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Analysis of *Ascaris lumbricoides* Infection with Total Ig E Levels in Elementary School Children of Lifuleo Village, East Kupang District

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Abstract. *Worm infections are a health concern in Indonesia and all other tropical nations. A number of species, including roundworms (Ascaris lumbricoides), whipworms (Trichuris trichiura), hookworms (Hookworms), Necator americanus, Ancylostoma duodenale, and Strongyloides stercoralis, are members of the Soil Transmitted Helminth nematodes. Children in elementary school are among the age groups most vulnerable to Ascaris lumbricoides. Without a clean and healthy lifestyle, an Ascaris lumbricoides infection might have major repercussions. Research on the correlation between Ascaris lumbricoides infection and total immunoglobulin E levels in primary school students is therefore required. This study uses a cross-sectional, observational analytical design. Children at Lifuleo Village's elementary schools served as the research subjects. The ECLIA method was used to assess total immunoglobulin E levels, and the Kato-Katz method was used to diagnose Ascaris lumbricoides infection under a microscope. An aberrant total immunoglobulin E level of 12.5% was observed in 42.5% of Ascaris lumbricoides eggs, according to the examination results.*

Keywords *: Ascaris lumbricoidest, Primary School Children, Immunoglobulin E Total*

1. INTRODUCTION

Worm infestations are a health concern in Indonesia and all other tropical nations. Worm infections fall under the category of neglected diseases, which are infections that receive insufficient attention, are chronic, do not exhibit obvious clinical symptoms, and only show symptoms over an extended period of time. This disease frequently affects those who live in poverty and have inadequate access to clean water and sanitation. Among the soil-transmitted helminths (STH) is Ascaris lumbricoides. Humans that consume the eggs of STH intestinal nematode worms through the oral-fecal pathway become infected. Ascaris lumbricoides, Trichuris trichiura, Necator americanus, Ancylostoma duodenale, and Strongyloides stercoralis are the species that make up this worm. Although STH infections are found in tropical and subtropical regions, the highest concentrations are found in Sub-Saharan Africa, America, China, and East Asia. STH affects almost 1.5 billion people, or 24% of the global population. Preschoolers, school-age children, women of childbearing age, including those who are pregnant, and individuals working in high-risk occupations like farming (Ensink, Hoek, Mukhtar, Tahir, & Amerasinghe, 2005) or tea pickers or miners (Sastry & Bhat, 2019) are all considered to be high-risk. It is challenging to determine Indonesia's current complete prevalence rate of STH. Poor personal hygiene has a greater prevalence rate in children than in adults (Lee & Ryu, 2019).

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The infection of STH is one of the neglected tropical diseases—lack of maximum effort and monitoring by health workers to prevent the disease. The STH-infected persons usually do not realize that they have been infected. The infection can be diagnosed by finding eggs and adult worms in the feces (Juhairiyah, Annida, & Indriyati, 2015). The incidence of helminthiasis in the Province of East Nusa Tenggara (NTT) is at the third position in Indonesia with a percentage of 28% (2018 NTT Health Service) after Banten Province, Which was 60.7%, and Nanggroe Aceh Darussalam Province (NAD) which was 59.2% (Zulhifah, 2016).

An intestinal parasite infection is indicated by elevated total immunoglobulin E levels. Higher immunoglobulin E disease levels can be caused by recent or pre-existing parasite infections and are frequently linked to soil-transmitted helminth worm infections. An inflammatory reaction brought on by environmental allergens is the early result of intestinal worm infections. CD4+ T lymphocytes are primarily responsible for controlling the inflammatory response brought on by intestinal worms and helminth parasites. Immunoglobulin E levels in children or patients with atopic dermatitis or asthma are more than 40,109/L(Yolazenia et al., 2017).

2. METHODS

Location

The research as conducted in the Manusak Village, East Kupang Subdistrict, Kupang District, East Nusa Tenggara (NTT) Province. Kupang District is situated in Timor island of Indonesia (Figure 1).

