**Original study**

# Body Fluid Analysis by Automated Sysmex XN 1000 – The way ahead

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Submitted: 31-May-2024
Revised: 22-Jun- 2024
Accepted: 28-Jun-2024
Published: 28-Jun-2024

**Abstract**

Body fluids are those excreted or secreted from the body. In the laboratory, the use of the term "body fluids" designates a category that excludes blood and urine. Determining the etiologic cause of fluid accumulation in various body cavities (i.e., joints, chest, and abdomen) is critical for proper treatment of various disorders. To evaluate the correlation and agreement of leucocyte and erythrocyte count in body fluids among Improved Neubauer Chamber and automated method using Sysmex XN-1000 body fluid module. A total of 500 routinely collected body fluid samples (CSF, ascitic fluid, pericardial fluid and pleural fluid) were included in the study. Total WBC counts were performed manually using improved Neubauer chamber and microscopic differential counts were performed using Leishman stained slides. Corresponding results of Sysmex XN-1000 body fluid module analysis were matched to these particular count categories. Absolute WBC cell counts showed a high correlation and agreement between methods (r> 0.9, p value=<0.001). However, a fair correlation existed between the lymphocyte percentage and neutrophil percentage measured through the two methods (r= 0.436 and r=0.447 respectively, p < 0.001). Body fluid module application of Sysmex XN-1000 serves as an acceptable alternative to manual method for measuring the total nucleated cell counts and for total RBC count. However, it cannot be recommended as a suitable alternative for manual differential cytologic workup.

**Keywords:** Sysmex XN 1000, Body fluid analysis, Cerebrospinal fluid analysis, Improved Neubauer Chamber



*How to cite this article*: *Chaturvedi S,Gupta GN, Faujdar M, Jain R, Rateesh S. Body Fluid Analysis by Automated Sysmex XN1000-The way Ahead, Int J Clinicopathol Correl. 2024; 8(1):22-28. 10.56501/intjclinicopatholcorrel.v8i1.1075. Copyright © 2024 Swetha chaturvedi*

## Introduction

Body fluids include fluids that are excreted or secreted from the body. In the laboratory, the use of the term "body fluids" designates a category that excludes blood and urine.

Determining the etiologic cause of fluid accumulation in various body cavities (i.e., joints, chest, and abdomen) is critical for proper treatment of various disorders. The microscopic analysis of the body fluids,

particularly the cell count provides valuable diagnostic information to the clinicians. For instance, elevated white blood cell (WBC) counts in a CSF sample (a mononuclear cell count of >5/µL in adults or >30/µL in newborns) can indicate one of several serious medical conditions, such as meningitis, encephalitis, neurologic disorders, and leukemic CSF infiltrations[1]. Spontaneous bacterial peritonitis is suspected when a large number of polymorph nucleated cells (PMNs) (PMN >250 x $10^6$/L) are present in ascites. Similarly, a high number of WBCs (WBC >100x$10^6$/L with >50% PMN) is indicative of peritonitis [2]. Cell counts above 10000/µL in pleural fluids are usually associated with Para pneumonic effusions. RBC counts are important for the diagnosis of intracerebral hemorrhage and for the exclusion of a traumatic tap as the cause of an elevated WBC count [1].

For peripheral blood, the automated cell count and leucocyte differential count are well established and precise, whereas for body fluids the cellular components have conventionally been enumerated microscopically by using a hemocytometer counting chamber. However, performing a manual body fluid count is time consuming, depends on skilled personnel, shows inter-observer variability, and can produce errors. Therefore, introduction of automated methods of analysis was sought after to improve accuracy, precision and turnaround time.

Sysmex XN-1000 (Sysmex America, Inc.) is one such multi-parameter automated hematology analyzer which is equipped with body fluid mode and offers to provide easy to use, flexible and simplified work flow module with better precision and accuracy. However, these methods are often hampered by electronic back ground noise, which might falsely elevate the cell count. So the present study has been undertaken to compare the performance of automated analyzer (Sysmex XN-1000 Body fluid module) with the manual improved Neubauer Chamber count in terms of accuracy and easier to use.

