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Characteristics of forest honey from several areas in Riau Province, Indonesia

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Abstract

Honey quality is influenced by several factors, including transportation conditions and length of storage. This study aimed to investigate the composition, characteristics, and quality of honey from several regions in Riau and Riau honey sent to Java Island. Honey samples were obtained from five districts in Riau Province, namely, Inderagiri Hulu, Kuantan Singingi, Pelalawan, Bengkalis, and Kampar Regencies. The honey parameters observed were based on the Indonesia National Standard (SNI) of honey. Data were analyzed descriptively by comparing the characteristics of honey samples from Riau Province. Results showed a significant difference between honey sent and those not sent between islands. Significant differences were mainly observed in hydroxymethylfurfural (HMF) levels. Almost all honey samples sent to Java Island had HMF levels above 50 mg/kg. Meanwhile, all Riau honey samples from the five regions had HMF levels below 50 mg/kg. The diastase enzyme activity was slightly higher in the honey samples from the five regions. Land transportation to the island decreased the quality of honey as indicated by the high levels of HMF and decreased diastase enzyme activity. Therefore, mileage and storage conditions must be considered in the delivery of Riau honey to other regions.

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1. Introduction

Honey is a natural product with a sweet taste produced by honeybees from flower nectar and consumed due to its high nutritional value and health benefits, such as antioxidants, anti-inflammatory, antimicrobial, and wound healing. The composition of honey depends on the type of flower, season, environmental factors, and postharvest treatment by beekeepers (1,2).

Riau Province is one of the largest honey producers in Indonesia. Honeybee cultivation is prospective enough to be developed in Riau Province because of the gap between supply and demand for honey production. The supply of honey is about 8,000 tons/year, and the demand for honey is 20,000 tons/year. Honey shortage is addressed by importing from other countries and domestic industries. Problems often faced by honeybee farmers include simple technology, unsustainable honey harvesting, and inability of the product to meet the standards or criteria set by the market (3).

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Land transportation is a necessary route for the delivery of honey outside Riau Province. Mileage that takes days, especially when shipping between the islands of Sumatra and Java, can reduce the quality of honey. Packing and high temperatures during land trip can also damage the composition of honey, thereby reducing its quality and consequently its health efficacy. For instance, hydroxymethylfurfural (HMF) and diastase activity are commonly used as parameters of honey quality, and both are greatly affected by temperature and storage conditions (4.5).

No data are available on the quality of Riau honey samples and the differences in the quality and composition of honey from Riau Province sent by land transportation to islands in Sumatra and Java with those of honey that only circulates in Riau Province. Therefore, this study aimed to determine the characteristics and quality of Riau honey samples, and compare the composition of honey from several regions in Riau Province with that of honey sent to Java Island.

2. Materials and Methods

A descriptive method was used to determine the characteristics of Riau honey and the effect of inter-island transportation on the composition of honey samples from five districts in Riau Province, namely, Indragiri Hulu (Inhu), Kuantan Singingi (Kuansing), Pelalawan, Bengkalis, and Kampar. This research was conducted from March to June 2019.

Forest honey samples consisted of one from Indragiri Hulu (Inhu) District, one from Kuantan Singingi (Kuansing), one from Bengkalis, two from Pelalawan, and one from honey farms in Kampar (kelulut honey). The honey sample from Pelalawan consisted of one sample from the Sorek area and one commercial honey sample.

In the first phase, honey samples from five districts in Riau Province were packaged in plastic bottles and sent to the Riau Provincial Trade Office for quality analysis. Testing parameters were adopted from SNI (Indonesia National Standard) 3545:2013.

In the second phase, honey samples from the five districts in Riau Province were sent to Java Island using land expedition services for 4 days. Honey was packaged in plastic bottles and then in boxes made of pieces of wood. Honey testing was conducted at the Agro-Industry Center (BBIA), Bogor. Testing for diastase activity, HMF, water, and acidity was conducted following SNI 3545:2013. Analysis of reducing sugar, sucrose sugar, total insoluble solids, and ash content was performed as per SNI 01-2892-1992. For metal contamination, MU/MO/10 (AAS) was adopted for lead and cadmium, MU/MO/12 (AAS) for mercury, MU/MO/13 (AAS) for arsenic, AOAC.999.11 (9.1.09.2005) for copper, and AOAC 985.35 (50.1.14.2005) for calcium. Total plate number (TPN) was tested following ISO 4833:2003(E) and ISO 7218:2012. Coliforms were tested with the 2002 BAM method, and mold and yeast were examined using BAM 2001. The laboratories were accredited for honey sample testing.

