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Molecular detection of some virulence factors genes and biofilm formation in clinical isolates of *Yersinia enterocolitica*

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Abstract

Objective

Yersinia enterocolitica is a foodborne pathogen responsible for gastrointestinal infections, notably diarrhea, with its virulence attributed to genetic factors enabling host cell invasion, immune evasion, and environmental persistence. This investigation aimed to detect the 16S rRNA gene, evaluate virulence-related genes (*yadA* and *hreP*), and evaluate biofilm formation in *Y. enterocolitica* isolates from diarrheal samples gathered in Babylon Province, Iraq, to elucidate their genetic diversity and pathogenic potential.

Materials and Methods

A total of 200 stool samples from patients with diarrhea were gathered from multiple hospitals across Babylon Province. Isolates were identified applying standard biochemical experiments and polymerase chain reaction (PCR) targeting the 16S rRNA gene of *Y. enterocolitica*, followed by gene sequencing. Molecular diagnosis of virulence genes *yadA* and *hreP* was carried out applying specific primers. Biofilm formation was evaluated through quantitative assays to identify the isolates' capability to adhere to surfaces, reflecting their potential for persistence in clinical environments. Sequence data were analyzed applying multiple sequence alignment and phylogenetic tree construction to evaluate genetic relatedness.

Results

Genetic diversity was evaluated via 16S rRNA gene sequencing, revealing high conservation with six single nucleotide polymorphisms (SNPs), primarily transitions, located in non-coding or structurally neutral regions. Iraqi isolates demonstrated close relatedness among some strains but

phylogenetic diversity in others, clustering into three major clades: First Genetically Unified (FGU), First New Diverging (FND), and Final Unique (FU). Sequence alignment included two Iraqi sequences (GenBank: PV628219, PV628221) and 17 reference sequences from GenBank, with conserved nucleotides color-coded (A: green, T: red, G: purple, C: blue) and SNPs highlighted. Iraqi strains were highly similar to the reference sequence PV628226, with SNPs at positions 19 and 141 denoting close relatedness. In total, six SNPs were identified, with strains PV628219 and PV628221 each exhibiting four SNPs. Phylogenetic analysis affirmed diverse genetic profiles among Iraqi *Y. enterocolitica* isolates, with some strains closely related and others more divergent. Virulence genes *yadA* and *hreP* were detected in 87.5% of isolates (7/8), suggesting meaningful pathogenic potential. Biofilm formation assays revealed that most isolates exhibited moderate to strong biofilm production, denoting their capacity to persist in clinical settings.

Conclusions

This study highlights the genetic diversity and pathogenic potential of *Y. enterocolitica* isolates from Babylon Province, Iraq. The identification of SNPs in the 16S rRNA gene and the presence of virulence genes *yadA* and *hreP* in most isolates underscore their molecular basis for pathogenicity. The observed biofilm formation suggests a mechanism for environmental persistence, posing challenges for infection control. These results contribute to understanding the epidemiology and molecular characteristics of *Y. enterocolitica* in Iraq, informing targeted diagnostic and therapeutic strategies.

Keywords: 16S rRNA, biofilm formation, clinical isolates, genetic diversity, virulence genes

Paper Type: Research Paper.

