Oxidative and Chlorinative Stress in Children with Dengue Hemorrhagic Fever Tahun Terbit 2016

by Edi Hartoyo

Submission date: 15-Nov-2020 10:03AM (UTC+0700)
Submission ID: 1446236479
File name: in_Children_with_Dengue_Hemorrhagic_Fever_Tahun_Terbit_2016.pdf (537.79K)
Word count: 3704
Character count: 19518

Available online at www.ijpcr.com
International Journal of Pharmaceutical and Clinical Research 2016; 8(8): 1186-1191

ISSN- 0975 1556

Research Article

Oxidative and Chlorinative Stress in Children with Dengue Hemorrhagic Fever

Edi Hartoyo[1*], Iskandar Thalib[2], Eko Suhartono[3], Ari Yunanto[1]

[1]*Department of Pediatric, Ulin General Hospital/Faculty of Medicine, Lambung Mangkurat University, Banjarmasin, South Kalimantan, Indonesia*
[2] *Research Unit of Mutiara Bunda Mother and Child Hospital, Martapura of South Kalimantan, Indonesia*
[3]*Department of Medical Chemistry/ Biochemistry, Faculty of Medicine, Lambung Mangkurat University, Banjarmasin, South Kalimantan, Indonesia*

Available Online:10th August, 2016

ABSTRACT

In this present study, we try to investigate the oxidative and chlorinative stress in children with Dengue Hemorrhagic Fever (DHF). The level of hydrogen peroxide (H_2O_2) and malondyaldehyde (MDA), myeloperoxidase (MPO) activity, and chlorinative index (CI), were measured in 61 confirmed dengue hemorrhagic (DHF) patients. Subjects were classified into 3 grades of DHF according to the Wold Health Organization (WHO) 1997 guidelines: grade I (DHF-1, n-22); grade II (DHF-2, n= 36); and grade III (DHF-3, n= 3). H_2O_2 and MDA level, and MPO activity were measured spectrophotometrically. CI was calculated by dividing the level of H_2O_2 and MPO activity. The results shows that the levels of H_2O_2 and MDA, MPO activity, and CI significantly different between group. The all parameters that investigated in this present study seems more higher with the higher grade of DHF, except for MPO activity. From this result, it can be concluded that both oxidative and chlorinative stress pathways might be involved in the pathomechanism of DHF.

Keywords: Dengue Hemorrhagic Fever, Oxidative Stress, Chlorinative Stress

INTRODUCTION

Dengue Hemorrhagic Fever (DHF) is a major public health issue in developing countries, especially Indonesia[1]. Indonesia is one of the nation in Southeast Asia that had the highest incidence of DHF. Since first discovered in 1968 in Surabaya, DHF cases in Indonesia have increased time by time[2]. The national incidence rate has been 27 cases per 100,000 populations with a case fatality rate of 1.5%[1]. Nowadays, all provinces in Indonesia have reported dengue cases, including South Kalimantan[2,3]. The occurrence number of DHF in South Kalimantan was 3-4%, while the mortality rate was 5 -8 %[3]. The DHF is caused by the infection of dengue virus (serotypes 1-4) that transmitted by the *aedes aegypti* mosquito[4,5]. Dengue virus is an RNA virus which belongs to the family Flaviviridae[6]. The clinical spectrum of dengue fever ranges from a non-specific afebrile illness, a mild-form DHF, and dengue shock syndrome (DSS)[7]. The pathomechanism of DHF is not clearly understood. Several hypotheses for the pathomechanisms of DHF have been proposed. Among them, oxidative stress might play a role in the pathomechanism of DHF[9,10]. Several studies have reported that dengue virus mediated oxidative stress[10,12]. It is well known that viral infection could activate neutrophils and others cells as a mechanism for protection[11,13]. This activated neutrophils induced a process called respiratory burst. During this process, several reactive oxygen species (ROS) were produced. It is caused by the increasing activity of NADPH oxidase and the releasing of myeloperoxidase (MPO)[14,15]. NADPH oxidase was involved in the formation of several ROS such as, hydrogen peroxide (H_2O_2), while MPO was involved in the formation of another ROS such as, HOCl[16,17]. The formation of HOCl will lead to a condition, known as chlorinative stress. It can be indicated by the increasing of ratio between H_2O_2 level and MPO activity known as Chlorinative Index (CI)[18]. Both oxidative and chlorinative stress will lead to a process called lipid peroxidation[18]. This process was characterized by the formation of some compounds, such as malondialdehyde (MDA)[19]. MDA is an indicator for ROS-induced lipid peroxidation[20,21]. Since oxidative and chlorinative stress might be involved in the pathomechanism of DHF. Thus, our present study aimed to investigate the involvement of oxidative and chlorinative stress by measuring H_2O_2 and MDA level, MPO activity, and calculate the CI in the serum of DHF patient.

