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Role Of Vitamin D-Binding Protein As A Diagnostic Biomarker For Patients With Acute Meningitis

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ABSTRACT

Background *Acute inflammation of the pia, arachnoids, and fluid in the brain's subarachnoid space is known as meningitis. It is challenging to diagnose because there is no diagnostic biomarker. The purpose of this study was to assess the viability of utilizing vitamin-D binding protein (VDBP) in cerebrospinal fluid (CSF) as a novel possible diagnostic for meningitis diagnosis.* ***Patients and methods:*** *A cross-section study was conducted on 48 patients with manifestations suggesting an acute meningitis, 28 patients with an acute meningitis who were divided into bacterial group containing 10 patients and viral group containing 18 patients confirmed by laboratory investigations and 20 patients who were clinically suspected as an acute meningitis, but excluded by laboratory investigations, the study wasconducted within the period from April 2022 to November 2022. CSF and blood samples were obtained in pairs. CSF and serum VDBP were measured in the 3 groups. CSF VDBP concentrations were compared versus serum VDBP concentrations according to disease (viral meningitis vs. bacterial meningitis vs non-meningitis). Receiver operating characteristic (ROC) analysis for diagnosing meningitis using CSF VDBP concentration was performed.* ***Results:*** *There was a statistical significant difference as regard the CSF VDBP (P<0.01) was found between viral, bacterial andcontrol groups (2.49±.65, 2.43±.55 and 1.74±.25µg/mL, respectively). There was a statistical significant difference (P<0.05) in serum VDBP between the viral, bacterial and control groups (214.5±36.5, 197.4±54.8 an[1]d 174.3±40.4 µg/mL, respectively). ROC curve analysis showed that the optimum cut-off level of CSF VDBP for diagnosing meningitis was 1.94 µg/mL with a sensitivity of 82.1% and specificity of 85%. AUC of CSF VDBP was 0.865 (95% CI: 0.761–0.969).* ***Conclusion:*** *The amount of Vitamin-D Binding Protein (VDBP) in*

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cerebrospinal fluid (CSF) showed outstanding diagnostic performance. It might be used as a diagnostic marker for acute meningitis.

Keywords: *Cerebrospinal fluid, Vitamin D-binding protein, Meningitis Biomarker.*

INTRODUCTION

Acute inflammation of the pia, arachnoids, and fluid in the brain's subarachnoid space is known as meningitis [1]. Meningitis comes in two primary varieties: septic and aseptic [2], with aseptic meningitis being the most prevalent. Meningitis can have several non-infectious causes, although viral, bacterial, fungal, and parasite infections account for the majority of cases [3].
Meningitis affects 20 persons per 100,000 worldwide, or around 1.2 million cases; the cause and incidence of the disease differ throughout geographical areas. If there is meningeal inflammation and no indications of bacterial growth by CSF culture, aseptic meningitis is different from bacterial meningitis [4].

With an estimated frequency of 1.38 cases per 100,000 inhabitants and a fatality rate of 14.3% in western countries, acute bacterial meningitis (ABM) is a medical emergency [5]. 10% of ABM patients pass away if their diagnosis or course of therapy is delayed [6].