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Introduction

Yersinia enterocolitica is a facultatively anaerobic bacterium which is one of the members Enterobacteriaceae (Doshi et al., 2024). It has a 69,704 base-pair Plasmid for Yersinia Virulence (pYV). The *Yersinia* genus has three pathogenic to humans including *Y. enterocolitica, Y. pseudotuberculosis* and *Y. pestis* (Al-Rawe et al., 2023). This bacterium is a meaningfu foodborne pathogen causes gastroenteritis, leading to high temperature, acute diarrhea and some cases lead to septicemia (Karlsson et al., 2021; Wang et al., 2021; Cheng et al., 2023). Post- infectious complications, like reactive arthritis, may also occur (Karlsson et al., 2021). Moreover, immunocompromised individuals, Infants and children are exclusively susceptible to infection (Meng et al., 2019). Contamination occurs through food sources, including dairy products, meat and vegetables. However, its global prevalence, *Y. enterocolitica* is frequently underestimated compared to well-known pathogens like *Escherichia coli* and *Salmonella spp.* (Ma et al., 2022). Under-reporting of yersiniosis is most attributed to the misconception that it poses a minimal food safety risk (Lou et al., 2025). Particularly, *Y. enterocolitica* can proliferate at low temperatures (4°C), making food contamination stored in refrigerators exclusively hazardous (Tian et al., 2021). This bacterium is diverse and classified into six biotypes (1A, 1B, 2, 3, 4, and 5) based on biochemical and immunological tests, furthermore, over 70 serotypes identified by O-antigen composition (Łada et al., 2023). The pYV plasmid encodes some virulence genes, like *yadA*, which generates an adhesin, and *yop*, which encodes outer membrane proteins responsible for , immune evasion, bacterial invasion and survival within the host environment (Al-Rawe et al., 2023). The pathogenicity of this bacterium is primarily driven by the expression of virulence genes, containing *ail*, *ystA*, *ystB*, *yadA*, and *virF*, which facilitate tissue invasion and evasion of the host immune response (Zheng et al., 2008). Additionally, biofilm formation enhances its persistence in diverse environments, increasing transmission and infection risks (Kote et al., 2023). Biofilms have a role to bacterial survival, chronic infections, and reduced antibiotic susceptibility, necessitating further investigation into their regulatory mechanisms (Meusken et al., 2022). The traditional biochemical tests are often inadequate for characteristic, detection and identification *Y. enterocolitica* from other microorganisms, like bacteria, parasites , fungi and viruses (Ahsani et al., 2010; Mohammadabadi et al., 2004; Khabiri et al., 2025). Polymerase chain reaction (PCR) has emerged as a consistent and rapid technique, providing results within hours compared to traditional methods (Mohammadabadi et al., 2011; Khabiri et al., 2023; Mohammadabadi et al., 2025). PCR enables the direct detection of *Y. enterocolitica* from clinical samples, exclusively through the diagnosis of 16S rRNA, which is critical for understanding its spread and pathogenicity (Shahdadnejad et al., 2016; Mohammadabadi et al., 2024). PCR facilitates the detection of virulence genes and their distribution among *Y. enterocolitica* strains

(Shoaib et al., 2019). In Iraq, previous investigations have documented isolation of *Y. enterocolitica* from diarrheal cases, highlighting its public health significance. In Mosul Kanan and Abdulla (2009) announced a 1.6% prevalence in stool samples from pediatric patients. Furthermore, Al-Rudh et al. (2021) identified a 3.75% prevalence in raw milk, emphasizing the potential for zoonotic transmission. This sudy aimed to molecularly identify *Y. enterocolitica* in clinical isolates from diarrheal patients by detecting 16S rRNA applying PCR, evaluating the presence of some virulence-related genes (*yadA* and *hreP*), and evaluating their roles in bacterial adaptation and pathogenicity, alongside investigating biofilm formation.

Materials and methods

Sample collection and preparation of bacterial inoculum: A total of 200 stool samples from patients with diarrhea were gathered from multiple hospitals in Babylon Province, Iraq, between 01/10/2024 and 31/01/2025 2025. This investigation was managed in accordance with the ethical standards of the Department of Biology, College of Education, University of Al-Qadisiyah, Iraq, and in compliance with the principles of the Declaration of Helsinki. All samples were screened for *Yersinia enterocolitica* applying standard biochemical experiments and affirmed by polymerase chain reaction (PCR) targeting the 16S rRNA gene, followed by 16S rRNA sequencing. For bacterial inoculum preparation, 3-5 well-extracted colonies were suspended in 4-5 mL of brain heart infusion broth and incubated at 37°C for 8 hours. The turbidity of the actively growing broth culture was adjusted with sterile broth to match the 0.5 McFarland standard, corresponding to almost 1.5×10^8 cells/mL.

Molecular detection of *Yersinia enterocolitica* and virulence-related genes: Genomic DNA was extracted from *Y. enterocolitica* isolates applying the genomic DNA purification Kit (Geneaid, Turkey) based on the manufacturer's instructions. Primers specific for the *yadA*, *hreP*, and 16S rRNA genes (Integrated DNA Technologies, Canada) are listed in Table 1. Conventional PCR was carried out in 50 µL reaction volumes containing 5 µL of DNA template, 25 µL of PCR Taq Master Mix (Abm, Korea), 3 µL of each forward and reverse primer, and 14 µL of nuclease-free water (Bioneer, Korea). Amplification was managed applying a Techne thermocycler (Techne, UK). PCR conditions for each gene are detailed in Table 2. PCR products were analyzed by electrophoresis on a 1.5% agarose gel stained with 1% ethidium bromide (Bio Basic, Canada) and visualized under UV illumination (UVP, USA).

