



The Relationship Between NS1 Examination and the Examination of Hemoglobin, Hematocrit, Leukocytes, Platelets, and Erythrocytes in Dengue Fever Patients in The Pediatric Ward

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Info Article	Abstract
Article History: Received: 18 December 2024 Accepted: 18 January 2025 Published: 31 January 2025 Keywords: NS1 Dengue Hemorrhagic Fever Hemoglobin Erythrocyte	The NS1 antigen test is developed to detect dengue virus infection during the acute phase, a stage marked by various hematological abnormalities. An increase in hemoglobin levels greater than 20% can support the diagnosis of Dengue Hemorrhagic Fever (DHF), often caused by hemoconcentration due to plasma leakage, which also raises erythrocyte levels in the bloodstream. This study aimed to examine the relationship between NS1 test results and hemoglobin, hematocrit, leukocyte, platelet, and erythrocyte levels in pediatric DHF patients at Dr. Soedarso Regional Hospital. Using a cross-sectional design with total sampling, 203 samples were collected from suspected pediatric DHF patients between May and July 2023. Data included NS1 results and hematology parameters. The analysis showed a significant association between positive NS1 results and hemoglobin levels ($p = 0.025$) as well as platelet counts ($p = 0.035$), but no significant relationship with hematocrit ($p = 0.872$), leukocytes ($p = 0.269$), or erythrocytes ($p = 0.060$). In conclusion, NS1 positivity is significantly associated with hemoglobin levels and platelet count, but not with hematocrit, leukocytes, or erythrocytes in pediatric DHF patients.

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1. INTRODUCTION

Fever is a condition characterized by an increase in body temperature. Clinically, fever is defined as a rise in body temperature of 1°C (1.8°F) or more above the average normal body temperature. The average normal body temperature in humans varies depending on the site of temperature measurement (Sari and Ariningpraja, 2021). Dengue Hemorrhagic Fever (DHF) is a type of illness caused by the dengue virus, which is transmitted through mosquitoes of the *Aedes* genus, such as *Aedes aegypti* or *Aedes albopictus*. Infected individuals typically experience symptoms ranging from mild to high fever, accompanied by headache, pain behind the eyes, muscle and joint pain, and in more severe cases, spontaneous bleeding (Yulianto, Santosa, and Handoyo 2023). The diagnosis of Dengue Hemorrhagic Fever (DHF) can be established based on clinical and laboratory criteria. The clinical criteria include sudden high fever with no apparent cause that persists for 2–7 days, bleeding manifestations, liver enlargement, and signs of shock. The laboratory criteria consist of thrombocytopenia (platelet count <100,000/μL) and hemoconcentration, indicated by an increase in hematocrit of ≥20%, which should be assessed according to the patient's age and gender (Ayu et al., 2012).

The NS1 antigen test has been developed to detect dengue virus infection during the acute phase, with various studies showing that NS1 has greater sensitivity compared to viral culture, Polymerase Chain Reaction (PCR), and anti-dengue IgM and IgG antibody tests. This is because the NS1 antigen can be detected early in the infection—within the first nine days of fever, before antibodies appear. In terms of specificity, the NS1 antigen is equally as accurate as the standard viral culture and PCR tests, reaching 100% specificity (Wowor 2011). An increase in haemoglobin levels by more than 20% can support the diagnosis of Dengue Hemorrhagic Fever (DHF). This rise in hemoglobin is accompanied by hemoconcentration, which is caused by plasma leakage and is one of the earliest haematological abnormalities found in DHF patients. Plasma leakage occurs because dengue virus infection triggers an inflammatory response that damages the capillary walls, making them unable to retain fluids. As a result, plasma fluid seeps out of the blood vessels, leading to hemoconcentration (Vebriani et al., 2016).

