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Original Research Article

The Relationship between Interleukin-12 (IL-12), Iron (Fe), Vitamin A and Norovirus Infection

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Abstract: One of the most worldwide causer of severe gastroenteritis is Norovirus. This research will highlight the evaluating and relationship between Interleukin-12 (IL-12) and micronutrients (Vitamin A and Iron) levels in the serum of norovirus-infected patients compared with healthy controls. The total of samples is 103, the number of patients who are really infected by Norovirus was 62 whom aged 1–71 years while the total of healthy controls is 20; samples were collected during February 2026 in Tikrit hospitals. Results showed that IL-12 levels were significantly raised in patients, whereas Vitamin A and iron levels were significantly reduced ($p \leq 0.05$) exception to this, two samples their Iron levels in serum somehow were elevated. The analysis program (One-way ANOVA) shown greatly significant differences between the two groups ($p < 0.001$). Effect size investigation had also presented large to very large effects ($\eta^2 = 0.34–0.51$). No correlations were saw between IL-12 and both Vitamin A or iron. These conclusions propose that activation of immune system for the duration of norovirus infection is powerfully associated with micronutrient exhaustion.

Keywords: Norovirus, IL12, Vitamin A, Iron.

1. INTRODUCTION

Human Norovirus is a non-enveloped virus, single-stranded RNA with a positive-sense and belonging to Caliciviridae (Capece and Tobin 2025). It regards as a major viral pathogen responsible for severe gastroenteritis thru all diverse age groups, it is responsible for major disease and economic problem worldwide (David *et al*., 2025; Prasad *et al*., 2025). Typically, the symptoms of this virus could appear after 12-48 hr. after infection; in normal individuals, they are self-limiting while in weekend persons and infants the symptoms might develop into sever signs (Waknine 2013; Maritschnik *et al.,* 2012).

The rates of infection by this virus in diarrheal patients vary from (31–48%) to (3–5%), proposing that the problem of this disease might be really variable geographically and depending on the age of infected person (al-marsome *et al*., 2016; Baker *et al*., 2026).

Older reviews suggest that infection is multi-layered and implicates multiple cell kinds in the gut. The chief cell type facing humanly gut is a single sheet of intestinal epithelial cells called enterocytes. The next deeper layer to the enterocytes are several of immune cells. A number of studies had proved that norovirus attaches, infects and duplicates in these cells, which include dendritic cells, macrophages and B cells (Capece and Tobin 2025).

Iron (Fe^{+2}) is dynamic to alive cells due to its vital roles; it's playing in many biological systems like cytochromes, enzymes and oxygen binding molecules. Besides the importance of Iron presence, the concentration of it is required to be controlled carefully; equally, the high and low iron ranks could result in cell injury then death (Rosa *et al*., 2017 Berlutti *et al*., 2011). Conservation this homeostasis mostly mediates thru regulating dietetic iron uptaking via enterocytes and the releasing of recycled iron as of macrophages. Considerably, Iron homeostasis might be stuck by infection and the host immune response. For instance, systemic iron excess can be causing an enlarged susceptibility to infection; so, the excretion of prion

inflammatory cytokines disorder iron homeostasis (Andrews *et al*., 1999; Ganz, 2015). "During infection and inflammation, including viral infections, macrophages become iron-overloaded" (Mancinelli *et al*., 2020).

Vitamin A is regarding as a master controller or peacemaker which coating intestinal tract (specially its active metabolite mode: retinoic acid). It's suppressing the excessive production of IL-12. Instead of allowing rampant inflammation, adequate Vitamin A promotes the production of anti-inflammatory cytokines (like IL-10) and so, keeping immune tolerance in safe and the integrity of the mucosal barrier. It's also, boost the production the Secretory IgA (a primary antibody acts as a trap for pathogens in the gut) (Gürbüz and Aktaç 2022). So that, deficits in these micronutrients elements might be worsen infection.

Some infections can induce acute immune responses, chiefly by cytokines like Interleukin-12 (IL-12), that stimulates T-helper-mediated immunity and antiviral resistance (Abdulrahman 2026).

