

Supporting information for

Single-Layer HNb₃O₈ with Strong and Nearby Lewis and Brønsted Acid Sites Boosts Amide Bond Hydrolysis for Urease Mimicking

Guohan Sun,^{a,#} Bo Yuan,^{a,#} Xinyu Wu,^a Shun Ying Lau,^a Linyuan Tian,^a Jung-Hoon Lee,^{b,*} Keizo Nakagawa^c, Yung-Kang Peng^{a*}

^aDepartment of Chemistry, City University of Hong Kong, Hong Kong, Hong Kong SAR.

^bDepartment of Chemistry, Soonchunhyang University, Asan, Korea.

^cResearch Center for Membrane and Film Technology, Graduate School of Science, Technology and Innovation, Kobe University, Kobe, Japan.

#Equal contribution to this work.

*Correspondence: ykpeng@cityu.edu.hk; jhlee67@sch.ac.kr

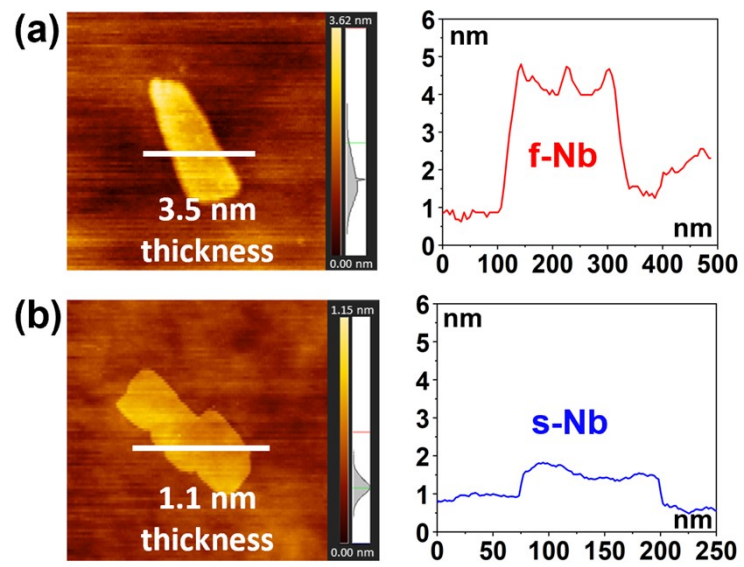


Figure S1. AFM images and thickness analysis of (a) f-Nb and (b) s-Nb.

