IRAQI Academic Scientific Journals
العراقية المجلات الأكاديمية العلمية
University of Kut Journal
ISSN (E): 2616 - 7808 II ISSN (P): 2414 - 7419
js.uokut.edu.iq
k.u.c.j.sci@alkutcollege.edu.iq
Vol. 11 , Issue 1 , June 2026

Prevalence of Bacterial Enteropathogens in Transfusion-Dependent Beta-Thalassemia Children with Iron Overload

Shaimaa M. Hadid [1] , Salem R. Arian AL-Aidy [2] , Safa A. Faraj [3]

Abstract

Background: Transfusion-dependent beta-thalassemia (TDT) patients are highly susceptible to repeated infections. Iron overload and disruptions in the immune system are the main causes. Gastroenteritis symptoms are a significant cause of morbidity in this group. **Objective:** This study aimed to determine the prevalence of bacterial enteropathogens in TDT children presenting with gastroenteritis and to evaluate the association between bacterial growth and clinical/laboratory parameters. **Methods**: A cross-sectional study was conducted on 100 TDT children (≤15 years) at the Center for Genetic Blood Disorders, Al-Kut Hospital, Iraq (August 2024–January 2025). Stool samples were analyzed using culture, VITEK® 2, and 16S rRNA sequencing. **Results:** Bacterial growth was confirmed in 20% of patients. Isolates were predominantly Gram-negative (95%), with Escherichia coli (50%) being the most frequent. A significant association was found between bacterial growth and younger age (<10 years, P=0.001) and higher serum ferritin levels (P=0.034). **Conclusion:** Younger age (<10 years) and iron overload are major predictors of bacterial gastroenteritis in TDT children**.**

Keywords: Beta-Thalassemia, Gastroenteritis, Iron Overload, Ferritin, Escherichia coli

Affiliation of Authors
[1, 2, 3] College Medicine, Univ. Wasit, Iraq, Wasit, 52001

[1] std2023201.shaimaamousah@uowasit.edu.iq
[2] S.r.a.al-Aidy@Uowasit.edu.iq
[3] safaafaraj@uowasit.edu.iq

[1] **Corresponding Author**

Paper Info.
Published: Jun. 2026

انتشار الممرضات البكتيرية المعوية لدى الأطفال المصابين بالتلاسيميا بيتا المعتمدة على نقل الدم والذين يعانون من فرط الحديد

شيماء موسى هديد[1] ، سالم رهيف عريان العايدي[2] ، صفاء عبد الاله فرج[3]

المستخلص

الخلفية :المرضى المصابون بالثلاسيميا المعتمدة على النقل الدموي (TDT) معرضون بشكل كبير للإصابة بالعدوى المتكررة بسبب زيادة الحديد واضطراب جهاز المناعة. تُعد التهاب الأمعاء سببًا مهمًا للمراضة في هذه الفئة.

الهدف :هدفت هذه الدراسة إلى تحديد انتشار مسببات الأمراض البكتيرية المعوية لدى الأطفال المصابين بالثلاسيميا المعتمدين على النقل الدموي والذين يعانون من التهاب الأمعاء، وتقييم العلاقة بين نمو البكتيريا والعوامل السريرية والمخبرية.

الطرق :أُجريت دراسة مقطعية شملت 100 طفل مصاب بـ TDT (≤15 سنة) في مركز اضطرابات الدم الوراثية، مستشفى الكوت، العراق (أغسطس 2024 – يناير 2025). تم تحليل عينات البراز باستخدام الزراعة، نظام VITEK® 2 ، وتسلسل جين S rRNA.16

النتائج :تم تأكيد نمو البكتيريا في 20% من المرضى. كانت العزلات في الغالب سالبة الغرام (95%)، وكان *Escherichia coli* هو الأكثر شيوعًا (50%). وُجد ارتباط معنوي بين نمو البكتيريا والعمر الأصغر <10 سنوات، (P=0.001) وارتفاع مستويات الفيريتين في الدم (P=0.034).

الخلاصة :يُعد العمر الأصغر <10 سنوات وزيادة الحديد من العوامل الأساسية المتنبئة بالتهاب الأمعاء البكتيري لدى الأطفال المصابين بالثلاسيميا المعتمدين على النقل الدموي.