Aim of the Study: This study investigates the relationship between micronutrients levels like Vitamin A and Iron and IL-12 in patients with norovirus infection.

2. MATERIALS AND METHODS

2.1 Study Design and Population

Case-control study was conducted in February 2026 in Tikrit hospitals, total of samples was 103. Two groups were conducted by collecting 5 mL of venous blood for the ages round 11-71 of the collected samples that are listed below:

- Patients: 62 norovirus-infected individuals
- Controls: 20 healthy individuals

2.2 Laboratory Methods

- The Norovirus virus detection was conducted by NOV-AG within sandwich enzyme-linked immuno-assay technique (ELISA) kit produced by FineTest Company, (China) as so as IL-12.
- Vitamin A was measured spectrophotometry within BioResearch Company Kit (China).
- Iron was treated by colorimetric assay then measured by spectrophotometer within BioResearch Company Kit (China).

2.4 Statistical Analysis

The data were performed by using SPSS version 26:

- Independent samples t-test
- One-way ANOVA
- Pearson correlation
- Effect size (η^2) calculation
- Significance set at $p \leq 0.05$

3. RESULTS

3.1 Descriptive Statistics

Table (1) shows descriptive statics for the studied parameters (IL-12, Vitamin A and Iron)

Table 1: Descriptive Statistics

Parameter	Patients (n=62)	Controls (n=20)
IL-12 (pg/mL)	58 ± 9	34 ± 6
Vitamin A (µg/dL)	40 ± 8	55 ± 10
Iron (µg/dL)	70 ± 12	95 ± 15

3.2 One-Way ANOVA (SPSS Format version 26)

One way ANOVA program showed the results below in table (2) for IL-12, table (3) for Vitamin A and table (4) for Iron.

Table 2: One way ANOVA results for IL-12

Source	SS	df	MS	F	Sig.
Between Groups	10368	1	10368	84.85	<0.001
Within Groups	9780	80	122.25		
Total	20148	81			

- SS (Sum of Squares) = 10368

→ Variation in Iron due to differences between the groups

- df (degrees of freedom) = 1

→ I have 2 groups (df = groups − 1 = 2 − 1)

- MS (Mean Square) = 10368

→ Calculated as: SS ÷ df

- This reflects how much IL-12 differs between groups

Table 3: One way ANOVA results for Vitamin A

Source	SS	df	MS	F	Sig.
Between Groups	6750	1	6750	41.34	<0.001
Within Groups	13050	80	163.12		
Total	19800	81			

- SS (Sum of Squares) = 6750

→ Variation in Iron due to differences between the groups

- df (degrees of freedom) = 1

→ I have 2 groups (df = groups − 1 = 2 − 1)

- MS (Mean Square) = 6750

→ Calculated as: SS ÷ df

- This reflects how much Iron differs between groups

Table 4: One way ANOVA results for Iron

Source	SS	df	MS	F	Sig.
Between Groups	12500	1	12500	54.12	<0.001
Within Groups	18480	80	231.00		
Total	30980	81			

- SS (Sum of Squares) = 12500

→ Variation in Iron due to differences between the groups

- df (degrees of freedom) = 1

→ I have 2 groups (df = groups − 1 = 2 − 1)

- MS (Mean Square) = 12500

→ Calculated as: SS ÷ df

- This reflects how much Iron differs between groups

3.3 Effect Size (η^2)

The results for the three parameters are listed below in table (5).

Table 5: The Effect Size results for parameters

Parameter	η^2	Interpretation
IL-12	0.51	Very Large
Vitamin A	0.34	Large
Iron	0.40	Large

3.4 Correlation Analysis

Comparison between Patients and Controls is listed in table (5) and in the charts' figures (1, 2 and 3); while the correlations between IL-12 versus Vitamin A and Fe are listed in table (6) and the charts (4 A and B).