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Table 1. Primers applied in this investigation for amplification of the *yadA*, *hreP*, and 16S rRNA genes

Target Gene	Primer Sequence (5'-3')	Product Size (bp)
yadA	F: TAAGATCAGTGTCTCTGCGGC	747
	R: TAGTTATTTGCGATCCCTAGCAC	
hreP	F: GCCGCTATGGTGCCTCTGGTGTG	757
	R: CCCGCATTGACTCGCCCGTATC	
16S rRNA	F: AGAGTTTGATCCTGGCTCAG	1500
	R: CTACGGCTACCTTGTTACGA	

Table 2. PCR thermocycling conditions for amplification of the *yadA*, *hreP*, and 16S rRNA genes

Gene	Step	Cycles	Temperature (°C)	Time
yadA	Initial denaturation	1	95	2 min
	Denaturation	35	95	30 s
	Annealing	35	58	30 s
	Extension	35	72	45 s
	Final extension	1	72	7 min
	Hold	-	4	Indefinite
hreP	Initial denaturation	1	95	5 min
	Denaturation	35	95	30 s
	Annealing	35	61	30 s
	Extension	35	72	45 s
	Final extension	1	72	7 min
	Hold	-	4	Indefinite
16S rRNA	Initial denaturation	1	95	5 min
	Denaturation	35	95	30 s
	Annealing	35	55	30 s
	Extension	35	72	90 s
	Final extension	1	72	7 min
	Hold	-	4	Indefinite

Results and discussion

Molecular characterization of *Yersinia enterocolitica* by 16S rRNA Gene Sequencing: Genetic relatedness of *Yersinia enterocolitica* isolates was evaluated through 16S rRNA gene sequencing. Multiple sequence alignment revealed high conservation of the 16S rRNA gene, with six single nucleotide polymorphisms (SNPs) identified, primarily transitions located in non-coding or structurally neutral regions. Iraqi isolates showed close genetic relatedness among some strains while exhibiting phylogenetic diversity in others, classified into three major clades: First Genetically Unified (FGU), First New Diverging (FND), and Final Unique (FU). The alignment of 16S rRNA gene sequences, showed in Figure 1, includes two Iraqi isolates (PV628219 and PV628221) and 17 reference sequences retrieved from GenBank. Conserved nucleotides are color-coded as follows: A (green), T (red), G (purple), and C (blue), with SNPs highlighted.

Compared to global strains, Iraqi isolates are highly similar to PV628226, with minor differences. Notably, both Iraqi strains share SNPs at positions 19 and 141, suggesting close genetic relatedness. The high conservation of the 16S rRNA gene enables its apply for identifying and grouping *Y. enterocolitica* strains.

Table 3. Single nucleotide polymorphisms (SNPs) detected in Iraqi strains compared to reference sequences

SNP position (Alignment)	Reference nucleotide	Variant nucleotide (Iraqi strain)	Strains affected	Type of change	Predicted effect
19	C	T	PV628219, PV628221	Transition	Silent – non-coding region
47	G	A	PV628219	Transition	Likely neutral – 16S conserved
112	T	G	PV628221	Transversion	Possible minor structural impact
141	C	T	PV628219, PV628221	Transition	No functional region affected
223	G	A	PV628221	Transition	Neutral – variable loop region
319	A	C	PV628219	Transversion	Rare SNP, unknown significance

Table 3 details the six SNPs identified in the 16S rRNA gene, with Iraqi strains PV628219 and PV628221 each exhibiting four SNPs. Remarkably, positions 19 and 141 share identical variants in both strains, denoting a close genetic association. Among the SNPs, four were transitions (C↔T, G↔A), which typically create less meaningful exchanges, while two were transversions (A↔C, T↔G), which may alter the DNA sequence more substantially. None of the SNPs were located in critical functional regions or universal primer-binding sites of the 16S rRNA gene, suggesting no impact on ribosomal function or bacterial viability. Previous investigations have noted that the 16S rRNA gene's conservation impressions ribosome structure and function (Woese, 1987). The presence of SNPs in non-functional regions indicates that Iraqi *Y. enterocolitica* strains belong to the same genetic family as global strains, supporting the utility of 16S rRNA sequencing for diagnosing and tracking bacterial diseases. Phylogenetic analysis, shown in Figure 2, elucidates the evolutionary relationships among *Y. enterocolitica* strains from Iraq and other regions. All Iraqi isolates were categorized into three clades: FGU, FND, and FU. The FGU clade includes three Iraqi strains with high similarity, suggesting recent local emergence

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within a short period. An Iraqi isolate (PV628226) in the FND clade shows relatedness to strains from India, Russia, and the USA, denoting historical interactions or shared geographic origins. Four Iraqi FU strains (PV628220, PV628223, PV628225, and PV628228) form a distinct clade, suggesting unique traits or environmental pressures specific to these isolates.