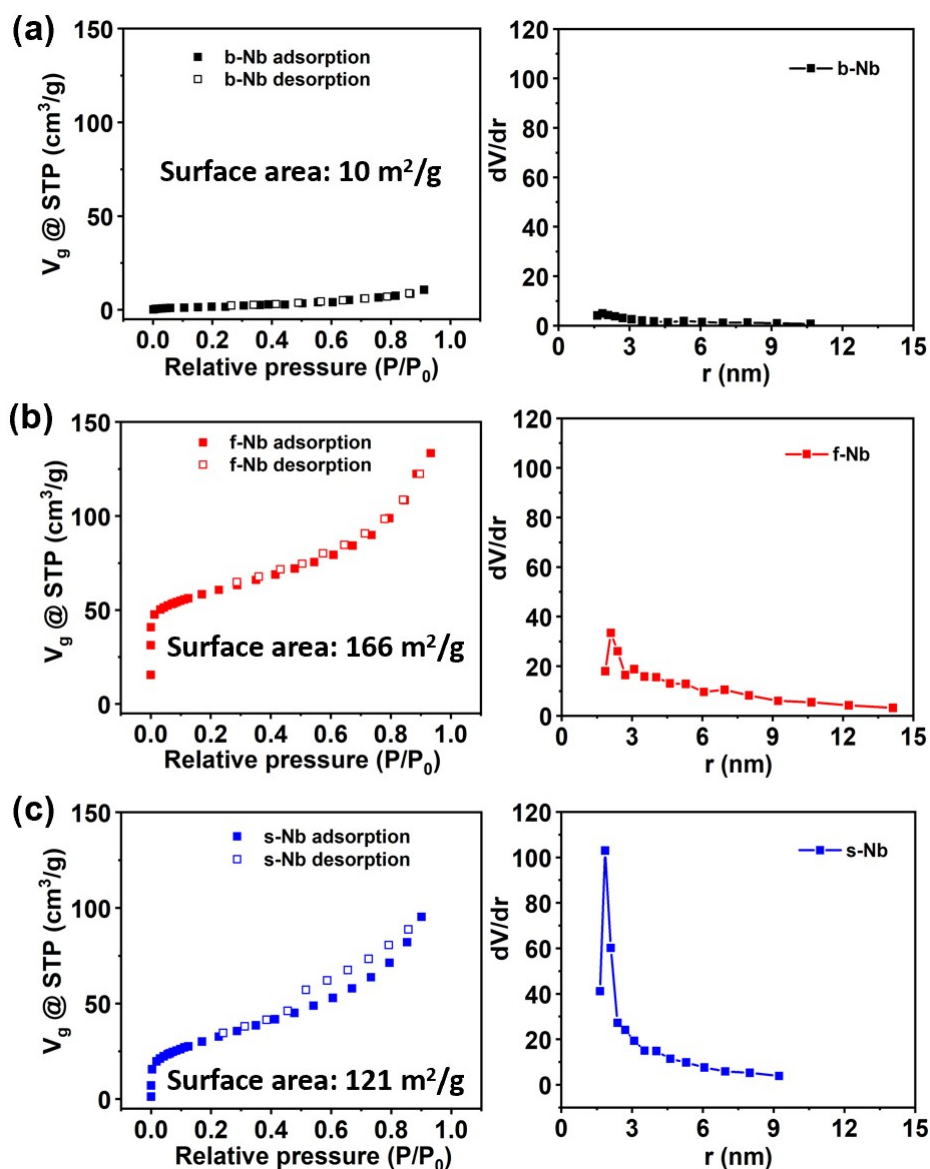


Figure S2. N_2 adsorption-desorption isotherms (left) and pore size distribution (right) of (a) b-Nb, (b) f-Nb, and (c) s-Nb samples.

According to IUPAC classification^[1], Type II isotherm observed for b-Nb and f-Nb here implies that both samples are nonporous powders or with pore diameters larger than micropores. While a hysteresis loop and a pore size at around 2 nm can be seen on s-Nb suggests a serious re-stacking between s-Nb samples.

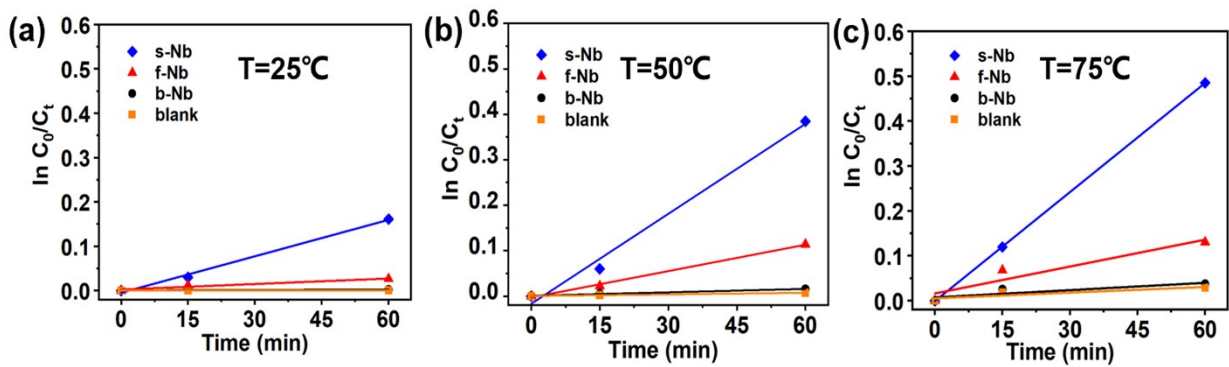


Figure S3. Plots of kinetic study over HNb_3O_8 samples at elevated temperatures: (a) 25°C , (b) 50°C , and (c) 75°C . See **Table S2** for the obtained k constants.

