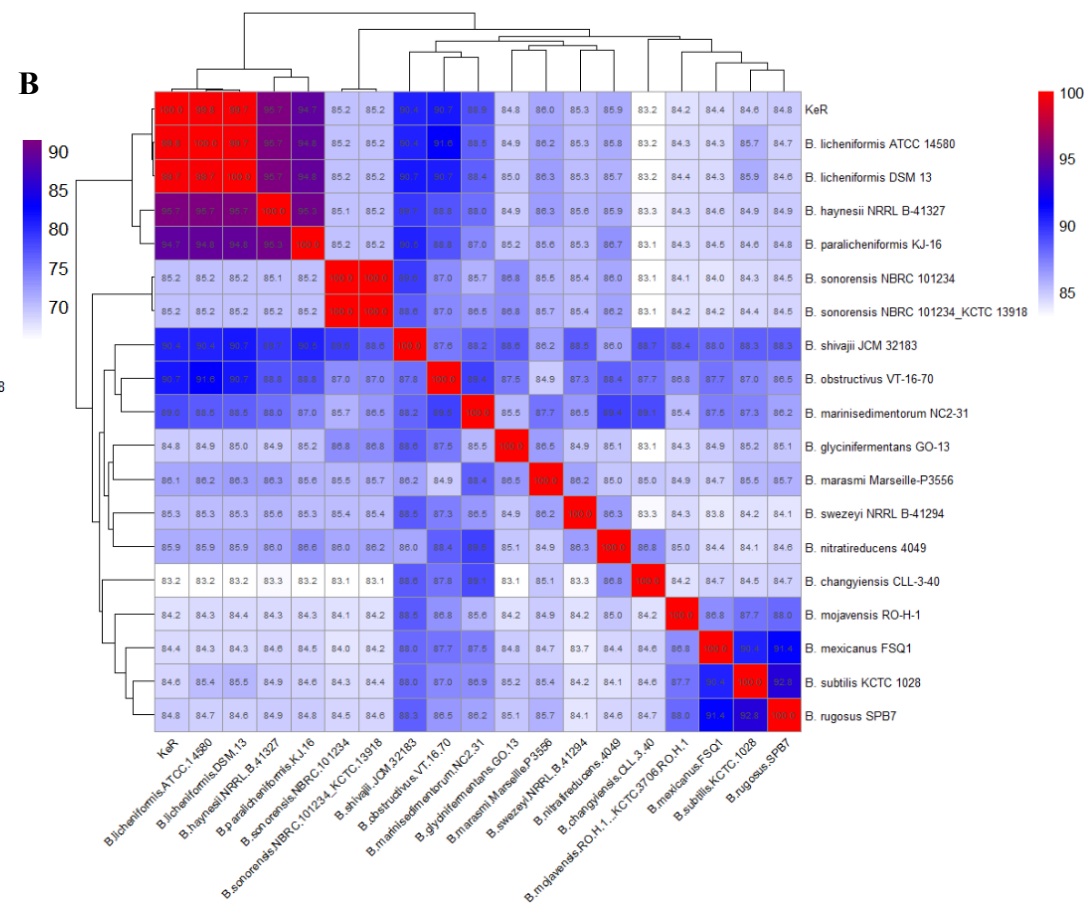
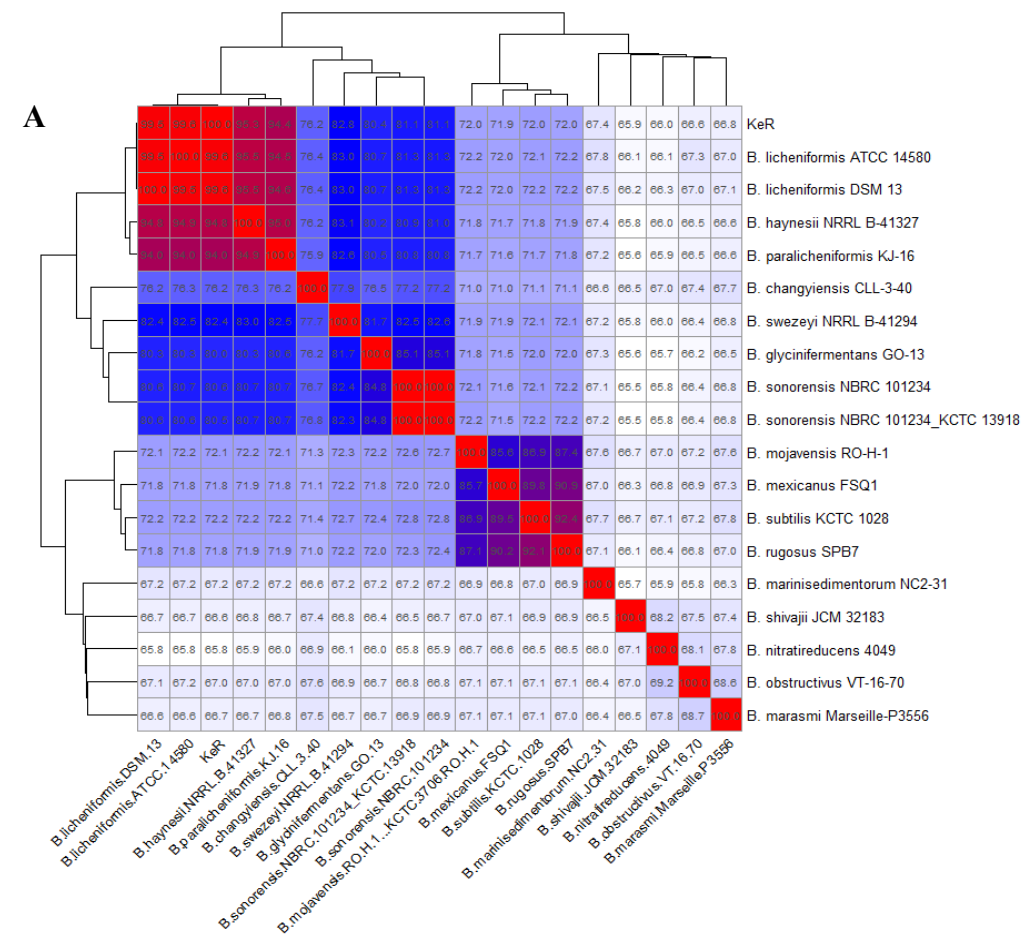