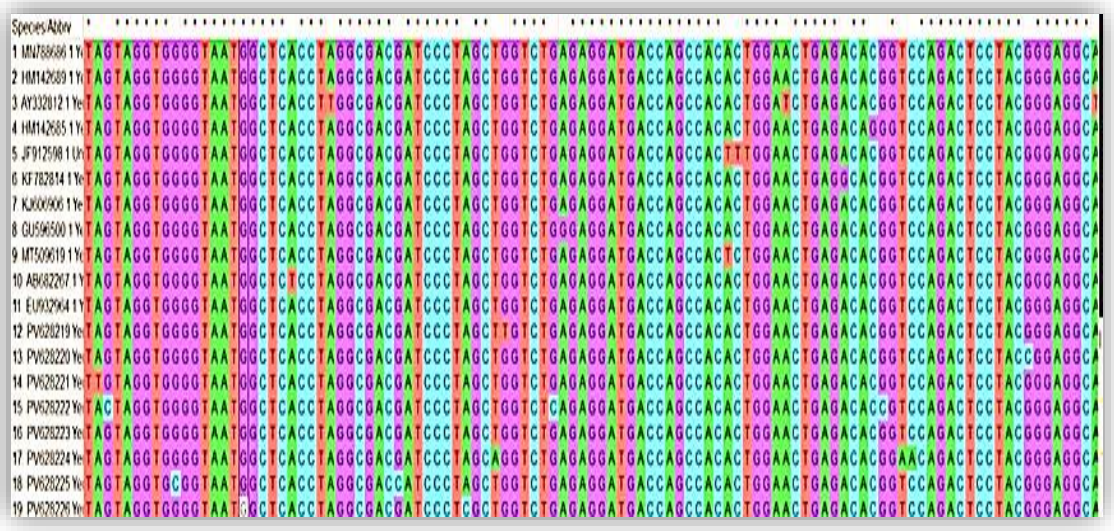

Figure 1. Multiple sequence alignment of 16S rRNA gene sequences of *Y. enterocolitica* isolates

Compared to phenotypic diagnosis methods, molecular approaches based on 16S rRNA sequencing prepare greater consistency and stability. The 16S rRNA gene is highly conserved, with minimal variability within species, making it a preferred marker for phylogenetic investigations and bacterial classification (Woese, 1987; Younis et al., 2021). The current investigation identified six SNPs in the 16S rRNA gene, with Iraqi strains PV628219 and PV628221 each exhibiting four SNPs, containing shared variants at positions 19 and 141.

Figure 2. Phylogenetic tree (Constructed with MEGA) displaying evolutionary relationships of *Y. enterocolitica* strains based on 16S rRNA gene sequences

These results suggest a close genetic association between these isolates. The minor genetic differences compared to global strains, exclusively the high similarity to PV628226, reinforce the idea that Iraqi strains share a high degree of genetic uniformity while maintaining distinct genetic markers. Kislichkina et al. (2025) announced an average 16S rRNA gene homology of 98.76% among *Yersinia* isolates, with a maximum variability of 2.85%, affirming the reliability of 16S

rRNA sequencing in spite of occasional intragenomic heterogeneity that may limit discrimination potential. Similarly, Sotohy et al. (2024) observed high genetic similarity among *Y. enterocolitica* strains from human and animal clinical cases in Egypt, with regional isolates clustering while maintaining distinct SNP variations. The phylogenetic tree in this investigation further clarifies the evolutionary relationships, with some Iraqi isolates closely connected and others showing a broader range of genetic differences. Kislichkina et al. (2025) noted that phylogenetic trees based on 16S rRNA sequences may differ from those constructed applying core SNPs, as the latter prepare higher resolution for genetic relationships.

Molecular detection of *yadA* and *hreP*: Polymerase chain reaction (PCR) analysis of eight *Y. enterocolitica* isolates detected the *yadA* gene (747 bp) in seven isolates (87.5%), as shown in Figure 3, and the *hreP* gene (757 bp) in seven isolates (87.5%), as shown in Figure 4.

