

Whole Genome Analysis of *Pediococcus acidilactici* XJ-24 and Its Role in Preventing *Listeria monocytogenes* ATCC[®] 19115[™] Infection in C57BL/6 Mice

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> L. Replication, recombination and n M.Cell welf/membrane/envelope bi N.Gell motify O. Postnersiational modification, p

> > econdary metabolites biosynthesis.

R General function prediction on S:Function Unknown

T Signal Invinduction + U Intracel Ular trafficial V Defense mechanism

Extracelular structu

Mobilione: prophag

Nuclear structure Cytoskolotori ISS / RNA

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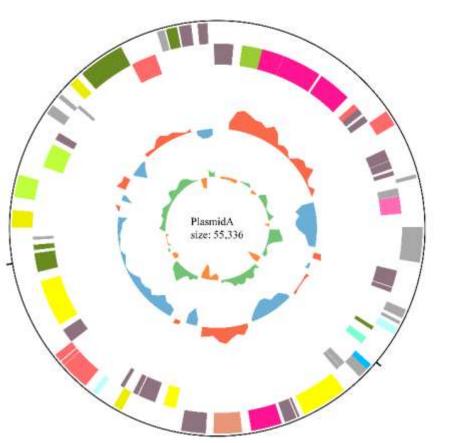


Introduction

Listeria monocytogenes is a Gram-positive foodborne pathogen that poses serious health risks, especially to vulnerable populations. It disrupts the intestinal barrier, causing systemic infection with a high mortality rate. Currently, there is no clinically approved vaccine for listeriosis, and severe cases depend largely on antibiotic therapy, which is increasingly challenged by rising antibiotic resistance. This underscores the urgent need for innovative, safe, and cost-effective strategies to prevent and treat listeriosis. Numerous studies have demonstrated that lactic acid bacteria hold great promise in preventing and alleviating *L. monocytogenes* infections, providing a strong foundation for the development of novel biotherapeutic approaches.

Objectives

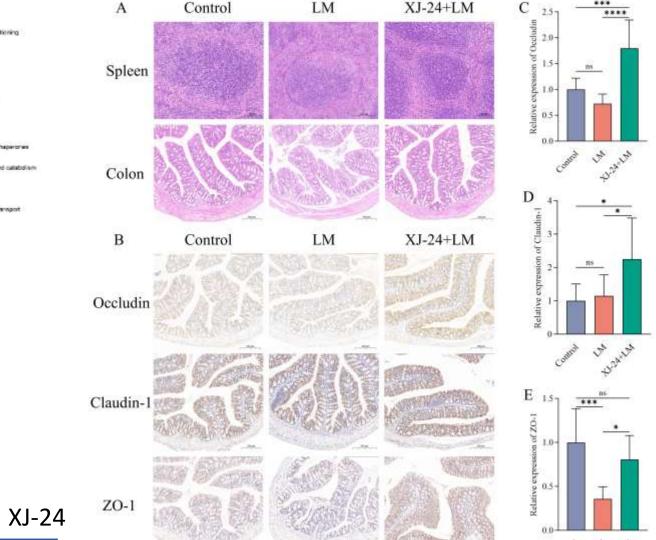
In our previous study, the strain Pediococcus acidilactici XJ-24

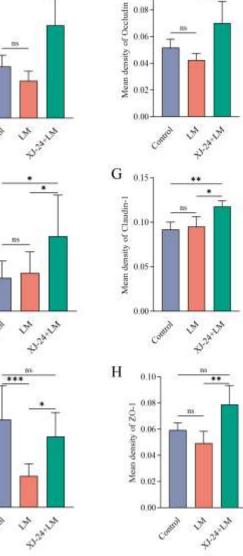


A circular genomic map of the *P. acidilactici* XJ-24 plasmid

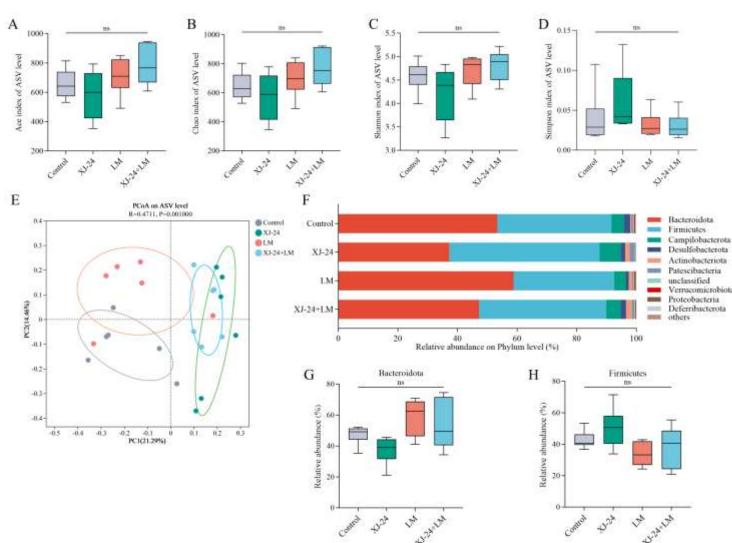
Probiotic characteristic-associated genes in *P. acidilactici* XJ-24