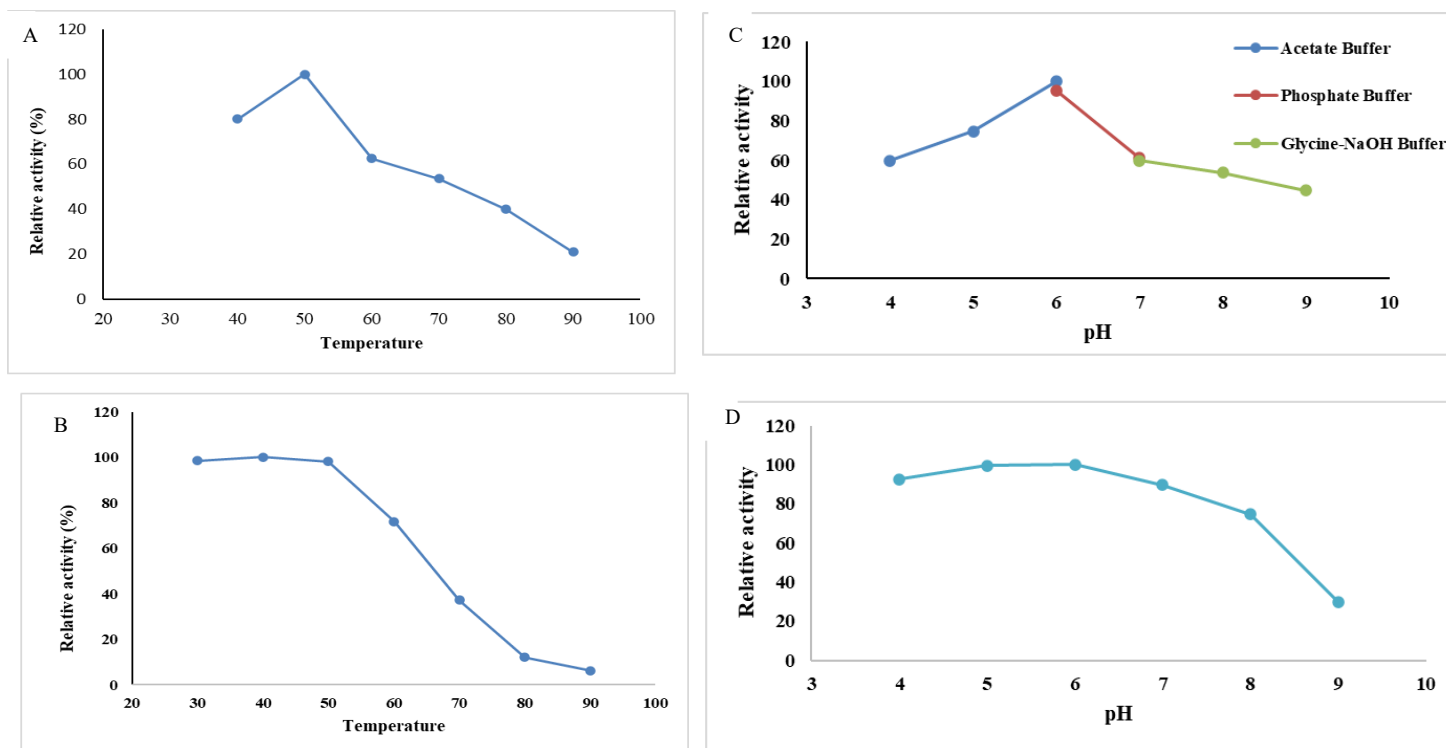