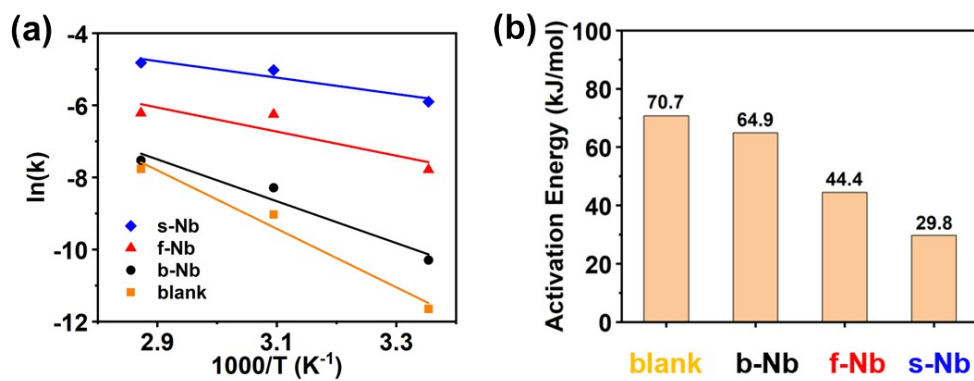


Figure S4. (a) Arrhenius plots for HNb_3O_8 samples and (b) the comparison of their activation energies ($E_{A(\text{acetamide})}$) with blank.

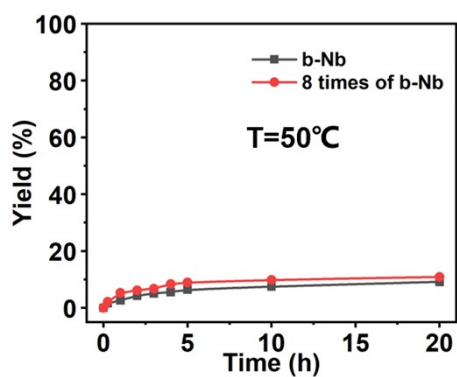


Figure S5. Acetamide hydrolysis at 50 °C over 8 times amounts of b-Nb as a function of time.

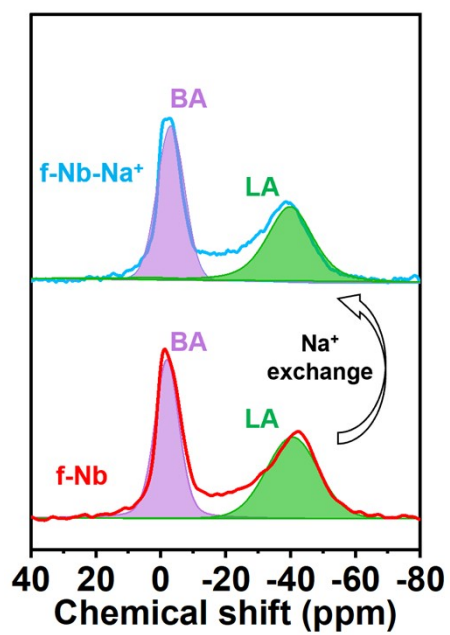


Figure S6. The comparison of TMP-³¹P NMR spectra of f-Nb-Na⁺ with that of f-Nb.

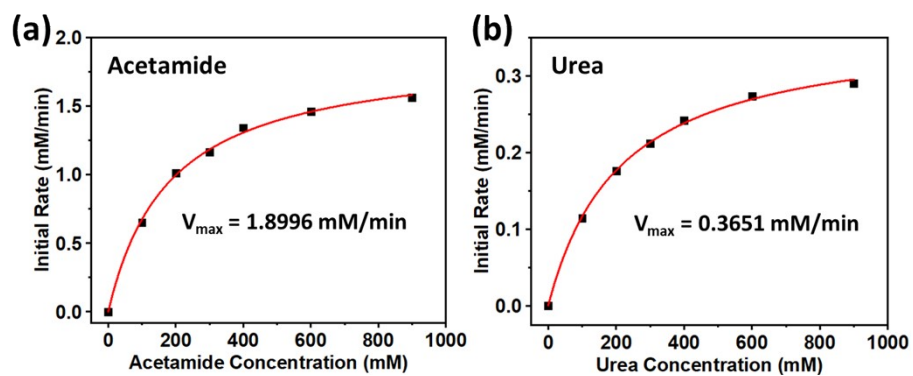


Figure S7. The concentration effect of (a) acetamide and (b) urea for s-Nb sample at 50 °C.