A decrease in leukocyte count or leukopenia occurs due to the nature of the dengue virus, which causes immunological changes at the cellular level. During the acute phase of infection, leukopenia can develop. This happens because the dengue virus damages the precursor cells that form leukocytes in the bone marrow, leading to a reduction in the number of white blood cells, resulting in leukopenia in dengue fever patients (RSUD dr. Soedarto, 2022). Thrombocytopenia in dengue infection occurs through mechanisms such as bone marrow suppression, platelet destruction, and a shortened lifespan of platelets. In Dengue Hemorrhagic Fever (DHF) patients, thrombocytopenia results from the formation of virus-antibody complexes, which stimulate platelet aggregation. These platelet aggregates are then filtered through the Reticuloendothelial System (RES) and destroyed. Additionally, increased platelet destruction in the peripheral blood can also contribute to the development of thrombocytopenia in DHF patients (Masihor, Mantik, and Mongan, 2013).

When hemolysis occurs, hemoglobin is released into the plasma. Plasma leakage not only causes an increase in hemoglobin levels and hematocrit values but also leads to an increase in red blood cells (erythrocytes) in the bloodstream. This increase in erythrocytes can serve as an indication of dengue infection (Kewo, Rambert, and Manoppo 2015). According to the Ministry of Health of the Republic of Indonesia report in 2023, during week 22, or the period from January to May 2023, there were 35,694 cases of Dengue Hemorrhagic Fever (DHF) reported across Indonesia. During the same period,

the Ministry of Health reported a total of 270 deaths due to dengue in Indonesia. (Kemenkes RI, 2023). In West Kalimantan, according to data from the West Kalimantan Health Office in 2023, Dengue Hemorrhagic Fever (DHF) cases have continued to rise from January to the end of October 2023. By week 43 in October, the reported number of DHF cases in West Kalimantan had reached 4,034 cases, with a total of 48 deaths recorded (Dinas Kesehatan Kalimantan Barat, 2023).

Dr. Soedarso Regional General Hospital (RSUD) in Pontianak handled 702 cases of Dengue Hemorrhagic Fever (DHF) from January to December 2023, with the majority of patients being children. According to Minister of Health Regulation No. 25 of 2015 regarding child health efforts, a child is defined as an individual up to 18 years old. Children were the most affected group, accounting for the highest number of DHF cases. The peak of cases occurred from May to June 2023, with a total of 203 cases recorded during that period. The aim of this study is to determine the relationship between NS1 antigen examination and haematological parameters specifically haemoglobin, hematocrit, leukocytes, platelets, and erythrocytes in pediatric patients with Dengue Hemorrhagic Fever (DHF) treated at Dr. Soedarso Regional General Hospital. Data analysis using the Correlation test Chi Square test using a computer program.

2. METHOD

This study is a observational quantitative research with a cross-sectional design. The study population consists of all paediatric patients suspected of having Dengue Haemorrhagic Fever (DHF) in the paediatric ward of Dr. Soedarso Regional General Hospital in 2023. The sample used in this study includes paediatric patients who were suspected of having DHF and were admitted to the paediatric ward of Dr. Soedarso Regional General Hospital between May and June 2023, totalling 203 patients who met the inclusion and exclusion criteria.

The data collected for this study is secondary data, which refers to data that already exists or is readily available for use in research and analysis (Swarjana, 2023). In this study, the secondary data includes medical records of the patients, which contain the results of the NS1 antigen test and haematological examinations (hemoglobin, hematocrit, leukocytes, platelets, and erythrocytes). These medical records serve as the basis for analysis, as they provide valuable information regarding the health condition and laboratory results of the patients.

The data will be analysed using both univariate and bivariate statistical analyses. Univariate analysis will be used to describe the characteristics of each variable, such as the distribution of hemoglobin, platelet levels, and NS1 antigen results. Meanwhile, bivariate analysis will examine the relationships between the NS1 antigen test results and the haematological parameters, allowing for a deeper understanding of how these factors may be interrelated in the context of paediatric DHF. This study received ethical approval from the Ethics Commission of the Poltekkes Kemenkes Pontianak with number: 323/KEPK-PK.PKP/VI/2024.