## Materials and Methods

The present study was conducted on the patients attending outpatient and inpatient department of Brig.T.K.Narayanan department of pathology, in a tertiary care hospital in western India, for a period of one year. The body fluids included-CSF, Pleural, pericardial and ascitic fluid. The study excluded clotted, extremely viscous and mucoid samples.

The gold standard reference method for body fluid cell counts and differential cell types was manual method using the improved Neubauer chamber. The samples were centrifuged at 2500rpm for 5 minutes and sediment was used for making smear for Leishman stain for differential count examination. All the body fluid samples after being examined by microscopic method were further run on Sysmex XN 1000 automated analyzer (body fluid mode). No sample pre preparation was required prior to sample analysis by Sysmex XN 1000 automated analyzer. While the RBCs are counted in the RBC channel using the sheath flow impedance technology; the WBC, mononuclear and polymorphonuclear cells are determined by flow cytometry in the differential channel. The differential channel combines forward scatter (size of cell) with side scatter (inner complexity of the cell) and the fluorescence intensity (DNA/RNA content) to identify and cluster each cell [3]. Statistical analysis was performed with the SPSS, version 20 for Windows statistical software package (SPSS inc., Chicago, il, USA). The mean, SD, and CV were calculated for each sample included in the study and regression analysis was performed. Pearson's correlation for normally distributed data and spearman correlation for non normal distributed data

was determined. Probability P value <0.05 was considered statistically significant.

## Results

The study population included 500 samples, out of which maximum samples were of CSF (51%), followed by Pleural Fluid (27.2%), Ascitic Fluid (19.4%) and least were of Pericardial Fluid (2.4%). (Figure 1)

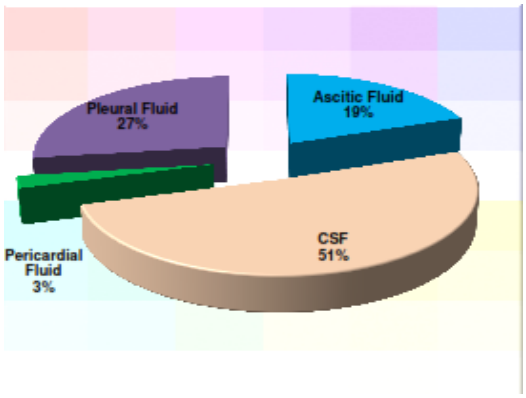


**Figure 1: Distribution of study population according to the type of fluid**

Table-1 shows various parameters measured by Sysmex XN 1000 automatic hematology analyzer including WBC count, RBC count, mononuclear cell count & percentage and similarly polymorphonuclear cell count & percentage. Table-2 shows descriptive analysis of manual count for WBC and differential counts.

**Table 1: Descriptive analysis of Total WB cell count XN 100**

| | WBC /mm3 | MN# | MN% | PMN # | PMN% | RBC |
|---|---|---|---|---|---|---|
| N | 500 | 500 | 500 | 500 | 500 | 500 |
| Mean | 2165.23 | 0.675 | 55.928 | 1.669 | 43.652 | 0037 |
| SD | 7288.25 | 1.782 | 31.948 | 7.64 | 31.861 | 0.458 |
| Median | 125 | 0.064 | 57.70 | 0.028 | 1.845 | 0.001 |

A significant positive correlation was seen between the WBC count measured by manual (improved Neubeaur chamber)method and Sysmex XN 1000 automatic hematology analyzer (r=.935, p < .001) by using pearson's coefficient of correlation. However, the differentiation of WBC into polymorphonulcear cells and mononuclear cells showed poor correlation- 0.199 & 0.439 respectively (Table-3).