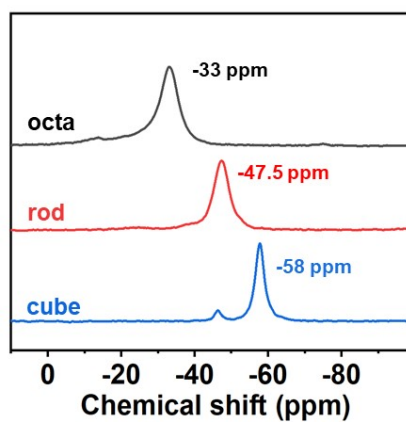


Figure S8. TMP-³¹P NMR of CeO₂ with different morphologies^[2].

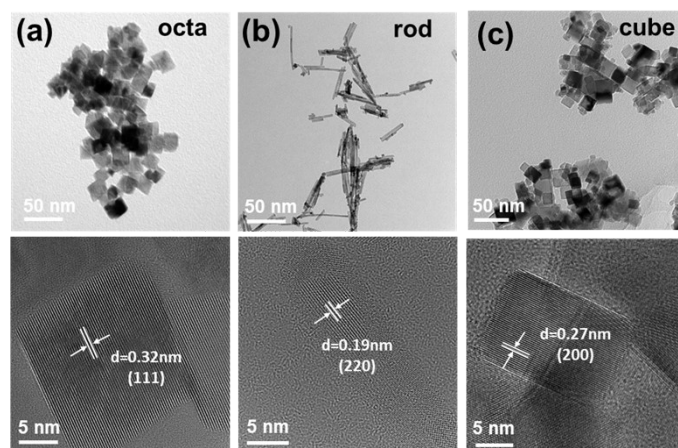


Figure S9. TEM and high-resolution TEM of (a) octahedron-, (b) rod-, and (c) cube-shaped CeO₂ samples.

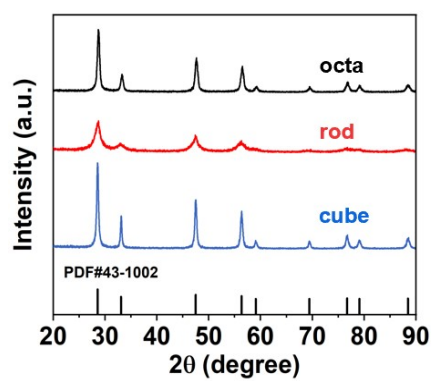


Figure S10. XRD spectra of CeO₂ samples.

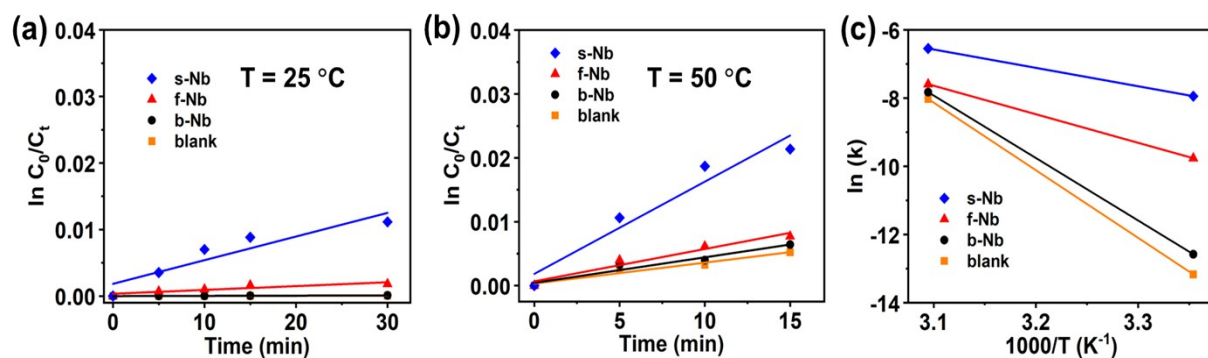


Figure S11. (a, b) Plots of kinetic study over HNb_3O_8 samples at elevated temperatures for urea (See **Table S4** for the obtained k constants) and (c) their Arrhenius plots.