Figure 1. Location of Lifuleo Village, Kupang District, East Nusa Tenggara Province is located in Timor Island and Lifuleo Villageis a part of Kupang District

Geographically, Kupang District has a landarea of 7,178.26 Km^2 and is the largest areain East Nusa Tenggara (ENT) Province.Kupang District covers 15.16 % of the total land area of ENT Province. Geographically, Kupang District is located at 123º16’10.66”-124º13’42.15” east longitude and 9º15’11.78”-10º.22 ‘14.25”south latitude.The territory is

bordered by the Savu Sea and Ombai Strait to the north, the Indian Ocean and the Timor Sea to the south, District of South Central Timor and the District Oecusi of the country of Timor Leste to the south, and Kupang City, RoteNdao District, Sabu Raijua District and Sawu Sea to the west (Pemerintah Daerah KabupatenKupang, 2013). Lifuleo Village is one of 10 villages in West Kupang District, Kupang Regency, East Nusa Tenggara Province. This village has an area of 6.8 km² with a population of 1191 people. Most (67.4 percent) of the people of Lifuleo Village earn their living as seaweed farmers, while others (32.6 percent) work as side fishermen.This village is a rural and slum area with low social and economic level, limited education level, limited hygienic criteria inthe availability of latrines, that resulting in the disposal of sewage in any place such as in the bushes around the place of residence (BPK RI Perwakilan Prop NTT, 2019).

Research design

The analytic observational design with thecross-sectional study was applied to this research. Samples collections were doneduring January-February 2024. Stool sampleswere collected using stool pots which were distributed on a day before sample collection,followed by an explanation on how to collect stool samples, and the subjects who agreeto give the sample were asked to sign the informed concern. The parents of the childrenare ethically represented to sign the informedconcerns. The sampling must meet inclusion criteria that the subjects who had signed the inform concern and gave their stool samples, the children who did not take the anthelminthicmedication within the last 6 months whenthe sampling was ongoing, and the samples which contained STH on microscopy examination. The exclusion criteria werethe children who did not give the samples during the data collection and who had taken anthelminthic medication within the last six months when the sampling was ongoing. Stool pots contained stool samples were then labeled with the identity of the subjects. The collected samples were then transported to theLaboratory of Parasitology, Study Program onMedical Technology, Institute of Polytechnicof Ministry of Health, Kupang District prior to microscopy examination and total Immunoglobulin E levels were measured using the ECLIA method.

3. RESULTS

Characteristics of subjects

Research subjects were 40 children, consisted of 27 females (67.5%) and 13 males (32.5%). Primary data on thecharacteristic of subjects were obtained by direct interviews using a questionnaire. Theresults were presented in Table 1.

Tabel 1. Prevalence of A. lumbricoides Infection Based on Age and Gender in Elementary School Age Children

Variable	Number of the sample (%)		
	Positive	**Negative**	**Total**
Age (years)			
6-12	18 (45)	22(55)	40(100)
Gender			
Female	12(44.5)	15(55.5)	27(100)
Male	6(46.2)	7(53.8)	13(100)

Results of Examination of Total Immunoglobulin E Levels

Normal levels of total IgE (IU/ml) are categorized based on age, namely for children aged 6-9 years is <90 IU/ml and aged 10-15 years is <200 IU. The number of children who were positively infected with A. lumbricoides had normal IgE levels, namely 5 children (27.8%), while those above normal were 13 children (72.2%). Children who were not infected with A. lumbricoides all had normal IgE levels and none were above normal as shown in Table 2 .

