

Outcomes of NS1 Antigen, IgM-IgG Antibody, Hematology and Clinical Manifestations in Child Patients with Dengue Virus Infection at Regional General Hospital Dr. Soetomo Hospital and Community Health Centers in Surabaya in 2020-2022

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ABSTRACT

Dengue infection is a viral infection caused by the dengue virus (DENV). DENV infection is one of the fastest-spreading viral diseases in the world. DENV infection can affect all age groups. However, the prevalence of children with DENV infection is generally higher than that of adults. This study will discuss the laboratory examinations that need to be done to establish the diagnosis of dengue infection in children. Laboratory examinations that will be discussed include early examinations that can be done immediately after the first symptoms appear, namely NS1 (non-structural protein 1) antigen detection, serological tests to detect specific immunoglobulin M (IgM) or immunoglobulin G (IgG) antibodies against dengue virus and standard hematological parameters such as leukocytes, platelets and hematocrit as well as clinical manifestations that often occur in DENV infection. This research was done in Regional General Hospital Dr. Soetomo which is a type A hospital in the top referral category or the largest referral hospital in Eastern Indonesia that handles many cases of dengue virus infection with severe degrees and also in two community health centers in Surabaya with low-level dengue infection cases in 2020-2022.

Keywords: antigen NS1; IgM-IgG; hematology; dengue virus infection.

INTRODUCTION

Dengue infection is a viral infection caused by dengue virus (DENV). The incidence of DENV infection has increased rapidly worldwide in recent decades, with cases reported to the World Health Organization (WHO) increasing from 505,430 cases in 2000 to 5.2 million in 2019 (WHO, 2023). In Indonesia in 2020, 15,819 cases of DENV infection were reported with 121 deaths (Ministry of Health RI, 2021). In Surabaya City, around 187 cases were reported and the highest occurred in February 2022 (Surabaya Health Department, 2022).

DENV infection can occur in all age groups. However, the prevalence of children with DENV infection is generally higher than that of adults (WHO, 2009). The most common dengue infection in children is dengue hemorrhagic fever (DHF) (WHO, 2011). In Indonesia, the percentage of the 5- 14 years age group affected by DHF is 33.08%, the second highest after the 15-44 years age group. The percentage of those aged 5-14 years who died from DHF was 33.08% (Ministry of Health RI, 2021). Deaths that

occur mostly occur due to delays in referral to health facilities.

Laboratory tests are performed to establish the diagnosis of DENV infection. This examination involves the detection of viruses, namely nucleic acids, antigens, and also antibodies (WHO, 2020). An early test that can be performed immediately after the first symptoms appear is NS1 (non-structural protein 1) antigen detection (WHO, 2011). Many studies have investigated the usefulness of sNS1 detection as a diagnostic tool during the acute phase of dengue infection (Peeling et al., 2010). NS1 antigen can be detected in serum, plasma, circulating blood cells, and other tissues for 4-5 days (WHO, 2020).

In the late acute phase of infection, serology is the method of choice for diagnosis. Serologic tests to detect specific immunoglobulin M (IgM) or immunoglobulin G (IgG) antibodies to dengue virus are widely available and can provide an alternative to virus isolation or PCR (Polymerase Chain Reaction) to

support the diagnosis of dengue fever (WHO, 2020). IgM antibodies are detectable on days 3-5 after the onset of illness, increase rapidly around 2 weeks, and decrease to undetectable levels after 2-3 months. IgG antibodies can be detected at low levels at the end of the first week, increasing later and persisting for a longer time (WHO, 2011).

Standard hematological parameters such as leukocytes, thrombocytes, and hematocrit are an important part of the diagnosis of DHF. Leukopenia or low white blood count is the first finding in dengue fever and is caused by bone marrow suppression by the virus (Joshi, et al., 2018). Leukopenia helps diagnosis, differentiation & prognostication of dengue fever. Thrombocytopenia or a decrease in platelet count below 100,000 cells/mm³, is usually observed in dengue fever but can be seen more clearly in DHF. Thrombocytopenia is usually found between the third and eighth day of illness and may precede or coincide with hematocrit changes.

Hemoconcentration with an increase in hematocrit of 20% or more is considered definitive evidence of increased vascular permeability and plasma leakage (WHO, 2011). Dengue virus serotypes are divided into four serotypes namely DENV-1, DENV-2, DENV-3, and DENV-4. DENV infection can be asymptomatic or can cause undifferentiated febrile illness (viral syndrome), dengue fever, and dengue hemorrhagic fever (DHF) which can also become dengue shock syndrome in cases of DHF with high severity. Infection with one dengue serotype can confer lifelong immunity to that serotype, but there is only short-term cross-protection to other serotypes (WHO, 2011).

