P33775; ScPmt2,P31382; 8 ScPmt3,P47190; ScPmt4,P46971; ScPmt5,P52867; ScPmt6,P42934; CaPmt1,O74189; 9 *Ca*Pmt2,O5ADM9; *Ca*Pmt4,059X23; *Ca*Pmt5,O5ACU3; *Ca*Pmt6.O5A688; 10 AfPmt1,B0XYZ3; AfPmt2,B0XPY7; AfPmt4,B0YA13; AnPmtA,Q96WN5; 11 AnPmtB,Q5B3W9; AnPmtC,Q5BDC1; AaPmtA,Q96VV1; *Nc*Pmt1,Q7SH94; 12*Nc*Pmt2,Q1K4Z4; *Nc*Pmt4,Q7SD53. *Cryptococcus neoformans, Schizosaccharomyces* 13pombe, Saccharomyces cerevisiae, Candida albicans, Aspergillus fumigatus, A. 14nidulans, A. awamori, and Neurospora crassa are abbreviated as Cn, Sp, Sc, Ca, Af, 15An, Aa, and Nc, respectively.

16

17 Supplementary Figure 2.

18 Disruption of Anpmt genes. Schematic representation of the disruptions of AnpmtB

19 (A), AnpmtC (B), and AnpmtApmtB (C) and recomplementation of AnpmtB (D) and

20 AnpmtC (E). Southern blot analysis of total DNA isolated from strain AKU89 (lanes 1

and 3) or from the respective disruptants (lanes 2 and 4) are shown in the panels on the

22 right (A, B, C). Probes were derived from retained regions of AnpmtA (A and C) and

23 AnpmtB (B and C) as indicated. PCR using total DNA isolated from strain AKU89

24 (lane 2), from the $\Delta Anpmt$ (lane 2), or the recomplemented strains (lane 4) are shown

25 in the panels on the right (D, E). λ DNA digested with *Hin*dIII was used as a size

29

1 marker (lane1).