Moreover, there is a higher chance of cognitive impairment and other neurological disorders in ABM survivors. Owing to the limited effectiveness of clinical indicators in the diagnosis of meningitis, a lumbar puncture (LP) must be done as soon as feasible on any patient exhibiting symptoms of meningitis in order to assess the CSF and confirm the diagnosis. Low and mostly lymphocytic pleocytosis, normal glucose levels, and a normal to modest increase in protein levels are among the CSF findings associated with aseptic meningitis [7].
Reduced glucose levels, an increased protein level, and a strong pleocytosis that is primarily neutrophilic are the characteristics of ABM [8]. Not every patient experiences this, though. In older patients or those with immunosuppression or partially cured meningitis, it varies. The differential cell count of CSF can also be impacted by the examiner's level of expertise. Therefore, in order to identify meningitis more reliably, more new markers are needed [9].
The 58-kDa multifunctional protein known as vitamin-D binding protein (VDBP) is produced in the liver and travels throughout the bloodstream. Acute phase reactant is what it is. Consequently, the VDBP level may vary based on the circumstances [10–13]. Previously identified as group-specific component globulin, VDBP is known to have a significant role in the metabolic transport of vitamin D. However, in recent years, it has also been shown to have additional activities, including actin sequestration and immune response control [14].
The gene that encodes VDBP (GC) is polymorphic. Its genotype frequency differs according on the ethnic group. Moreover, genotype-based differences in VDBP affinity for 25(OH)D have been discovered. Three primary VDBP polymorphic variations (GC1F, GC1S, and GC2) are caused by two single-nucleotide polymorphisms (SNPs), rs7041 and rs4588. The frequency of these variations differs according on the ethnic group. The variants with the highest affinity for vitamin D include GC1F, GC1S, and GC2 [15–16].
Blood, urine samples, breast milk, CSF, saliva, seminal fluid, ascitic fluid, and the surfaces of neutrophils, monocytes, and lymphocytes have all been found to contain VDBP. The brain, heart, lung, kidney, placenta, spleen, testis, and uterus express VDBP mRNA differently [14–15]. Other body fluids were shown to have lower quantities of VDBP than blood, which has the greatest concentrations [16]. Although VDBP has been found in the CSF, its production within the CNS is unknown. VDBP immunoreactivity was recently discovered in the supraoptic and paraventricular nuclei in an animal experiment [17].

Previous research has correlated alterations inVDBP concentrations to the pathophysiology of a several illnesses, such as multiple sclerosis [13, 17, 18]. Additionally, it was discovered that severe neurodegeneration, such as Alzheimer's disease, increased intrathecal VDBP production [19]. This finding suggests that elevated VDBP may function as an actin scavenger.

Recently CSF VDBP level was reported that it could act as a new diagnostic marker for acute meningitis **[20].** The aim of the current study was to evaluate the validity of using CSF VDBP as a new potential marker for diagnosing meningitis.

PATIENTS AND METHODS

Study design: A cross-section study included 48 patients with suspected an acute meningitis who underwent LP to confirm or rule out acute meningitis. within the period from April 2022 to November 2022

Study subjects: This study was conducted in Makkah hospital on 48 patients with manifestations suggesting an acute meningitis, 28 patients with acute meningitis who were divided into bacterial group containing 10 patients and viral group containing 18 patients confirmed bylaboratory investigations, and 20 patients who were clinically suspected as acute meningitis but excludedby laboratory investigations. Patients aged more 18years old, from both sexes who were presented with clinical pictures of acute meningitis (fever, vomiting, neck stiffness, headache and convulsion) were enrolledin this study. Patients with other causes of increased intra cranial tension or other neurological disease e.g., multiple sclerosis (MS), other causes of fever, coma and chronic infection. Patients with clinical pictures suggestive of cerebro-vascular diseases, patients of chronic kidney diseases (CKD), patients with acuteand moderate to severe chronic liver diseases (CLD) and pregnancy, were excluded from this study.

Complete history taking and thorough clinical examination were done including manifestations suggesting an acute meningitis (fever, headache, convulsion, neck rigidity, positive kerning's and Brudzinsky signs and coma).

Investigations: CSF and blood samples were obtained.Serum and leukocytes were separated and kept at – 80^{o}C. CSF analysis including CSF color, aspect, pressure, chemical analysis including protein, glucose, and cellular CSF analysis including total leucocyticcount, cellular differential count, gram stain, and culture were done for each patients' CSF sample.

VDBP Assay: CSF and serum VDBP concentrations were measured utilizing an enzyme-linkedimmunosorbent assay (ELISA) kit (Solarbio, China) inaccordance to the manufac-turer's guidelines.

Other laboratory investigations included: CBC, CRP, RBS, PT. PTT, INR, Liver function tests and kidney function tests.

Imaging included: Brain CT and/or Brain MRI.

Ethical approval:
This study was ethically approved. Written informed consent was obtained from all participants. Thisstudy was executed according to the code of ethics of the World Medical Association (Declaration of Helsinki) for studies on humans.

Statistical analysis

Statistical analyses were carried out by IBM SPSS Statistics software, v 25.0. Numbers and percents (%) are calculated for categorical variables while medians and ranges are pre•sented for continuous variables. The significance of normally distributed variables was determined using one-way analysis of variance (ANOVA) followed by post hoc Tukey's test. The

correlation between VDBP levels in the CSF and in the serum was examined using simple correlation analysis. ROC curve and AUC wereutilized to examine the performance of CSF VDBP. Reference interval was calculated in accordance to guidelines of the Clinical and Laboratory Standards Institute. After ruling out outliers using Tukey method,data set was shown using nonparametric analysis (2.5– 97.5th percentile interval). P value $\leq$0.05 was set as statistically significant.