MATERIAL AND METHODS

Subjects

The study was conducted on DHF patients after informed consent was obtained from them. It was approved by the Ethics Commission of the Faculty of Medicine, Lambung Mangkurat University, Banjarbaru, South Kalimantan, Indonesia. A total of 61 subjects (aged 6 months–18 years) were recruited for this study. Subjects were divided into 3

**Author for Correspondence: ekoantioxidant@gmail.com*

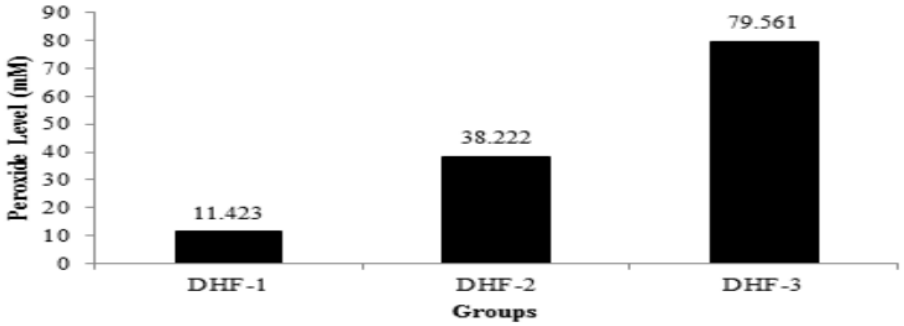

Figure 1: H_2O_2 level in the different stage of DHF (DHF: dengue hemorrhagic fever)

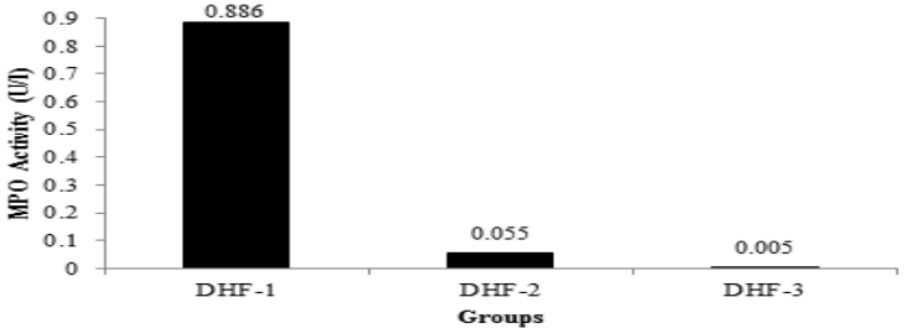

Figure 2: MPO activity in the different stage of DHF (MPO: myeloperoxidase; DHF: dengue hemorrhagic fever)

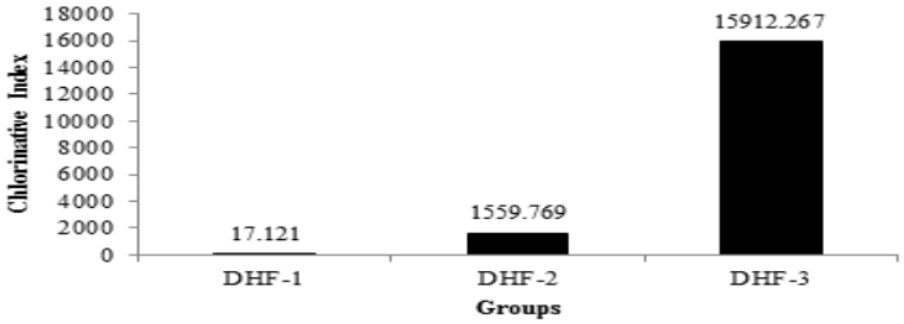

Figure 3: CI in the different stage of DHF (CI: chlorinative index; DHF: dengue hemorrhagic fever)