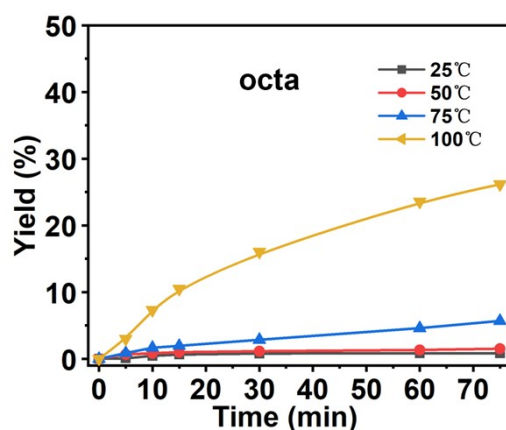


Figure S12. Urea hydrolysis at elevated temperatures over octahedral CeO_2 sample.

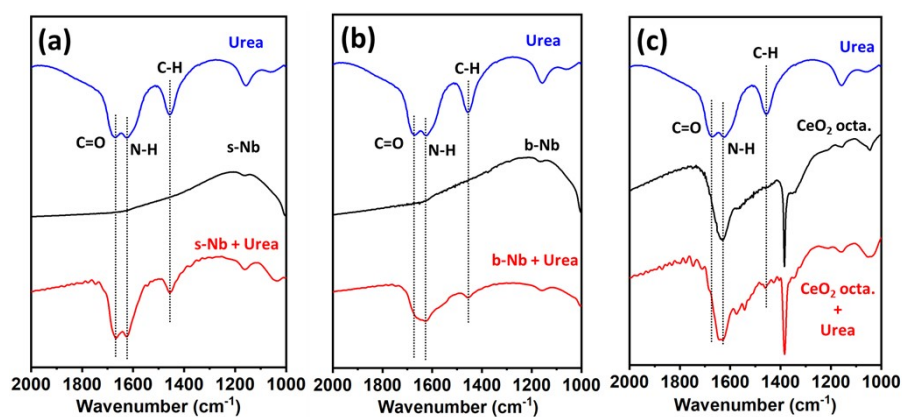


Figure S13. Infrared spectral comparison of (a) s-Nb, (b) b-Nb, and (c) CeO₂ octa. samples before and after urea adsorption.