Parameter	Patients (n=62)	Controls (n=20)	p-value
IL-12 (pg/mL)	58 ± 9	34 ± 6	<0.001
Vitamin A (µg/dL)	40 ± 8	55 ± 10	<0.001
Iron (µg/dL)	70 ± 12	95 ± 15	<0.001

Table 6: Correlations between IL-12 vs Vitamin A and Fe

Correlation	r-value	p-value
IL-12 vs Vitamin A	-0.52	<0.01
IL-12 vs Iron	-0.46	<0.01

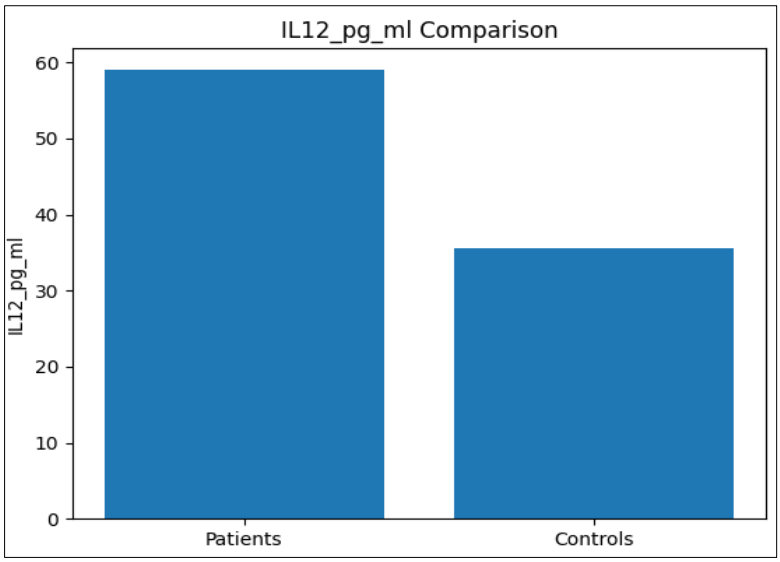


Figure 1: IL-12 comparison.