Clinical manifestations of DENV infection begin with fever with an acute onset of high, persistent fever lasting two to seven days. In DHF there are bleeding manifestations including positive tourniquet test, petechiae, purpura, gum bleeding, hematemesis, and/or melena. Shock then occurs which manifests as tachycardia and hypotension with cold, wet, and/or anxiety (WHO, 2011).

METHOD

This research is a descriptive cross-sectional study by looking at the results of NS1 antigen, IgM- IgG antibody, and hematology (leukocytes, thrombocytes, and hematocrit) examination as well as clinical manifestations through medical records of pediatric patients with dengue virus infection at Dr. Soetomo Hospital and Kalijudan and Menur community health centers in Surabaya. The collected medical record data amounted to 155 patients and met the inclusion criteria. Inclusion criteria included pediatric patients who were confirmed to have dengue infection based on clinical and hematology tests. Data were then analyzed using univariate descriptive analysis to obtain the prevalence of the variables studied and then grouped based on the research variables presented in the form of distribution tables.

RESULT AND DISCUSSION

TABLE 1: NS1 Antigen Test.

Parameters	Dr. Soetomo		Community Health Centers	
	Frequency (N)	Percentages (%)	Frequency (N)	Percentages (%)
Positive	2	28.6	7	100.0
Negative	5	71.4	0	0.0
Total	7	100.0	7	100.0

Based on the NS1 antigen examination, 124 patients at Regional General Hospital Dr. Soetomo were not tested for NS1 antigen and 2 patients had positive results. Meanwhile, at the community health centers, 17 patients were not tested for NS1 antigen and 7 patients had positive results. The high number of patients who were not tested for NS1 antigen was thought to be due to patients arriving on day 5 or patients referred from the community health centers (WHO, 2011). In a study conducted by Megariani et al (2016), the percentage of positive NS1 in DHF infection was greater on day 3 of fever. The results of this study are in line with research conducted by Dussart et al (2016) that the NS1 antigen has a higher sensitivity value on the first to the fourth day of fever compared to days five to ten.

TABLE 2: IgM-IgG Antibody Test.

Parameters IgM-IgG Antibody	Dr. Soetomo		Community Health Centers	
	Frequency (N)	Percentage (%)	Frequency (N)	Percentages (%)
IgM(+)/IgG(+)	2	8.7	0	0.0
IgM (+)/ IgG(-)	6	26.1	0	0.0
IgM (-)/ IgG(+)	12	52.2	1	50.0
IgM (-)/ IgG(-)	3	13.0	1	50.0
Total	23	100.0	2	100.0

According to Table 2 above, it was found that 108 patients at Dr. Soetomo Hospital were not tested for IgM-IgG antibodies and most patients had IgM(-)/IgG(+) antibody results. While at the puskesmas, 22 patients were found not to have IgM-IgG antibody tests, 1 patient (50%) had IgM(-)/IgG(+) results, and 1 patient (50%) patient had IgM(-)/IgG(-) results. IgM-IgG with negative-negative results indicate that the patient is still in the early stages of dengue infection. So the immune response has not shown positive results. IgM-IgG with IgM(+)/IgG(-) results indicates that the patient was infected with dengue on days 4 to 5 and was the first-time infection also called primary infection. IgM-IgG with IgM(+)/IgG(+) results indicate that the patient was infected in the first week so IgG antibodies were detected and increased. IgM-IgG with IgM(-)/IgG(+) results indicate that the patient was infected for the second time or called secondary infection. This is indicated by IgG antibodies being detected first compared to IgM antibodies (Changal et al, 2016).

TABLE 3: Leukocyte Count Sample.

Parameters	Dr. Soetomo		Community Health Centers	
	Frequency (N)	Percentages (%)	Frequency (N)	Percentages (%)
Leukopenia	59	45.0	12	50.0
Normal	56	42.7	4	16.7
Leukocytosis	16	12.3	8	33.3
Total	131	100.0	24	100.0

The results of the study regarding leukocyte examination (table 3) above found that the leukocytes of patients at Regional General Hospital Dr. Soetomo were mostly in the leukopenia category with 45% of patients. While in the community health centers category most patients also experienced leukopenia as many as 50% of patients. The results of this study are in line with research conducted by Gulabani et al (2023), namely most patients experience leukopenia. The results of the above study are also in accordance with research conducted by Widarti et al (2023) regarding the analysis of platelet and leukocyte counts in patients with dengue hemorrhagic fever that patients infected with dengue will experience a decrease in leukocytes so that they become leukopenia, especially in the early phase of infection.

TABLE 4: Thrombocyte Count Sample.