الكلمات المفتاحية: بيتا ثلاسيميا، التهاب المعدة والأمعاء، فرط الحديد، الفيرتين، الإشريكية القولوني

انتساب الباحث
[1، 2، 3] كلية الطب، جامعة واسط، العراق، واسط، 52001

[1] std2023201.shaimaamousah@uowasit.edu.iq
[2] S.r.a.al-Aidy@Uowasit.edu.iq
[3] safaafaraj@uowasit.edu.iq

[1] **المؤلف المراسل**

معلومات البحث
تأريخ النشر : حزيران 2026

Introduction

β-thalassemia is a genetic blood disease that affects hemoglobin production. These disorders are caused by mutations in the β-globin gene. As a result, patients suffer from ineffective erythropoiesis and chronic anemia. [1]. Patients with transfusion-dependent β-thalassemia (TDT) need regular blood transfusions. These transfusions help maintain safe hemoglobin levels and prevent problems caused by anemia. [2]. However, frequent transfusions can lead to iron overload. Excess iron can damage multiple organs and weaken the immune system. It also promotes bacterial growth and reduces the effectiveness of immune cells, such as neutrophil chemotaxis and phagocytosis, or activates the complement system. As a result, TDT patients have an increasing susceptibility to infections. [3].

Patients with transfusion-dependent beta-thalassemia (TDT) need lifelong blood transfusions, leading to iron overload and complications like splenectomy. Excess iron weakens the immune system, increasing infection risk. This vulnerability is caused by anemia and treatment-related factors such as iron overload and asplenia [4].

Thalassemia is also associated with broad immune defects. Patients may have defective phagocyte function, reduced lymphocyte differentiation, and an altered CD4/CD8 ratio [5]. In an iron-rich environment, bacteria can proliferate faster. For example, *Yersinia enterocolitica* becomes more virulent when iron is abundant. Excess iron is one of the main factors that make it so aggressive [6].

In summary, frequent transfusions and iron overload in children with TDT create an environment for infections and disrupt normal immune defenses. This often manifests as gastrointestinal symptoms. Patients may experience abdominal pain, diarrhea, and other gut problems. These symptoms likely reflect their compromised immunity and iron imbalance [5].

Enteric pathogens are often the main cause of infections in thalassemia patients. In one large series, 23.8% of thalassemia patients had culture-confirmed infections, and *Escherichia coli* was by far the most commonly isolated pathogen [4].

Despite this, there is limited data specifically on bacterial gastroenteritis in TDT children. The current study analyzed stool cultures from TDT children with recurrent gastroenteritis and correlated bacterial findings with clinical and laboratory markers such as age and serum ferritin.

Materials and Methods

A cross-sectional study was conducted from August 2024 to January 2025 at the Center for Genetic Blood Disorders, Al-Kut Hospital, Wasit, Iraq. The study included 100 TDT children aged ≤15 years suffering from recurrent gastroenteritis, selected using simple random sampling from patients who visited the hospital during the study period.

Stool specimens were collected in sterile containers and transported to the laboratory within 2 hours of collection. Venous blood (3–5 mL) was collected for CBC, ferritin, and biochemical analysis. Stool samples were cultured on MacConkey, CIN, and blood agar and incubated at 37°C for 48 hours.

Isolates were identified through colony morphology, Gram staining, and conventional biochemical tests, followed by automated identification using the VITEK® 2 system and confirmed by PCR targeting the 16S rRNA gene (band size 372 bp), which was amplified using specific primers under the following PCR conditions: initial denaturation at 95°C for 3

minutes; 35 cycles of denaturation at 95°C for 30 seconds, annealing at 56°C for 30 seconds, and extension at 72°C for 1 minute; with a final extension at 72°C for 5 minutes.

PCR products were visualized on a 1.5% agarose gel stained with ethidium bromide (Figure 1). PCR products were purified using the HiPure DNA cleanup kit (Magen, China), following the manufacturer's instructions, followed by Sanger sequencing.

The sequences were compared to the SILVA rRNA gene database for taxonomic identification based on 16S rRNA gene similarity.

Exclusion Criteria

Exclusion criteria: Patients older than 15 years, those who were not transfusion-dependent, and patients diagnosed with other hemoglobinopathies were excluded from the study.

Statistical Analysis

Data were analyzed using SPSS Statistics version 26 and Microsoft Excel. Continuous variables are presented as mean ± standard deviation (SD), and categorical variables as frequencies and percentages. Chi-square test and Student's t-test were used as appropriate, and a P-value < 0.05 was considered statistically significant.