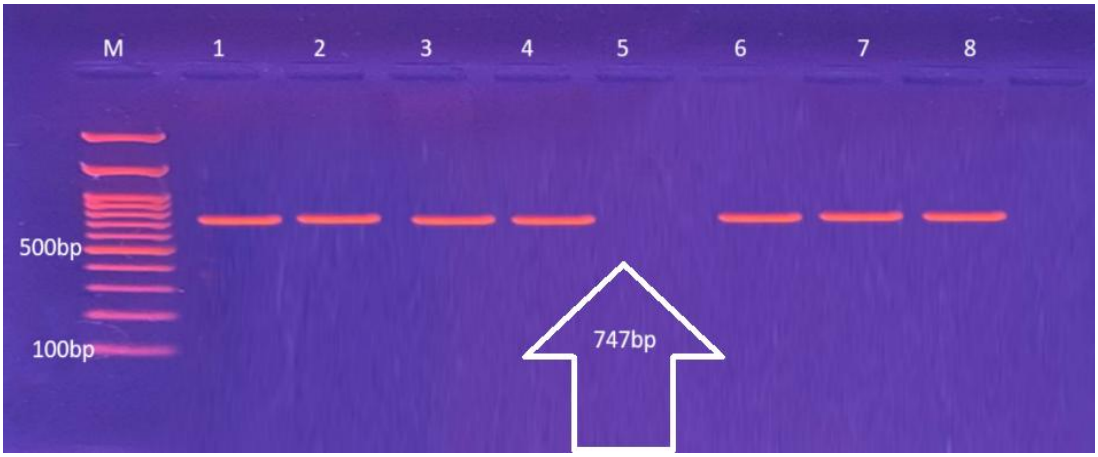

Figure 3. Agarose gel electrophoresis of *yadA* gene amplification in *Y. enterocolitica* isolates (747 bp). Lane M: DNA marker; Lanes 1–8: DNA from *Y. enterocolitica* isolates. The 747 bp band is visible in seven lanes, indicating the presence of *yadA*

The high prevalence of *yadA* and *hreP* genes in clinical *Y. enterocolitica* isolates prepares critical insights into their pathogenicity, exclusively in human infections. The *yadA* gene encodes a trimeric autotransporter adhesin (YadA), which plays a principal role in bacterial adhesion to host extracellular matrix proteins, a fundamental step in initiating infection (Meusken et al., 2022). YadA also facilitates colonization and evasion of host immune defenses, contributing to infection persistence (Schindler et al., 2012). Meusken et al. (2022) demonstrated that YadA's interaction with glycan moieties enhances adhesion and immune evasion, further boosting *Y. enterocolitica*'s capability to adhere to host tissues. Schütz et al. (2010) showed that YadA trimer stability is crucial for virulence, as mutations affecting its structure meaningfully reduce bacterial

adhesion and resistance. The 87.5% prevalence of *yadA* in this investigation is consistent with Zheng et al. (2008), who detected *yadA* in 89% of 160 pathogenic *Y. enterocolitica* isolates from 2600 stool samples applying real-time PCR. However, some investigations announced lower *yadA* prevalence: Tadesse et al. (2013) found *yadA* in 12.8% of porcine isolates, and Shabana et al. (2015) did not detect *yadA* in *Yersinia* isolates from chicken meat. Kot et al. (2017) noted that while *yadA* was shown in most *Y. enterocolitica* strains from children, it was not universal across all biotypes. Younis et al. (2021) announced *yadA* in only 2.4% of meat-derived isolates in Egypt. These discrepancies may be attributed to differences in sample sources, diagnosis methodologies, molecular approaches, strain diversity, or environmental and evolutionary factors.

