

## Supplementary Information

**Table S1.** Daily body weight change of SD rats after oral administration of rPhy-P for 7 days.

	Dose (U/ kg)			
	0	500	1000	1500
Day 0	167.33 ± 3.055 <sup>a</sup>	154.67 ± 18.148 <sup>a</sup>	166.00 ± 5.292 <sup>a</sup>	153.33 ± 17.243 <sup>a</sup>
Day 1	171.33 ± 2.517 <sup>b</sup>	157.33 ± 17.673 <sup>b</sup>	171.00 ± 5.568 <sup>b</sup>	161.67 ± 17.559 <sup>b</sup>
Day 2	177.33 ± 1.155 <sup>c</sup>	162.67 ± 16.653 <sup>c</sup>	180.00 ± 7.211 <sup>c</sup>	166.67 ± 17.088 <sup>c</sup>
Day 3	179.33 ± 2.517 <sup>d</sup>	166.00 ± 18.028 <sup>d</sup>	181.67 ± 9.238 <sup>d</sup>	170.33 ± 18.230 <sup>d</sup>
Day 4	186.33 ± 5.508 <sup>e</sup>	171.33 ± 19.425 <sup>e</sup>	186.00 ± 8.718 <sup>e</sup>	178.67 ± 18.903 <sup>e</sup>
Day 5	180.67 ± 5.774 <sup>f</sup>	167.33 ± 19.425 <sup>f</sup>	180.00 ± 6.000 <sup>f</sup>	170.67 ± 20.817 <sup>f</sup>
Day 6	178.67 ± 7.572 <sup>g</sup>	166.67 ± 18.037 <sup>g</sup>	178.00 ± 8.718 <sup>g</sup>	169.33 ± 16.653 <sup>g</sup>
Day 7	180.67 ± 2.517 <sup>h</sup>	169.67 ± 18.230 <sup>h</sup>	183.33 ± 8.145 <sup>h</sup>	172.33 ± 15.948 <sup>h</sup>

No significance observed between normal and treated in any days. <sup>a-h</sup> showing no significant difference in days 0, 1, 2, 3, 4, 5, 6, 7, respectively.

**Table S2.** Organ weights of rats after oral administration of rPhy-P for 7 days.

Organs	Dose (U/ kg)			
	0 U	500 U	1000 U	1500 U
Heart	0.34 ± 0.014	0.37 ± 0.269	0.31 ± 0.386	0.35 ± 0.016
Liver	3.47 ± 0.164	3.36 ± 0.155	3.35 ± 0.540	3.57 ± 0.563
Spleen	0.24 ± 0.028	0.25 ± 0.017	0.25 ± 0.022	0.27 ± 0.051
Brain	1.00 ± 0.056	1.04 ± 0.138	1.03 ± 0.113	1.04 ± 0.172
Lung	0.49 ± 0.032	0.50 ± 0.033	0.53 ± 0.017	0.52 ± 0.060
Kidney	Left	0.39 ± 0.003	0.36 ± 0.037	0.38 ± 0.018
	right	0.37 ± 0.005	0.38 ± 0.054	0.39 ± 0.014

**Table S3.** Incidence of mortality and clinical signs after 7 days oral administration of rPhy-P into rats.

Dose (U/kg)	Mortality	Clinical Signs						
		Salivation	PRONE position	Decrease of Locomotive Activity	Loss of Fur	Bite/Scratch Wounds	Lacrimation	Closed Eyes
0	0/3 <sup>1</sup>	0/3	0/3	0/3	0/3	0/3	0/3	0/3
500	0/3	0/3	0/3	0/3	0/3	0/3	0/3	0/3
1000	0/3	0/3	0/3	0/3	0/3	0/3	0/3	0/3
1500	0/3	0/3	0/3	0/3	0/3	0/3	0/3	0/3

<sup>1</sup> Total number of rats treated.

**Figure S1.** Strategy for the site-directed mutagenesis (SDM) of pRTL2 vector sequences to insert *Aspergillus nidulans phy* gene. **(a)** Gene expression cassette in the pRTL2 vector contains 35S promoter, TEV leader sequence, *Nco*I site for the cloning and terminator. AT nucleotides in *Nco*I site were modified into GC nucleotides by SDM-PCR, changing to *Sac*II site; **(b)** *Aspergillus nidulans phy* gene was cloned into mutagenized pRTL2-MS vector, resulting in pRTL2-MS-Phytase. The resultant vector was used as a template to perform the secondary SDM-PCR to recover the start codon of *phy* gene, changing from *Sac*II to *Nco*I site; **(c)** *Phy* gene contains three *Nco*I sites.