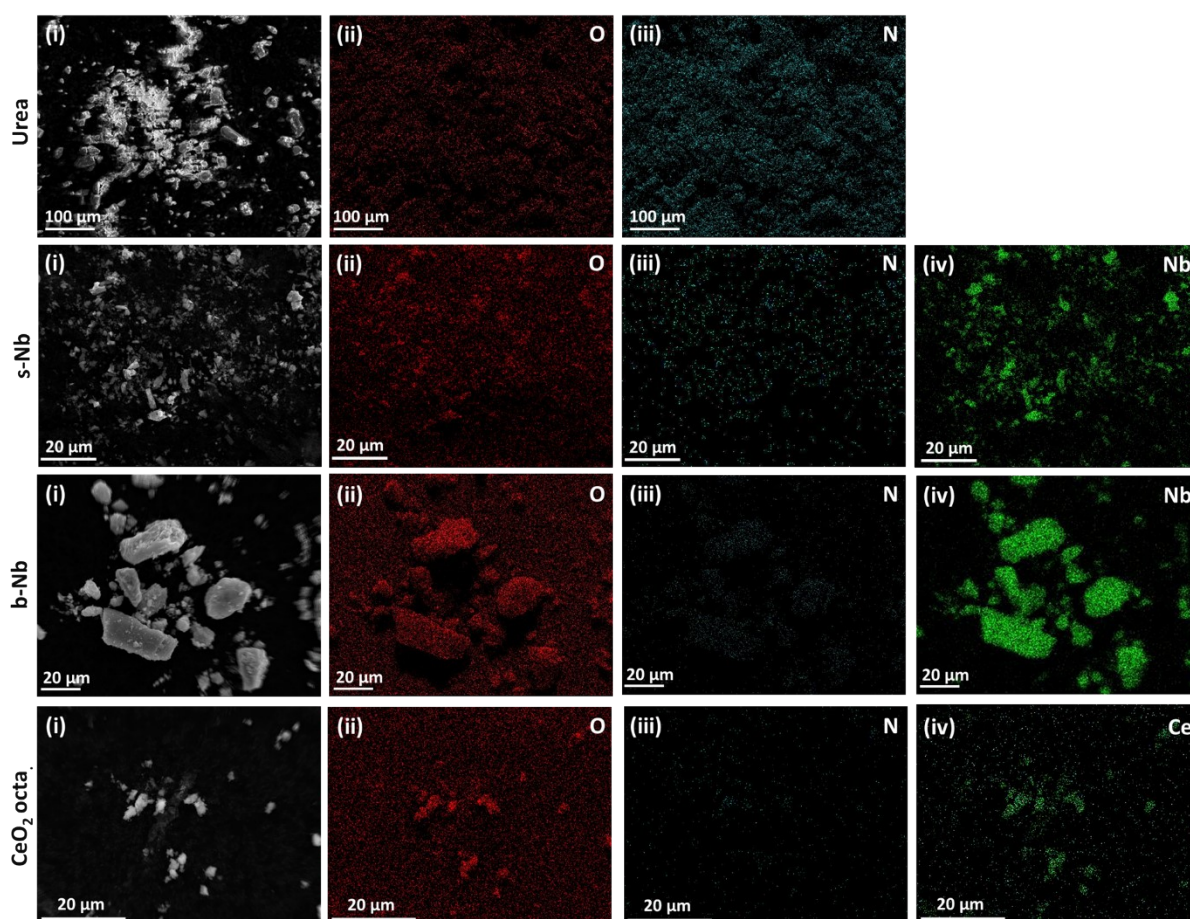


Figure S14. The scanning electron microscopy (i) image and elemental (ii) O, (iii) N, (iv) Nb/Ce mapping of urea and urea-adsorbed s-Nb, b-Nb, and CeO₂ octa. samples.

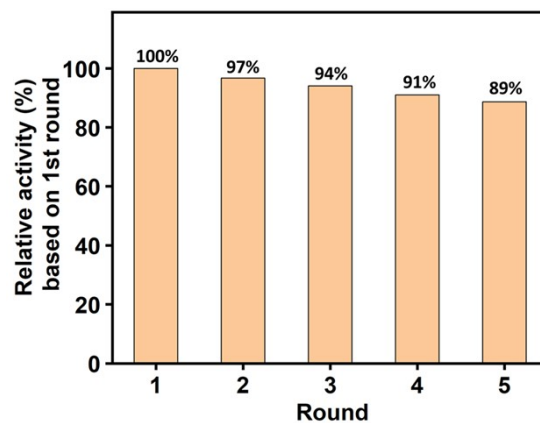


Figure S15. (a) The recyclability test for s-Nb in urea hydrolysis at 50 °C for five consecutive rounds. The activity was analyzed after 75 mins of reaction and the activity of the first round was set as 100% for comparison.

Table S1. The time-dependent yield (%) of NH₃ from acetamide hydrolysis over HNb₃O₈ samples at elevated temperatures.