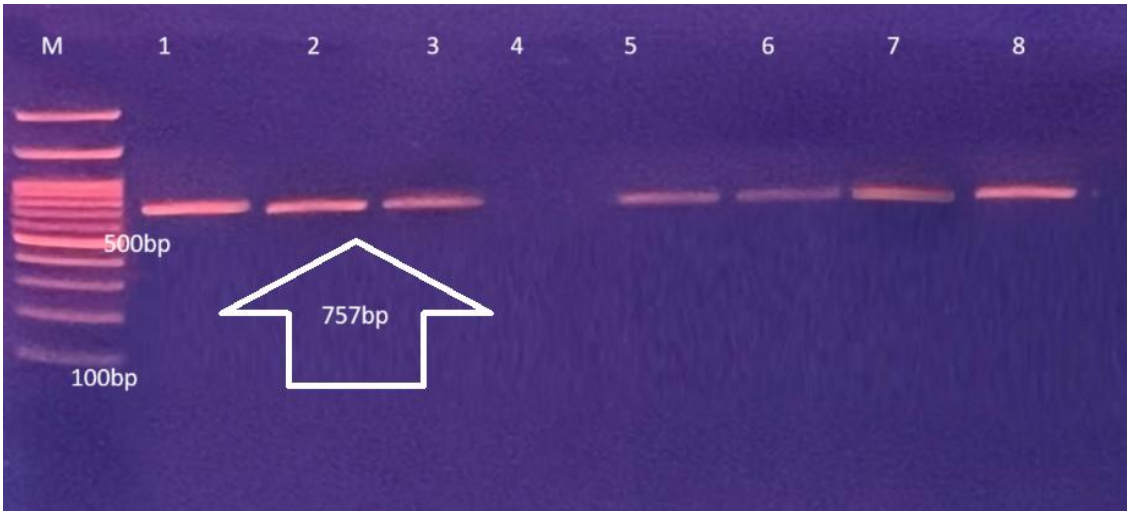

Figure 4. Agarose gel electrophoresis of *hreP* Gene amplification in *Y. enterocolitica* isolates (757 bp). Lane M: DNA marker; Lanes 1–8: DNA from *Y. enterocolitica* isolates. The 757 bp band is visible in seven lanes, indicating the presence of *hreP*

The *hreP* gene, a chromosomal virulence-related factor, contributes to bacterial stress responses and survival under adverse situations, located within a cluster of flagellum biosynthesis and chemotaxis genes (Heusipp et al., 2001). This positioning suggests roles in bacterial motility and environmental adaptation, regulated by a complex network of transcriptional regulators responsive to environmental stressors (Wagner et al., 2009). The chromosomal location of *hreP* underscores its impression on bacterial survival and adaptation, independent of plasmid-encoded virulence factors like *yadA*. Previous investigations indicate that *hreP* is exist in both pathogenic and non-pathogenic *Y. enterocolitica* strains, suggesting that its presence alone may not confer pathogenicity, exclusively in non-diarrheal isolates (Peruzy et al., 2017; Ventola et al., 2023). The 87.5% prevalence of *hreP* in this investigation highlights its potential significance in clinical diarrheal isolates, warranting further investigation into its role in pathogenesis.

Biofilm formation in *Y. enterocolitica* strains: All *Y. enterocolitica* isolates (100%) exhibited biofilm-forming capability, as demonstrated in Figure 5.

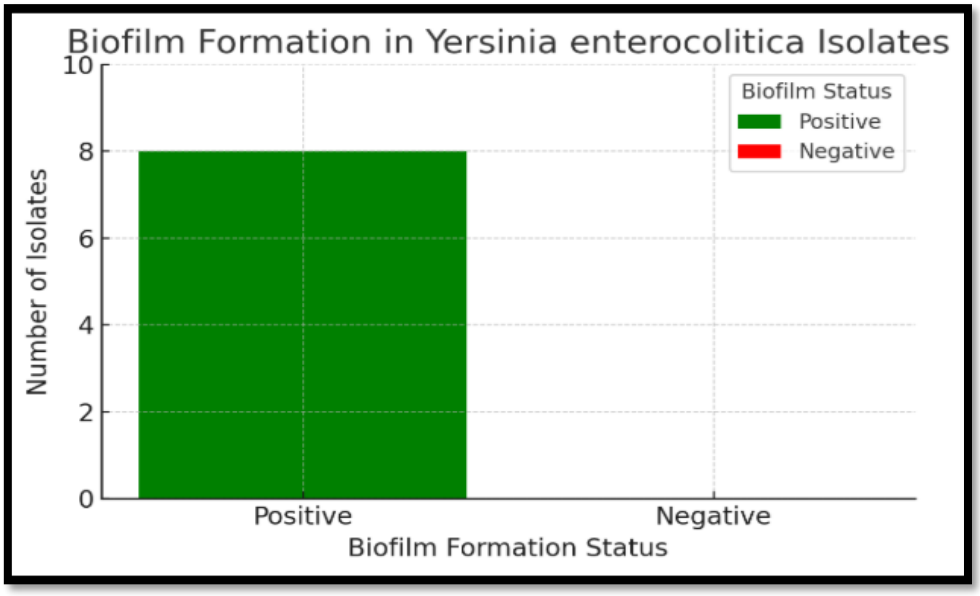

Figure 5. Bar plot demonstrating biofilm-forming capability of *Y. enterocolitica* isolates