RESULTS

Forty-eight subjects were included with a median age of 23 years. Percentage of females was 45.83%. Among these 48 patients.10 patients were in bacterial meningitis group (20.83%), 18 patients were in viral meningitis group (37.5%) and 20 patients were in control group (41.66%). A significant differenceexisted between groups as regards convulsion(P<0.01), also regarding neck stiffness (Table 1).

Variables		**acterial infection group (N=10)**		**Viral infection group (N=18)**		**Control(N=20)**		**Kruskal-Wallis H**	**Sig.**
		Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**		
Age		26.50	22.38	20.27	16.41	26.35	16.32	1.658	0.437
N			**%**	**N**	**%**	**N**	**%**	**X^2**	**Sig.**
Gender	**Male**	4	40.0%	11	61.1%	11	55%	1.164	0.559
	Female	6	60.0%	7	38.9%	9	45%		
Residence:		100% urban							
Clinical findings								**P-value**	**Sig.**
Fever		7 (70%)		12 (66.7%)		12 (60%)		0.859	0.390
Headache		7 (70%)		10 (55.6%)		5 (25%)		0.34	2.16
Vomiting		6 (60%)		9 (50%)		6 (30%)		0.41	1.79
Convulsion		5 (50%)		2 (11.1%)		0 (0%)		**0.039***	6.49
Neck stiffness		10 (100%)		12 (66.7%)		2 (10%)		**0.002***	23.04
Kering's sign		9 (90%)		10 (55.6%)		0 (0%)		**0.003***	18.84
Brudzinski's sign		10 (100%)		9 (50%)		0 (0%)		**0.005***	15
Pulse Mean ±SD		84.5±8.2		78.8±7.8		84.68±5.6		0.052	5.898
Temperature Mean ±SD		37.8±1.2		37.4±0.6		37.6±0.9		0.737	0.611
Systolic PB Mean ±SD		125±10		124±12		120±5		0.411	1.781
Diastolic PB Mean ±SD		83±6		82±7		80±2		0.570	1.125

A statistical significant difference was found between groups regarding CRP (P<0.01). However, there were no statistical significant differences (P-value >0.05) regarding Hb, WBCs, Platelets, RBS, ALT, AST, Albumin, PT, PTT,INR, Creatinine, Urea (Table 2).

Groups **Variables**	**Bacterial infection group (n= 10)**	**Viral infection group(n=18)**	**ıtrol group (n= 20)**	**Test of significance Kruskal-WallisTest**	**P-value**
HB(g/d), Mean ±SD	12.3±1.5	13±1.2	12.7±1.0	0.902	0.637
WBCs(/L), Mean ±SD	12.8±2.9	12.2±2.4	10.8±2.4	3.483	0.175

Platelet (/µL), Mean ±SD	270.5±62.6	282.3±35.1	278.0±43.1	0.500	0.779
CRP (mg/dL), Mean ±SD	62.8±15.3	50.9±12.6	24.9±5.1	18.406	**<0.010****
RBS (mg/dL), Mean ±SD	101.0±24.3	105.0±25.1	107.0±26.3	1.216	0.545
ALT(u/L), Mean ±SD	22.1±5.4	22.5±5.5	21.4±5.2	0.306	0.858
AST(u/L), Mean ±SD	36.7±8.8	40.1±9.8	33.8±8.3	0.285	0.867
Albumin (gm/dl)Mean ±SD	4.3±0.43	3.9±0.27	4.0±0.39	4.743	0.093
PT (Seconds)Mean ±SD	16.5±3.0	18.0±4.3	16.5±3.9	1.190	0.552
PTT (Seconds)Mean ±SD	28.0±2.9	28.3±2.0	28.8±2.2	2.204	0.332
INR, Mean ±SD	1.27±0.30	1.35±0.32	1.22±0.30	0.666	0.717
Creatinine (mg/dL) Mean ±SD	0.9±0.1	1.0±0.2	0.98±0.23	1.209	0.546
Urea (mg/dL)Mean ±SD	22.3±5.4	30.3±7.4	27.3±6.7	4.073	0.130

\A significant difference in CSF TLC was found among the bacterial, viral and control groups (P<0.001). As regards the CSF pressure, a significant difference existed among bacterial, viral and control groups. The mean value ofCSF pressure was significant in the bacterial group and higher than the CSF pressure mean of the control group andthe mean of the viral group**.** As regards the CSF Protein, a significant difference was reported between bacterial, viral and control groups. The mean value of CSF Protein was significant in the bacterial group and higher than the in CSF protein mean of the viral group and the mean of the control group (Table 3).