3. RESULTS AND DISCUSSION

This study aims to determine the average frequency distribution based on the gender of patients who underwent the NS1 antigen test in the pediatric ward of Dr. Soedarso Regional General Hospital. Below is the result of the frequency distribution of individuals based on their gender. The patients based on gender who underwent the NS1 antigen test in the pediatric ward of Dr. Soedarso Regional General Hospital. Among the 203 patients, 106 (52.5%) were male and 97 (47.8%) were female. This distribution

indicates that a slightly higher proportion of male patients were tested for NS1 compared to female patients.

Table 1. Relationship Between NS1 Antigen Test and Hematological Parameters in DHF Patients at Dr. Soedarso Hospital

Parameter			Hemoglobin			Total	p-value
			Low	Normal	High		
NS1	Positive	N	17	66	10	93	0.025
		%	18.3%	7.10%	10.8%	100.0%	
	Negative	N	7	93	10	110	
		%	6.4%	84.5%	10.8%	100.0%	
Parameter			Hematokrit			Total	
			Low	Normal	High		
NS1	Positive	N	25	62	6	93	0.0872
		%	26.9%	66.7%	6.5%	100.0%	
	Negative	N	27	77	6	110	
		%	6.4%	84.5%	10.8%	100.0%	
Parameter			Leukosit			Total	
			Low	Normal	High		
NS1	Positive	N	53	36	4	93	0.296
		%	57.0%	38.7%	4.3%	100.0%	
	Negative	N	53	47	10	110	
		%	6.4%	84.5%	10.8%	100.0%	
Parameter			Trombosit			Total	
			Low	Normal	High		
NS1	Positive	N	64	26	3	93	0.035
		%	68.8%	28.0%	3.2%	100.0%	
	Negative	N	83	17	10	110	
		%	75.5%	15.5%	9.1%	100.0%	
Parameter			Eritrosit			Total	
			Low	Normal	High		
NS1	Positive	N	11	62	20	93	0.060
		%	11.8%	66.7%	21.5%	100.0%	
	Negative	N	5	88	17	110	
		%	75.5%	15.5%	9.1%	100.0%	

Table 1 presents the relationship between the NS1 antigen test results and various hematological parameters, including hemoglobin, hematocrit, leukocyte count, platelet count, and erythrocyte count, along with the associated p-values. For hemoglobin, a significant association was found with a p-value of 0.025, indicating a relationship between NS1 positivity and increased hemoglobin levels. However, no significant association was observed between hematocrit ($p = 0.0872$), leukocytes ($p = 0.296$), and erythrocytes ($p = 0.060$), as the p-values were higher than 0.05, suggesting no strong correlation with NS1 results. For platelets, a significant association was identified with a p-value of 0.035, indicating that NS1 positivity is related to lower platelet counts. These findings suggest that while NS1 positivity is significantly linked to hemoglobin and platelet levels, its relationship with other hematological parameters like hematocrit, leukocytes, and erythrocytes is not statistically significant.

DISCUSSION

One of the latest diagnostic methods being developed for diagnosing dengue fever is the examination of a specific dengue virus antigen, namely the non-structural protein 1 antigen, also called NS1. The NS1 antigen test is used as an early diagnostic test for dengue fever. The NS1 antigen appears earlier than anti-dengue antibodies, making detection using the NS1 antigen much more effective compared to antibody detection using IgG/IgM (Immunoglobulin G/Immunoglobulin M). In diagnosing dengue fever, not only the NS1 examination but also hematological tests should be conducted. According to WHO criteria, hematological criteria for diagnosing dengue fever are indicated by a platelet count $\leq 100,000$ cells/mm³ and plasma leakage with signs of increased hematocrit levels $\geq 20\%$. A study by Kambu and Samaran (2023) found that changes in leukocyte counts also occur in dengue fever patients, specifically ≤ 5000 cells/mm³, which is useful for predicting the critical phase of plasma leakage. These findings even precede the findings of thrombocytopenia and increased hematocrit levels. The NS1 antigen test provides quicker and more accurate results for the serodiagnosis of dengue virus infection because NS1 antigen is secreted in relatively high concentrations in the plasma of dengue fever patients. Therefore, this study was conducted to explore the relationship between the NS1 test results and hematological test results such as hemoglobin, hematocrit, leukocytes, platelets, and erythrocytes in dengue fever patients treated at the pediatric ward of RSUD dr. Soedarso, Pontianak.