Tabel 2. Total IgE levels in elementary school age children in Manusak Village who were infected with A. Lumbricoides

Variable	Levels IgE Total (%)		
Ascaris lumbricoides	**Normal**	**Abnormal**	**Total**
Positive	5(27.8)	13(72.2)	18(100)
Negative	22	0	22(100)
Total	27	13	40

Discussion

The STH infection in children has garnered more focus than among farmers, evident in the deworming initiatives and the distribution of albendazole treatment to students in Indonesia (Sungkar, Ridwan, & Kusumowidagdo, 2017). According to the microscopy analysis results of 130 samples, only A. lumbricoides species (100%) were detected, affecting 50 children (38.4%) positively (Bria et al, 2021). The species A. lumbricoides is the most common STH found in South Asia and Southeast Asia, although notable geographic differences exist (Silver et al., 2018). The focused characteristics of A. lumbricoides were linked to pH and soil texture, which are factors that affect the distribution of STH infections. Additionally, A. lumbricoides has been reported to favor acidic soil. Clay and loam soils support the development of A. lumbricoides eggs better than sandy loam soil, which is more suitable for hookworm (Wardell et al., 2017). Such soil types were identified in Kupang District (Pemerintah Daerah Kabupaten Kupang, 2013).

The largest nematode found in the human digestive system is Ascaris lumbricoides. This nematode is found across the world, and the jejunum is usually where the adult worms live. The larvae undergo a pulmonary migration phase to mature after ingesting the embryonated eggs, which is how the infection is contracted (Khuroo, Zargar, & Mahajan,

1990). Using unprocessed sludge or biosolids from sewage treatment systems where the eggs accumulate, or unclean hands and food contaminated with A.lumbricoides-infected human feces, the infection is spread through the fecal-oral route (Asaolu & Ofoezie, 2018). The majority of intestinal nematode infections occur in rural areas with poor sanitation infrastructure and warm, humid tropical climates. Infection happens in cities as well. There may be tiny, isolated patches of high prevalence even within low prevalence areas. Infection is only absent from cold or hot, dry regions (Brooker, 2010). Approximately 818 million (771.7-891.6 million) persons are thought to have contracted A. lumbricoides in 2010 (Pullan et al, 2014).

An increase in total Immunoglobulin E levels in worm infections due to the Helper2 cell response has also been proven by many experts. Medeiros (2006) stated that worm infections transmitted through soil (geohelminth) resulted in increased production of polyclonal immunoglobulin E through the induction of interleukin-4 (IL-4) and interleukin-13 (IL-13). The polyclonal immunoglobulin E formed could reduce allergic reactions. in populations with a fairly high degree of parasite infection. The process by which polyclonal Immunoglobulin E suppresses allergic responses involves its binding to the Fel receptor on mast cells, which prevents allergen-specific Immunoglobulin E from attaching to these mast cells, thereby stopping degranulation. Another mechanism that may clarify the protective role of intestinal worm infections against allergies is the theory of IgG4 inhibition (blocking IgG4) and Modified Helper2. In helminths, TG4 antibodies are generated that can prevent the degranulation of effector cells, thus diminishing allergic responses. In the Modified Helper2 theory, Treg (regulatory T) cells play a role. These Treg cells will produce interleukin 10 (IL-10) and transforming growth factor (TGF-a). This cytokine reservoir can suppress immune responses and allergic inflammation (Rusjdi & Harminarti, 2011).

Molecular diagnostic methods possess high sensitivity and specificity, including multiplex PCR and real-time PCR (qPCR). Multiplex PCR allows for the identification of several parasite species in one reaction, streamlining diagnostics by substituting multiple individual tests with a single molecular test. Multiplex PCR displayed encouraging outcomes for A. lumbricoides, T. trichiura, and N. americanus (Phuphisut et al, 2014). The qPCR, in contrast to traditional PCR that merely shows the presence of infection, allows for the quantification of parasites and the intensity of the associated infection. qPCR exhibits greater sensitivity compared to both the Kato-Katz method and the flotation technique (FS7) for identifying A. lumbricoides infections and co-infections. Multiplex qPCR assays have effectively identified A. lumbricoides infections along with various intestinal parasites

(Basumi et al, 2012; Mejia et al, 2013).