Gene locus	Gene name	Gene function
Universal stress family protein		
ACG4EF_00285	-	universal stress protein
ACG4EF_01025	-	universal stress protein
ACG4EF_01030	-	universal stress protein
ACG4EF_01595	-	universal stress protein
ACG4EF_03305	-	universal stress protein
ACG4EF_07140	-	universal stress protein
Proteases and chaperones		
ACG4EF_02365	clpP	ATP-dependent Clp endopeptidase proteolytic
ACG4EF_05205	hslU	ATP-dependent protease ATPase subunit HslU
ACG4EF_05210	hslV	ATP-dependent protease subunit HslV
ACG4EF_05335	-	ATP-dependent Clp protease ATP-binding subunit
ACG4EF_05700	сlpВ	ATP-dependent chaperone ClpB
ACG4EF_06340	clpX	ATP-dependent Clp protease ATP-binding
-	•	subunit ClpX
ACG4EF_07860	-	ATP-dependent Clp protease ATP-binding subunit
Temperature stress		
ACG4EF_02195	groES	co-chaperone GroES
	groL	chaperonin GroEL
	<u> </u>	heat-inducible transcriptional repressor
ACG4EF_04750	hrcA	HrcA
	dnaK	molecular chaperone DnaK
	dnaJ	molecular chaperone DnaJ
	cspA	cold-shock protein
ACG4EF_07950	-	cold-shock protein
Bile tolerance		
ACG4EF_01095	arcD	arginine-ornithine antiporter
ACG4EF_01470	nagB	glucosamine-6-phosphate deaminase
ACG4EF_08565	pyrG	CTP synthase
Acid tolerance		
ACG4EF_07220	atpC	ATP synthase subunit epsilon
ACG4EF_07225	atpD	ATP synthase subunit beta
ACG4EF_07230	atpG	ATP synthase subunit gamma
ACG4EF_07235	atpA	ATP synthase subunit alpha
ACG4EF_07240	atpH	ATP synthase F1 subunit delta
ACG4EF_07245	atpF	ATP synthase subunit B
ACG4EF_07250	atpE	ATP synthase subunit C
ACG4EF_07255	atpB	ATP synthase subunit A
ACG4EF_06250	-	Na+/H+ antiporter NhaC family protein
ACG4EF_09950	nhaC	Na+/H+ antiporter NhaC



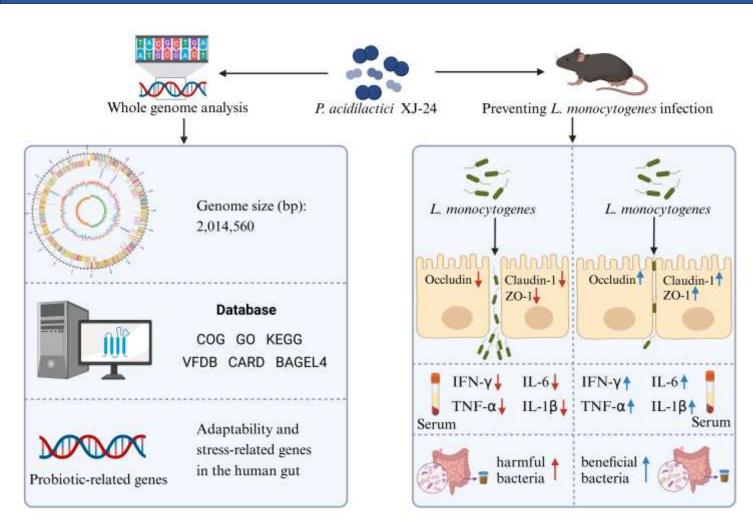


Effect of *P. acidilactici* XJ-24 on the histological structure and intestinal barrier integrity in the LM infected mice.



