J. Microbiol. Biotechnol.
https://doi.org/10.4014/jmb.2007.07003

Supporting information

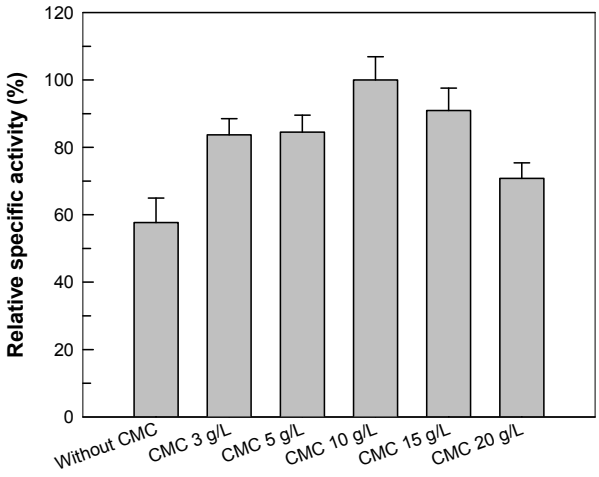
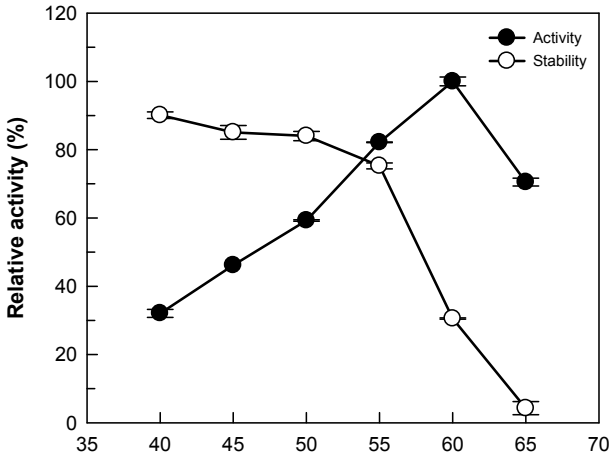


Fig. S1. Effect of the concentration of carboxymethyl cellulose added during cultivation on the hydrolytic activity for ginsenoside Rb2. The reactions were performed at 50°C for 6 h in 0.2 M citrate-phosphate buffer (pH 5.5) containing 0.4 mg/mL ginsenoside Rb2 and 1.0 mg/mL extracellular enzyme. Data are expressed as the means of three experiments and the error bars represent standard deviations.

A



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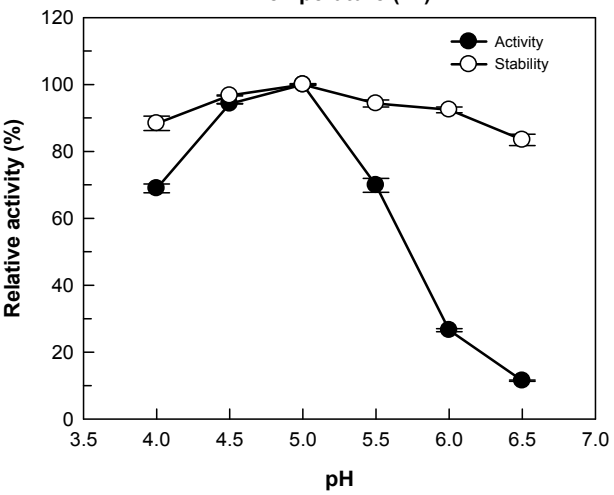
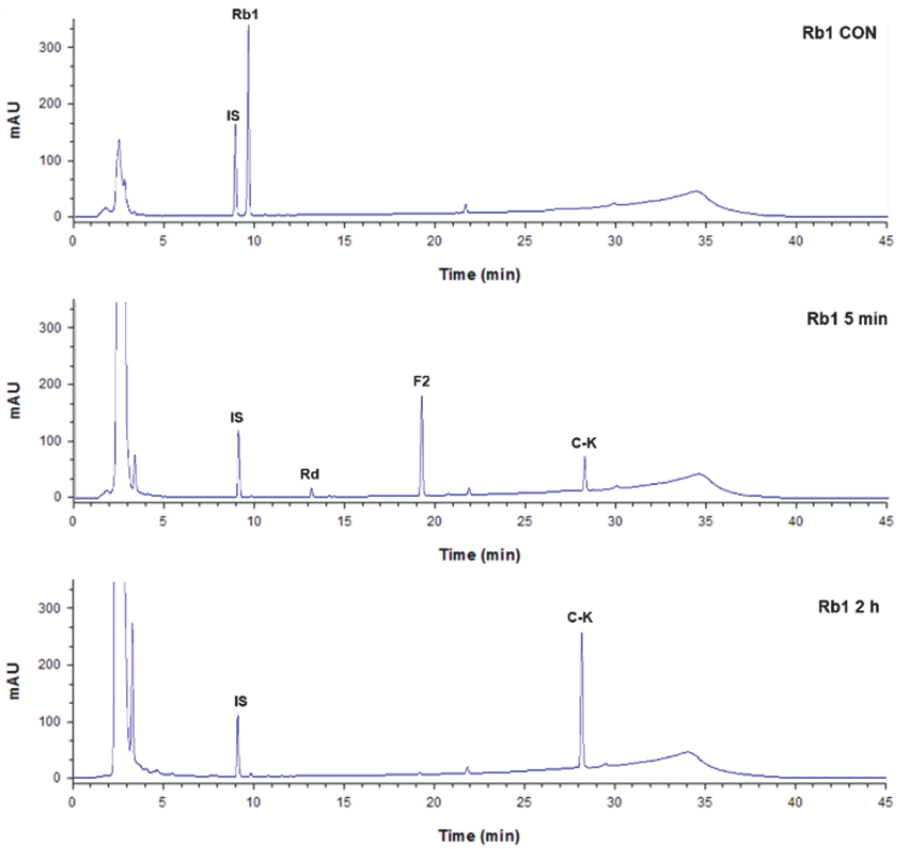