The honey samples were examined in terms of 13 parameters such as diastase activity, HMF, water content, reducing sugar (glucose), sucrose, acidity, total insoluble solids, ash, metal contamination (Pb and Cd), microbial contamination (total plate count, coliform, mold, and yeast), and calcium levels tested at BBIA using SNI Parameter in 2013. Each sample was measured twice to ensure the validity and reproducibility of the data. The results were qualitatively analyzed to describe the characteristics of several honey samples in Riau and to determine the changes in HMF levels and diastase activity after the honey was sent to Java.

This study was approved by the ethical clearance committee of the Faculty of Public Health Universitas Indonesia (No.: 332/UN2.F10/PPM.00.02/2019, date of issue: May16, 2019).

3. Results and Discussion

3.1. Characteristics of Riau Forest Honey

Riau is a province in Indonesia and is located on Sumatra Island. The climate is tropical with a maximum temperature of 35.1 °C and a minimum temperature of 21.8 °C. The average annual rainfall ranges between 1,700 and 4,000 mm (6).

The honey samples were analyzed at the Riau Provincial Trade Office using SNI 2013 parameters. The characteristics of some Riau honey samples are presented in Table I. The highest diastase activity was found in the Kuansing honey sample (8.16), and an average of 0 was obtained for the honey samples from other regions. The requirement for diastase activity according to SNI is a minimum of 3 DN. The diastase enzyme activity of the honey in this study was lower than that of the honey from India and Ethiopia varying from 36.7 DN to 57.5 DN and from 3.91 DN to 13.6 DN, respectively (7,8) but was higher than that of the honey from Malaysia ranging between 0 and 0.75 DN (9). The variation of diastase activity could be caused by the honey's floral origin.

Table 1. Characteristics Of Riau Honey Samples.

Characteristics	Kuansing	Inhu	Bengkalis	Kampar	Sorek	Commercial
Diastase Activity (DN)	8.16	0	0.38	0.34	0.86	0
Hidroxiimethylfurfural (Mg/Kg)	0.49	9.05	11.94	0.58	9.44	27.84
Moisture (% B/B)	> 25	24.2	20.6	> 25	24.6	19.4
Reducing Sugar (% B/B)	63.86	64.18	63.37	37.77	67.14	63.37
Sucrose (% B/B)	0	2.53	6.38	0.35	0	6.14
Acidity (1 N/Kg)	42.65	49.70	94.73	198.47	39.86	82.04
Total Insoluble solid (% B/B)	0.81	1.02	0.37	1.54	1.08	1.90
Ash (% B/B)	0.48	0.20	0.54	0.44	0.20	0.42
Metal Contamination:						
Lead (Mg/Kg)	< 0.34	< 0.34	< 0.34	< 0.34	< 0.34	< 0.34
Cadmium (Mg/Kg)	< 0.16	< 0.16	< 0.16	< 0.16	< 0.16	< 0.16
Microbial Contamination:						
Total Plate Count (Col/G)	1.0×10^6	2.5×10^3	6.2×10^2	2.8×10^3	6.9×10^3	3.7×10^3
Coliform (Apm/G)	< 3.0	< 3.0	< 3.0	< 3.0	< 3.0	< 3.0
Mold And Yeast (Col/G)	2.8×10^2	3.0×10^1	2.0×10^1	1.0×10^2	7.0×10^1	2.0×10^1

The diastase activity in honey is affected by several factors. The temperature and heating process may decrease the diastase content. Biological differences among species of bees also affect the diastase content of honey (10).

The enzyme content in honey is one of the characteristics that are considered beneficial for health. The main enzymes found in honey are invertase (saccharase), diastase (amylase), and glucose oxidase. Diastase activity is an essential factor that determines the quality of honey. Although enzymes are a small part of honey's composition, their presence is

associated with health advantages, including nutritional benefits, carbohydrate digestion, and antimicrobial activity (11,12).

