



Supplementary Table

Biological properties and potential probiotic bacteria isolated from honey collected in Chiang Mai, Thailand

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Supplementary Tables

Table S1 Molecular identification of honey isolates from each honey sample

Code	Type	Food source	Numbers of isolates (with different colony morphology)	Identification based on 16S rRNA gene sequencing (% identity)
SH1	Stingless bee honey	Wildflowers	2* (3)*	<i>Bacillus subtilis</i> , <i>Bacillus tequilensis</i> (100%) <i>Bacillus subtilis</i> , <i>Bacillus tequilensis</i> (97.76%)
SH2	Stingless bee honey	Wildflowers	4* (5)*	<i>Priestia aryabhatai</i> (100%) <i>Bacillus megaterium</i> (100%) <i>Priestia megaterium</i> (100%) <i>Priestia megaterium</i> (99.79%)
SH3	Stingless bee honey	Longan and Wildflowers	2* (4)*	NA <i>Bacillus stercoris</i> , <i>Bacillus spp.</i> 99.59%
SH4	Stingless bee honey	Wildflowers	4* (5)*	<i>Bacillus spp.</i> (85.49%) <i>Bacillus subtilis</i> (100%) <i>Bacillus licheniformis</i> , <i>Bacillus spp.</i> (100%) NA
HH5	Honeybee honey	Wildflowers	1* (1)*	<i>Bacillus velezensis</i> (100%)
HH6	Honeybee honey	Wildflowers	0* (0)*	-
HH7	Honeybee honey	Longan	2* (3)*	<i>Bacillus spp.</i> (i.e., <i>Bacillus aerius</i> , <i>Bacillus altitudinis</i> , <i>Bacillus stratosphericus</i> , <i>Bacillus pumilus</i> , <i>Bacillus aerophilus</i>) (100%) <i>Priestia megaterium</i> , <i>Bacillus spp.</i> (99.76%)
HH8	Honeybee honey	Wildflowers	0* (0)*	-

ND = unable to assign; * = numbers of isolates with different colony morphology on MRS agar plates (microaerobic environment); ♦ = numbers of isolates with products based on amplification using universal 16S rRNA primers.

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Table S2 Auto-aggregation and cell surface hydrophobicity evaluation

Strain tested	% Auto-aggregation			% MATS
	1 hr	2 hr	3 hr	
<i>Escherichia coli</i>	8.27±5.74 ^{de}	11.53±2.62 ^d	21.82±2.31 ^e	47.71±1.38 ^b
H001 (<i>Bacillus</i> sp.)	22.47±9.93 ^{bc}	48.16±1.01 ^a	60.60±0.76 ^{bc}	23.74±0.85 ^d
H002 (<i>Bacillus</i> sp.)	24.63±16.47 ^{bc}	33.01±2.24 ^c	63.46±1.22 ^b	20.53±4.15 ^d
H003 (<i>Bacillus</i> sp.)	43.30±0.51 ^a	48.45±0.46 ^a	89.69±0.09 ^a	80.90±0.80 ^a
H004 (<i>Bacillus</i> sp.)	28.68±1.16 ^b	38.87±0.99 ^b	59.25±0.66 ^c	26.50±4.30 ^{cd}
H005 (<i>Bacillus</i> sp.)	clumping	clumping	clumping	86.00±0.86 ^a
H006 (<i>Bacillus</i> sp.)	18.67±1.77 ^{bcd}	34.94±1.41 ^c	43.07±1.24 ^d	33.06±7.18 ^c
H007 (<i>Bacillus</i> sp.)	12.66±1.13 ^{cde}	12.66±1.13 ^d	17.80±1.07 ^f	19.79±5.92 ^d
<i>p</i> Value	< 0.001	< 0.0001	< 0.0001	< 0.0001

Clumping = cells clumped at bottom of test tubes from beginning; MATS = microbial adhesion to solvents, used to evaluate hydrophobicity and electron exchange.

^{a-f} = mean ± SD values with different lowercase superscripts have significantly ($p \leq 0.05$) different inhibition zones according to Duncan's multiple range test.

Table S3 Antibiotic susceptibility (based on disc diffusion assay), hemolytic phenotype, biofilm and spore formation of honey bacterial isolates (H001–H007)

Disk content (µg)	Antibiotic susceptibility											Hemolytic phenotype	Biofilm	Spore formation
	10 AMP	30 CHL	5 CIP	2 CLI	15 ERY	10 GEN	30 KAN	5 RIF	10 STR	30 TET	30 VAN			
isolates														
H001	S	R	S	S	S	S	S	S	S	R	S	γ-hemolytic	B	Sp
H002	S	S	S	S	S	S	S	S	S	S	S	γ-hemolytic		
H003	S	R	S	S	S	S	S	S	S	S	S	γ-hemolytic	B	
H004	S	S	S	S	S	S	S	S	S	S	S	α-hemolytic		
H005	S	S	S	S	S	S	S	S	S	S	S	γ-hemolytic		
H006	S	S	S	S	S	S	S	S	S	S	S	γ-hemolytic		Sp
H007	S	S	S	S	S	S	S	S	S	S	S	β-hemolytic		Sp

AMP = ampicillin; CHL = chloramphenicol; CIP = ciprofloxacin; CLI = clindamycin; ERY = erythromycin; GEN = gentamicin; KAN = kanamycin; RIF = rifampicin; STR = streptomycin; TET = tetracycline; VAN = vancomycin; S = sensitive; R = resistant; B = biofilm-forming post-stationary phase; Sp = spore-forming by induction.