Hemoglobin is a molecule that contains heme (iron) and globin polypeptide chains (alpha, beta, gamma, and delta), found in erythrocytes and serves as an oxygen transport mechanism. Hemoglobin levels at the onset of the disease are usually normal or slightly reduced. They then increase following hemoconcentration, which is the earliest hematological abnormality found in cases of dengue fever (Kewo et al. 2015). The Chi-Square test yielded a p-value of 0.025 ($p < 0.05$), meaning there is a significant relationship between the NS1 test results and hemoglobin levels in dengue fever patients in the pediatric ward of RSUD dr. Soedarso. This result contrasts with a study by Charisma (2020), which found no relationship between the NS1 test results and hemoglobin levels in dengue fever patients. Hemoglobin levels on the first day of dengue fever are typically normal or slightly decreased. However, they increase along with the occurrence of hemoconcentration, which indicates plasma leakage caused by dengue virus infection (Wowor 2011).

An increase in hematocrit levels, also known as hemoconcentration, is generally accompanied by a decrease in platelet count. In dengue infection, hematocrit levels typically begin to increase on day 3 of the disease and rise in line with the progression of dengue. The increase in hematocrit levels occurs due to plasma leakage into the extravascular space through damaged capillaries. As a result of this leakage, plasma volume decreases, leading to hypovolemic shock and circulatory failure. In severe cases with bleeding, hematocrit levels generally decrease instead of increasing (Megariani et al. 2014). The Chi-Square test yielded a p-value of 1.000 ($p > 0.05$), meaning there is no significant relationship between NS1 test results and hematocrit levels in dengue fever patients in the pediatric ward of RSUD dr. Soedarso. These results are consistent with research by Putra et al. (2016), which found no significant relationship between NS1 test results and hematocrit levels. Similarly, research by Ayu et al. (2012) also found no relationship between hematocrit levels and NS1 test results.

Leukocytes are white blood cells that function as the body's defense against bacteria or viruses. The normal leukocyte count ranges between 4,000–10,000 cells/mm³. Leukocytes also play an important role in the body's immune function, and an increased

leukocyte count in the blood indicates the presence of an infection. At the onset of dengue fever (when the fever first occurs), the leukocyte count is typically normal or decreased, with neutrophil cells predominating. Leukopenia, primarily caused by the failure to produce mature leukocytes (also known as polymorphonuclear leukocytes), occurs. During the final phase of the fever, lymphoblastoid cells are found. Leukopenia peaks just before the fever subsides and returns to normal 2-3 days after the temperature drops (Prayoga 2017). The Chi-Square test yielded a p-value of 0.269 ($p > 0.05$), meaning there is no significant relationship between NS1 test results and leukocyte counts in dengue fever patients in the pediatric ward of RSUD dr. Soedarso. This finding differs from research by Wulandari et al. (2023), which stated that there is a significant relationship between NS1 test results and leukocyte levels. Leukopenia can occur in dengue fever patients due to the dengue virus's effect on the leukocyte precursor cells in the bone marrow, leading to leukopenia. This leukopenia may occur when dengue fever is in its acute phase (Soedarto 2012).

Thrombocytopenia is one of the criteria used by the World Health Organization (WHO) as an indicator to diagnose and assess the severity of dengue fever infection. Thrombocytopenia is a hematological abnormality found in the majority of dengue virus infection cases. Platelet counts begin to decrease during the fever phase and reach their lowest point during the shock phase. Platelet counts rise rapidly during the convalescence phase, and normal levels are usually reached 7-10 days after the onset of illness (Putra et al. 2016). The Chi-Square test yielded a p-value of 0.035 ($p < 0.05$), meaning there is a significant relationship between NS1 test results and platelet counts in dengue fever patients in the pediatric ward of RSUD dr. Soedarso. These results align with research by Apriliana et al. (2019), which found a significant relationship between NS1 test results and platelet count in dengue fever patients. Research by Sari and Yasa (2020) also found a significant relationship between NS1 test results and platelet count.