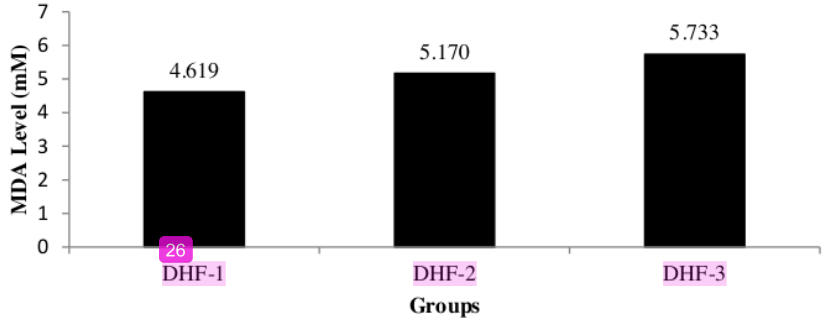

Figure 4: MDA level in the different stage of DHF (MDA: malondialdehyde; DHF: dengue hemorrhagic fever)

groups: group I (DHF-1, n = 22) with a grade I of DHF, while group II (DHF-2, n = 36) with grade II of DHF, and group (DHF-3, n = 3) with grade III of DHF according to the World Health Organization (WHO) 1997 guidelines for control and prevention of DHF[11]. Dengue infection was confirmed by IgM/IG g and NS-1 detection.

Samples collection

Samples of blood were collected from the patients using vacutainers containing EDTA. The samples were centrifuged for 15 min at 2000 rpm and stored at -2^{0}C until analysis.

H_2O_2 level analysis

H_2O_2 level was calculated by the FOX2 method with slight modification. Solutions measured spectrophotometrically at λ = 505 nm. Standard and test solutions consisted of 1 M H_2O_2 200 µL and 200 µL serum, respectively, with the addition of 160 µL phosphate buffer solution pH 7.4, 160 µL $FeCl_3$ (251.5 mg $FeCl_3$ dissolved in 250 ml distilled water) and 160 µL o-fenantroline (120 mg o-phenantroline dissolved in 100 ml distilled water) for both solutions. The composition of the blank solution was identical to that of the test solution, except for the absence of $FeCl_3$ in the blank. Subsequent to preparation, all solutions were incubated for 30 minutes at room temperature, then centrifuged at 12,000 rpm for 10 minutes, and the absorbance of the standard (As), test (Au) and blank (Ab) solutions measured at λ=505 nm, using the supernatant of each solution[21].

MPO activity analysis

MPO activity was measured spectrophotometrically using o-dianisidine (Sigma-Aldrich) and H_2O_2. In the presence of H_2O_2 as oxidizing agents, MPO catalyses the oxidation of o-dianisidine yielding a brown coloured product, oxidized o-dianisidine, with a maximum absorbance at 470 nm. One unit (U) of MPO activity was defined as that degrading 1 μmol of H_2O_2 per minute at 25^{o}C[22].

CI analysis

CI was a ratio between H_2O_2 level and MPO activity. CI was calculated following to equation:

$$CI = \frac{Hydrogen\ Peroxide\ Level}{MPO\ Activity}$$

MDA level analysis

MDA level was calculated by thiobarbituric acid reactive substances (TBARS) by the technique already proposed by Buege and Aust[23]. The supernatant were put into a Pyrex tube that contained 10% of trichloracetic acid and 0.67% of TBARS and incubated at 100^{o}C for 15 min. Then chill the mixture on ice for 5 min and add the 5 ml of n-butyl-liquor. Let the mixture stand for 40 s and centrifuged at 1000 rpm for 15 min. The TBARS value was calculated by the spectrophotometer at the absorbance of 532 nm and figured utilizing the coefficient 1.56 × 105 mol/cm. The MDA concentration expressed in µmol MDA. As a standard solution we used commercially MDA.

Statistical analysis

Data are represented as mean±SEM. For comparing H_2O_2 MDA levels, MPO activity, and CI between groups, Kruskal-Wallis followed by Mann-Whitney test were used. Statistical significance was set at p<0.05. The software used for the analysis of the data was the Statistical Package for the Social Sciences (SPSS) version 16.0 and Microsoft Excel 2010 for Windows 10.

RESULTS

To observe the involvement of oxidative stress pathway in the dengue infection, the level of H_2O_2 was measured as an indicator of the oxidative stress status. The result shows that in all grades of DHF, H_2O_2 was produced (Figure 1). Kruskal-Wallis test result shows that the level of H_2O_2 is significantly different from each other (p<0.05). Mann-Whitney test results show that there are a significant difference of H_2O_2 level between DHF-1 and DHF-2 group, DHF-1 and DHF-3 group, and DHF-2 and DHF-3 group (p<0.05) (Table 1). The result indicated that oxidative stress might be involved in DHF and the response is higher in severe cases.

[a]p-Values were calculated using the Kruskal–Wallis test followed by Mann-Whitney test; p < 0.05 was considered statistically significant.

[b] Indicates p-value when compared between DHF-1 and DHF-2.