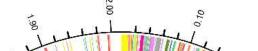
demonstrated strong *in vitro* inhibitory activity against *L. monocytogenes* and exhibited promising probiotic properties. However, the lack of complete whole-genome information for this strain and the unexplored *in vivo* preventive effects against *L. monocytogenes* infections hinder any further applications for it. This study aims to thoroughly analyze the genome of *P. acidilactici* XJ-24 using whole-genome sequencing, identify key functional genes, and explore its *in vivo* mechanisms and effectiveness in preventing *L. monocytogenes* infection

Graphical Abstract



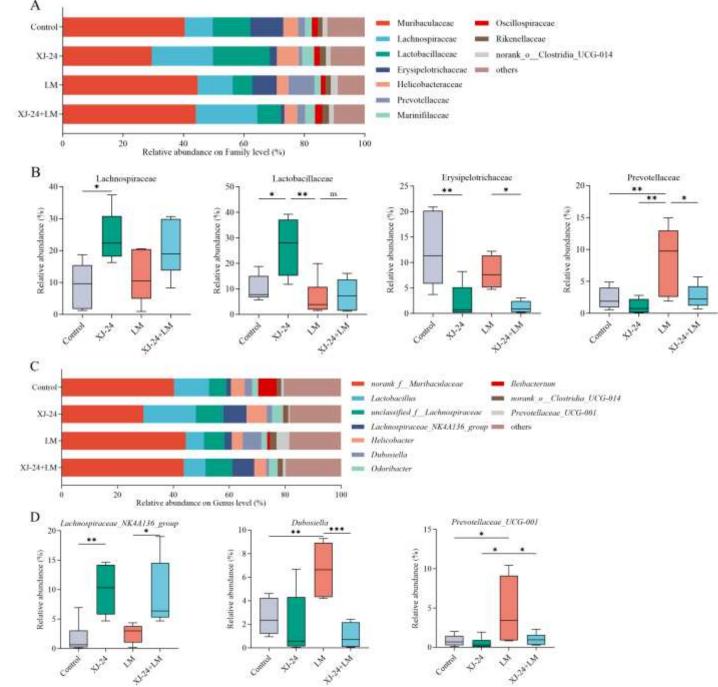
Results: Whole-genome analysis

GenBank accession numbers: CP172259-CP172260





Mice fecal microbiota diversity and relative abundance of the intestinal microbiome in each group at the phylum level

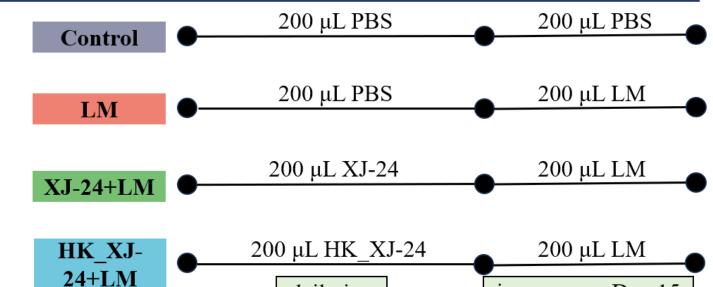


no acid transport and metabolis uclectide transport and metabolish Carbohydrate transport and metabolis enzyme transport and metabolish ipid transport and metabolis Replication, reco A:Cell wall/men N:Cell motility General function prediction on S:Function unknow T:Signal transduction mechanism P. acidilactici XJ-24 U:Intracellular trafficking, secretion, a size: 2,014,560 bp V:Defense mechan WExtracellular struct Mobilome: prophages. Nuclear structure Z:Cytoskeleto 16S rRNA 235_rRNA 55_rRNA

A circular genomic map of the *P. acidilactici* XJ-24 chromosome

anchor protein AmaP

Results: Animal study



daily i.g.i.g. once on Day 15prevenXJ-24 administration72 h LM infectionlevels,

Day 18

Day 15

The study design for the animal experiments

Adaption

Day -7

Day 0

Relative abundance of intestinal microbiome in each group at the family and genus level

Conclusion

Whole-genome analysis revealed that *P. acidilactici* XJ-24, which exhibits strong inhibitory activity against *L. monocytogenes*, possesses favorable probiotic properties and a high degree of safety. Animal experiments further demonstrated that *P. acidilactici* XJ-24 can prevent *L. monocytogenes* infection by reducing pro-inflammatory cytokine levels, alleviating tissue damage in the colon and spleen, enhancing the expression of tight junction proteins, and modulating the gut microbiota.

References

Acknowledgements

[1] Hu, W.; Zhou, S.; Ibrahim, A.; Li, G.; Awad, S.; Ramos-Vivas, J.; Kan, J.; Du, M. Whole Genome Analysis of *Pediococcus acidilactici* XJ-24 and Its Role in Preventing *Listeria monocytogenes* ATCC[®] 19115[™] Infection in C57BL/6 Mice. *Antibiotics* 2025, 14, 323.

A:RNA processing and modification

B:Chromatin structure and dynamic

[2] Hu, W.; Li, H.; Shi, Z.; Yang, X.; Yi, Z.; Zhou, S.; Kan, J.; Du, M. Screening and in situ inhibitory effects of probiotic lactic acid bacteria against Listeria monocytogenes. Food Ferment. Ind. 2024. Available online: https://doi.org/10.13995/j.cnki.11-1802/ts.039915

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