Erythrocytes, or red blood cells, are produced in the red bone marrow and are responsible for transporting hemoglobin. When hemolysis occurs, hemoglobin is released into the human plasma, and about 3% of this hemoglobin passes through the glomerular membrane into the glomerular filtrate. Therefore, for hemoglobin to remain in the human bloodstream, it must stay within the erythrocytes. Increased hemoglobin levels and hematocrit values occur due to plasma leakage caused by dengue virus infection, and the presence of many red blood cells in the blood vessels indicates dengue fever infection (Guyton and Hall 2007). The Chi-Square test yielded a p-value of 0.060 ($p > 0.05$), meaning there is no significant relationship between NS1 test results and erythrocyte counts in dengue fever patients in the pediatric ward of RSUD dr. Soedarso. This result is consistent with research by Hidayatullah & Aisyah (2017), which found no significant relationship between NS1 test results and erythrocyte counts.

4. CONCLUSION

The results of the data analysis revealed a significant relationship between positive NS1 antigen test results and hemoglobin levels as well as platelet counts. However, no significant relationship was found between positive NS1 results and hematocrit levels, leukocyte counts, or erythrocyte counts in dengue fever patients treated in the pediatric ward of Dr. Soedarso Regional General Hospital. Future studies are recommended to explore the longitudinal changes in hematological parameters throughout the different phases of dengue infection, particularly in relation to NS1 antigen dynamics. Additionally, investigating the correlation between NS1 positivity and clinical outcomes such as severity of plasma leakage, duration of hospitalization, and response to treatment could provide a more comprehensive understanding of its prognostic value. Expanding the study

population to include multi-center data or adult patients could also enhance the generalizability of findings. Furthermore, integrating virological profiles, such as dengue serotypes or viral load, may help clarify the underlying mechanisms driving hematological changes in NS1-positive patients.