Total immunoglobulin E is a form of antibody present in the bloodstream. Antibodies are proteins produced by the immune system to defend the body against dangers like bacteria or viruses. In the meantime, total Immunoglobulin E refers to the quantity of Immunoglobulin E antibodies present in the bloodstream. When foreign substances enter the body and attach to mast cells, they will be bound by Immunoglobulin E, leading to the release of histamine, which triggers an allergic reaction. Blood has antibodies produced by the immune system to defend the body against assaults from bacteria, viruses, and allergens. A Total Immunoglobulin E (IgE) test is a blood examination that quantifies the levels of immunoglobulin E antibodies. The standard value for overall IgE levels is <100 IU/ml. This kind of antibody is typically present in low amounts in the body. When the figure exceeds the norm, it may indicate that the body is excessively responding to the allergen. In other terms, measuring total Immunoglobulin E can detect an allergic response. The complete Immunoglobulin E test is commonly known as an allergy test, as it is frequently utilized to identify allergic responses (Permatasari, 2019).

The prevalence of ascariasis in Manusak Village was notably higher than in other regions of Indonesia, with 38.4% of 50 children testing positive in Manusak Village of West Kupang Subdistrict, Kupang District, whereas in Sorong District, Papua Province, it was only 3.4% (Yuwono, Husada, & Basuki, 2019), and 0.8% in Karang Asem District, Bali Province (Bayu, Pradinata, Sudarmaja, & Ariwati, 2019). Meanwhile, no cases of ascariasis were reported in Banjarmasin City, South Kalimantan Province (Juhairiyah et al., 2015). The occurrence of ascariasis in Manusak village was lower than in the Raja Basa District of Lampung Province, where the prevalence in children reached 88.2% (Wintoko, 2014).

The high incidence of ascariasis might relate to the anthelminthic medication program from the local health center, which was not implemented properly by the children in Manusak village. In Indonesia, according to Decree Minister of Health, No. 424 / Menkes/ SK / VI / 2006 regarding Guidelines for Worms Control mass treatment of intestinal worms, is if the prevalence of an area above 30% is carried out mass treatment two times a year. For a prevalence below 30%, treatment is carried out selectively for subjects who are positively found worm's eggs in their stools and carried out at the current health facility come for treatment. However, according to WHO 2006, for areas with high infection rates, the medication can be implemented every three times a year (Kementerian Kesehatan Republik Indonesia, 2006).

The direct details about the anthelmintic treatment obtained from the participants indicated that they were given albendazole in August, but none ingested the medication due to its large size making it difficult to swallow. A suggestion made to a community to crush the anthelminthic medication for easier swallowing could resolve this issue (Vitamine angels, 2020). Nonetheless, there is no available publication regarding the distribution of anthelmintics, alongside the application of albendazole and its resistance in Kupang District. Similarly, the Health Profile of Kupang District 2018 did not include any information about albendazole distribution. The distribution of tablets containing vitamin A and iron was explained clearly (Dinas Kesehatan Kupang, 2018). Infections caused by A. lumbricoides and malnutrition adversely affect the growth and development of the infected individual (Quihui-Cota et al, 2004).

Numerous research efforts have taken place concerning alterations in the immune reaction to parasites. Research by experts indicates that infections from intestinal worms lead to a shift in the Th2 balance favoring Th2 cells (The polarized). Polarization: Th2 in helminth infections parallels Th2 polarization in allergic diseases. The Th2 response is marked by the notable proliferation and differentiation of Th2 cells that secrete interleukin-4 (IL-4), interleukin-5 (IL-5), and interleukin-13 (IL-13). These interleukins promote eosinophil proliferation and stimulate B cells to generate IgE (Rusjdi & Harminarti, 2011).