Biofilm formation is a critical survival plan for *Y. enterocolitica*, enhancing resistance to environmental stressors and antimicrobial factors. The universal biofilm-forming capability among all studied isolates suggests improved virulence, persistence in natural environments, and resistance to antimicrobial treatments, posing meaningful risks to public health, exclusively in healthcare and food settings. This result aligns with Ioannidis et al. (2014), who announced that all clinical *Y. enterocolitica* strains formed biofilms and exhibited meaningfully higher resistance to antimicrobial agents compared to planktonic forms, emphasizing the role of biofilms in bacterial persistence. Lenchenko et al. (2019) evaluated biofilm formation in *Y. enterocolitica* and found that S-form colonies were more capable of forming biofilms than R-form colonies, suggesting strain-dependent variation. Özdemir and Arslan (2022) observed that *Y. enterocolitica* isolates from food exhibited exceptional biofilm-forming capability at numerous temperatures, denoting that environmental factors like temperature impression but do not suppress biofilm formation. Kim et al. (2008) demonstrated that flagellar mutations impairing structure or rotation meaningfully reduced biofilm production under stable situations, suggesting that genetic factors impression biofilm formation. Wang et al. (2017) found that Ca^{2+} ions and the virulence plasmid enhance biofilm formation in *Y. enterocolitica*. In contrast, Lou et al. (2025) announced that 52.7% of *Y. enterocolitica* isolates from poultry meat lacked biofilm-forming capability,

potentially due to differences in environmental situations, strain-specific factors, or methodological techniques.

Conclusions: This study identifies *Y. enterocolitica* in clinical samples, detecting unique SNPs in Iraqi strains, which display minor genetic variations whereas maintaining close relatedness to global strains. The high prevalence 87.5% of virulence genes *hreP* and *yadA*, shared with the universal biofilm-forming capability 100%, highlights the pathogenic potential and adaptability of *Y. enterocolitica*. These findings emphasize the importance of molecular diagnosis techniques, like PCR and 16S rRNA sequencing, to understand the epidemiology of this bacterium and to improve diagnostic strategies in clinical settings.

Author contributions

Baneen Maan Kareem: Conceptualization, sample collection, experimental work, data analysis, manuscript drafting. Hadaf Mahdi Kadhim: Supervision, methodology design, manuscript review and editing.

Data availability statement

The datasets generated through and/or analyzed in the current investigation are available from the corresponding author on reasonable request.

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Ethical approval

This investigation was managed in accordance with the ethical standards of the Department of Biology, College of Education, University of Al-Qadisiyah, Iraq, and in compliance with the principles of the Declaration of Helsinki. Ethical approval was granted under reference number 42–11/10/2014. Written informed consent was achieved from all participants or their legal guardians prior to specimen collection.

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Conflict of interest

The authors declare no conflicts of interest related to this research. No financial, personal, or professional relationships exist that could have biased the design, conduct, or reporting of this investigation.

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تشخیص مولکولی برخی ژن‌های عوامل بیماری‌زا و تشکیل بیوفیلم در جدایه‌های بالینی *Yersinia enterocolitica*

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چکیده

هدف: Yersinia enterocolitica یک پاتوژن منتقل‌شده از طریق غذا است که مسئول عفونت‌های گوارشی، به‌ویژه اسهال، می‌باشد و بیماری‌زایی آن به عوامل ژنتیکی مرتبط است که امکان تهاجم به سلول‌های میزبان، فرار از سیستم ایمنی و پایداری در محیط را فراهم می‌کنند. این مطالعه با هدف شناسایی ژن 16S rRNA، ارزیابی ژن‌های مرتبط با بیماری‌زایی (yadA و hreP) و بررسی تشکیل بیوفیلم در جدایه‌های Yersinia enterocolitica از نمونه‌های اسهالی جمع‌آوری‌شده در استان بابل، عراق، انجام شد تا تنوع ژنتیکی و پتانسیل بیماری‌زایی آن‌ها روشن شود.