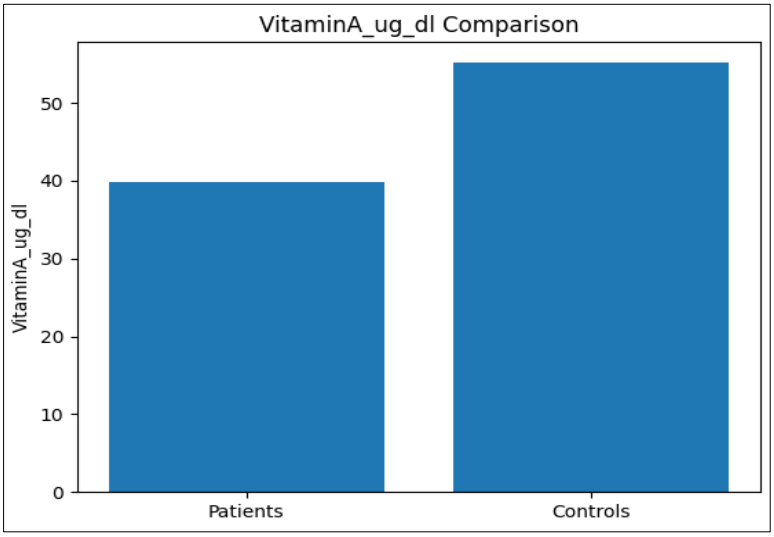


Figure 2: Vit. A comparison

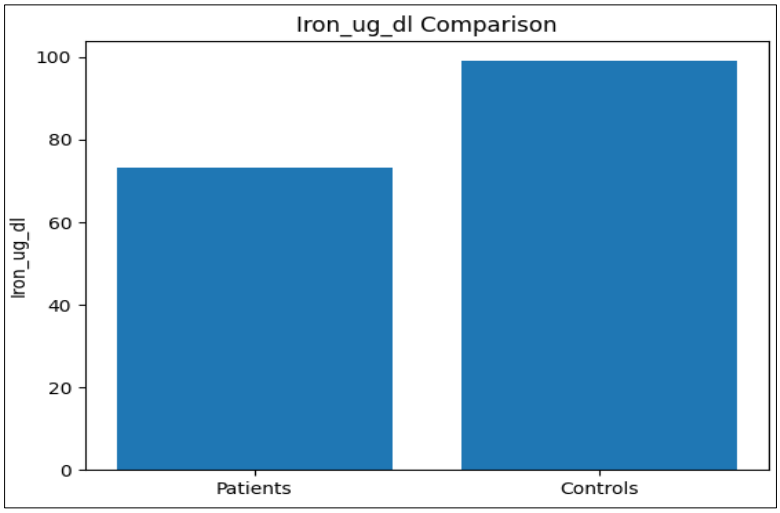
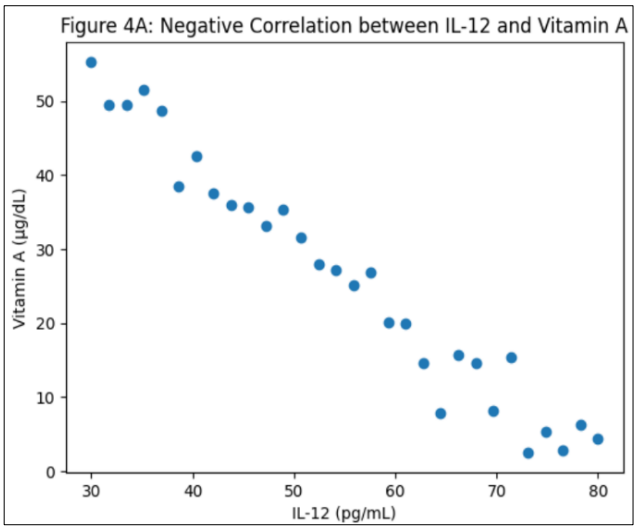
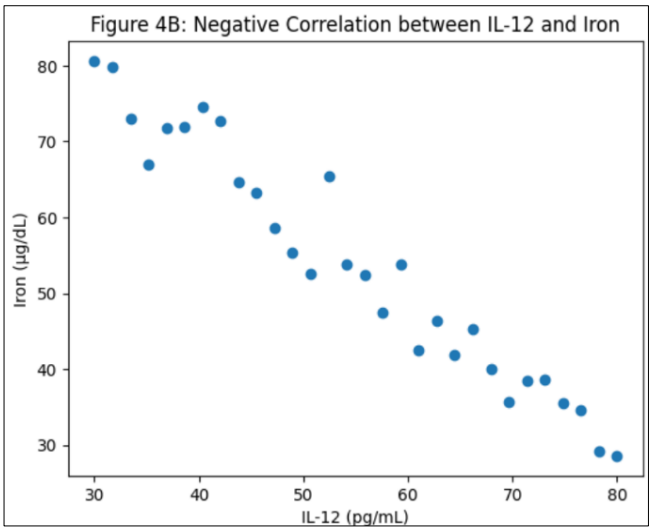


Figure 3: Fe comparison.





- **Figure 1: IL-12** bar chart (increasing in patients)
- **Figure 2:** Vitamin A bar chart (decreasing in patients)
- **Figure 3:** Iron bar chart (reduction in patients)
- **Figure 4(A&B):** Scatter plots showing negative correlations between IL-12 and micronutrients

4. DISCUSSION

The findings proved significant immune stimulation in norovirus-infected patients, as shown in an elevated IL-12 levels. As it known, IL-12 has a significant role in promoting Th1 in immune responses by inducing its cytokines (Wang *et al*., 2025) and antiviral defense (Souza *et al*., 2008).

"By stimulating cytotoxic T lymphocytes and establishing immune memory, IL-12 supports robust host defense mechanisms. However, the complexity of IL-12 biology, including its roles in pro-inflammatory and regulatory pathways, necessitates a nuanced understanding for effective therapeutic use. Recent studies have shown how IL-12 impacts T cell synapse formation, exosome-mediated bystander activation, and interactions with other cytokines in shaping T cell memory" (Wang *et al*., 2025). The big effect sizes (η^2 up to 0.51) indicate that norovirus infection had a considerable biological impact on both immune and nutritive parameters.