 $\mathbf{2}$

3 Supplemental Table. Oligonucleotides used in this study

Oligonucleotide	Sequence	Restriction site
primers		(underlined)
pmtB-F	5'-ATTYWTIGAYGTICAYCCICC-3',	
pmtB-R	5'-AGGRAARTARTGRTGIARRAA-3'	
pmtC-F	5'-TTYGAYTNCAYCCICCITT-3'	
pmtC-R	5'-TANCCNGTIACYTGYTGICC-3'	
An-pmtB-RT-F	5'-ATGGCGAAGGACACCCTGGA-3'	
An-pmtB-RT-R	5'-TTCATTGAGACCGGGAATCGGAGT-3'	
An-pmtC-RT-F	5'-ATGGGATTATCGGCTGGCCATTACCAT-3'	
An-pmtC-RT-R	5'-TTTCGCGAAGTGCAAGTCATAGCCAAG-3'	
F1-PnkuB	5'-GATATCGGACCTTCTTGACTGGACC-3'	EcoRV
R1-PnkuB	5'-CGCAGAGGTGACCGAAGTCGATATGTGTGAGACGCAAACA-3'	
F2-aur	5'-TGTTTGCGTCTCACACATATCGACTTCGGTCACCTCTGCG-3'	
R2-aur	5'-CTCCTTAGGAACATTACTCCTTCAAGGGCTGAAAGTATGCC-3'	
F3-TnkuB	5'-GGCATACTTTCAGCCCTTGAAGGAGTAATGTTCCTAAGGAG-3'.	
R3-TnkuB	5'-CGATATCAGTGCCCTTCCC-3'	EcoRV
pmtB-around-F	5'-AATCTAGAAGGCCTAGGACCGTAGGTGTATTCCC-3'	20010
pmtB-around-R	5'-AAAAAACAGCTGAATTCTCCCCTAAATTCG-3'	
ptrA-KpnI-F	5'-AAAGGTACCGGGGTGACGATGAGCCGCTC-3'	KpnI
ptrA-KpnI-R	5'-TGGTACCGGGCAATTGATTACGGGATCC-3'	KpnI
pmtB-pr-F	5'-GGTTTTCCTGGCGATGCAAA-3'	1
pmtB-pr-R	5'-TGAATTCTTATTCATTGAGACCGGGAATC-3'	
F2-AnpmtB	5'-TTAAGTCGACATGACACCTTCCGGAGCGTG-3'	SalI
R2-AnpmtB	5'-CACCGTCGACGTATGACTTTTAGATAGTGTCAG-3'	SalI
F1-AnpGpB	5'-GGATTTGATGTAAATGTAGTCGACATGACACCTTCCG-3'	
R1-AnpGpB	5'-CGGAAGGTGTCATGTCGACTACATTTACATCAAATCC-3'	
F2-AnpGpB	5'-TCTAAAAGTCATACGTCGACGAGTGGAAATGTGTAACGG-3'	
R2-AnpGpB	5'-CCGTTACACATTTCCACTCGTCGACGTATGACTTTTAGA-3'	
F-AnpmtBPr	5'-CCGAGATCACCGTCAGGTT-3'	
R-AnpmtBPr	5'-GCGGGGAAATAGTGGTGTAA-3'	
pmtC-around-F	5'-AATCTAGAAGGCCTACGCGTGCTTATACCGGAAT-3'	
pmtC-around-R	5'-TTTCGCGAAGTGCAAGTCATAGCCAAG-3'	
pmtC-pr-F	5'-ATAAAGCTTCCATCACTATGCGCCAC-3'	
pmtC-pr-R	5'-TTTCGCGAAGTGCAAGTCATAGCCAAG-3'	
F1-AnnyrG	5'-CACCATGCATGATTGGTATGTTTCCTCGGA_3'	NsiI
R1-AnpyrG	5'-TAGGGCATGCTTCCTCTAGCGCAAACAAGTTC-3'	SphI
F3-AnpmtC	5'-CACAGTCGACGTCTTCACTTGGTGGAGATATA-3'	SalI
R3-AnpmtC	5'-CCAAGTCGACATTTCGGGCGCTATATCAGG-3'	SalI
F-AnpmtCPr	5'-ATGTCTTCATCGCCCTCTCTG-3'	
R-AnpmtCPr	5'-ATGGCGAAGCTGGACGATATC-3'	
F1-PglaA	5'-AAGCTTGTCAAGGGACAGCAAGTAT-3'	HindIII
R1-PglaA	5'- <u>GGTACC</u> TCGCTTTTCTTTGAGGGAC-3'	KpnI
argB-ApaI-F	5'- <u>GGGCCC</u> GCATGCAATAATTGCAGCAA-3'	ApaI
argB-ApaI-R	5'- <u>GGGCCC</u> GTCGACCTACAGCCATTGCG-3'	ApaI
F2I-TglaA	5'- <u>GATATCAGATCT</u> TAGACTATTCATTCCTATTGAT-3'	EcoRV
		BglII (double)
K2I-PglaA	5- <u>GAATTCGCGGCCGC</u> TGGGGGGGGGGGGGGTAACGATGTTGA-3'	EcoRI
E1-5660.2 eve	5'- A A G A CC ATGGGGTCGTTC A CCCTATCC A C 3'	Ncol
R1-5660 2-exp	5'-ACA AGTCGACGTGCCGGTCAGGATTCGT3'	Sall
F1-3HA	5'-TTGACCATGGGTCGACGCTAGCTACCCATACGATGTT-3'	Sall

R1-3HA	5'-GATT <u>CTGCAG</u> CTAAGCGTAATCCGGTACATCGTAT-3'	PstI
NotI-wscA-F	5'-AAAA <u>GCGGCCGC</u> ATGAGGTCGTTCACG-3'	NotI
BglII-3HA-R	5'-AAAA <u>AGATCT</u> CTAAGCGTAATCCGG-3'	BglII

¹ I, Y, W, R, and N indicates Inosine, C or T, A or T, Aor G, and A, C, T, or G, respectively.



Supplementary Figure 1. Goto, M. et al.



Supplementary Figure 2. Goto, M., et al.



a →	6.3 kb	-	b
_	<i>ptrA</i>		$ \Delta pmtB$
c 🕨	2.3 kb	◀ d	





F2-†AnpmtB (a) F-pmtBpr (c) R2-†AnpmtB (b) R-pmtBpr (d)



Primer pairs used



Supplementary Figure 2. Goto, M., et al.