Fig. S2. Effects of temperature and pH on the activity and stability of extracellular enzyme from *Aspergillus niger* for ginsenoside Rb1. (A) Temperature effect. The reactions were performed for 10 min in 0.2 M citrate-phosphate buffer (pH 5.0) containing 0.4 mg/mL ginsenoside Rb1 and 1.0 mg/mL extracellular enzyme by varying the temperature from 40 to 65°C. (B) pH effect. The reactions were performed at 55°C for 10 min in 0.2 M citrate-phosphate buffer containing 0.4 mg/mL ginsenoside Rb1 and 1.0 mg/mL extracellular enzyme by varying the pH 4.0 to 6.5. The reactions of temperature and pH stability of the extracellular enzyme were determined by measuring the residual activity at 55°C and pH 5.0 after incubating the extracellular enzyme for 24 h. Data are expressed as the means of three experiments and the error bars represent standard deviations.

A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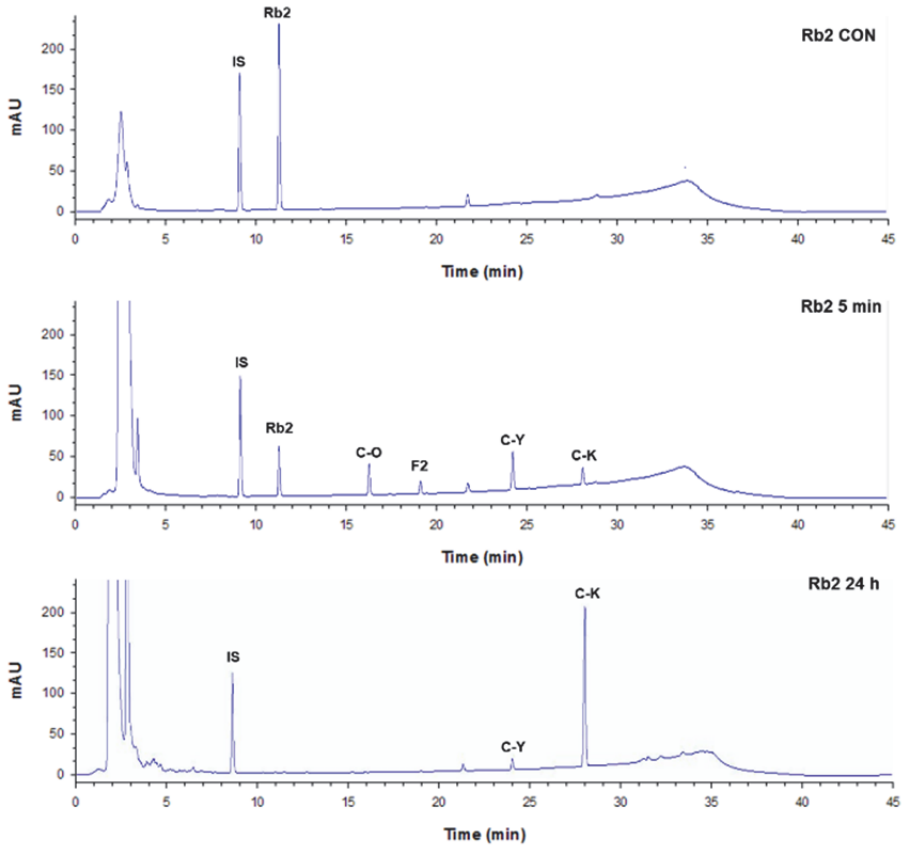
IS
Rb1 CON
mAU
Time (min)
IS
Rd
F2
C-K
Rb1 5 min
mAU
Time (min)
C-K
IS
Rb1 2 h
mAU
Time (min)