Groups **Variables**	**Bacterial infection Group (n= 10)**	**Viral Infection group (n=18)**	**Control group (n= 20)**	**Test of significance Kruskal-Wallis Test**	**P-value**
CSF TLC (/uL) Mean ±SD	8981.11±244.1	87.78±20.81	0.000	38.36	<0.001 **
CSF pressure (cmH_2O) Mean ±SD	3.8±0.63	2.8±0.4	3.1±0.74	4.93	0.012*
CSF protein (mg/dL) Mean ±SD	290.9±64.8	94.44±23.3	48.1±11.8	18.8	<0.001**
CSF glucose (mg/dL) Mean ±SD	43.7±10.61	78±17.4	84.6±20.42	5.29	0.07

A significant difference in CSF color was found among the bacterial, viral and control groups. The color was clear in 20% in bacterial, 72.2% in viral and 100% in control. As regards the CSF aspect, a significant difference was found between bacterial, viral and control groups. The aspect was clear in 30% in bacterial, 72.2% in viral and 100%in control (Table 4).

Variables	**Bacterial (10)**	**Viral (18)**	**Control (20)**	**Test of significance (X^2)**	**P-value**

CSF color Clear White Yellow Red	 2(20%) 2(20%) 4(40%) 2(20%)	 13(72.2%) 0(0%) 1(5.6%) 4(22.2%)	 20(100%) 0(0%) 0(0%) 0(0%)	17.6	**<0.001****
CSF aspect Clear Turbid	 3(30%) 7(70%)	 13(72.2%) 5(27.8%)	 20(100%) 0(0%)	23.6	**<0.001****
Bacterial infection S.aureus Strept/pneumonia E. Coli	 2(20%) 6(60%) 2(20%)	-	-	-	-

As regards the CSF VDBP, a significant difference was detected among bacterial, viral and control groups. There was a statistically significant difference in Serum VDBP between bacterial, viral and control groups (Table 5).

Groups **Variables**	**Bacterial infection group (n= 10)**	**Viral infection group (n=18)**	**ontrol group (n= 20)**	**Test of significance Kruskal-Wallis Test**	**P-value**
CSF VDBP (µg/mL) Mean ±SD	2.43± 0.55	2.49± 0.61	1.74± 0.25	18.301	** **0.01**
Serum VDBP (µg/mL) Mean ±SD	197.4±48.7	214.5±6.5	174.3±40.4	7.623	* **0.05**

At a cut off value 1.94 mg/dl of the marker (VDBP), the area AUC was 0.865 meaning that it has a strong diagnostic performance with sensitivity, specificity, PPV, NPV, accuracy, 95% CI, P (82.1%, 85%, 88.5, 77.3, 83.3, 0.761-0.969,
< 0.001 respectively) (Table 6)

Variable	**AUC**	**Cutoff**	**Sensitivity**	**Specificity**	**PPV**	**NPV**	**Accuracy**	**95% CI**	**P-value**
CSF VDBP level	0.865	1.94 mg/mL	82.1%	85%	88.5	77.3	83.3	0.761–0.969	0.001

Plot graphs for CSF VDBP levels **(Figure 1)** and for serum VDBP levels were obtained in the three groups (Figure 2). ROC curve analysis was used to calculate the optimum cut-off level or CSF VDBP in order to diagnose meningitis. The optimum cut-off of CSF VDBP was 1.94 µg/mL with a sensitivity of 82.1% and specificity of 85%. AUC of CSF VDBP was 0.865 (95% CI: 0.761–0.969) (Figure 3).