The worm A. lumbricoides can adjust the immune system to favor T helper 2 (Th2) cells and can affect regulatory T cells (Treg) to secrete the cytokine interleukin 10 (IL-10). Consequently, it alters the equilibrium of the Th1/Th2 cell ratio. Due to the presence of IL-10, STH worms are able to persist in the host (Kind et al., 2007; Anthony et al., 2007; Everst et al., 2010; Maizels et al., 2003). Numerous studies have been carried out concerning alterations in the immune reaction to parasites. Expert research indicates that intestinal worm infections lead to a shift in the Th1/Th2 balance favoring Th2 cells (Th2-polarized). Th2 polarization during worm infections mirrors that of allergic diseases. This Th2 activity is marked by the prevalence of Th2 cell proliferation and differentiation, leading to the secretion of interleukin 4 (IL-4), interleukin-13 (IL-13), and interleukin-5 (IL-5). Interleukin 4 (IL-4) and interleukin 13 (IL-13) will promote eosinophil growth and stimulate B cells to generate IgE (Romagnani et al., 2006).

The immunological response to STH infection involves IgE, eosinophils, mast cells, and basophils. IgE activates mast cells via FceRI, triggering the release of mediators that affect gut mechanisms to expel and clear worms from the intestine. Efector cell recruitment like Th2, eosinophils, and macrophages is facilitated by cytokines, which then localize in the infected cell to eliminate the parasite (Cooper, 2009). IgE levels correspond with the quantity of worms

in the intestine, although the exact mechanism remains unclear. In chronic infection, B cells change class to produce IgG4, leading to a reduction in IgE levels. In acute infections, the eosinophil count is greater than that in chronic infections. Assessing IgE levels and the severity of infection allows us to analyze the impact of STH infection on IgE levels and their protective function in the immune system against helminth infections (Acevedo et al, 2011).

A response from the host to STH infection involves a rise in IgE levels, as well as an elevation in eosinophils, mast cells, and basophils (Bradley et al., 2004). Nonetheless, the rise in overall IgE levels during STH infection and its protective role against infection remains a phenomenon that lacks thorough explanation (Apsari et al., 2019). Multiple studies have indicated a connection between A. lumbricoides infection and allergic disorders (Takeuchi et al., 2008; Hunninghake et al., 2007). Elevated concentrations of specific IgE against A. lumbricoides were identified in 5-year-old children displaying bronchial asthma symptoms in rural Bangladesh (Takeuchi et al., 2008) and also in Venezuela (Hagel et al., 2007). This indicates that elevated anti-A. lumbricoides IgE levels correlate with a higher likelihood of asthma symptoms in 5-year-old children having a considerable count of A. lumbricoides worms (Takeuchi et al., 2008). This implies that individuals with asthma have elevated anti-A. lumbricoides IgE since the rise in IgE levels is driven by hypersensitivity to an A. lumbricoides infection. Worm infections are among the factors that elevate IgE levels, as IgE specific to worm infections is a crucial part of the immune defense against these parasites (Lynch et al., 1998). Nonetheless, elevated total IgE levels were observed in patients with STH infection compared to those without the infection. STH infection is linked to allergic conditions like asthma and eczema, but not to allergic rhinitis (Wijaya et al., 2014).

4. CONCLUSION AND LIMITATION

Conclusion

A significant prevalence of ascariasis cases in the examined region stemmed from the children's adherence to proper personal hygiene and sanitation practices. The educational attainment of their parents, which was linked to the infection, affected their children's habits. The detection of Ascaris lumbricoides eggs in soil and children's fecal samples confirmed the transmission pathway through those environments. By raising awareness regarding Ascaris lumbricoides infection, its transmission and prevention methods, along with emphasizing the necessity of using PEE while working in the field, followed by treating children infected with Ascaris lumbricoides, transmission will be reduced or halted. Infection with Ascaris lumbricoides is expected to lead to increased antibody production due to the buildup of antigens

that activate the host's immune response. Consequently, the increased level of antigen led to a greater quantity of IgE.

Limitation

The constraint of this research was that discovering a single species was crucial to determine if there was truly just one species or if other undetected species were present that the Kato Katz method missed. Given that the practice of going shoeless is linked to A. lumbricoides infection, hookworm infection may also be present in the community. The PCR technique could be employed alongside microscopy analysis to ensure that samples are preserved in the field, thereby maintaining the DNA of the parasite before additional laboratory processing.

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