[c] Indicates p-value when compared between DHF-1 and DHF-3.

[d] Indicates p-value when compared between DHF-2 and DHF-3.

Plasma MPO activity was found to be lower in the case with the severe forms (DHF-2 and DHF-3) than in DHF-1 cases (Figure 2). Analysis of the mean comparison between MPO activity in different grade of DHF revealed a significant difference between all grades of DHF (p<0.05). Mann-Whitney test results shows that there are a significant difference of MPO activity between DHF-1 and DHF-2 group, DHF-1 and DHF-3 group, and DHF-2 and DHF-3 group (p<0.05) (Table 1). The result indicated that chlorinative stress might be involved in DHF. Fig. 3 represented the mean values±standard error (mean±SE) of CI. Dispersion of measured values around each mean varied from 17.121 to 15912.267. That data also suggests that the highest CI is in the DHF-3 group and the lowest is the DHF-1 group (Figure 3). Statistical analysis test results show that all groups of treatment significantly (p<0.05) different from each other. Mann-Whitney test results show that there are a significant difference of CI between DHF-1 and DHF-2 group, DHF-1 and DHF-3 group, and DHF-2 and DHF-3 group (table 1). The result is in line with the result in figure 2. It is indicated that chlorinative stress might be involved in DHF. Fig. 4 represented the mean values±standard error (mean±SE) of plasma MDA levels. Dispersion of measured values around each mean varied from 4.619 to 5.733. That data also suggests that the highest MDA level is in the DHF-3 group and the lowest is the DHF-1 group (Figure 4). Statistical analysis test results show that all groups of treatment significantly (p<0.05) different from each other. Mann-Whitney test results show that there are a significant difference of MDA level between DHF-1 and DHF-2 group, DHF-1 and DHF-3 group, and DHF-2 and DHF-3 group (p<0.05) (Table 1). The result indicated that DHF increased lipid peroxidation.

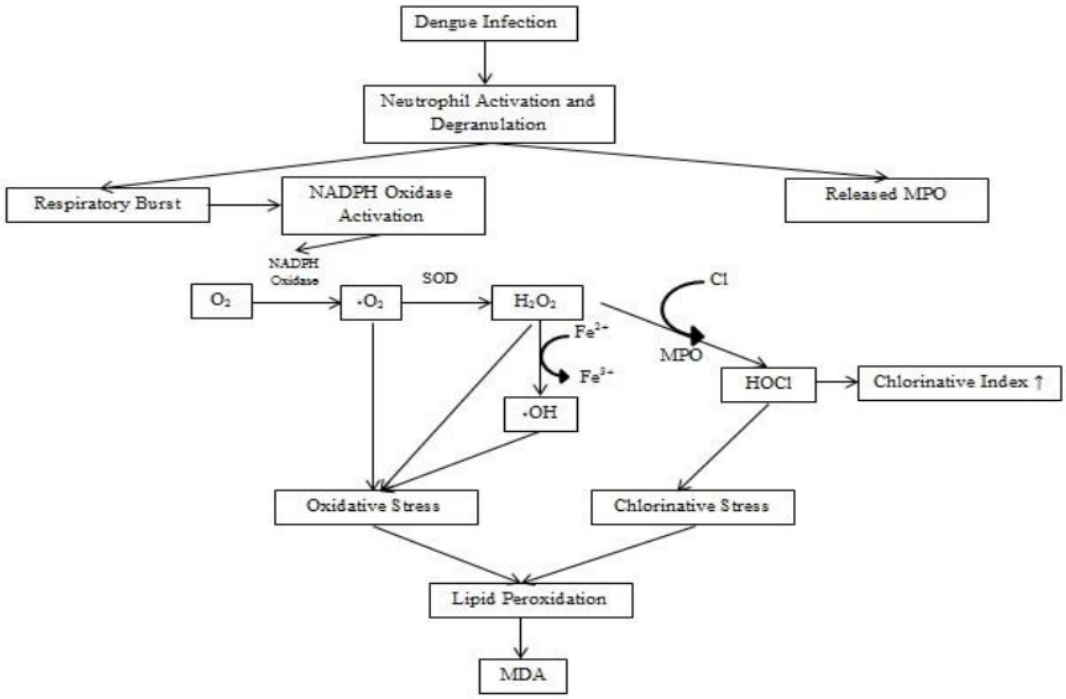

Figure 5: Oxidative and chlorinative stress in DHF pathomechanism.