Figure 1

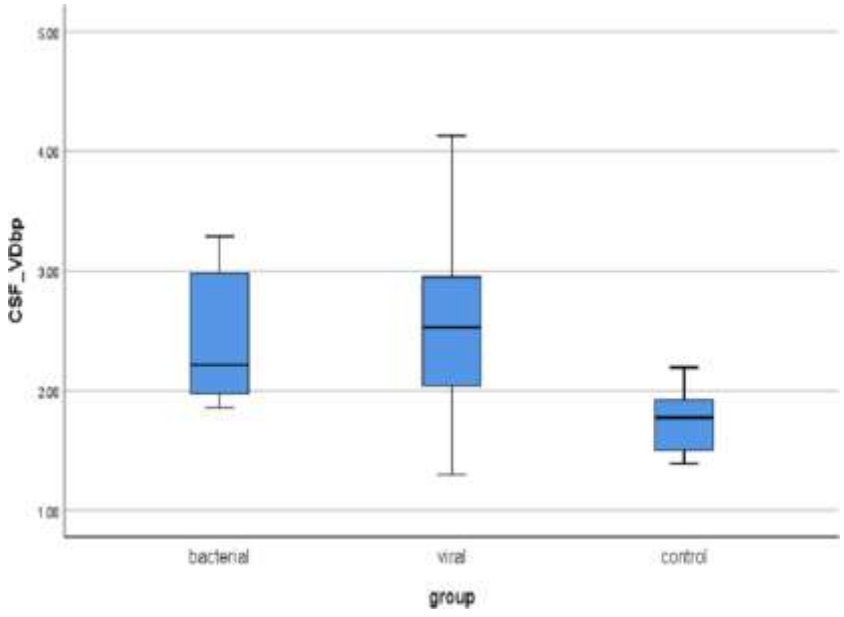


Figure 2

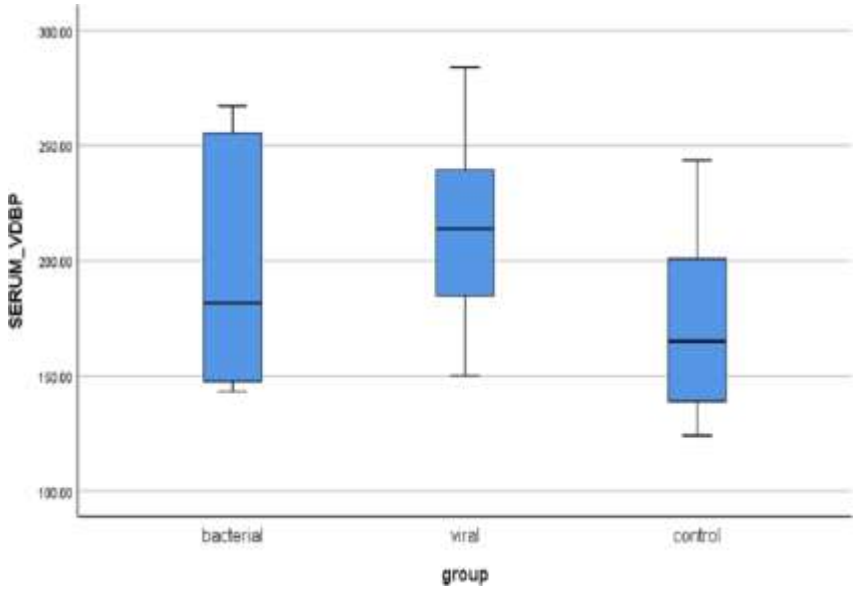
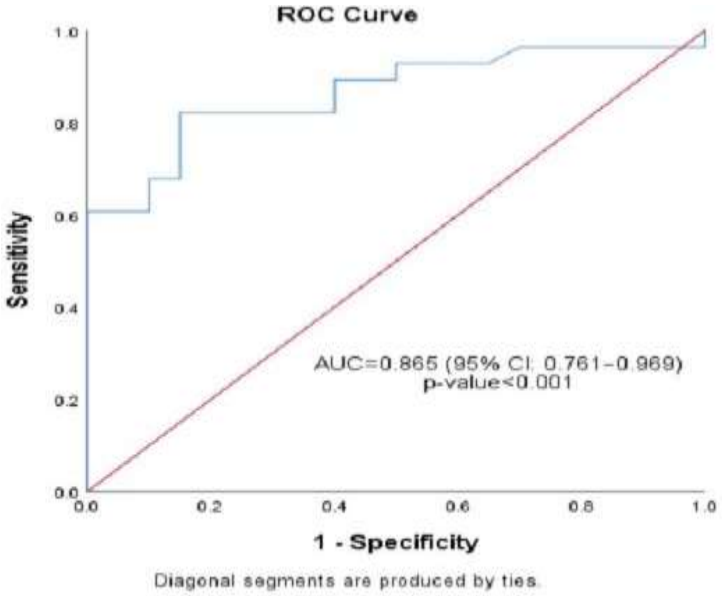