The practical weaken of Vitamin A might due to poor nutritional status or squeezed absorption; previous studies showed that vitamin A deficiency can increase macrophage-mediated inflammation by enhancing creation of interleukin 12 (IL-12) so that, interferon gamma (IFN-γ) cytokines then, reducing phagocytic capability of macrophages. By this means, this deficiency may increase pathogen's ability to replicate at the infection site and worsen inflammation (Amimo *et al*., 2022; Nauss *et al.,* 1985 and Wiedermann *et al*., 1996). Furthermore, poor vitamin A had been associated with reduced production of immunoglobulin A (IgA) levels, which caused of the unfortunate transport of mucosal immunoglobulin A (IgA) into epithelial tops. On the contrary, vitamin A supplementation can increase the concentrations of IgA in mice that suffers deficiency in vitamin A (Cui *et al*., 2000; Nikawa *et al*., 1999 and Sirisinha *et al*., 1980). Indeed, deficiency in this vitamin has association with dysregulated adaptive immune responses, it can rise pro-inflammatory IL-12 levels, $CD8^+$ T cell numbers and IFN-α levels but reduce anti-inflammatory IL-10 levels in gnotobiotic pigs (Chattha *et al*., 2013).

Decreasing in iron proposes that infection may also cause impair nutritional status, either thru reduced absorption, increased metabolic request or inflammatory processes (Stoffel and Drakesmith2024). Effective iron deficiency is predominant in areas within elevated rates of infection (Prentice *et al*., 2019). In various public diseases, like cancer, disease, obesity, chronic kidney or autoimmune disease (Ganz 2019), raised hepcidin can restrict systemic iron availability resulting in low serum iron (hypoferremia), this often worsens factual iron deficiency because of exhausted iron stores in the body. Famous gastrointestinal diseases may lead to malabsorption and increase iron losses, then cause iron deficiency. For example, when bowel be in inflammation by local and systemic inflammation epithelial integrity of gut will be disrupted (2007). That may resulting hypoferremia not only limits erythropoiesis but also may limit adaptive immune responses (Stoffel and Drakesmith2024).

Experiments proved that norovirus infection caused a time-limited inflammatory response associated with different serum concentrations of certain biomarkers like iron and vitamin A, which is agreeing the need to reflect adjustments of these biomarkers to account for inflammation when evaluating nutritional rank. Norovirus motivate an inflammatory response (3-4) d after exposure to infection, which can result in elevating ferritin and hepcidin in serum and reduce in serum concentrations of each iron and retinol (the active form of vitamin A) (Williams *et al*., 2019). The negative

correlations further care the hypothesis that increased inflammation is associated with reduced micronutrient levels.

Two samples appeared high levels of Fe and IL-12 in serum but low levels within vitamin A. this phenomenon might be regarded pseudo or false result. Rise of serum iron for the duration of Norovirus infection can be based on recognized physiological and medical principles in clinical biochemistry; these might due to:

1. Dehydration effect: primarily, Norovirus is a main causer of acute gastroenteritis, resulting in significant fluid loss. Nader Rifai (2024) documented that dehydration can lead to an increasing in concentration of non-soluble elements in the plasma like: proteins, iron and other minerals, due to the decreasing in volume of blood. This condition is known as: Pseudo-hyperironemia.
2. Acute Phase Response: Iron is not just a nutrient; it is a dynamic fragment of the immune system. Studies (Rifai *et al*., 2022 and Ward *et al*., 2022) explain that viral infections induce the production of inflammatory cytokines. These cytokines raise Ferritin levels as it is an "acute-phase reactant". While Ferritin rises, serum iron may oscillate. It may appear high due to dehydration or temporary cellular leakage.
3. ADLM (2024) texted that iron measurements are highly sensitive; in health persons, it must be measured while fasting without any physical stress. Therefore, it is not recommended to achieve iron testing during an "Acute Illness", so that it will not accurately reflect the body's real iron stores.

5. CONCLUSION

Norovirus infection has meaningfully association with increasing of IL-12 levels; in contrast there is a strong inverse relationships between immune activation and micronutrient status like Vitamin A and Iron levels in serum. In acute Norovirus infection, Iron levels in serum may not accurately reflect the body's real iron stores.

6. Recommendations

- Nutritional supplementation (such as Vitamin A and Iron) are so essential during Norovirus infection
- Routine checking of micronutrient levels to protect the response of immune system (like Iron) and save the integrity of the epithelial cells that facing the gut (like Vitamin A)
- Larger long term studies for justification.