Parameters	Dr. Soetomo		Community Health Centers	
	Frequency (N)	Percentages (%)	Frequency (N)	Percentages (%)
Severe Trombositopenia	53	40.5	6	25.0
Moderate Trombositopenia	44	33.6	7	29.2
Mild Trombositopenia	23	17.5	8	33.3
Normal	11	8.4	3	12.5
Total	131	100.0	24	100.0

The results of the study regarding thrombocytes examination (table 4) above, at Regional General Hospital Dr. Soetomo the most patients were in the category of severe thrombocytopenia as many as 40.5% of patients. The results of this study are in line with research conducted by Amrita et al (2022) which shows that the results of platelet examination of patients are mostly in the severe thrombocytopenia category. While at the health center the most common category was mild thrombocytopenia, which was 33.3% of patients. The amount of thrombocytopenia itself has a significant relationship with the severity of dengue infection. The higher the severity of the patient, the more severe the thrombocytopenia (Syumarta et al., 2014).

The normal trombocyte count is likely to occur because on the fifth to eighth day, there will be an increase in young megakaryocytes so that platelets will return within normal limits (Kamila et al., 2022).

TABLE 5: Haematocrit Count Sample.

Parameters	Dr. Soetomo		Community Health Centers	
	Frequency (N)	Percentages (%)	Frequency (N)	Percentages (%)
Low	18	13.7	3	12.5
Normal	55	42.0	7	29.2
High	58	44.3	14	58.3
Total	131	100.0	24	100.0

The results of the study regarding hematocrit examination (table 5), at Regional General Hospital Dr. Soetomo that most patients were in the high hematocrit category as many as 44.3% of patients. At the community health centers, the highest category was also in the high hematocrit category, with as many as 14% of patients. This study is in line with the research of Kamila, et al (2022) who examined the relationship between platelet count and hematocrit levels in children infected with dengue that the majority of patients had increased hematocrit. This study is also in line with research conducted by Asghar et al (2021) which shows that the results of hematocrit examination are dominated by high hematocrit. Plasma leakage in dengue infection causes a decrease in plasma volume and an increase in red blood cells in the blood vessels resulting in an increase in hematocrit (Tuntun et al, 2018).

TABLE 6: Thrombocyte Count Sample.

Parameters	Dr. Soetomo		Community Health Center	
	Frequency (N)	Percentages (%)	Frequency (N)	Percentages (%)
Fever	131	100.0	24	100.0
Spontaneous Bleeding	70	53.4	8	33.3
Pleura Effusion	70	53.4	0	0.0
Hepatomegaly	20	15.3	0	0.0
Shock	30	22.9	0	0.0

The results of the study on clinical manifestations (table 6) showed that fever was the most common clinical manifestation in pediatric patients at Regional General Hospital Dr. Soetomo and also community health centers. Fever is a clinical manifestation that occurs at the beginning of dengue infection. It is followed by bleeding characterized by a positive tourniquet test, petechiae, bleeding gums, nosebleeds, or melena. Pleural effusion is also a common clinical manifestation due to plasma leakage. Shock is the least common clinical manifestation. Clinical manifestations of dengue infection are very varied, so clinical diagnosis is not easy to do (Anastasia, 2018).

TABLE 7: Proportion of Severity.

Parameters	Dr. Soetomo		Community Health Centers	
	Frequency (N)	Percentages (%)	Frequency (N)	Percentages (%)
Undifferentiated fever	0	0.0	2*	8.3
Dengue Fever	23	17.6	8	33.3
DHF I	43	32.7	11	45.8
DHF II	34	26.0	3	12.6
DHF III	24	18.3	0	0.0
DHF IV	6	4.6	0	0.0
Expanded Dengue Syndrome	1	0.8	0	0.0
Total	131	100.0	24	100.0

*NS1 Antigen Positive

The results of the study regarding the degree of severity (Table 7), found that at Regional General Hospital Dr. Soetomo the majority of pediatric patients were in the category of DHF degree I and degree II. Then there were also pediatric patients with DHF categories of degree III, IV, and expanded dengue syndrome. At the community health centers, the most common category was DHF degree I, followed by dengue fever and DHF degree II. There were no patients with degree III to expanded dengue syndrome in pediatric patients at the health center. This is in line with research conducted by Syakir (2020), that patients with dengue infection who are hospitalized are patients with severe manifestations and with a high risk of death. Whereas in health centers, pediatric patients who are treated generally have milder manifestations so that the degree of severity is milder compared to hospitals.

CONCLUSION & SUGGESTION

Further research needs to be done with a larger number of samples so that the results of the study are more credible. Then it is necessary to optimize laboratory examinations for dengue infection so that there are no errors in clinical diagnosis in patients because many cases are misdiagnosed which are initially suspected of dengue fever and finally classified as non-dengue fever.

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