Figure 3



DISCUSSION

A meningeal infection brought on by a bacterial, viral, or fungal infection is known as meningitis. Serious consequences, including death and brain damage, may result from it [1]. In order to increase the accuracy of meningitis diagnosis, additional biomarkers are required, as clinical symptoms and CSF findings are not trustworthy enough to rule out meningitis [21].
The liver produces VDBP, which is then released into the bloodstream. Acute phase reactant is what it is. Its amount can therefore vary based on the situation and be employed as a diagnostic biomarker [10,11].

Convulsions were present in 50% of cases of bacterial meningitis, 11.1% of cases of viral meningitis, and 0% of cases in the control group. There was a significant statistical difference in convulsions between the bacterial, viral, and control groups (p-value < 0.05). This results was in partial agreement with **Brouwer et al.** [22] who found that convulsions occur in approximately 20% to 40% of adults with bacterial meningitis.
There was a significant statistical difference (P-value <0.05) between the bacterial and viral groups when it came to meningeal irritation signs, such as neck rigidity, positive Kerning's, and Brudzinsky signs. These signs were present in 100%, 90%, and 100% of cases of bacterial meningitis, 66.7%, 55.6%, and 50% of viral cases, and 10%, 0%, 0% in the control group, respectively. Our results were consistent with the Ndreu et al. [23] study, which found that 73.1%, 55.2%, and 56.7% of patients with meningitis had neck rigidity, positive Kerning's, and Brudzinsky symptoms, respectively.
A significant statistical difference (P-value <0.01) existed amongst groups regarding CRP. Themean of CRP in the bacterial group (62.8±24) was higher compared with the viral group (50.9±22.5), and the control group (24.9±9.4). Our findings corroborated those of Streharova et al. [24], who discovered that CRP is present in 3.6% of viral meningitis and 19.4% in bacterial meningitis.

Regarding the physical CSF characters, significant differences were detected between cases with bacterial meningitis and cases with viral meningitis regarding tension, color and aspect of CSF:

Regarding the CSF color, a significant difference existed among bacterial, viral and control groups (Chi-square Test=17.6, P<0.01). Change in CSF color was found in 80% in

bacterial, 27.8% in viral and 0% in control. These results were in agreement with **Lebel et al.** [25] who found that CSF color was significantly different between subjects with bacterial meningitis and subjects with viral meningitis. In the bacterial meningitis group, CSF color was more likely to be cloudy or purulent, while in the viral meningitis group, CSF color was more likely to be clear or slightly cloudy.
Regarding the CSF component, the bacterial, viral, and control groups showed a statistically significant difference (Chi-square Test=23.6, P<0.01). 70% of the bacterial, 27.8% of the viral, and 0% of the control groups had turbidity. These outcomes were in line with those of Wang et al. [26], who discovered that bacterial meningitis had much higher CSF turbidity than viral meningitis.

A there was a statistical significant differenceamong the bacterial, viral and control groups as regards the CSF pressure (Kruskal-Wallis Test=4.93, P<0.05). The mean pressure was significant in the bacterial group (μ = 3.8±0.63) and higher than the CSFpressure mean of the control group (μ = 3.1±0.74) and the mean of the viral group (μ = 2.8±0.78). The TLC was higher in the bacterial group (μ= 8981.11±31539.10) than that of the viral group (μ = 87.78±97.51), and control groups (Kruskal-Wallis Test=38.36, P<0.001) and this was statisticaly significant, this was in agreement with **Jyoti et al.** [27] who found a significant difference in CSF TLC between the bacterial, viral, and control groups. The mean CSF TLC was highest among the bacterial meningitis cases (258 cells/mm3), followed by the viral meningitis cases (85 cells/mm3), and controls (2 cells/mm3).