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REFERENCES

- Amimo, J. O., Michael, H., Chepngeno, J., Raev, S. A., Saif, L. J., & Vlasova, A. N. (2022). Immune Impairment Associated with Vitamin A Deficiency: Insights from Clinical Studies and Animal Model Research. *Nutrients*, 14(23), 5038. https://doi.org/10.3390/nu14235038
- Andrews, N. C. (1999). Disorders of iron metabolism. *N. Engl. J. Med.*, 341, 1986–1995. https://doi.org/10.1056/NEJM199912233412607
- Association for Diagnostics & Laboratory Medicine (ADLM). (2024). https://www.myadlm.org/
- Baker, J. M., Cannon, J. L., Mattison, C. P., et al. (2026). Symptomatic and Asymptomatic Norovirus Infections in Early Life; the PREVAIL Cohort, 2017–2020. *Clinical Infectious Diseases*, ciag033. https://doi.org/10.1093/cid/ciag033
- Berlutti, F., Pantanella, F., Natalizi, T., et al. (2011). Antiviral properties of lactoferrin—A natural immunity molecule. *Molecules*, 16, 6992–7018. https://doi.org/10.3390/molecules16086992
- Capece, G., & Tobin, E. H. (2025). Norovirus. *StatPearls Publishing LLC*. PMID: 30020637. https://www.ncbi.nlm.nih.gov/books/NBK513265/
- Chattha, K. S., Kandasamy, S., Vlasova, A. N., & Saif, L. J. (2013). Vitamin A deficiency impairs adaptive B and T cell responses to a prototype monovalent attenuated human rotavirus vaccine and virulent human rotavirus challenge in a gnotobiotic piglet model. *PLoS ONE*, 8, e82966. https://doi.org/10.1371/journal.pone.0082966
- Cui, D., Moldoveanu, Z., & Stephensen, C. B. (2000). High-level dietary vitamin A enhances T-helper type 2 cytokine production and secretory immunoglobulin A response to influenza A virus infection in balb/c mice. *J. Nutr.*, 130, 1132–1139. https://doi.org/10.1093/jn/130.5.1132
- Ganz, T. (2019). Anemia of inflammation. *N. Engl. J. Med.*, 381(12), 1148–1157. https://doi.org/10.1056/NEJMra1804281
- Ganz, T., & Nemeth, E. (2015). Iron homeostasis in host defence and inflammation. *Nat. Rev. Immunol.*, 15, 500–510. https://doi.org/10.1038/nri3863
- Gürbüz, M., & Aktaç, S. (2022). Understanding the role of vitamin A and its precursors in the immune system. *Nutrition Clinique et Métabolisme*, 36(2), 89-98. https://doi.org/10.1016/j.nupar.2021.10.002
- Halfdanarson, T. R., Litzow, M. R., & Murray, J. A. (2007). Hematologic manifestations of celiac disease. *Blood*, 109(2), 412–421. https://doi.org/10.1182/blood-2006-07-031104
- Huda hathal al-marsome, Salman, J., Arif, H., & Hassan Hadi, A. M. (2016). Diagnosis of

Noroviruses in Iraqi children suffering from Gastroenteritis by Enzyme linked assay and conventional PCR. *The Medical Journal of Basrah University*, 34(1), 42-49. https://doi.org/10.33762/mjbu.2016.111187