Ethical approval

The study was approved by the Ethics Committee of Wasit Health Directorate (Reference No. 1523, dated 06-08-2024). Verbal consent was obtained from the patients' guardians before joining the study. The study followed the ethical rules of the 1964 Helsinki Declaration and its later updates. Participants' information was kept private using anonymous codes.

Results and Discussion

Isolation Rate and Gram Stain Results

In this study, bacterial cultures were positive in only 20% (n=20) of the 100 samples.

All isolates from thalassemic patients (n=20) showed a positive band of 372 bp on a 1.5% agarose gel, as shown in Figure 1. Table (1) presents the Gram stain results of these positive cultures, showing a stark predominance: 95% (n=19) were Gram-negative, and only 5% (n=1) were Gram-positive.

All isolates from thalassemic patients (n=20) showed a positive band of 372 bp on 1.5% agarose gel, as shown in Figure (1).

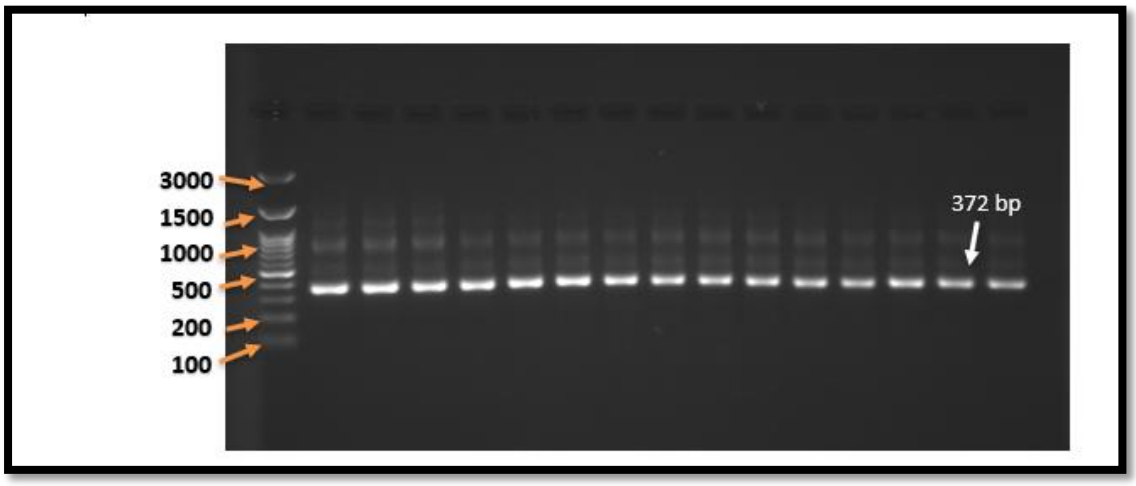

Figure (1): Agarose gel electrophoresis of PCR products of the amplification of partial region of gene 16s rRNA of Bacteria (Band size 372 bp)

This finding is highly significant and expected in the context of gastrointestinal infections. Gram-negative bacteria, particularly members of the Enterobacteriaceae family, are the primary enteric pathogens and opportunistic invaders in patients with compromised immunity.

Table (1): Gram Stain Results of Bacterial Isolates from Positive Culture

Gram Stain Result	Frequency (n)	Percentage (%)
Negative	19	95.0%
Positive	1	5.0%
Total	**20**	**100.0%**

Reference: Statistical Analysis Results using SPSS.

Distribution of Bacterial Isolates

In our study, we observed significant bacterial growth in stool samples from transfusion-dependent β-thalassemia (TDT) patients. The most common of the isolates, *Escherichia coli*, was detected in 50% (10/20) of positive samples. Other bacteria were also detected, including *Klebsiella pneumoniae* (20%, 4/20), *Enterobacter cloacae* (15%, 3/20), and Proteus mirabilis (5%, 1/20), as detailed in Table (2).

Table (2): Distribution of Bacterial Isolates from Stool Samples of TDT Patients.

Bacteria Species	Frequency	Percent (%)
***Enterobacter cloacae* complex (E. cloacae)**	3	15.0%
***Escherichia coli* (E. coli)**	10	50.0%
***Enterococcus faecium* (E. faecium)**	1	5.0%
***Klebsiella pneumoniae* (K. pneumoniae)**	4	20.0%
***Proteus mirabilis* (P. mirabilis)**	1	5.0%
***Roseomonas gilardii* (R. gilardii)**	1	5.0%
Total	**20**	**100.0%**

Reference: Statistical Analysis Results using SPSS.