**Table 2: Descriptive analysis of nucleated cells manual count**

| | WBC/mm3 | Lymphocytes % | Neutrophils % |
|---|---|---|---|
| N | 500 | 500 | 500 |
| Mean | 1400.31 | 73.80 | 26.04 |
| SD | 6007.621 | 28.012 | 27.871 |
| Median | 200 | 90 | 10 |

**Table 3: Correlation of WBC count by XN 1000 Sysmex and manual count**

| | Correlation |
|---|---|
| N | 500 |
| WBC | 0.935 |
| Neutrophils | 0.199 |
| Lymphocytes | 0.439 |
| P value <.001 | |

The total WBC count showed a good correlation between both manual & Sysmex XN 1000 method. (Figure 2) The differential count determination of type of WBC population (polymorphs, lymphocytes) did not yield good correlation between the two methods as shown in figure 3 and figure 4 respectively.

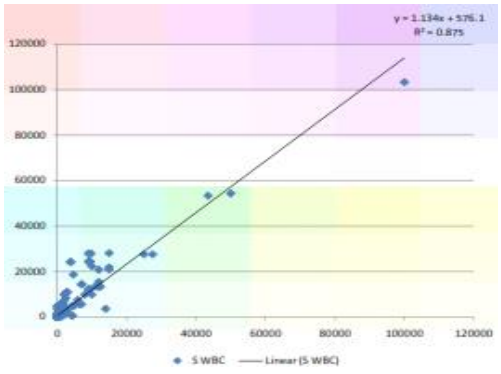

**Figure 2: Correlation of Total WBC count by manual method and Sysmex XN 1000 (6 part)**

Sysmex XN 1000 automatic hematology analyzer correctly detected 0-5 cell range in 70% of cases however overestimated the more than 5 cell range.

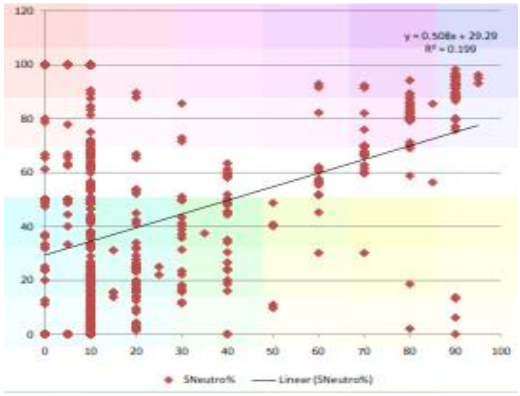

**Figure 3: Correlation of Total Neutrophil % by manual method and Sysmex XN 1000 (6 part)**

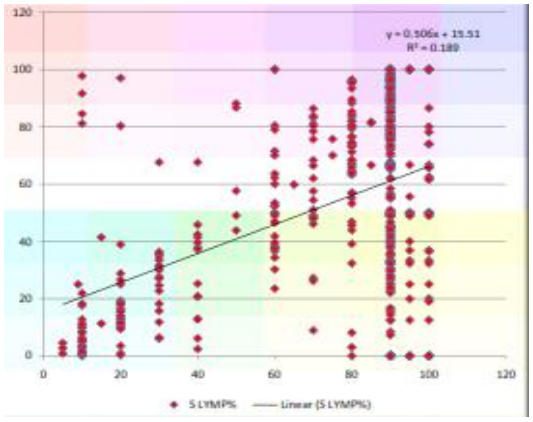

**Figure 4: Correlation of Total Lymph% by manual method and Sysmex XN 1000 (6 part)**

On further classifying the body fluid as CSF and non-CSF fluid the coefficient of correlation for WBC count by manual and automated method was 0.946 & 0.947 each. [ p value <.001] (Table-4).

Table-5 shows WBC count measured by two different methods in CSF. The manual method estimated less than 5 cells / cumm in 150 samples and more than 5 cells / cumm in 105 samples of CSF.

Table-6 shows WBC count for fluids other than CSF where XN 1000 correctly identified cells in range of <500/cumm and 500-1000 / cumm cell range whereas above 10,000 /cumm cell range were overestimated by Sysmex XN 1000 automatic hematology analyzer. The findings were statistically significant.