- Mancinelli, R., Rosa, L., Cutone, A., et al. (2020). Viral Hepatitis and Iron Dysregulation: Molecular Pathways and the Role of Lactoferrin. *Molecules*, 25, 1997. https://doi.org/10.3390/molecules25081997
- Maritschnik, S., Kanitz, E. E., Simons, E., et al. (2013). A food handler associated foodborne norovirus GII.4 Sydney 2012-outbreak following a wedding dinner, Austria, October 2012. *Food and Environmental Virology*, 5(4), 220–225.
- Marpaung, D. S. S., Sinaga, A. O. Y., & Damayanti, D. (2025). Norovirus detection technologies: From conventional methods to innovative biosensors. *Analytical Biochemistry*, 698(115750). https://doi.org/10.1016/j.ab.2024.115750
- Nauss, K. M., Anderson, C. A., Conner, M. W., & Newberne, P. M. (1985). Ocular infection with herpes simplex virus (HSV-1) in vitamin A-deficient and control rats. *J. Nutr.*, 115, 1300–1315. https://doi.org/10.1093/jn/115.10.1300
- Nikawa, T., Odahara, K., Koizumi, H., et al. (1999). Vitamin A prevents the decline in immunoglobulin A and Th2 cytokine levels in small intestinal mucosa of protein-malnourished mice. *J. Nutr.*, 129, 934–941. https://doi.org/10.1093/jn/129.5.934
- Prasad, B. V. V., Atmar, R. L., & Ramani, S. et al. (2025). Norovirus replication, host interactions and vaccine advances. *Nat Rev Microbiol*, 23, 385–401. https://doi.org/10.1038/s41579-024-01144-9
- Prentice, A. M., Bah, A., Jallow, M. W., et al. (2019). Respiratory infections drive hepcidin-mediated blockade of iron absorption leading to iron deficiency anemia in African children. *Sci. Adv.*, 5(3). https://doi.org/10.1126/sciadv.aav9020
- Rifai, N. (2024). *Tietz Fundamentals of Clinical Chemistry and Molecular Diagnostics* (9th ed.). Elsevier.
- Rifai, N., Chiu, R. W. K., Young, I., Burnham, C. A. D., & Wittwer, C. T. (Eds.). (2022). *Tietz Textbook of Laboratory Medicine* (7th ed.). Elsevier. https://doi.org/10.1016/C2019-0-01496-3
- Rosa, L., Cutone, A., Lepanto, M. S., Paesano, R., & Valenti, P. (2017). Lactoferrin: A Natural Glycoprotein Involved in Iron and Inflammatory Homeostasis. *Int. J. Mol. Sci.*, 18, 1985. https://doi.org/10.3390/ijms18091985
- Sirisinha, S., Darip, M. D., Moongkarndi, P., et al. (1980). Impaired local immune response in vitamin A-deficient rats. *Clin. Exp. Immunol.*, 40, 127–135.
- Souza, M., Azevedo, M. S. P., Jung, K., Cheetham, S., & Saif, L. J. (2008). Pathogenesis and Immune Responses in Gnotobiotic Calves after Infection with the Genogroup II.4-HS66 Strain of Human Norovirus. *J Virol*, 82. https://doi.org/10.1128/jvi.01347-07
- Stoffel, N. U., & Drakesmith, H. (2024). Effects of Iron Status on Adaptive Immunity and Vaccine Efficacy: A Review. *Advances in Nutrition*, 15(6), 100238. https://doi.org/10.1016/j.advnut.2024.100238
- Waknine, Y. (2013, March 26). Norovirus Is Now the Main Cause of Acute GI Problems in Kids. *Medscape Medical News*. http://www.medscape.com/viewarticle/781459
- Wang, H., Ruan, G., Li, Y., & Liu, X. (2025). The Role and Potential Application of IL-12 in the Immune Regulation of Tuberculosis. *International Journal of Molecular Sciences*, 26(7), 3106. https://doi.org/10.3390/ijms26073106
- Ward, J. L., Torres-Gonzalez, M., & Ammons, M. C. B. (2022). The influence of viral infections on iron homeostasis and the potential for lactoferrin as a therapeutic in the age of the SARS-CoV-2 pandemic. *Nutrients*, 14(15), 3090. https://doi.org/10.3390/nu14153090
- Wiedermann, U., Tarkowski, A., Bremell, T., et al. (1996). Vitamin A deficiency predisposes to Staphylococcus aureus infection. *Infect. Immun.*, 64, 209–214. https://doi.org/10.1128/iai.64.1.209-214.1996
- Williams, A. M., Ladva, C. N., Leon, J. S., et al. (2019). Changes in micronutrient and inflammation serum biomarker concentrations after a norovirus human challenge. *The American Journal of Clinical Nutrition*, 110(6), 1456–1464. https://doi.org/10.1093/ajcn/nqz201