For HMF level parameters, all Riau honey samples met SNI requirement of below 50 mg/kg (from 0.49 mg/kg to 27.84 mg/kg). The lowest HMF level was found in Kuansing honey sample at 0.49 mg/kg. The average HMF level in this study was lower than that of Tunisian honey (12.07 mg/kg to 27.43 mg/kg) (13) and higher than that of Ethiopian honey (0 to 3.37 mg/kg) (8). The high water content of honey could increase its HMF level.

HMF is a cyclic aldehyde produced from the breakdown of sugar through the Maillard reaction (a non-enzymatic browning reaction) during food processing or long storage. The content of simple sugars (glucose and fructose), acidic compounds, and minerals can increase the production of HMF. HMF affects the freshness of honey because freshly harvested honey contain minimal and even no HMF. HMF concentrations tend to increase during processing or storage (4). High acid content, moisture content, sugars, amino acids, and minerals could increase HMF level (14).

The moisture content of Riau honey samples ranged from 19.4% to >25%. The maximum water content according to SNI is 22. The honey samples from Bengkalis had a moisture content of 20.6, and commercial honey had a water content of 19.4. The Bengkalis honey sample has undergone dehumidification, a process of decreasing water content. Commercial honey is also suspected to have undergone this procedure.

The moisture content of Riau honey samples was higher than that of Tunisia honey (17.27% to 19.80%) and Turkish honey (15.1% and 20.4%) (13,15). The high moisture content in Riau honey samples can be caused by honey harvesting in the rainy season.

Honey contains water, fructose, and acidic compounds. Mold can grow in honey when the water content of honey is high. Fermentation is a problem in honey. If the water content increases, then fermentation and decomposition could rapidly occur (16). Water content in honey is affected by plant species, geographical origin, season at harvest time, and postharvest processing, such as honey extraction, processing, and storage conditions (14).

The highest reducing sugar content was detected in Sorek honey samples at 67.14 (minimum SNI requirement of 65). The honey samples from other regions had reducing sugar levels below 65. These values were lower than the reducing sugar content of Saudi honey (72.36 ± 0.32 g/100 g) (1) and Croatian honey with an average of 67.69 ± 7.05 g/100 g for acacia honey and 77.82 ± 12.25 g/100 g for chestnut honey (17). The reducing sugar content is determined by the duration of storage and honey collection time (18).

Honey is rich in reducing sugars, notably fructose (38%) and glucose (31%). Both types of sugar do not require hydrolysis by enzymes in the digestive tract to directly be absorbed in the small intestine. Fructose may lower hyperglycemia or sugar levels in experimental mice, healthy subjects, and patients with diabetes. Therefore, fructose does not enhance blood glucose. Furthermore, its metabolism does not require insulin. Glucose also increases fructose absorption in the gut (11).

The requirement for sucrose content in honey is a maximum of 5% according to SNI. The honey from Kuansing, Inhu, Kampar, and Sorek achieved this requirement. However, the honey from Bengkalis and Pelalawan (commercial) did not reach the SNI standard and had sucrose contents of 6.38% and 3.14%, respectively. Tunisian honey has a sucrose level below 5% (0.20% to 4.60%) (13). Multifloral honey from Poland and Slovakia has a sucrose level of $4.40\% \pm 1.95\%$ and $4.47\% \pm 2.15\%$, respectively (19). Source of plant, immaturity of honey, and artificial feeding of bees affect the sucrose content in honey (14).

The sucrose in honey is turned into glucose and fructose. High levels of sucrose indicate premature honey harvesting. Hence, invertase cannot fragment sucrose into glucose and fructose. Sugar content in honey is affected by the ratio of fructose–glucose and sucrose content. Crystallized honey suggests a high level of glucose. Therefore, the glucose in honey is hardly soluble in water. Honey also contains other types of sugar that can impede crystallization (13,20).

The maximum acidity level in honey is 50 N/Kg according to SNI 2013. Bengkalis, Kampar, and Commercial honey samples had a high acidity above 50 N/kg. Sudan honey has an acidity of 51.80 ± 1.95 meq/kg for *Apis mellifera* honey and 98.40 ± 1.82 meq/kg for *Apis florea* honey (21). Meanwhile, Tunisian honey has a low acidity ranging from 0.0071 N/kg to 0.0272 N/kg. Different levels of acidity in honey may be caused by the varied botanical origin or the difference in harvest season (13).