**Table 4: Correlation of WBC count by XN 1000 Sysmex and manual count for CSF fluid only**

| WBC Count | XN 1000 Sysmex | Manual count ( Gold standard) |
|---|---|---|
| 0-5 / mm3 | 150 (58.82%) | 105 (41.18%) |
| >5 / mm3 | 105 (41.8%) | 150 (58.82%) |
| XN 1000 success rate – 105/150 = 70%<br>Failure – overestimation for WBC >5/mm3 (30%) | | |

**Table 5: Comparative analysis of WBC count by XN 1000 Sysmex and manual count for body fluids other than CSF**

| WBC Count | XN 1000 Sysmex | Manual count ( Gold standard) |
|---|---|---|
| <500 / mm3 | 131 (53.47%) | 105 (42.86%) |
| 500-10,000 / mm3 | 99 (40.41%) | 113 (46.12%) |
| >10,000 / mm3 | 15 (6.12%) | 27 (11.02%) |

**Table 6: Correlation of WBC count by XN 1000 Sysmex and manual count CSF & Other fluids**

| | CSF | Other fluids |
|---|---|---|
| Pearson's coefficient | 0.946 | 0.947 |
| n | 255 | 25 |
| P <.001 | | |

The RBC count of the total 180 samples belonging to group A (Non zero group) out of these samples 152(84.44%) showed RBC count more than zero on Sysmex XN 1000 automatic hematology analyzer, the remaining 28 (15.56%) showed zero RBC count. Similarly, of 320 samples in group B (Nil RBC group) 227 (70.945) showed zero RBC count by XN 1000 analyzer and 93 (29.06%) showed RBC count of more than zero (Table-7).

The agreement rate of XN 1000 for group A (Zero group) was 84.4% and for group B (other than zero) was 71%.

**Table 7: Correlation of RBC count by XN 1000 Sysmex and manual count CSF & Other fluids**

| | Number of samples - 500 | |
|---|---|---|
| | Any number (Group A) 180 | Nil (Group B) 320 |
| Positive | 152 (84.44%) | 93 (29.06%) |
| Negative | 28 (15.56%) | 227 (70.94%) |

## Discussion

The prospective study on body fluids at the tertiary care hospital in India showed good correlation between WBC counts measured by manual (Improved Neubauer chamber) and Sysmex XN 1000 automatic hematology analyzer for CSF (r- 0.946) and for fluids other than CSF (r- 0.947). The differentiation of WBC into polymorphs and lymphocytes by Sysmex XN 1000 automatic hematology analyzer was not satisfactory and hence mandates manual microscopy for identification of WBC cell types.

The findings of our study are similar to those by Paris et al[4] who did body fluid study using XE 5000 and found coefficient of correlation for WBC count r= >0.99.

Nagisa Nakazamia[5] in their study on XN 2000 obtained good correlation for WBC count by manual and automatic methods (r=0.85-1). Similarly, Fleming [6] on XN 1000 Sysmex obtained good correlation between WBC and RBC count by manual and automatic methods. TM Lehto [7] in their study on XT 4000i obtained poor correlation of CSF WBC count between manual and automatic methods. The differential classification of WBC into Polymorphs and Lymphocytes was not satisfactory in our study. Our findings are supported by studies from Hoffman [8], Aulesa [9], Andrea Perrne et al [10], De Jong et al [11] and Conner et al[12] on XE 5000 system.

The study by James William[13] showed good correlation between polymorph and lymphocyte identification by Automatic and Manual methods. The differentiation of WBC into polymorphs and lymphocytes is vital in diagnosis. The present study finding are in favor of using manual differential than automated as a current generation of automated body fluid analyzers will require future improvement before one could confidently rely up on automation in body fluid analysis.