Association with Clinical Characteristics

Analysis of clinical characteristics Table (3) showed a highly significant association between younger age (<10 years) and bacterial growth (P = 0.001), with children under 10 years old having a bacterial growth rate of 80% compared to 20% in the ≥ 10 years group.

Table (3): Comparison of Clinical Characteristics between Bacterial Growth and Non-Growth Groups

Variable	Category	Growth (n=20)	Non-Growth (n=80)	P value
Sex	Male	13 (65.0%)	41 (51.2%)	0.270
	Female	7 (35.0%)	39 (48.8%)	
Age group	< 10	16 (80.0%)	24 (30.0%)	0.001
	≥ 10	4 (20.0%)	56 (70.0%)	
Splenectomy	Yes	1 (5.0%)	8 (10.0%)	0.485
	No	19 (95.0%)	72 (90.0%)	
Iron chelator used	Exjade	16 (80.0%)	71 (88.8%)	0.433
	Desferal	1 (5.0%)	4 (5.0%)	
	None	3 (15.0%)	5 (6.3%)	
Hepatitis Infection	HCV	1 (5.0%)	5 (6.3%)	0.833
	No infection	19 (95.0%)	75 (93.8%)	
Hypothyroidism	Yes	2 (10.0%)	9 (11.3%)	0.873
	No	18 (90.0%)	71 (88.8%)	

Reference: Statistical Analysis Results using SPSS.

Association with Laboratory Parameters

Patients with bacterial growth showed a significantly higher mean serum ferritin level compared to those without (3422.9 ± 2487.1 ng/mL vs. 2470.8 ± 1545.1 ng/mL, p = 0.034).

Furthermore, both SGPT (ALT) and SGOT (AST) levels were significantly elevated in the bacterial growth group (p = 0.010 and p = 0.012, respectively).

This is supported by the significantly higher White Blood Cell (WBC) count in the growth group (10.80 ± 4.93 × 10^3/µL vs. 8.45 ± 4.30 × 10^3/µL; p = 0.037).

The significantly lower Total Serum Bilirubin (TSB) in the growth group (p = 0.009) is an interesting finding.

No statistically significant differences were observed in platelet (PLT) count or hemoglobin (Hb) levels between the two groups. Table (4).

Table (4): Comparison of Laboratory Parameters (Mean ± SD)

Variable	Growth (Mean ± SD)	Non-Growth (Mean ± SD)	P-value
TSB (mg/dL)	1.02 ± 0.77	1.53 ± 0.76	0.009
SGPT (U/L)	43.10 ± 37.23	26.99 ± 20.46	0.010
SGOT (U/L)	45.30 ± 19.07	32.79 ± 19.65	0.012
WBC (×10³/µL)	10.80 ± 4.93	8.45 ± 4.30	0.037
PLT (×10³/µL)	353.7 ± 113.3	359.1 ± 161.1	0.887
Hb (g/dL)	8.99 ± 0.71	8.65 ± 1.01	0.162
Serum Ferritin (ng/mL)	3422.90 ± 2487.10	2470.84 ± 1545.19	0.034

Reference: Statistical Analysis Results using SPSS.

Discussion

This relatively low isolation rate, when compared to molecular detection methods or other culture-based studies on enteropathogens in immunocompromised patients, may be partly due to the effect of frequent antibiotic use, which can reduce bacterial load and limit the ability of pathogenic bacteria to grow in conventional cultures [7].

The systemic administration of antibiotics, especially during early life, can rapidly and persistently disrupt the gut microbiota composition [8], suppressing the culturable bacterial load and limiting the ability of pathogenic bacteria to grow in conventional culture [9].

Conventional culture methods have several limitations. Many gut microorganisms depend on specific growth conditions, which make them difficult or impossible to isolate using routine culture techniques [10]. In contrast, molecular methods, such as PCR and next-generation sequencing (NGS), are more sensitive and can identify a wider range of bacterial species. This suggests that the actual prevalence of intestinal dysbiosis or bacterial carriage may be higher than what is detected using culture alone [11].

