

Supporting information for

Characterization of urease from *Providencia* sp. LBBE and its application in degrading urea and ethyl carbamate in rice wine

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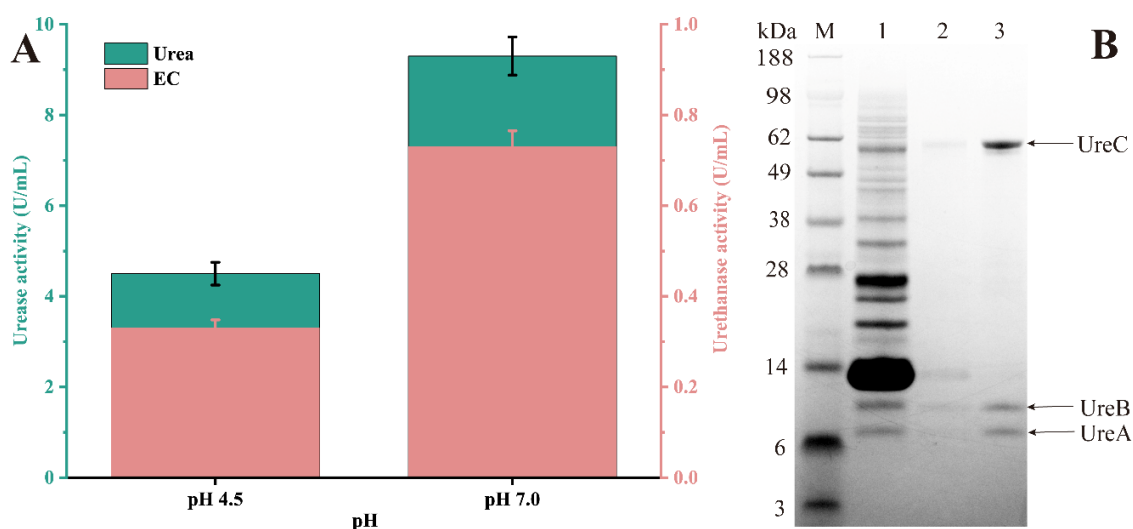


Figure S1 Heterologous expression and purification of recombinant Ps_Urease.

(A) Crude enzyme activity of Ps_Urease towards EC and urea. (B) SDS-PAGE analysis of recombinant Ps_Urease samples. M: protein molecular weight standard. Line 1: cell lysis supernatant. Line 2: cell lysis precipitation. Line 3: purified Ps_Urease.

Table S1 Enzymatic properties of bifunctional urease.

| Source | pH _{opt} for EC | pH _{opt} for urea | K _m for EC (mM) | K _m for Urea (mM) | pH Stability | Ethanol Stability | Urea elimination efficiency in alcoholic beverages | EC elimination efficiency in alcoholic beverages | Reference |
|--|--------------------------|----------------------------|----------------------------|------------------------------|--|--|---|--|-----------|
| <i>L. reuteri</i> CICC6124 | 5.0 | 3.0 | 41.32 ^a | 0.7147 ^a | More than 90.0% of its activity was retained after treatment at pH 4.5 or pH 5.0, and 37°C for 0.5 h. | More than 60% of its activity was retained in the presence of 15% (v/v) ethanol or after treatment with 15% ethanol at 37°C for 0.5 h. | Rice wine, 50 U/L enzyme, 95.8 % of urea was decomposed at 20 °C for 60 h. | No notable degradation of the EC was observed in the same condition. | [17, 18] |
| <i>B. paralicheniformis</i> ATCC 9945a | 7.0 | 5.0 | 1018 ^a | 118 ^a | Retaining more than 60.0% of its activity after treatment at pH 5.0 and 4°C for 6 h. | Retaining more than 70% of its activity in the presence of 15% (v/v) ethanol or after treatment with 15% (v/v) ethanol at 37°C for 2 h | Rice wine, 6000 U/L enzyme, 92.0 % of urea was decomposed at 37 °C for 20 h | No notable degradation of the EC was observed in the same condition. | [19] |
| <i>B. amyloliquefaciens</i> JP-21 | 6.0 | 6.0 | 207.07 ^b | 3.41 ^b | Retaining about 50.0% of its activity after treatment at pH 4.5 and 4°C for 6 h. | Retaining more than 50% of its activity after treatment with 20% (v/v) ethanol at 37°C for 2 h | Rice wine, 6000 U/L enzyme, 97.0 % of urea was removed at 37 °C for 50 h | 12% of EC was removed under the same condition. | [22] |
| <i>P. rettgeri</i> JNB815 | 5.0 | 4.5 | 386 ^c | 9.14 ^a | Retaining about 100.0% of its activity after treatment at pH 5.0 for 0.5 h | NP | NP | NP | [20, 21] |
| <i>Providencia</i> LBBE | sp. 7.0 | 7.5 | 515.6 ^a | 32.0 ^a | Retaining 41.3% and 59.4% of its activity after treatment at pH 4.5 and 5.0, respectively, at 4°C for 6 h. | Retaining more than 58% of its activity in the presence of 15% (v/v) ethanol or after treatment with 15% (v/v) ethanol at 37°C for 1 h, respectively | Rice wine, 6000 U/L enzyme, 49.8 % of urea was removed at 30 °C for 9 h. | No notable degradation of the EC was observed in the same condition. | This work |

Note: a, measured at pH 4.5; b, measured at pH 6.0; c, measured at pH 5.0; NP, not reported.