Sample	Time (h)	0.25	1	2	3	4	5	10	20
	Temperature (°C)								
blank	25	0.00014	0.04878	0.35173	1.28883	1.45576	1.61853	2.10418	2.25249
	50	0.25034	0.73454	1.2669	1.52771	1.60133	1.78039	2.31459	2.47774
	75	1.90526	2.86776	3.35173	3.58883	3.65576	3.81853	4.30418	4.45249
	100	3.68126	4.34776	4.83173	5.06883	5.13576	5.29853	5.78418	5.93249
b-Nb	25	0.11600	0.22177	0.84983	2.02536	3.01495	4.0493	5.90798	8.92722
	50	0.58013	1.55674	2.51349	3.10885	5.47559	6.24916	8.53267	10.12682
	75	2.55023	3.68052	3.68015	4.28601	5.82921	8.62594	11.07078	12.74352
	100	4.40368	5.56034	7.12067	8.23892	8.71393	9.02731	10.40835	15.36186
f-Nb	25	1.17300	2.61024	9.26354	14.53576	17.57849	20.24923	22.66994	23.71737
	50	2.22063	10.78190	19.23142	23.35812	25.25948	25.79973	30.43695	32.20002
	75	6.60734	12.25780	20.43735	22.40198	27.43096	30.89979	47.34301	53.56032
	100	16.02377	19.72947	26.25759	26.72187	27.93464	30.21385	36.71946	56.85613
s-Nb	25	2.95023	14.87722	20.60297	25.53202	27.20023	30.87591	40.55553	49.52687
	50	5.82231	31.92427	38.19994	41.78398	44.01576	46.73715	55.86686	65.62126
	75	11.30043	38.44515	51.91944	56.45697	63.26165	65.90023	73.48153	77.87113
	100	33.30041	42.00689	51.97733	61.86437	64.9194	66.18161	80.89721	95.04683

Table S2. The k constants obtained for HNb₃O₈ samples at elevated temperatures in acetamide hydrolysis.

Sample	k constant (min ⁻¹)		
	25°C	50°C	75°C
blank	8.7503E-06	1.1949E-04	4.2388E-04
b-Nb	3.3897E-05	2.5178E-04	5.4051E-04
f-Nb	4.1422E-04	1.9300E-03	2.0000E-03
s-Nb	2.7400E-03	6.5900E-03	8.0900E-03

Table S3. The comparison of activity of homo-/hetero-catalysts reported in acetamide and urea hydrolysis.

Entry	Substrate	Catalyst	Substrate Concentration	Amount of catalyst	Temp. (°C)	pH	Reported Activity	Rate (mM s ⁻¹)	Reference
1	Acetamide	Nb ₂ O ₅	1 mmol acetamide in 5 mmol water	50 mg	100	-	^[a] Initial rate: 1.63 mol g ⁻¹ h ⁻¹	4.538 x 10 ⁻²	Catal. Sci. Technol., 2021, 11, 1949–1960
2		Single layer HNb ₃ O ₈ (s-Nb)	0.3 M	1 mg/mL	100	-	^[b] k constant: 2.2 x 10 ⁻² min ⁻¹	1.11 x 10 ⁻¹	This work
3	Urea	Urease	0.02 M	8.39 x 10 ⁻⁶ mM	25	7.4	^[c] 2.2 x 10 ⁻² mM s ⁻¹	2.2 x 10 ⁻²	Nanoscale, 2018, 10, 13074
4		CeO _{2-x} nanorods	0.3 M	1 mg/mL	25	7.4	^[c] 0.9 x 10 ⁻⁴ mM s ⁻¹	0.9 x 10 ⁻⁴	
5		Single layer HNb ₃ O ₈ (s-Nb)	0.3 M	1 mg/mL	25	-	^[b] k constant: 3.554 x 10 ⁻³ min ⁻¹	1.777 x 10 ⁻³	This work
6		[Co(tren)Cl ₂]Cl (molecular catalyst)	0.01 M	4.5 x 10 ⁻⁴ M	45	9.0	^[b] k constant: 3.5 x 10 ⁻³ s ⁻¹	3.5 x 10 ⁻⁴	Eur. J. Inorg. Chem. 2018, 5058–5067
7		Ni ₂ L ₂ (OAc) ₂ (H ₂ O) (molecular catalyst)	0.05 M	2 x 10 ⁻³ M	25	-	^[b] k constant: 13.73 s ⁻¹	686.5	Dalton Trans., 2021, 50, 4848–4858

[a] The initial rate (mol g⁻¹ h⁻¹) was converted to Rate (mM s⁻¹) by multiplying the catalyst weight (50 mg).