The predominance of E. coli among isolates is consistent with its established role as a major commensal and opportunistic pathogen in the human gut. The finding that *E. coli* is the most frequently isolated organism, consistent with its established role as a major commensal and opportunistic pathogen in the human gut. More importantly, in the context of thalassemia and iron overload (hyperferritinemia), E. coli and other Enterobacteriaceae thrive. These bacteria have siderophore systems that allow them to scavenge and utilize the free iron in the body more effectively [4,12].

Our results are similar to previous studies, although there are variations in rates of bacterial detection. For example, Al-Salouki (2017, Babylon) [13] found *Escherichia coli* in most of the samples, 97.6% (46/58). *Klebsiella* and *Enterobacter* were also common, found in 46.3% (25/58) and 56.1% (33/58) of the isolates,

respectively. Even with different percentages, *E. coli* was still the most frequent organism, which is consistent with our results.

Another study by Al-Badri et al. (2016) in Thi-Qar [14] reported bacterial growth in 9 out of 20 stool samples, which is about 45%. In their study, *E. coli* accounted for 22.2% (2/9), *Enterobacter* for 11.1% (1/9), and *Proteus mirabilis* for 22.2% (2/9).

Although the percentages of specific bacteria differ between studies, these differences can largely be attributed to differences in sample size, patient demographics (particularly age groups), and methodological variations. The frequent isolation of Escherichia coli as the most common isolate agrees with our findings.

The isolation of a single Roseomonas gilardii isolate (5%) is significant. R. gilardii is an uncommon Gram-negative coccobacillus generally considered an opportunistic pathogen, almost exclusively isolated from immunocompromised patients with underlying illnesses or severe comorbidities [15]. Its presence supports the hypothesis of significant underlying immune compromise in TDT patients. [5].

The age-infection relationship (higher infections < 10 years) may be confounded by transfusion frequency, chelation compliance, or immune maturity, which were not statistically controlled for. Several studies have shown that children with thalassemia who receive frequent blood transfusions, have elevated ferritin levels, and demonstrate poor compliance with iron chelation therapy are more susceptible to infections [4].

Antibiotics in general, especially those administered during early childhood, have significant effects on the composition of the gut microbiome. Early childhood is considered a highly sensitive period for antibiotic use [16].

Additionally, immune maturity in younger children is inherently lower, contributing to their increased susceptibility. Immunity in younger children, rendering them more susceptible to bacterial infections. Several factors contribute to these observations, including weaker innate and adaptive immunity.

These findings agree with a previous Egyptian study [17], which found a mean age at diagnosis of 1.76 ± 0.659 years (ranging from 1 to 3.5 years), an age associated with incomplete immune maturation. That study also reported lower natural killer (NK) cell activity, reduced complement C3 levels, imbalances in T and B lymphocytes, and higher immunoglobulin levels. These factors may help explain why younger patients in our study showed more abnormal bacterial growth.

A study from Iraq found that both iron overload, as seen in thalassemia, and iron deficiency, as in iron deficiency anemia, are associated with increased risk of specific bacterial infections. In thalassemia patients, Escherichia *coli* was the most common pathogen, and its presence was related to serum iron levels [19].

Other research supports the idea that excess iron increases the risk of infections, especially in the bloodstream and urinary tract. For example, a study in Saudi Arabia [4] between 2007 and 2022 reported that higher iron levels are linked to a higher risk of infections and impaired immune system function in patients with thalassemia. Iron overload creates a favorable environment for bacteria to proliferate and disrupts normal immune responses.

Elevated liver enzymes in TDT patients are usually linked to chronic iron-related liver damage or viral hepatitis [20]. However, in our study, the

difference in liver enzymes suggests that active gut bacterial infection or the resulting systemic inflammatory response may contribute to liver injury.

This is supported by the higher White Blood Cell (WBC) count in patients with bacterial growth, indicating an active infection or inflammatory process. Interestingly, the lower Total Serum Bilirubin (TSB) in this group may point to a different pattern of hemolysis or liver function, which requires further study.

Limitations

The detection of bacterial enteropathogens mainly depended on culture-based methods, which might not fully reflect the actual prevalence of difficult-to-culture or non-culturable organisms. Furthermore, the study did not fully control for antibiotic exposure before sample collection, which could have reduced the chances of positive cultures. Additionally, the study was conducted at a single center with a relatively small sample size, which may limit the ability to generalize the findings.