The characteristics of flower nectar sucked by bees affect the acidity in honey. Low acidity indicates freshness, and high acidity indicates the fermentation of glucose into organic acids. The consistency of honey also affects its acidity. Liquid texture has a higher acidity than solid texture because the production of free acid from fermentation easily occurs in liquid honey texture. In addition, acidity is affected by the length of storage. Free acids produced by fermentation during storage will increase the acidity of honey (20).

The total insoluble solids in this study ranged from 0.37% to 1.90%. SNI 3545:2013 imposed a maximum of 0.5%, which was only achieved by Bengkalis honey. By contrast, Turkish honey has a weak total insoluble solid content between 0.001% and 0.080% (22), and Ethiopian honey has a total insoluble solid content of 0.005%–0.22%. Honey requires a proper procedure to avoid contamination with insoluble solids (23). A high number of insoluble solids in honey, such as impurities or other particles, could indicate poor hygiene during postharvest honey process.

All honey samples met the maximum ash content according to SNI (not more than 0.5%), except for Bengkalis honey that had a slightly high ash content (0.54%). Sudan honey contains 0.26% (*Apis mellifera*) and 1.16% (*Apis florea*) ash content (21), and Brazil honey has an ash content between 0.11% and 0.95% (24).

Ash levels indicate mineral content and are affected by honey extraction methods and bee food (21,25). Other factors can influence ash content in honey, such as environment, geography, and vegetation (24).

For metal contamination, all Riau honey samples met the SNI requirements (<2.0 mg/kg for lead and <0.2 mg/kg for cadmium). Turkey honey has low lead (mean of 0.04 ± 0.09) and cadmium (mean of 0.0002 ± 0) contents. In Chinese honey, the average contents of lead and cadmium are 1.34 and 33.98 $\mu\text{g}/\text{kg}$, respectively (26,27). Most of Riau honey samples were derived from multifloral honey or forest honey distant from industrial areas. For this reason, the concentration of heavy metals was low in Riau honey samples.

Bees can fly up to a radius of 4 km from the hive, allowing them to access an area of 50 km². Contact of bees with air, soil, and water cause their honey to contain heavy metals. The level of heavy metals in honey can indicate the number of heavy metals found in the environment. Contaminated air, water, and soil and honey processing influence the composition of heavy metals in honey (11,28).

Although heavy metals constitute a small part of honey, they are crucial in the quality of honey. Honey can contain heavy metals such as lead and cadmium that can damage health (11). Furthermore, heavy metals such as Pb, Cd, Hg, Cr, Cu, Mn, Ni, and Zn are

nonbiodegradable substance that can accumulate in the human body and cause health problems, including metabolic and respiratory disorders, headaches, nausea, and vomiting (28).

The maximum TPC according to SNI is $<5 \times 10^3$ cfu/g. Kuansing honey and Sorek honey had TPC above SNI, namely, 1.0×10^6 and 6.9×10^3 cfu/g, respectively. Similarly, Nigeria honey has a TPC ranging from 1.0×10^4 cfu/ml to 1.2×10^5 cfu/ml, and Ghana honey has a TPC between 6.0×10^4 and 1.1×10^5 cfu/ml (29,30).

TPC indicates the number of microorganisms (pathogenic and nonpathogenic) in honey. For example, *Bacillus* sp., *Clostridium* sp., *Micrococcus* sp., lactic acid bacteria, yeast, and fungi are present in honey. These microorganisms could originate from the air, soil, water, dust, pollen, and nectar. In addition, unhygienic conditions during honey extraction, processing, and packaging may cause microorganism contamination in honey (30,31).

Honey can load microorganisms, such as bacteria, mold, and yeast. Contamination in honey can occur either through primary sources (pollen, flower nectar, dust, soil, body, and digestive tract of bees) or secondary sources during postharvest processing, such as humans and equipment (32,33).

All honey samples fulfilled coliform requirements i.e., <3 APM/g but failed to meet the requirements for mold and yeast because they exceeded SNI requirements, i.e., $<1 \times 10^1$ APM/g. Similarly, the amounts of mold and yeast found in Croatian honey are above the accepted standard for mold at 18–182 cfu/g and for yeast at 18–1,300 cfu/g. The molds in Croatian honey originated from primary sources such as the digestive tract of bees (34). Microorganisms are found in honey because they can survive in high sugar solutions and acid conditions (29).