DISCUSSION

The present study compared the plasma levels of H_2O_2 in the different grade of dengue infection (grade I, II, and III). The result shows that the H_2O_2 level was found to be higher in grade III than grade I and II. This result of this present study suggests that dengue virus infection induced the formation ROS. ROS seems to be higher in the grade III more than grade I, and II. This result indicated that the level of ROS will be increase with the increasing of DHF severity. The result is similar to those reported previously in dengue-21ected Cuban adult, in whom serum level of total H_2O_2 was significantly higher in DHF patients than in normal healthy individuals[11]. The increasing of ROS formation may be due to a process known as respiratory burst. Respiratory burst is a term used when neutrophils become activated through the actions of NADPH oxidase and generate ROS production that can be very effective to kill a microorganism, inducing virus[24]. 18s mechanism has been described in several studies for several viral infections, such as hepatitis C virus, rhinovirus, HIV, and dengue virus[25]. Besides ROS, activated neutrophils several components and chemokines. One of a major component that found in azurophilic granules of neutrophils is MPO. Our result study indicated that MPO is involved in the pathomechanism of DHF. The result shows that the MPO activity is lower in the higher grade of DHF. This is in contrast to previous reports, but there has been limited data available on the effect of DHF on MPO activity[26]. This different result could be due to the severity of the disease. It is well known that DHF is charazterized by trombocytopenia and leucopenia. It has also been reported that dengue infections can caused neutropenia. The severity of neutropenia is directly proportional to the severity of 29F. Khan *et al* result study shows that neutropenia was seen more commonly in patients with DHF than patients with DF[27]. Since MPO activity reflects the neutrophil functions, lower neutrophil count might be affected the MPO activity[28]. The MPO is a glycosylated haem enzyme that participates in innate immune defense by forming microbicidal reactive oxidants and diffusible radical species[17,29]. MPO catalyzes the reaction between H_2O_2 and chloride ions to form HOCl, a highly oxidising agent, to kill intracellular microorganisms[29]. HOCl can cause a condition known as chlorinative stress[30]. This condition can be seen by an index known as CI. Our result suggests that CI is increasing with the increasing grade of DHF. This result indicated that in the higher degree of DHF, the chlorinative stress that occurred will be higher than the lower grade. Dengue viral infections will lead to an activation and degranulation of neutrophils. This activation and degranulation induced the activity of NADPH oxidase to catalyst O_2 to form $\cdot O_2$ via respiratory burst. $\cdot O_2$ will dismutated by SOD to form H_2O_2 and with the presence of some transition metals, H_2O_2 were breakdown to $\cdot OH$ via fenton reaction. All ROS that produced during respiratory burst can cause oxidative stress. Besides activated NADPH oxidase, the activation and degranulation of neutrophil can induce the releasing of MPO. Then, MPO catalyst the reaction between H_2O_2 and Cl to form HOCl. This will lead to a condition known as

Table 1:Comparison of H_2O_2 and MDA levels, MPO activity, and CI in the different stage of DHF[a]

Parameters	DHF-1	DHF-2	DHF-3	p-Value[b]	p-Value[c]	p-Value[d]
Peroxide (mM)	11.423	38.222	79.561	0.000	0.006	0.005
MDA (mM)	4.619	5.170	5.733	0.000	0.006	0.007
MPO (U/l)	0.886	0.055	0.005	0.000	0.006	0.004
CI	17.121	1559.77	15912.267	0.000	0.006	0.004

H_2O_2: hydrogen peroxide; MDA: malondialdehyde; MPO: myeloperoxidase; CI: chlorinative index; DHF: dengue hemorrhagic fever.

chlorinative stress which is charazterized by an increase of Chlorinative Index (CI). Both oxidative and chlorinative stress can induce a lipir peroxidation which is marked by the formation of MDA. Oxidative and chlorinative stress, both has a consequence. The consequence is macromolecul damage, such as lipid. Lipid damage caused by both oxidative and chlorinative stress known as lipid peroxidation[18,30]. One of the most well studied markers of lipid peroxidation is MDA[19-20,31]. Consistent with this idea, our result data suggest that MDA was formed during dengue infection. In higher grade, the level of MDA seems to be more higher. It may be cause this MDA level are correlated with the level of H_2O_2 and CI rate, although this study does not correlated these variables. The whole process was illustrated in figure 5. In conclusion, the present study demonstrated that oxidative and chlorinative stress might be involved in pathomechanism of DHF. It seems the oxidative and chlorinative stress is more higher with the higher grade of DHF. From the results, it also can be concluded that both oxidative and chlorinative stress could cause a further reaction to damage macromolecule, such as lipid.

CONFLICT OF INTEREST

We declare that we have no conflict of interest.