B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Rb2 CON
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Time (min)
IS
Rb2
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C-K
IS
C-Y
Rb2 24 h
mAU
Time (min)

C

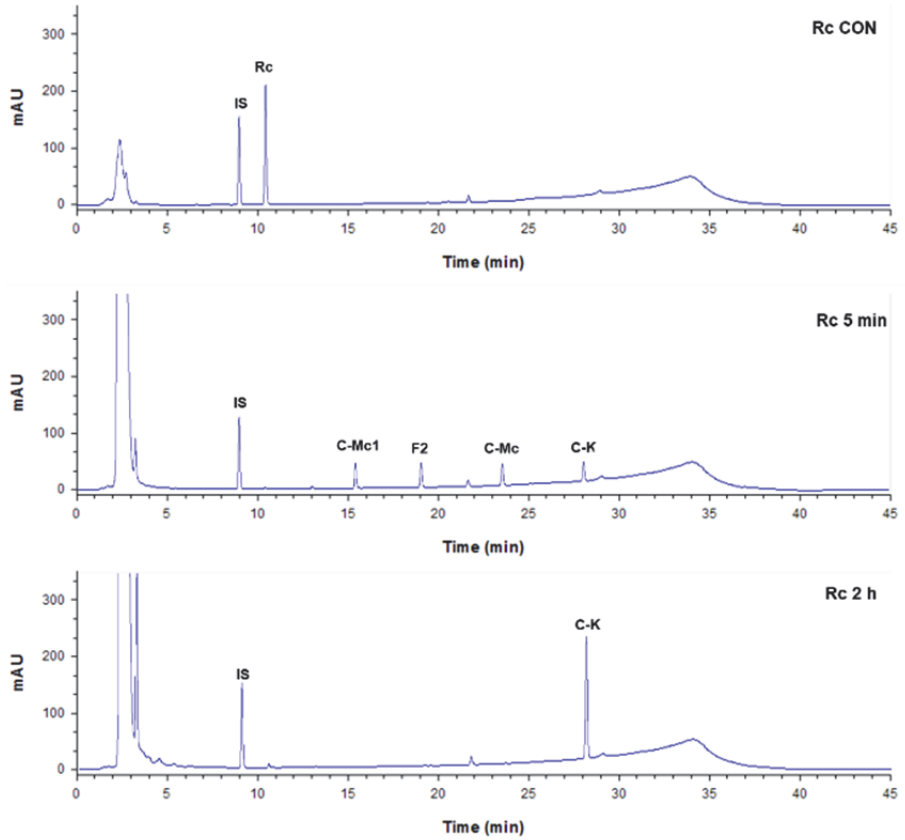


Fig. S3. High-performance liquid chromatography profiles during the conversion of ginsenosides Rb1, Rb2, and Rc into compound K by extracellular enzyme from *Aspergillus niger*. (A) Conversion of Rb1 to compound K (C-K). (B) Conversion of Rb2 to C-K. (C) Conversion of Rc to C-K. The reactions were performed at 55°C for 5 min and 2 h or 24 h in 0.2 M citrate-phosphate buffer (pH 5.0) containing 1.0 mg/mL of each ginsenoside and 2.5 mg/mL extracellular enzyme. Control (CON) indicated high-performance liquid chromatography profile of substrate at initial time.