Most microorganisms in honey are not harmful to health. The antimicrobial properties in honey can inhibit the growth of microorganisms. However, honey may contain *Clostridium botulinum*, which leads to infant botulism. Consequently, honey is not recommended for children under 1-year-old (31).

Table II shows the honey quality test results with 13 parameters based on SNI 3545:2013. None of the Riau honey samples met all the honey requirements based on SNI 3545:2013. Honey samples from Kuansing, Inhu, and Sorek met eight SNI requirements, and honey samples from Bengkalis, Kampar, and commercial honey samples met only seven SNI 3545:2013 requirements.

Table 2. Honey Quality Test Results.

Honey Samples	Test Parameters												
	1	2	3	4	5	6	7	8	9	10	11	12	13
Kuansing	√	√	X	X	√	√	X	√	√	√	X	√	X
Inhu	X	√	X	X	√	√	X	√	√	√	√	√	X
Bengkalis	X	√	√	X	X	X	√	X	√	√	√	√	X
Kampar	X	√	X	X	√	X	X	√	√	√	√	√	X
Sorek	X	√	X	√	√	√	X	√	√	√	X	√	X
Comersial	X	√	√	X	X	X	X	√	√	√	√	√	X

√ = Qualified, X = Not Qualified, 1 = Diastase Activity, 2 = HMF, 3 = Total Moisture, 4 = Reduced Sugar Content, 5 = Sucrose Content, 6 = Acidity, 7 = Total insoluble solid, 8 = Ash, 9 = Lead Contamination, 10 = Cadmium Contamination, 11 = Total Plate Count, 12 = Coliform, 13 = Mold and yeast

Each of the three honey samples that achieved the eight SNI requirements had priorities. Kuansing honey samples showed high diastase activity, Sorek honey samples had high levels of reducing sugar, and Inhu honey samples contained low total plate numbers.

3.2. Changes in Riau Honey Composition after Inter-Island Shipping

The honey samples sent to Java were analyzed at BBIA, Bogor. Changes in the composition of honey samples were observed, primarily in the level of diastase activity and HMF levels. These two parameters are affected by the heating process and have a negative correlation. Diastase activity tends to decrease when the heating process is prolonged, and HMF level tends to increase with heating time (7). Table III provides the changes in the composition of Riau honey sent by land transportation to Java Island.

Table 3. Change In Honey Composition After Inter-Island Delivery.

Characteristics	Kuansing	Inhu	Bengkalis	Kampar	Sorek	Commercial
Diastase Activity (Dn)	0	0	0	0	0	1.83
Hydroxymethylfurfural (Mg/Kg)	128	153	0	129	213	0
Moisture (% B/B)	24.0	23.6	19.1	33.9	23.3	17.5
Reducing Sugar (% B/B)	64.0	65.7	57.4	28.7	58.4	61.2
Sucrose (% B/B)	1.71	0	7.68	0	10.1	7.70
Acidity (1 N/Kg)	31.5	33.3	88.9	62.8	37.3	38.3
Total Insoluble Solid (% B/B)	0.04	0.23	0.76	0.65	0.39	0.16
Ash (% B/B)	0.08	0.09	0.40	0.09	0.11	0.42
Metal Contamination:						
Lead (Mg/Kg)	< 0.034	< 0.034	< 0.034	< 0.034	< 0.034	< 0.034
Cadmium (Mg/Kg)	< 0.007	< 0.007	< 0.007	< 0.007	< 0.007	< 0.007
Copper (Mg/Kg)	0.09	0.02	0.04	0.34	0.09	0.15
Mercury (Mg/Kg)	< 0.005	< 0.005	< 0.005	< 0.005	< 0.005	< 0.005
Arsenic (Mg/Kg)	< 0.013	< 0.013	< 0.013	< 0.013	< 0.013	< 0.013
Microbial Contamination:						
Total Plate Count (Kol/G)	10	25	< 10	< 10	50	5
Coliform (Apm/G)	< 3	< 3	< 3	< 3	< 3	< 3
Mold (Kol/G)	< 10	27	< 10	< 10	< 10	< 10
Yeast (Kol/G)	4.9 X 10 ²	1.8 X 10 ²	< 10	< 10	7.4 X 10 ²	< 10
Calcium (Mg/100 G)	9.74	18.6	27.0	18.5	12.1	12.7

Table 3 shows a significant decrease in the quality of honey after inter-island delivery, especially in the diastase activity. Almost all honey samples had 0 (zero) activity of diastase, except for the commercial honey from Pelalawan with a diastase level of 1.83 DN. This value is below the SNI requirement, that is, a minimum of 3 DN.