REFERENCES

1. Hutagalung J, Halim W, Koto A. Dengue hemorrhagic fever in West Sumatera, Indonesia, 2009. Outbreak Surveillance and Investigation Reports 2011; 4 (2): 1-5.
2. Karina A, Sari SYI, Sumardi HU, Setiawati EP. Incidence of dengue hemorrhagic fever related to annual rainfall, population density, larval free index and prevention program in Bandung 2008 to 2011. Althea Medical Journal 2015; 2 (2): 262-267.
3. Hartoyo E, Indra R, Widajanto E, Hadinegoro SR. Interleukin-17 role on the occurrence of plasma leakage dengue hemorrhagic fever through cytokine proinflamasi activation: (Study of in-vivo and in-vitro macrophage culture and cell endothelia). International Journal of Science and Research 2014; 3 (11): 1711-1717.
4. Tsai JJ, Chokephaibulkit K, Chen PC, Liu LT, Hsiao HM, Lo YC, Perng GC. Role of cognitive parameters in dengue hemorrhagic fever and dengue shock syndrome. Journal of Biomedical Sciences 2013; 20 (88): 1-11.
5. Roopashri G, Vaishali MR, David MP, Baig M, Navneetham A, Venkataraghavan K.Clinical and oral implications of dengue fever: A review. Journal of International Oral Health 2015; 7 (2): 1-5.
6. Sermugapandian I, Kumar SV, Jayashree V, Manjuladevi K,Ravichandiran V. Dengue virus: A review. World Journal of Pharmacy and Pharmaceutical Sciences 2015; 4 (2): 151-160.
7. Koshy JM, John M. Myocarditis complicating pregnancy in dengue hemorrhagic fever. Indian Journal of Clinical Medicine 2012; 3: 17-20.
8. Lee IK, Lee WH, Liu JW, Yang KD. Acute myocarditis in dengue hemorrhagic fever: a case report and review of cardiac complications in dengue-affected patients. International Journal of Infectious Diseases 2010; 14 (2010): e919-e922.
9. Lei HY, Huang KJ, Lin YS, Yeh TM, Liu HS, Liu CC. Immunopathogenesis of dengue hemorrhagic fever. American Journal of Infectious Disease 2008; 4 (1): 1-9.
10. Soundravally R,Hoti SL, Patil SA, Cleetus CC, Zachariah B, Kadhiravan T, Narayanan P, Kumar BA. Association between proinflammatory cytokines and lipid peroxidation in patients with severe dengue disease around defervescence. International Journal of Infectious Disease 2014; 18 (2014): 68-72.
11. Gil L, Martinez G, Tapanes R, Castro O, Gonzalez D, Bernardo L, Vasquez S, Kouri G, Guzman M. Oxidative stress in adult dengue patients. American Journal of Tropical Medicine and Hygiene 2004; 71 (5): 652-657.
12. Castro R, Pinzon HS, Guzman NA. A systematic review of observational studies on oxidative/nitrosative stress involvement indengue pathogenesis. Colombia Medica 2015; 46 (3): 135-143.
13. Galani IE, Andreakos E. Neutrophils in viral infections: Current concepts and caveats. Journal of Leukocyte Biology 2015; 98: 557-564.
14. Yunanto A, Firdaus RT, Triawanti, Suhartono E. Advanced Oxidation Protein Products (AOPPs) of newborn at risk of sepsis as novel parameter for early-onset neonatal sepsis. International Journal of Bioscience, Biochemistry, and Bioinformatics 2014; 4 (2): 90-93.
15. Metzler KD, Fuchs TA, Nauseef WM, Reumaux D, Roesler J, Schulze I, Wahn V, Papayannopoulos V, Zychlinsky A. Myeloperoxidase is required for neutrophil extracellular trap formation:implications for innate immunity.
16. Yunanto A, Firdaus RT, Triawanit, Suhartono E. Salivary antioxidative index in newborns at risk of sepsis as novel parameter for early-onset neonatal