In our study, XN 1000 could estimate correctly 70% of CSF samples with low WBC count (0-5/cumm). There was over estimation of WBC count for CSF samples having more than 5 WBC per cumm. The results were not statistically significant between the two methods. Our findings are contrary to the findings by Takemura et al [14] who on XE 5000 found that body fluid samples with high WBC count had better correlation between two methods of WBC cell counting. The findings of Boer

et al [15] were similar to those of the present study as they concluded that samples with low cell count in CSF had better correlation between manual and automatic methods.

As for the red cell count our study had concordat findings with other literature studies by Fleming[6], Aune[16] & De Jonge[17]. It is possible that with further advancement in technology the differential counting by automated body fluid analysis are done with better accuracy and efficiency with greater emphasis on identification of malignant cells as highlighted by high fluorescence cells. However, the undisputed advantage in terms of shorter turnaround time as also supported in studies by T M Lehto[7] and Giuseppe et al[18]. The conventional microscopic analysis of body fluid include time consuming steps like centrifugation of samples preparing of slide, Neubauer chamber charging and then review by pathologist whereas XN 1000 analyzer requires only small sample volume 88 uL and interprets result within 2 minutes. We observed differences in various literature studies related to correlation between manual and automatic analyzer body fluid as analysis are attributed to different types of automatic analysis, different sample size, different technique of slide staining and expertise as well as experience of pathologist .

To summarize, based on our study and other available literature evidence we may conclude that nucleated cells and RBC count in emergency situation or in situation where expertise of pathologist is not available the automated body fluid analyzer using X N 1000 Sysmex provides a good alternative. It could be used as screening tool where high blood fluid samples are received in laboratory as the cell count serve as vital information for screening and decision making. As with all advancements it is up to the user to utilize the instrument judicially without compromising the quality of reporting, improving TAT and ultimately patient care.

The limitation of instrument well known to the uses so that the application in areas at done in a prudent manner.

## Conclusion

XN 1000 body fluid mode offers quickly cell counting (WBC & RBC) for body Fluids. The typing into polymorphs and lymphocytes however needs to be more efficient and future analysis and more literature studies with up gradation of the technology are needed as this part of analysis is neglected in smaller Laboratories and health care setting**.**

## Reference

1. Kjeldsberg CL, Knight JA. Cerebrospinal fluid. In: Body Fluids. 3rd ed. Chicago, IL: ASCP Press; 1993:65-157.

2. Piraino B, Bailie GR, Bernardini J, Boeschoten E, Gupta A, Holmes C, et al. Peritoneal dialysis-related infections recommendations: 2005 update. Perit Dial Int.2005;25:107–31.

3. Linssen J, Jennissen V, Hildmann J, Reisinger E, Schindler J, Malchau G, et al. Identification and quantification of high fluorescence stained lymphocytes as antibodies synthesizing/ secreting cells using the automated routine hematology analyser XE-2100. Cytometry B Clin Cytom 2007;72:157-66. https://doi.org/10.1002/cyto.b.20150.

4. Paris A, Nhan T, Cornet E, Perol JP, Malet M, Troussard X . Performance evaluation of the body fluid mode on the platform Sysmex XE-5000 series automated hematology analyzer. Int J Lab Hematol. 2010;32(5):539–547.https://doi.org/10.1111/j.1751-553X.2010.01220.x

5. Nagisa Nakazawa, Noriko Wada, Yuzo Tanaka, Kazumi Gondo, Yumiko Tanaka, Takayuki Seto, et al. Evaluation of Assay Performance of the Body Fluid Mode on the Automated Hematology Analyzer XN-2000.Sysmex J Int. 2012;22(1):1-11.