Conclusions

This study concludes that bacterial gastroenteritis is a significant complication in children with transfusion-dependent β-thalassemia. Younger age (<10 years) is a significant risk factor due to immune system immaturity. Additionally, iron overload (hyperferritinemia) is strongly associated with increased susceptibility to bacterial gastroenteritis.

Recommendations

1. Ensure strict adherence to iron chelation therapy to reduce the risk of iron-facilitated bacterial infections in TDT patients.
2. Consider routine screening for bacterial enteropathogens in TDT children under 10 years presenting with gastrointestinal symptoms.

Suggestions

Future studies should employ metagenomic sequencing to investigate gut microbiome dysbiosis in TDT patients and its relationship with iron metabolism in greater detail.

References

[1] Y. Ginzburg and S. Rivella, β-thalassemia: A model for elucidating the dynamic regulation of ineffective erythropoiesis and iron metabolism. Blood, 2011; 118:4321–30.

[2] S. Gardenghi, R. W. Grady and S. Rivella, Anemia, ineffective erythropoiesis, and hepcidin: Interacting factors in abnormal iron metabolism leading to iron overload in β-thalassemia. Hematology/Oncology Clinics of North America, 2010; 24:1089–107.

[3] C. Asadov and G. Aliyeva, Beyond anemia: unraveling neutrophil defects and infection susceptibility in β-thalassemia. Blood Research, 2025; 60.

[4] E. Mansory, L. Abdulrahman, B. Osman, S. Sawsan, A. Ruckn, M. Aljedaani, et al., Predisposing factors to infections in thalassemia syndrome patients. Mediterranean Journal of Hematology and Infectious Diseases, 2025; 17(1): e2025055.

[5] S. Fakeh, A. Masoud, R. Abuqtaish, B. Salman, L. Al-Ramahi, O. AlWahkyan, et al., Gastrointestinal pathologies associated with

thalassemia: A systematic review. Gastroenterology Insights, 2025; 16.

[6] M. D. Cappellini, C. A., E. A., et al., Infections in Thalassaemia Major. In: Guidelines for the Clinical Management of Thalassaemia, 2nd revised ed. Nicosia, Cyprus: Thalassaemia International Federation, 2008.

[7] G. Cusumano, G. A. Flores, R. Venanzoni and P. Angelini, The impact of antibiotic therapy on intestinal microbiota: Dysbiosis, antibiotic resistance, and restoration strategies. Antibiotics, 2025; 14.

[8] H. Neuman, P. Forsythe, A. Uzan, O. Avni and O. Koren, Antibiotics in early life: dysbiosis and the damage done. FEMS Microbiology Reviews, 2018; 42:489–99.

[9] G. Chen, Y. Li, S. Wei, X. Wang, Z. Kuang, W. Guo, et al., Role of gut microbiota in thalassemia: a review of therapeutic prospects. Frontiers in Physiology, 2025; 16.

[10] C. Su, L. Lei, Y. Duan, K. Q. Zhang and J. Yang, Culture-independent methods for studying environmental microorganisms: Methods, application, and perspective. Applied Microbiology and Biotechnology, 2012; 93:993–1003.

[11] L. R. Macfarlane-Smith, S. Ahmed and M. H. Wilcox, Molecular versus culture-based testing for gastrointestinal infection. Current Opinion in Gastroenterology, 2018; 34:19–24.

[12] F. A. Obed, A. M. Omran and K. Sh. Jebur, Effect of iron overload on prevalence of common bacterial infection in thalassemia patients. Iraqi Journal of Hematology, 2024; 13(1):38–43.

[13] I. Noori Dawood Alsaluki, I. Kerim Abd Alshibly and M. Hassen Noor, An immunological evaluation and study of dysbiosis associated with thalassemic patients in Babylon Province. World Journal of Pharmaceutical Research, 2017; 6:1334–48.

[14] B. J. Al Badry, A. A. Hussin, I. N. Abid, A. Sh. Jabber and A. M. Thmen, Bacterial infections in thalassemia patients at Thi-Qar Province, South Iraq. American Scientific Research Journal for Engineering, Technology and Sciences, 2016; 19(1):199–205.

[15] C. Schlappi, J. D. Bernstock, W. Ricketts, G. A. Nix, C. Poole, J. Lebensburger, et al., Roseomonas gilardii bacteremia in a patient with HbSβ0-thalassemia: Clinical implications and literature review. Journal of Pediatric Hematology/Oncology, 2020; 42: e385–7.