sepsis. Journal of Medical and Bioengineering 2014; 3 (1): 63-66.
17. Yunanto A, Iskandar, Suhartono E. In-vitro effects of some antibiotic drugs on saliva thiocyanate and oxidation protein products levels on newborn at risk of sepsis. International Journal of Pharmaceutical and Clinical Research 2016; 8 (1): 86-89.
18. Suhartono E, Triawanti, Leksono AS, Djati MS. Effects of cadmium exposure on lipid peroxidation and chlorinative stress of rat kidney. Journal of Experimental Life Science 2015; 5 (1): 1-5.
19. Suhartono E, Nijka JA, Anhar VY, Sari RA, Edyson, Marisa D. Anti-lipid peroxidation activities of three selected fruits juices against cadmium induced liver damage *in vitro*. Journal of Tropical and Life Sciences 2015; 5 (2): 75-79.
20. Aflanie I, Muhyi R, Suhartono E. Effect of heavy metal on malondialdehyde and advanced oxidation protein produtcs concentration: A focus on arsenic, cadmium, and mercury. Journal of Medical and Bioengineering 2015; 4 (4): 332-337.
21. Suhartono E, Triawanti, Leksono AS, Djati MS. Oxidative stress and kidney glycation in rats exposed cadmium. International Journal of Chemical and Engineering and Applications 2014; *5*: 497–501.
22. Rao AM, Anand U, Anand CV. Myeloperoxidase in chronic kidney disease. Indian Journal of Clinical Biochemistry 2011; 26 (1): 28-31.
23. Suhartono E, Triawanti, Yunanto A, Firdaus RT, Iskandar. Chronic Cadmium Hepatooxidative in Rats: Treatment with Haruan Fish (Channa striata) Extract. APCBEE Procedia 2013; 5: 441-445.
24. Drescher B, Bai F. Neutrophil in viral infections, friend or foe?. Virus Research 2013; 171 (1): 1-7.
25. Olagnier D, Peri S, Steel C, Montfoort NV, Chiang C, Beljanski V, Slifker M, He Z, Nichols CN, Lin R, Balachandran S, Hiscott J.Cellular oxidative atress response controls the antiviral and apoptotic programs in dengue virus-infected dendritic cells. PLoS Pathogens 2014; 10 (12): 1-18.
26. Guven MK, Aydin H, Yildiz G, Engin A, Celik VK, Bakir D, Deveci K. The importance of myeloperoxidase enzyme activity in the pathogenesis of Crimean–Congo haemorrhagic fever F. Journal of Medical Microbiology 2013; 62: 441-445.
27. Khan E, Kisat M, Khan N, Nasir A, Ayub S, Hasan R. Demographic and clinical features of dengue fever in pakistan from 2003–2007: A retrospective cross-sectional study. PloS ONE 2010; 5 (9): 1-7.
28. Kothari N, Keshari RS, Bogra J, Kohli M, Abbas H, Malik A, Dikshit M, Barthwal MK. Increased myeloperoxidase enzyme activity in plasma is an indicator of inflammation and onset of sepsis. Journal of Critical Care 2011; 26: 435.e1-435.e7.
29. Ulgen S, Yaramis CP, Rayaman E, Gurer US, Or ME, Sehirli AO. Effects of inactivated *Parapoxvirus ovis* on polymorphonuclear leukocyte function and myeloperoxidase activity in horses. Veterinarni Medicina 2014; 59 (12): 631-636.
30. Marisa D, Rudito M, Zagita MG, Biworo A, Suhartono E. Hepatotoxicity effect of rifampicin and isoniazid via chlorinative stress pathway mechanism *In-vitro*. International Journal of Toxicological and Phamrmacological Research 2016; 8 (1): 18-22.
31. Ho E, Galougahi KK, Liu CC, Bhindi R, Figtree GA. Biological markersofoxidativestress:Applicationstocardiovascular researchandpractice. Redox Biology 2013; 1: 483-491.

Oxidative and Chlorinative Stress in Children with Dengue Hemorrhagic Fever Tahun Terbit 2016

ORIGINALITY REPORT

12%	8%	9%	1%
SIMILARITY INDEX	INTERNET SOURCES	PUBLICATIONS	STUDENT PAPERS

PRIMARY SOURCES

1	www.scielo.br Internet Source	1%
2	www.osirjournal.net Internet Source	1%
3	eprints.skums.ac.ir Internet Source	1%
4	www.frontiersin.org Internet Source	1%
5	www.omicsonline.org Internet Source	1%
6	theses.gla.ac.uk Internet Source	<1%
7	"IOF Regionals ANZBMS Annual Scientific Meeting, held with the JSBMR 2nd Asia-Pacific Osteoporosis and Bone Meeting", Osteoporosis International, 2011 Publication	<1%