6. C. Fleming, R. Brouwer, J. Lindemans, R. de Jonge. Validation of the

body fluid module on the new Sysmex XN-1000 for counting blood cells in cerebrospinal fluid and other body fluids. Clin Chem Lab Med 2013;50(10):1791-1798. https://doi.org/10.1515/cclm-2011-0927

7. Lehto T M, Leskinen P, Hedberg P and Vaskivuo T. Evaluation of the Sysmex XT-4000i for the automated body fluid analysis. Int J Lab Hematol.2014;36(2):114-123. https://doi.org/10.1111/ijlh.12134

8. Hoffmann JJ, Janssen WC. Automated counting of cells in cerebrospinal fluid using the CellDyn-4000 hematology analyzer. Clin Chem Lab Med 2002; 40:1168–73. https://doi.org/10.1515/CCLM.2002.204

9. Aulesa C, Mainar I, Prieto M, Cobos N, Galimany R. Use of the Advia 120 hematology analyzer in the differential cytologic analysis of biological fluids (cerebrospinal, peritoneal, pleural, pericardial, synovial, and others). Lab Hematol. 2003;9(4):214-24

10. Andrea Perné et al. Performance Evaluation of the Sysmex XE-5000 Hematology Analyzer for White Blood Cell Analysis in Cerebrospinal Fluid. Archives of Pathology & Laboratory Medicine.2012;136(2): 194- 198. https://doi.org/10.5858/arpa.2011-0030-OA

11. de Jonge R, Brouwer R, van Rijn M, van Acker BA, Otten HJ, Lindemans J. Automated analysis of pleural fluid total and differential leukocyte counts with the Sysmex XE-2100. Clin Chem Lab Med 2006;44:1367–71. https://doi.org/10.1515/CCLM.2006.242

12. Conner BD, Lee YC, Branca P, Rogers JT, Rodriguez RM,Light RW. Variations in pleural fluid WBC count and differential counts with different sample containers and different methods. Chest 2003; 123:1181–7. https://doi.org/10.1515/CCLM.2006.242

13. James E. Williams, Jeri Walters, Kimberley Kabb, Gaining Efficiency in the Laboratory – Automated Body Fluid Cell Counts: Evaluation of the Body Fluid Application on the Sysmex XE-5000 Hematology Analyzer. Lab Medicine.2011; 42(7):395-401. https://doi.org/10.1309/V1HMLAYJRY01RT

14. Busadee Pratumvinit, Noppawan Sivasariyanonds, Leatchai Wachirutmanggur, Wimol Chinsawangwatanakul, Sathien Sukpanichnant. Comparison of Conventional Manual Methods with the ADVIA 120 Automated Method for Counting of Red and White Blood Cells in Cerebrospinal Fluid. Siriraj Med J 2007; 59: 64-68.

15. Takemura H, Tabe Y, Ishii K , et al. Evaluation of capability of cell count and detection of tumor cells in cerebrospinal and body fluids by automated hematology analyzer [in Japanese]. Rinsho Byori. 2010;58(6):559–564.

16. Boer K, Deufel T, Reinhoefer M. Evaluation of the XE-5000 for the automated analysis of blood cells in cerebrospinal fluid. Clin Biochem. 2009;42:684–691.

17. Aune MW, Becker JL, Brugnara C, Canfield W, Dorfman DM,Fiehn W, et al. Automated flow cytometric analysis of bloodcells in cerebrospinal fluid: analytic performance. Am J ClinPathol 2004;121:690–700.https://doi.org/10.1016/j.clinbiochem.2009.01.025

18. de Jonge R, Brouwer R, van Rijn M, van Acker BA, Otten HJ, Lindemans J. Automated analysis of pleural fluid total and differential leukocyte counts with the Sysmex XE-2100. Clin Chem Lab Med 2006;44:1367–71.https://doi.org/10.1515/CCLM.2006.242

19. Giuseppe Lippi, Clarissa Cattabiani1, Anna Benegiamo, Daniela Gennari, Fernanda Pavesi, Alberta Caleffi et al. Evaluation of the Fully Automated Hematological Analyzer Sysmex XE-5000 for Flow Cytometric Analysis of Peritoneal Fluid. Journal of Laboratory Automation.2013:1-5. https://doi.org/10.1177/2211068212471833