[16] H. Neuman, P. Forsythe, A. Uzan, O. Avni and O. Koren, Antibiotics in early life: dysbiosis and the damage done. FEMS Microbiology Reviews, 2018; 42:489–99.

[17] M. R. Beshir, M. Bakry Abd-Alaziz, W. I. Ismail, E. Gamal Baz and M. Bakryabd-Alaziz, Evaluation of immune system alterations in children with β-thalassemia major: Single-center Egyptian study, 2024.

[18] B. R. Wilson, A. R. Bogdan, M. Miyazawa, K. Hashimoto and Y. Tsuji, Siderophores in iron metabolism: From mechanism to therapy potential. Trends in Molecular Medicine, 2016; 22:1077–90.

[19] F. A. Obed, A. M. Omran and K. Sh. Jebur, Effect of iron overload on prevalence of common bacterial infection in thalassemia patients. Iraqi Journal of Hematology, 2024; 13(1):38–43.

[20] K. M. Salama, O. M. Ibrahim, A. M. Kaddah, S. Boseila, L. A. Ismail and M. M. Abdel Hamid, Liver enzymes in children with beta-thalassemia major: Correlation with iron

overload and viral hepatitis. Open Access Macedonian Journal of Medical Sciences, 2015; 3(2):287–92.

Appendix

Figure (2): *E. coli* colonies appeared in pink color on MacConkey agar

Figure (3): K. pneumoniae on MacConkey agar the colonies appeared in pink color, mucoid

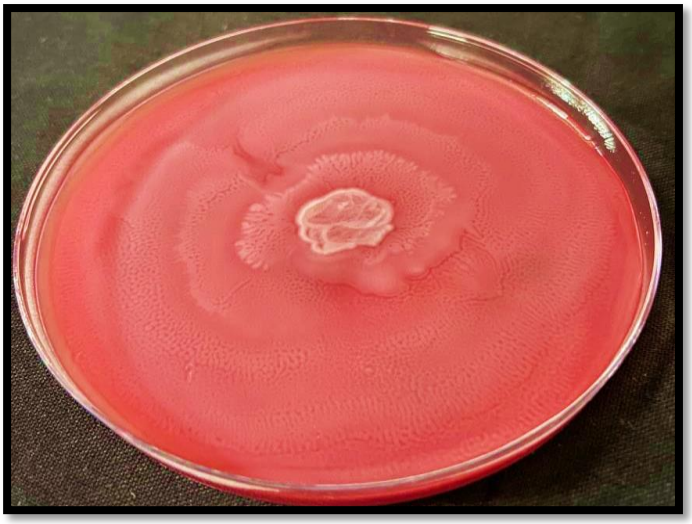

Figure (4) :P. mirabilis on blood agar the colonies appeared characteristic swarming motility

Figure (5): Enterobacter cloacae on MacConkey agar the colonies appeared in pink color

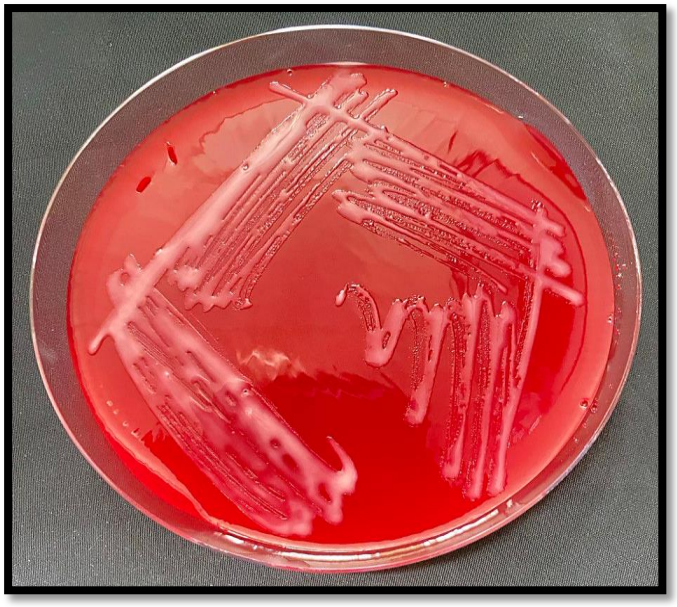

Figure (6) :Roseomonas gilardii on blood agar the colonies appeared dull white, mucoid, and glistening