As regards CSF protein, a significant difference existed among the three study groups (Kruskal-Wallis Test=18.8, P<0.01). The mean values of in CSF protein were in bacterial, viral and control groups (μ = 290.9±64.8, 94.44±108.44, and 48.1±24.8,
respectively). These results aligned with those of Brouwer et al. [28], who reported a substantial difference in mean CSF protein levels between the control group (0.28 g/L) and the bacterial meningitis group (2.99 g/L).

Our study demonstrated that, culture results in bacterial meningitis cases were positive in 10 cases (100%). The most commonly isolated microorganisms were St. pneumoniae in 6 cases (60%), Staph aureus in2 cases (20%) and E Coli in 2 cases (20%). This was comparable to the findings of Brouwer et al. [29], who discovered that 70–85% of cases of bacterial meningitis had a positive CSF culture.

In the current study, CSF VDBP was statistically significantly higher in meningitis groups than control group (P<0.01) and This was consistent with the findings of Lee et al. [20], who discovered that CSF VDBP was greater in cases of acute meningitis than in controls. In the present study ROC curve analysis revealed that CSF VDBP concentrations in meningitis cases had a high predictive value. CSF VDBP has a sensitivity of 82.1% and a specificity of 85 % at a cut- off level of 1.94 μg/mL for diagnosing meningitis and this was in agreement with **Lee et al.** [20] who found that at a cut off level of 1.96 μg/mL, the CSF VDBP has an excellent diagnostic performance, withsensitivity of 82.4% and specificity of 85.9%.

This suggests that CSF VDBP levels could serve as a valuable novel biomarker for diagnosingmeningitis.

CONCLUSION

Excellent diagnostic performance is demonstrated by the CSF VDBP cut-off level of 1.94 μg/mL, which has an AUC of 0.865, 82.1% sensitivity, and 85% specificity. Therefore, CSF VDBP may serve as a diagnostic marker for acute meningitis.

REFERENCES

1. **Wadei H, Mai M, Ahsan N et al. (2006):** Hepatorenal syndrome: pathophysiology and

management. Clinical Journal of the American Society of Nephrology, 5:1066-79.