jneuroinflammation.biomedcentral.com

8	Internet Source	<1%
9	Binita Koirala Sharma, David Charles Klinzing, John Donnie Ramos. "Modulatory activities of Zingiber officinale Roscoe methanol extract on the expression and activity of MMPs and TIMPs on dengue virus infected cells", Asian Pacific Journal of Tropical Disease, 2015 Publication	<1%
10	Noha M. Shafik, Ghada M. Abou-Fard. " Ameliorative Effects of Curcumin on Fibrinogen-Like Protein-2 Gene Expression, Some Oxido-Inflammatory and Apoptotic Markers in a Rat Model of -Arginine-Induced Acute Pancreatitis ", Journal of Biochemical and Molecular Toxicology, 2016 Publication	<1%
11	J.M. Koshy, M. John. "Myocarditis Complicating Pregnancy in Dengue Hemorrhagic Fever", Indian Journal of Clinical Medicine, 2012 Publication	<1%
12	www.jbiomedsci.com Internet Source	<1%
13	ijarcet.org Internet Source	<1%
14	www.scirj.org	

	Internet Source	<1%
15	Lei, H.Y.. "Transient Hemophagocytic Activity in Dengue Immunopathogenesis", Journal of the Formosan Medical Association, 200908 Publication	<1%
16	ebm.rsmjournals.com Internet Source	<1%
17	journal.waocp.org Internet Source	<1%
18	Olagnier, David, Suraj Peri, Courtney Steel, Nadine van Montfoort, Cindy Chiang, Vladimir Beljanski, Michael Slifker, Zhong He, Carmen N. Nichols, Rongtuan Lin, Siddharth Balachandran, and John Hiscott. "Cellular Oxidative Stress Response Controls the Antiviral and Apoptotic Programs in Dengue Virus-Infected Dendritic Cells", PLoS Pathogens, 2014. Publication	<1%
19	Phung Khanh Lam, Tran Van Ngoc, Truong Thi Thu Thuy, Nguyen Thi Hong Van et al. "The value of daily platelet counts for predicting dengue shock syndrome: Results from a prospective observational study of 2301 Vietnamese children with dengue", PLOS Neglected Tropical Diseases, 2017 Publication	<1%

20	Handbook of Experimental Pharmacology, 2015. Publication	<1%
21	Eu-Jin Cho, Ji-Sun Shin, Kyung-Sook Chung, Yong Sup Lee, Young-Wuk Cho, Nam-In Baek, Hae-Gon Chung, Kyung-Tae Lee. "Arvelexin Inhibits Colonic Inflammation by Suppression of NF-κB Activation in Dextran Sulfate Sodium-Induced Mice and TNF-α-Induced Colonic Epithelial Cells", Journal of Agricultural and Food Chemistry, 2012 Publication	<1%
22	doaj.org Internet Source	<1%
23	astro2005.abstractsnet.com Internet Source	<1%
24	www.pubmedcentral.nih.gov Internet Source	<1%
25	Ying-Jung Wu, Yi-Shan Li, Wei-Lung Tseng, Chi-Yu Lu. "Microextraction combined with microderivatization for drug monitoring and protein modification analysis from limited blood volume using mass spectrometry", Analytical and Bioanalytical Chemistry, 2018 Publication	<1%
	P. Manivannan, P. Isakki Devi. "Dengue fever	

26	prediction using K-means clustering algorithm", 2017 IEEE International Conference on Intelligent Techniques in Control, Optimization and Signal Processing (INCOS), 2017 Publication	<1%
27	Ing-Kit Lee, Wen-Huei Lee, Jien-Wei Liu, Kuender D. Yang. "Acute myocarditis in dengue hemorrhagic fever: a case report and review of cardiac complications in dengue-affected patients", International Journal of Infectious Diseases, 2010 Publication	<1%
28	Agieshkumar Balakrishna Pillai, Krishna Raja Muthuraman, Vignesh Mariappan, Srisha Srivatsan Belur et al. "Oxidative stress response in the pathogenesis of dengue virus virulence, disease prognosis and therapeutics: an update", Archives of Virology, 2019 Publication	<1%
29	Erum Khan, Mehreen Kisat, Nabil Khan, Amna Nasir, Salma Ayub, Rumina Hasan. "Demographic and Clinical Features of Dengue Fever in Pakistan from 2003–2007: A Retrospective Cross-Sectional Study", PLoS ONE, 2010 Publication	<1%
30	Cleetus Cherupanakkal, Vijayakumar	<1%

Ramachadrappa, Tamilarasu Kadhiravan, Narayanan Parameswaran et al. "Differential expression of NADPH oxidase-2 (Nox-2) and nuclear factor-erythroid 2-related factor 2 (Nrf2) transcripts in peripheral blood mononuclear cells isolated from dengue patients", VirusDisease, 2017

Publication

Exclude quotes	On	Exclude matches	< 2 words
Exclude bibliography	On		