REFERENCES

- Apriliana, E., Tjiptaningrum, A., & Prayoga, M. J. (2019). Hubungan hasil pemeriksaan antigen Non Struktural 1 (NS1) terhadap gejala, tanda klinis dan jumlah trombosit pada pasien suspek infeksi Dengue di RS Urip Sumoharjo. *Jurnal Agromedicine*, 6(1), 30-37.
- Ayu, P., Bahrur, U., & Arif, M. (2016). Platelet Demam Berdarah Dengue. *Indonesian Journal Of Clinical Pathology and Medical Laboratory*, 18(3), 157–160. <https://doi.org/10.24293/ijcpml.v18i3.383>
- Charisma, A. M. (2020). Relationship of Non Structural Antigen 1 (Ns1) Examination Results To Clinical Signs, Symptoms And Routine Blood Examination In Patients Suspected Dengue Infections At Inpatients Clinic of Vita Medika Kepung Kediri Districts. *Indonesian Journal of Tropical and Infectious Disease*, 8(1), 66-78.
- Dinas Kesehatan Kalimantan Barat. (2023). *Kasus Demam Berdarah Dengue di Kalimantan Barat [Internet]*. Dinas Kesehatan Kalimantan Barat
- Guyton, A. C., & J. E. Hall. (2007). *Sel-Sel Darah Merah, Anemia, dan Polisitemia*. Jakarta: *Buku Ajar Fisiologi Kedokteran EGC*.
- Hidayatullah, M. A. A. M., & Aisyah, R. (2017). Hubungan Jumlah Trombosit dengan Jumlah Eritrosit pada Pasien Infeksi Virus Dengue di RS X Surakarta. *Biomedika*, 9(2), 65-70. <https://doi.org/10.23917/biomedika.v9i2.5846>
- Kambu, Y., & Samaran, E. (2023). *Meningkatkan Kadar Trombosit Penderita Dengue Hemorrhagic Fever Dengan Rebusan Daun Petatas Ungu (Ipomea Batatas L. Poir.* Pekalongan: Penerbit NEM.
- Kemendes RI. (2023). *Infografis Demam Berdarah Dengue Di Indonesia*. Jakarta: Ditjen P2PM.
- Kewo, I. R. A., Rambert, G., & Manoppo, F. (2015). Karakteristik Eritrosit Pasien Anak Dengan Infeksi Virus Dengue Di Manado.” *Jurnal E-Biomedik* 3(2), 566–571. Retrieved from: <https://ejournal.unsrat.ac.id/v3/index.php/ebiomedik/article/view/8438>
- Masihor, J. J. G., Mantik, M. F. J., & Mongan, A. E. (2013). Hubungan Jumlah Trombosit dan Jumlah Leukosit Pada Pasien Anak Demam Berdarah Dengue. *Jurnal E-Biomedik (EBM)*, 1(1), 391–392.
- Megariani, R. Mariko, A. Alkayar, A., & Putra, A. E. (2014). Uji Diagnostik Pemeriksaan Antigen Nonstruktural 1 untuk Deteksi Dini Infeksi Virus Dengue pada Anak. *Sari Pediatri*, 16(2), 121-127. <http://dx.doi.org/10.14238/sp16.2.2014.121-7>
- Prayoga, M. J. (2017). Hubungan Hasil Pemeriksaan Antigen Non Struktural 1 (NS1) Terhadap Gejala, Tanda Klinis Dan Jumlah Trombosit Pada Pasien Suspect Infeksi Dengue. *Skripsi*. Universitas Lampung.
- Putra, I. A., Syauqy, A., Darmawan, A., & Rahman, A. O. (2016). Korelasi Pemeriksaan Ns 1 Ag Dan Pemeriksaan Darah Tepi Pada Anak Dengan Demam. *Jambi Medical Journal : Jurnal Kedokteran Dan Kesehatan*, 4(2), 106–118. <https://doi.org/10.22437/jmj.v4i2.3577>.
- RSUD dr. Soedarso. (2022). *Profil Rumah Sakit Umum Daerah dr. Soedarso*. RSUD dr. Soedarso, Pontianak.
- Sari, E. K., & Ariningpraja, R.T. (2021). *Demam: Mengenal Demam Dan Aspek Perawatannya*. Malang: UB Press.

- Soedarto, S. (2012). *Demam Berdarah Dengue*. Jakarta: CV. Agung Seto.
- Vebriani, L., Wardana, Z., & Fridayenti, F. (2016). Karakteristik Hematologi Pasien Demam Berdarah Dengue Di Bagian Penyakit Dalam RSUD Arifin Achmad Provinsi Riau Periode 1 Januari – 31 Desember 2013. *Jurnal Online Mahasiswa: Fakultas Kedokteran*, 3(1), 1-20. Retrieved from: <https://jom.unri.ac.id/index.php/JOMFDOK/article/view/9175>
- Wowor, M. F. (2011). Deteksi Dini Demam Berdarah Dengue Dengan Pemeriksaan Antigen NS1." *Jurnal Biomedik: JBM*, 3(1):1–9. <https://doi.org/10.35790/jbm.3.1.2011.853>
- Wulandari, M. F., Hadi, S., & Putri, D. E. (2023). Hubungan Jumlah Leukosit Terhadap Hasil Pemeriksaan Ns-1 Pasien Suspek DBD. *Binawan Student Journal*, 5(1), 1-7. <https://doi.org/10.54771/bsj.v5i1.675>
- Yulianto, B., Santosa, B. J., & Handoyo, S. (2023). *Memberdayakan Masyarakat Mencegah Dan Mengatasi DBD/DHF Dengan PSN 3M Plus*. Surabaya: Scopindo Media Pustaka.