مواد و روش‌ها: در مجموع ۲۰۰ نمونه مدفوع از بیماران مبتلا به اسهال از بیمارستان‌های مختلف در استان بابل جمع‌آوری شد. جدایه‌ها با استفاده از آزمایش‌های بیوشیمیایی استاندارد و واکنش زنجیره‌ای پلیمراز (PCR) با هدف‌گیری ژن 16S rRNA‌ی Yersinia enterocolitica شناسایی شدند و سپس توالی‌یابی ژن انجام شد. تشخیص مولکولی ژن‌های بیماری‌زایی yadA و hreP با استفاده از پرایمرهای اختصاصی انجام گرفت. تشکیل بیوفیلم از طریق آزمایش‌های کمی ارزیابی شد تا توانایی جدایه‌ها در چسبیدن به سطوح، که نشان‌دهنده پتانسیل پایداری آن‌ها در محیط‌های بالینی است، بررسی شود. داده‌های توالی با استفاده از هم‌ترازی چندگانه توالی و ساخت درخت فیلوژنتیک برای ارزیابی خویشاوندی ژنتیکی تحلیل شدند.

نتایج: تنوع ژنتیکی از طریق توالی‌یابی ژن 16S rRNA ارزیابی شد که نشان‌دهنده حفاظت بالا با شش پلی‌مورفیسم تک‌نوکلئوتیدی (SNP)، عمدتاً انتقالی، در مناطق غیرکدکننده یا ساختاری خنثی بود. جدایه‌های عراقی خویشاوندی نزدیک بین

برخی سویه‌ها را نشان دادند، اما در برخی دیگر تنوع فیلوژنتیک مشاهده شد و به سه خوشه اصلی تقسیم شدند: اولین گروه ژنتیکی یکپارچه (FGU)، اولین گروه جدید واگرا (FND) و گروه نهایی منحصربه‌فرد (FU). هم‌ترازی توالی شامل دو توالی عراقی (GenBank: PV628219, PV628221) و ۱۷ توالی مرجع از GenBank بود، با نوکلئوتیدهای حفاظت‌شده با کد رنگی (A: سبز، T: قرمز، G: بنفش و C: آبی) وSNPها برجسته شدند. سویه‌های عراقی شباهت زیادی به توالی مرجع PV628226 داشتند، با SNPهایی در موقعیت‌های ۱۹ و ۱۴۱ که نشان‌دهنده خویشاوندی نزدیک بود. در مجموع، شش SNP شناسایی شد، به‌طوری‌که سویه‌های PV628219 و PV628221 هر کدام چهار SNP داشتند. تحلیل فیلوژنتیک پروفایل‌های ژنتیکی متنوعی را در میان جدایه‌های Yersinia enterocolitica عراقی تأیید کرد، به‌طوری‌که برخی سویه‌ها ارتباط نزدیک و برخی دیگر واگرایی بیشتری داشتند. ژن‌های بیماری‌زایی yadA و hreP در ۸۷/۵ درصد جدایه‌ها (۷ از ۸) شناسایی شدند که نشان‌دهنده پتانسیل بیماری‌زایی قابل‌توجهی است. آزمایش‌های تشکیل بیوفیلم نشان داد که اکثر جدایه‌ها توانایی تولید بیوفیلم متوسط تا قوی را دارند که نشان‌دهنده ظرفیت آن‌ها برای پایداری در محیط‌های بالینی است.

نتیجه‌گیری: این مطالعه تنوع ژنتیکی و پتانسیل بیماری‌زایی جدایه‌های Yersinia enterocolitica از استان بابل، عراق را برجسته می‌کند. شناساییSNP ها در ژن 16S rRNA و حضور ژن‌های بیماری‌زایی yadA و hreP در اکثر جدایه‌ها، اساس مولکولی بیماری‌زایی آن‌ها را نشان می‌دهد. تشکیل بیوفیلم مشاهده‌شده، مکانیسمی برای پایداری محیطی را پیشنهاد می‌کند که چالش‌هایی برای کنترل عفونت ایجاد می‌کند. این یافته‌ها به درک اپیدمیولوژی و ویژگی‌های مولکولی Yersinia enterocolitica در عراق کمک می‌کند و استراتژی‌های تشخیصی و درمانی هدفمند را اطلاع‌رسانی می‌کند.

کلمات کلیدی: تشکیل بیوفیلم، تنوع ژنتیکی، جدایه‌های بالینی، ژن‌های بیماری‌زایی، 16S rRNA

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