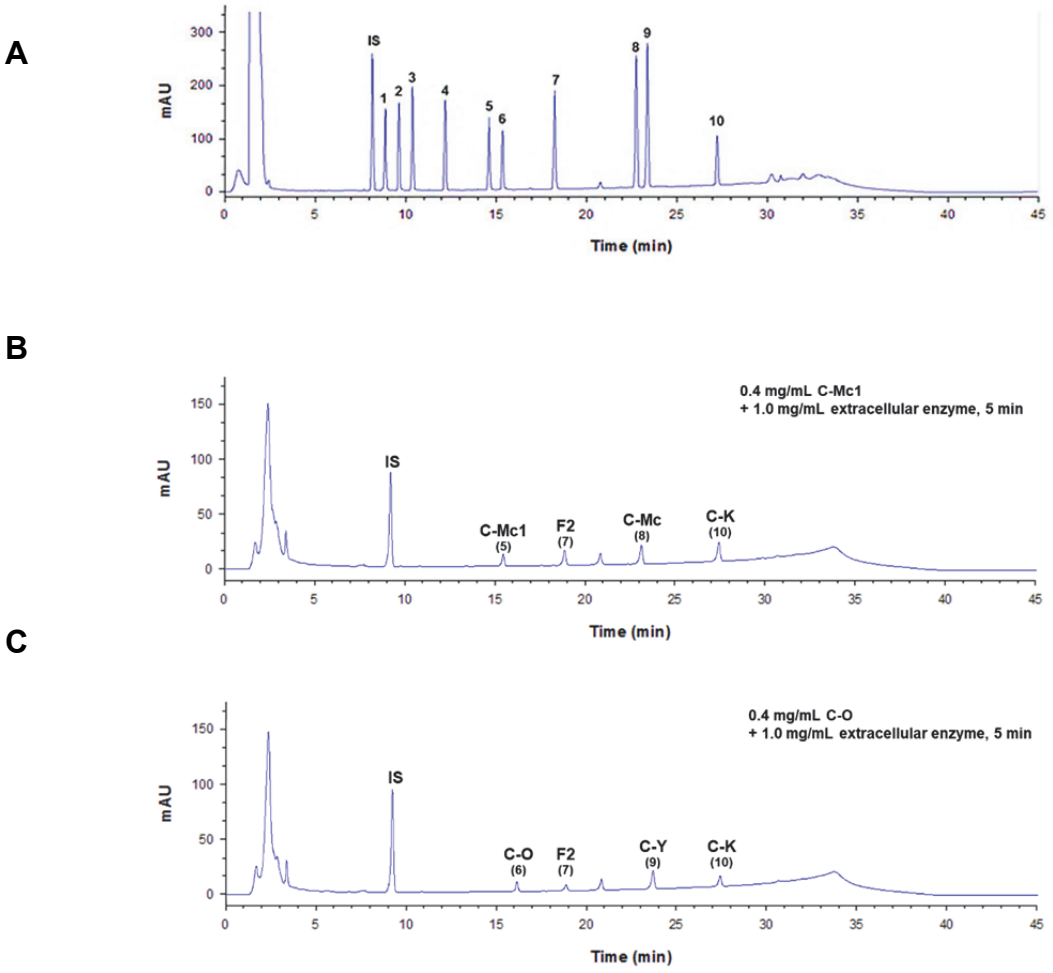
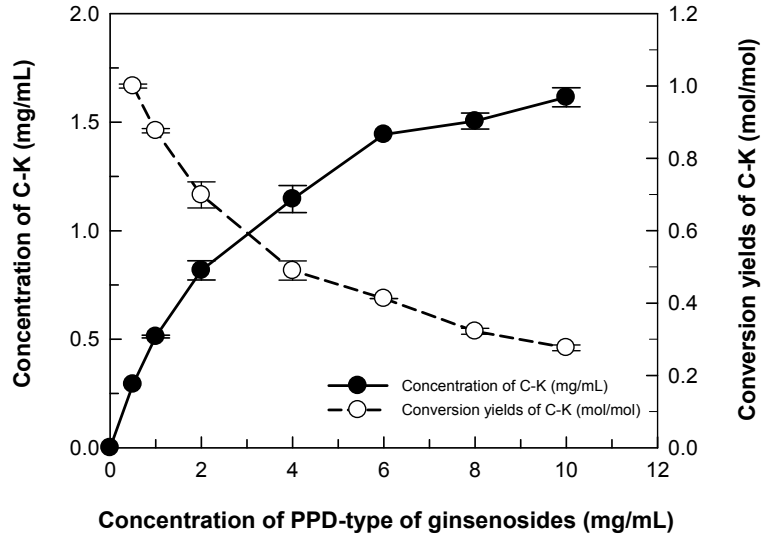