[b] The k constant (s⁻¹ or min⁻¹) was converted to Rate (mM s⁻¹) using “Rate = k [substrate]” as hydrolysis is a pseudo-first-order reaction.

[c] Extracted from the linear concentration-dependent range in the corresponding Michaelis–Menten plot.

Table S3 compares the activity of homo-/hetero-catalysts in acetamide (entries 1 and 2) and urea (entries 3–7) hydrolysis. Since urea can undergo the elimination pathway and thermal decomposition at temperature > 75 °C, we only considered catalysts used at low temperature (< 50 °C) for comparison. Based on the calculated rate (mM s⁻¹), s-Nb outperforms not only the reported solid catalysts Nb₂O₅ (entry 1) and CeO_{2-x} nanorods (entry 4) in acetamide and urea hydrolysis, respectively, but also the molecular catalyst, [Co(tren)Cl₂]Cl (entry 6), in the latter reaction. While this mimetic is still about 10 times less active than urease at 25 °C (entry 3) as discussed in the maintext. It is interesting to note that the other molecular catalyst, Ni₂L₂(OAc)₂(H₂O) (entry 7), exhibits a calculated rate (686.5 mM s⁻¹) much higher than that of urease (2.2 x 10⁻² mM s⁻¹). However, the authors found that this catalyst does not follow the hydrolysis pathway as urease rather they tend to decompose urea into ammonia and cyanate (i.e., the elimination pathway) at 25 °C.

Table S4. The time-dependent yield (%) of NH₃ from urea hydrolysis over HNb₃O₈ samples at elevated temperatures.

Sample	Time (min)	5	10	15	30	60	75
	Temperature (°C)						
blank	25	0.00100	0.00191	0.00288	0.00575	0.00661	0.00679
	50	0.25945	0.32725	0.52228	0.95945	0.99783	1.03774
	75	0.39292	0.65458	0.94988	1.8569	3.60069	4.67533
	100	2.00011	4.31436	6.49030	12.04813	18.58090	22.16311
b-Nb	25	0.00178	0.00345	0.00519	0.01037	0.01160	0.01168
	50	0.32693	0.39929	0.64055	1.01019	1.19672	1.28188
	75	0.51503	0.77956	1.13443	2.52887	4.86926	6.06303
	100	2.63528	4.99764	8.06326	15.18513	25.35662	26.92249
f-Nb	25	0.07142	0.10237	0.16344	0.18135	0.18191	0.18369
	50	0.39828	0.61406	0.76758	1.23505	1.25563	1.26975
	75	0.59993	1.09745	1.54793	2.84928	6.12511	7.39698
	100	3.94407	7.08741	10.50000	18.36034	28.18250	30.96721
s-Nb	25	0.35365	0.69846	0.87996	1.10862	1.82846	2.19666
	50	1.05517	1.85113	2.11393	2.49239	3.90141	4.38616
	75	3.17222	4.3458	4.94694	6.72607	10.08458	11.40415
	100	6.03636	9.99773	13.70618	22.82206	39.54293	44.39367

Table S5. The k constants obtained for HNb₃O₈ samples at temperatures < 75°C in urea hydrolysis.

Sample	k constant (min ⁻¹)	
	25°C	50°C
blank	1.9117E-6	3.2778E-4
b-Nb	3.4505E-6	4.0009E-4
f-Nb	5.7604E-5	5.0571E-4
s-Nb	3.5540E-4	1.4412E-3

References

- 1 K. S. W. Sing, D. H. Everett, R. A. W. Haul, L. Moscou, R. A. Pierotti, J. Rouquerol, T. Siemieniewska, *Pure & Appl. Chem.*, 1985, **57**, 603.
- 2 Z. Tan, G. Li, H. L. Chou, Y. Li, X. Yi, A. H. Mahadi, A. Zheng, S. C. E. Tsang and Y. K. Peng, *ACS Catal.*, 2020, **10**, 4003.