Supplementary figure 1. Pairwise average nucleotide identity (ANI) comparison between strain KeR9 and *Bacillus* genomes retrieved from TYGS results: (A) ANIb percentage identity and (B) ANIm percentage identity. ANI values were calculated using JSpeciesWS and visualized in R (v4.5.0) with the pheatmap package.



Supplementary figure 2. Effect of temperature and pH on the activity and stability of the purified α -amylase.

(A) Effect of temperature on α -amylase activity. (B) Effect of pH on α -amylase activity measured in acetate (pH 4.0–6.0), phosphate (pH 6.0–7.0), and glycine–NaOH (pH 7.0–9.0) buffers. (C) Thermal stability of α -amylase following incubation at different temperatures. (D) pH stability of α -amylase following incubation in buffers of different pH values. Enzyme activity was expressed as relative activity (%), with the maximum activity under each assay condition defined as 100%.

Supplementary table 1. Detection of Amylase activity from KR9, AT6 and AST14 strains

Strains	The diameter of the hydrolyzed zones (mm)	Enzymatic activity (mU/mL).
<i>Bacillus licheniformis</i> KeR9	19±0.2	331 ± 35
<i>Bacillus licheniformis</i> AT6	11±0.3	178±15
<i>Bacillus licheniformis</i> AS14	7±0.4	126±27

Supplementary table 2. Pairwise dDDH values (d4) between *Bacillus* strain KeR9 and type-strain genomes retrieved from the TYGS analysis, along with its corresponding confidence intervals (C.I.).

Subject	d4	C.I. d4
<i>Bacillus licheniformis</i> ATCC 14580	98.1	[97.2 -98.7]
<i>Bacillus licheniformis</i> DSM 13	98.1	[97.2 -98.7]
<i>Bacillus haynesii</i> NRRL B-41327	64.1	[61.2 -66.9]
<i>Bacillus paralicheniformis</i> KJ-16	58.0	[55.2 -60.7]
<i>Bacillus shivajii</i> JCM 32183T	37.1	[34.7 -39.7]
<i>Bacillus obstructivus</i> VT-16-70	30.2	[27.8 -32.7]
<i>Bacillus swezeyi</i> NRRL B-41294	26.2	[23.8 -28.7]
<i>Bacillus sonorensis</i> KCTC13918	24.3	[22.0 -26.8]
<i>Bacillus sonorensis</i> NBRC 101234	24.3	[22.0 -26.8]
<i>Bacillus glycinifermentans</i> GO-13	23.5	[21.2 -25.9]
<i>Bacillus nitratreducens</i> 4049	21.3	[19.1 -23.8]
<i>Bacillus marinisedimentorum</i> KCTC 33721	20.9	[18.6 -23.3]
<i>Bacillus marasmi</i> Marseille-P3556 T	20.7	[18.5 -23.1]
<i>Bacillus changyiensis</i> CLL-3-40T	20.3	[18.1 -22.7]
<i>Bacillus mojavensis</i> RO- H-1	18.8	[16.7 -21.2]
<i>Bacillus subtilis</i> ATCC 6051	18.7	[16.5 -21.0]
<i>Bacillus rugosus</i> SPB7	18.4	[16.3 -20.8]
<i>Bacillus mexicanus</i> FSQ1	18.1	[15.9 -20.4]

Supplementary table 3. Optimal factor levels for α -amylase production by *Bacillus licheniformis* KeR9 predicted by the Doehlert design and RSM

Parameter	Optimal Value
Starch (carbon source)	8.4718 g/L
KH ₂ PO ₄	1.0837 g/L
MgSO ₄ ·7H ₂ O	0.1842 g/L
Agitation speed	108.91 rpm
pH	7.04

Supplementary table 4. Relative activity of the purified α -amylase in the presence of various metal ions, surfactants, inhibitors, and other chemical reagents (10 mM).

Material (10mM)	Relative Activity (%)
None	100 $\pm$ 0.50
NaCl	126 $\pm$ 0.75
KCl	106 $\pm$ 1.01
MgCl ₂	118 $\pm$ 2.37
MnCl ₂	130 $\pm$ 1.20
CaCl ₂	135 $\pm$ 1.85
CuSO ₄	89 $\pm$ 1.54
FeSO ₄	114 $\pm$ 0.25
ZnSO ₄	68 $\pm$ 1.87
SDS	95 $\pm$ 2.70
Urea	98 $\pm$ 1.15
EDTA	72 $\pm$ 2.45
Triton X-100	92 $\pm$ 0.25
β -mercaptoethanol	93 $\pm$ 1.69