Fig. S4. High-performance liquid chromatography profiles for the conversion of compound O and compound Mc1 into compound K by extracellular enzyme from *Aspergillus niger* for 5 min with PPD-type ginsenoside standards. (A) High-performance liquid chromatography (HPLC) profile for ginsenoside standards. (B) HPLC profile for reaction products of compound Mc1 (C-Mc1). (C) HPLC profile for reaction products of compound O (C-O). The reactions were performed at 55°C for 5 min in 0.2 M citrate-phosphate buffer (pH 5.0) containing 0.4 mg/mL of C-Mc1 or C-O and 1.0 mg/mL extracellular enzyme. Internal standard (IS, digoxin); 1, Rb1; 2, Rc; 3, Rb2; 4, Rd; 5, C-Mc1; 6, C-O; 7, F2; 8, compound Mc (C-Mc); 9, compound Y (C-Y); and 10, compound K (C-K).

A



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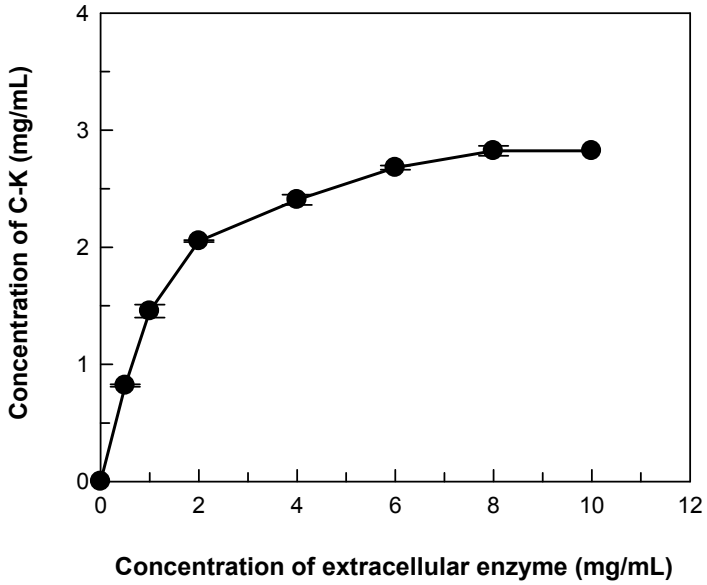

Fig. S5. Effects of the concentrations of (A) protopanaxadiol-type ginsenoside mixture from Korean ginseng (PPDKG) and (B) extracellular enzyme from *Aspergillus niger* on compound K production. (A) The reactions were performed at 55°C for 12 h in 0.2 M citrate-phosphate buffer (pH 5.0) containing 1.0 mg/mL extracellular enzyme by varying the concentration of PPDKG from 0.5 mg/mL to 10.0 mg/mL. (B) The reactions were performed at 55°C for 12 h

in 0.2 M citrate-phosphate buffer (pH 5.0) containing 6.0 mg/mL PPDKG by varying the concentration of extracellular enzyme from 0.5 mg/mL to 10.0 mg/mL. ●, Concentration of produced compound K (mg/mL); ○, Conversion yields of produced compound K (mol/mol). Data are expressed as the means of three experiments and the error bars represent standard deviations.

Table S1. Relative activity of extracellular enzymes from *A. niger* for aryl-glycosides.

Substrate[1]	**Specific activity** (μmol/min/mg)	**Relative activity** (%)
*o*NP[2]-*β*-D-glucopyranoside	0.5 ± 0.00	5.6 ± 0.03
*o*NP-*β*-D-xylopyranoside	4.2 ± 0.01	51.6 ± 0.14
*p*NP[3]-*α*-D-galactopyranoside	14.8 ± 0.33	181 ± 4.05
*p*NP-*β*-D-galactopyranoside	3.8 ± 0.04	46.4 ± 0.43
*p*NP-*α*-D-glucopyranoside	1.0 ± 0.00	12.0 ± 0.00
*p*NP-*β*-D-glucopyranoside	8.2 ± 0.07	100 ± 0.87
*p*NP-*α*-L-arabinofuranoside	18.9 ± 0.12	230 ± 1.45
*p*NP-*α*-L-arabinopyranoside	0.11 ± 0.00	1.3 ± 0.00
*p*NP-*β*-D-xylopyranoside	4.1 ± 0.01	50.4 ± 0.14
*p*NP-*β*-D-rhamnopyranoside	0.10 ± 0.01	1.3 ± 0.06

[1]Substrate concentration: 1 mM.

[2]*o*NP: *o*-nitrophenol

[3]*p*NP: *p*-nitrophenol