2. **Pellicoro A, Ramachandran P, Iredale J et al. (2014):** Liver f ibrosis and repair: immune regulation of woundhealing in a solid organ. Nat Rev Immunol., 14:181-94.
3. **Ginès P, Krag A, Abraldes J et al. (2021):** Liver cirrhosis. The Lancet, 398(10308):1359-76.
4. **Weng F (2021):** Comment on urinary NGAL as adiagnostic and prognostic marker for acute kidney injury in cirrhosis. Clin Transl Gastroenterol.,12(7):e00382. doi: 10.14309/ctg.0000000000000359.
5. **Carvão J, Carvão J, Pereira V et al. (2020):** Challenges in measuring renal function in liver cirrhosis: Are there implications in clinical practice? Acta GastroenterolBelg., 83(4):633-8.
6. **Montoliu S, Ballesté B, Planas R et al. (2010**): Incidence and prognosis of different types of functional renal failure in cirrhotic patients withascites. Clin Gastroenterol Hepatol., 8:616-22.
7. **Slack A, Yeoman A, Wendon J (2010):** Renal dysfunction in chronic liver disease. Crit Care, 14:214- 8.
8. **Walls A, Bengaard A, Iversen E et al. (2021):** Utility of SUPAR and NGAL for AKI risk stratification and early optimization of renal risk medications among older patients in the emergency department. Pharmaceuticals, 14(9):843-8.
9. **Reddy S, Wyawahare M, Priyamvada P et al. (2020):**Utility of urinary neutrophil gelatinase associated lipocalin (NGAL) in decompensated cirrhosis. Indian Journal of Nephrology, 30(6):391-7.
10. **Schmidt-Ott K, Mori K, Li J et al. (2007):** Dual action of neutrophil gelatinase-associated lipocalin. J Am Soc Nephrol., 18(2):407-13.
11. **Parikh C, Coca S, Thiessen-Philbrook H et al. (2011):** Postoperative biomarkers predict acute kidney injury and poor outcomes after adult cardiac surgery. J Am Soc Nephrol., 22(9):1748-57.
12. **Haase-Fielitz A, Bellomo R, Devarajan P et al. (2009):** The predictive performance of plasma neutrophil gelatinase-associated lipocalin (NGAL) increases with grade of acute kidney injury. Nephrol Dial Transplant., 24(11): 3349-54.
13. **Kellum J, Lameire N (2013):** KDIGO AKI guideline work group. Diagnosis, evaluation, and management ofAKI: a KDIGO summary (Part 1). Crit Care, 17:204. doi: 10.1186/cc11454.
14. **Angeli P, Gines P, Wong F et al. (2015**): Diagnosis and management of acute kidney injury in patients with cirrhosis: revised consensus recommendations of the international club of ascites. Gut, 64(4):531-7.
15. **Verna E, Brown R, Farrand E et al. (2012):** Urinary neutrophil gelatinase-associated lipocalin predicts mortality and identifies acute kidney injury in cirrhosis.Digestive Diseases and Sciences, 57:2362-70.
16. **Levy K, Meehan K, Kelly K et al. (2006):** Change in attachment patterns and reflective function in a randomized control trial of transference-focused psychotherapy for borderline personality disorder.Journal of Consulting and Clinical Psychology, 74(6):1027-40.
17. **Rimola A, Gracia-Tsao G, Navasa M et al. (2000):** Diagnosis, treatment and prophylaxis of spontaneous bacterial peritonitis: a consensus document. International Ascites Club. J Hepatol., 32:142-53.
18. **Kamath P, Wiesner R, Malinchoc M et al. (2001):** Amodel to predict survival in patients with end-stage liver disease. Hepatology, 33:464-70.
19. **Ruf A, Kremers W, Chavez L et al. (2005):** Additionof serum sodium into the MELD score predicts waitinglist mortality better than MELD alone. Liver Transpl., 11:336-41.
20. **Pugh R, Murray-Lyon I, Dawson J et al. (1973):** Transection of the oesophagus for bleeding oesophageal varices. Br J Surg., 60:646-9.
21. **Belcher J, Garcia-Tsao G, Sanyal A et al. (2013):** Association of AKI with mortality and complications inhospitalized patients with cirrhosis. Hepatology, 57(2):753-62.
22. **Garcia-Tsao G, Parikh C, Viola A (2008):** Acute kidney injury in cirrhosis. Hepatology, 48:2064-77.
23. **Mehta R, Bouchard J (2011):** Controversies in acute kidney injury: effects of fluid overload on outcome. Contrib Nephrol., 174:200-11.
24. **Gines P, Angeli P, Lenz K et al. (2010**): EASL clinical practice guidelines on the management of ascites, spontaneous bacterial peritonitis, and hepatorenal syndrome in cirrhosis. J Hepatol.,

53:397-417.

25. **Baboolal K, Jones G, Janezic A et al. (2002):** Molecular and structural consequences of early renal allograft injury. Kidney International, 61(2):686-96.
26. **Ronco C (2007):** NGAL: diagnosing AKI as soon as possible. Critical Care, 11(6):173. doi: 10.1186/cc6162.
27. **Bachorzewska-Gajewska H, Malyszko J, Pawla K etal. (2007):** Could neutrophil-gelatinase-associated lipocalin and cystatin C predict the development of contrast-induced nephropathy after percutaneous coronary interventions in patients with stable angina and normal serum creatinine values? Kidney and Blood Pressure Research, 30(6):408-15.
28. **Fagundes C, Pépin M, Guevara M et al. (2012):** Urinary neutrophil gelatinase-associated lipocalin as biomarker in the differential diagnosis of impairmentof kidney function in cirrhosis. Journal of Hepatology, 57(2):267-73.
29. **Qasem A, Farag S, Hamed E et al. (2014):** Urinary biomarkers of acute kidney injury in patients with liver cirrhosis. International Scholarly Research Notices, 14:376795. doi: 10.1155/2014/376795.
30. **Hamdy H, El-Ray A, Salaheldin M et al. (2018)**: Urinary neutrophil gelatinase-associated lipocalin in cirrhotic patients with acute kidney injury. Annals of Hepatology, 17(4):624-30.
31. **Gungor G, Ataseven H, Demir A et al. (2014):** Neutrophil gelatinase-associated lipocalin in prediction of mortality in patients with hepatorenal syndrome: a prospective observational study. Liver International, 34(1):49-57.
32. **Udgirkar S, Rathi P, Sonthalia N et al. (2020)**: Urinary neutrophil gelatinase-associated lipocalin determines short-term mortality and type of acute kidney injury in cirrhosis. JGH Open, 4(5):970-7.