Table S1: Test used in the study along with reference ranges

Index name	Abbreviations	Normal reference value (adult)
White blood cell count	WBC	(3.5-9.5) ×10⁹/L
platelet count	PLT	(125-350) ×10⁹/L
Alanine aminotransferase	ALT	Male: 9-50 u/l; Female: 7-40 u/l (serum)
Aspartate aminotransferase	AST	15-40 U/L (serum)
total bilirubin	Total Bilirubin (TBIL)	3.4-20.5 μmol/L (serum)
Hyaluronic acid	Hyaluronic Acid (HA)	reference value < 100 ng/mL
Type III procollagen	Procollagen Type III (PCIII)	reference value < 120 ng/mL
Type IV collagen	Collagen Type IV (CIV)	reference value < 80 ng/mL)
Laminin	Laminin (LN)	reference value < 115 ng/mL)
Quantification of hepatitis B virus surface antigen	HBsAg	negative (< 0.05 IU/mL)
Hepatitis B virus DNA (Quantitative)	HBV DNA	negative (usually < 10 IU/mL)

In this study, we employed advanced methodologies and equipment to carry out a series of diagnostic tests assessing various biomarkers indicative of liver function and viral presence. We used the Sysmex XN Series to count the white blood cells (WBC) and platelets (PLT) [1]. Blood samples were collected via venipuncture into Ethylenediaminetetraacetic acid (EDTA) and gel tubes, and then processed, which diluted the blood and suspended it in a sheath fluid. The gel tubes used to get clot blood for serum isolation through centrifugation [2]. The analyzer used electrical impedance and laser scattering to count WBCs and PLTs. The results were then automatically compared to known reference ranges (Table S1).

We used the Bromocresol Green (BCG) method, which is a well-known colorimetric assay, to test for Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST) [3]. Clot blood was centrifuged to separate the serum, the color change in a reaction with the BCG reagent was used to measure the enzymatic activity of ALT and AST. We used a spectrophotometer to measure absorbance and then compared the results to normal reference ranges (Table S1). Vanadate oxidation method was used do measure total bilirubin (TBIL) test; the color changed in direct proportion to the amount of bilirubin in the serum [4]. We measured absorbance and compared the results to standard curves to find out how much bilirubin was in the sample. This is important for checking liver health and finding out if there is a problem with the liver or bile duct. We used the Enzyme-Linked Immunosorbent Assay (ELISA) to measure the levels of Hyaluronic Acid (HA) in the serum [5]. Serum samples were incubated with specific antibodies for hyaluronic acid. The resulting color change, which was caused by enzyme activity, was measured spectrophotometrically to find out how much HA was present. This indicates tissue remodeling and liver fibrosis. Similarly, ELISA method for Procollagen Type III (PCIII), Collagen Type IV (CIV), and Laminin (LN) [6]. This method used specific antibodies to catch these biomarkers in serum, and then enzyme-linked secondary antibodies were added. The color changed was measured with a

spectrophotometer. High levels of these biomarkers are signs of liver fibrosis. Moreover, Chemiluminescence Immunoassay (CLIA) was used to find viral antigens and nucleic acids, for Hepatitis B Virus Surface Antigen (HBsAg) and Hepatitis B Virus DNA (HBV DNA) [7]. We measure HBsAg levels and check for viral replication by finding HBV DNA.

References

1. Chen, S., et al., *Micro–red blood cell, fragmented red blood cell, platelet distribution width, mean platelet volume, and platelet–large cell ratio on Sysmex XN series hematology analyzers can be used for the reflex test of impedance platelet count in clinical practice.* Archives of Pathology & Laboratory Medicine, 2024. **148**(1): p. 61-67.
2. Ayala-Lopez, N., et al., *Comparative evaluation of blood collection tubes for clinical chemistry analysis.* Clinica Chimica Acta, 2021. **520**: p. 118-125.
3. Soleimani, N., et al., *Termination of Repeat Testing in Chemical Laboratories Based on Practice Guidelines: Examining the Effect of Rule-Based Repeat Testing in a Transplantation Center.* Journal of Analytical Methods in Chemistry, 2021. **2021**(1): p. 9955990.
4. Kittanakom, S., et al., *Impact of switching total bilirubin assays on the classification of neonates at high risk for hyperbilirubinemia.* Clinical Chemistry and Laboratory Medicine (CCLM), 2023. **61**(9): p. e175-e178.
5. Liu, Y., et al., *Carbohydrate-binding module could integrate with ELISA and serve the simple and specific quantification of hyaluronic acid.* International Journal of Biological Macromolecules, 2024. **283**: p. 137528.
6. Chu, S., Y. Chen, and Y. Wang, *Enhancing liver fibrosis detection: a novel PIGR-utilizing approach in chronic hepatitis B injury assessment.* BMC gastroenterology, 2025. **25**(1): p. 82.
7. Shen, L., et al., *Performance evaluation of the MAGLUMI Hepatitis B virus surface antigen chemiluminescence immunoassay.* Journal of Medical Virology, 2024. **96**(7): p. e29817.