Diastase activity is a measurement of α -amylase and β -amylase activities and is often used as a quality parameter of honey. Diastase activity decreases due to heating (9). A decreased diastase activity can be due to transportation between the islands of Sumatra and Java. The storage conditions of honey samples prior to transport might have caused heating in honey during transport for 4 days.

HMF levels increased significantly (>50 mg/kg), except in Bengkalis honey samples and commercial honey samples with HMF 0 levels. This phenomenon occurred because HMF broke down into levulinic acid and formic acid. Hence, HMF was not detected.

The determination of HMF in honey samples aims to find out the freshness of honey. Increased HMF is associated with prolonged storage at high temperatures or excessive heating of honey. The HMF level acceptance limit varies by country and is high in the tropics

at 80 mg/kg (4,18). According to SNI 3545: 2013, HMF levels in honey must not be more than 50 mg/kg.

Factors that can affect the formation of HMF include high temperature, low pH, prolonged storage, high water content, and use of metal box (4). Honey samples sent by land transportation can increase HMF levels due to storage conditions that cause the heating of honey during a 4-day expedition. The high water content in four honey samples (Kuansing, Inhu, Kampar, and Sorek) can also induce the increase in their HMF levels.

HMF has negative effects on human health, such as cytotoxicity on mucous membranes, skin, upper respiratory tract, chromosomal aberrations, and carcinogenic effects on humans and animals. However, recent research showed that HMF has positive effects such as antioxidants, anti-allergies, anti-inflammatory, and anti-carcinogens. These results were obtained at the preclinical stage and therefore require further research (4).

For other parameters, no significant changes were observed in the honey sent to Java. Calcium levels were only measured in BBIA Bogor, and the highest of 270 mg/kg was found in Bengkalis honey. The calcium level in this study was higher than that in honey from natural hives in Nigeria that ranges between 37,76 and 40,90 mg/kg (35) and honey from Tunisia that ranges between 221,07 and 113,85 mg/kg honey (13).

The predominant minerals in honey are potassium, sodium, calcium, and magnesium (14). The mineral content of honey is manifested in its color, that is, dark honey has more minerals than light honey (35). Consumption of honey provides calcium that is easily absorbed and could strengthen bone mass. This mineral could reduce the risk of osteoporosis or low bone mass, a cause of fractures especially in the elderly (36).

4. Conclusions

None of the analyzed Riau's honey samples fulfilled all SNI 3545:2013 requirements. Only three honey samples, namely Kuansing, Inhu, and Sorek honey samples, could fulfill 8 out of 13 SNI requirements, and another three honey samples could meet 7 out of 13 SNI 3545:2013 requirements. A decrease in diastase activity and an increase in HMF were observed in the honey sent to Java. Therefore, distance and storage conditions must be considered in the land transportation of honey out of Riau Province to maintain honey quality.

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Author Contributions

Conceptualization, S.D., W.S., Y.Y., U.P.; methodology, S.D. and W.S.; formal analysis, W.S. and S.D.; investigation, S.D. and W.S.; data curation, W.S. and S.D.; writing—original draft preparation, S.D.; writing—review and editing, S.D., W.S., Y.Y., U.P., G.B.; project administration, S.D.; funding acquisition, S.D., W.S., Y.Y., and U.P. All authors have read and agreed to the published version of the manuscript.

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Institutional Review Board Statement

This study was conducted according to the guidelines of the Declaration of Helsinki, and approved by the Institutional Review Board (or Ethics Committee) of the Health Research Ethics Committee, Faculty of Public Health, Universitas Indonesia, Indonesia (protocol code 332/UN2.F10/PPM.00.02/2019, 16 May 2019).

Conflicts of Interest

The authors declare no conflict of interest.

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