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Morphological Profile Of Mycobacterium Tuberculosis Bacteria In Positive Acid-Fast Bacillus (Afb) Patients During 4 Months Of Treatment

SY DIDIK WIDIYANTO , TEGUH BUDIHARJO , KUNTJORO AP

Department of Health Analyst Poltekkes Kemenkes Semarang

Jl. Wolter Monginsidi Central Pedurungan Semarang

E-mail: sy.didik@yahoo.co.id

Abstract

Tuberculosis (TB) is a disease caused by the bacterium Mycobacterium tuberculosis. These bacteria are generally in the form of whole rods, but based on the experience encountered by several Health Analysts in microscopic examination of sputum, bacteria are often found that are not in general shape.Researchers are interested in knowing the differences in morphology of Mycobacterium tuberculosis bacteria microscopically in the sputum preparations of patients with positive smears every week for two months of treatment.This study used a quantitative type with an observational (non-experimental) research design. The research criteria are descriptive with the type of periodic series (time series). Subjects were sixteen TB patients with positive smears who were observed for bacterial morphology in sputum preparations every week, from suspect to 8th week after treatment by calculating the average size of bacteria and the percentage of bacterial damage.Bacterial size continued to decrease with increasing treatment time each week. Its size is reduced from 0.19 m – 0.49 m. Cell size decreased by an average of 46% with the smallest decrease at week 2 of 4% and the largest decrease at week six by 61%, before finally being undetectable again at week eight. every week from suspect to two months of treatment in the length range of 1.07 m – 1.40 m and the wide range of 0.35 m – 0.19 m. TB patients have experienced conversion or change from smear positive to smear negative after two months of treatment in the intensive stage. Bacterial cell damage from week to week, on average increased by 29% per week until finally negative at week 8 or two months after treatment.The conclusion from this study is that the size of the Mycobacterium tuberculosis bacteria showed changes with the time of intensive treatment with its size reduced from 0.19 m – 0.49 m. Cell size was reduced by an average of 46%. Bacterial cell damage from week to week, an average of 29% increase per week

Keywords: Morphology; Bacteria; AFB Positive

Introduction

On In the early 1900s the World Health Organization (WHO) and the International Union Against Tuberculosis and Lung Disease (IUATLD) developed a TB control strategy known as the Directly Observed Treatment Short-course (DOTS) strategy. The DOTS strategy consists of 5 components. Of the five DOTS components, one of the most important is case finding through quality-assured microscopic examination of sputum carried out by Health Analysts. Microscopic examination of sputum not only serves to find TB cases, but also serves to provide an assessment of the health progress of TB patients. Currently the role of the laboratory is no longer to support the diagnosis but is to determine the diagnosis of TB (Kemenkes RI, 2012). The cause of TB disease is the bacterium Mycobacterium tuberculosis. Mycobacterium tuberculosis is an acid-fast bacillus (AFB) bacterium. AFB is a trait that is not only possessed by the bacterium Mycobacterium tuberculosis, but AFB is a trait possessed by the genus Mycobacterium.

Morphology Mycobacterium tuberculosis bacteria are generally gram (+) rods, slightly bent, long or short stems (Soemarno, 2000). However, based on the experience encountered by several Health Analysts when conducting microscopic examinations, it is often found that AFB has a different shape than its normal shape. Under the microscope, AFB often has an incomplete rod shape such as a dotted or shortened line. According to Soemarno (2000), if there is a slight disturbance, the Mycobacterium tuberculosis bacteria will be cut off into short rods or cocci.

Those diagnosed with TB disease are then given Anti-TB Drugs according to the National TB control program. The drugs include Isoniazid, Rifampicin, Prazinamide, and Ethambutol. Isoniazid (INH) has the action of inhibiting the formation of mycolic acid, a component of the cell envelope (envelope) in mycobacteria. Rifampicin can cause inhibition of mycobacterial RNA synthesis. Pyrazinamide and Ethambutol are tuberculostatic against intracellular mycobacteria in macrophages (Nugroho, 2012). These drugs are the possible causes of damage to the Mycobacterium tuberculosis bacteria, because these drugs are a nuisance to the Mycobacterium tuberculosis bacteria. National TB Control Program, A good TB treatment is where two months after treatment there is no more smear positive on sputum smear microscopic examination. However, there are some who may be smear negative before two months, but also some who are more than two months smear positive are still found. Many factors influence it. So it is possible that within the estimated time of one month to four months there will be a change in the morphology of the Mycobacterium tuberculosis bacteria before it finally dies and becomes smear negative.

Based on the above background, researchers are interested in conducting research on the morphology of Mycobacterium tuberculosis bacteria microscopically in patients with smear positive for four months of treatment.

Method

Study carried out at the Semarang Health Laboratory Center and the Health Analyst Department of the Health Polytechnic of Semarang. Respondents and samples (sputum) of patients who were examined and found to be smear positive at the health service of the Semarang Health Center. Positive smear sputum during the period July to November 2020, then received intensive treatment by giving anti-TB drugs according to the National TB control program. The drugs include Isoniazid, Rifampicin, Prazinamide, and EthambutolIsoniazid (INH).

Respondent of18 people were those who had their sputum taken, were newly discovered patients on examination at the Semarang Health Center during the period July to November 2020. Microscopic observation of the morphology of Mycobacterium tuberculosis bacteria in the sputum preparations of suspected (initial examination) patients with positive smear and calculating the percentage of bacteria that were not damaged and the damaged one.

Observation microscopic morphology of Mycobacterium tuberculosis bacteria on sputum preparations of patients with positive smear for 2 weeks using ZeihlNeelsen staining, gram staining was observed using a Nikon 1000 multimedia microscope. After treatment and calculating the percentage ‘ bacteria were not damaged and changes in cell size, length, width and dimensions (length x width) was measured using a Hitachi 3000x electron microscope. Observing the morphology of Mycobacterium tuberculosis microscopically in the sputum preparations of patients with positive smear 4 weeks after treatment and calculating the percentage of undamaged and damaged bacteria, and so on for up to 10 weeks, carried out with a period of every 2 weeks. The morphology of the Mycobacterium tuberculosis bacteria that has been observed using a microscope is documented by saving the photo, and the percentage of the number of bacteria that are not damaged and those that are damaged every two weeks. The course of bacterial morphology is studied to change its shape from time to time and is discussed in more depth.

Results

Table 1. Negative smear test results

No	Week)	Number of people)	Percentage (%)
1	One	0	0
2	Two	0	0
3	Three	0	0
4	Four	2	11
5	Five	0	0
6	Six	9	50
7	Seven	0	0
8	Eight	7	39
Total		**18**	**100**

Data from the results of the examination showed that 2 subjects (11%) were smear negative in the fourth week and on average at the sixth week of examination, there were an additional 9 patients with 50% smear negative results. At the eighth week, all study subjects were declared smear negative (100%). The size is reduced from 0.19 m – 0.49 m. Cell size decreased by an average of 46% with the smallest decrease at week 2 of 4% and the largest decrease at week six by 61% before finally being undetectable in size again at week eight. The damage to Mycobacterium tuberculosis bacterial cells since week zero has occurred as much as 22.8% and continues to increase the level of cell damage. At the sixth week the level of cell damage reached 85% and at the eighth week it was 100% damaged.

Discussion

Data from the results of the examination showed that 2 subjects (11%) were smear negative at the fourth week and on average at week six the smear examination resulted in negative smears. In the eighth week, all research subjects were declared smear negative. These results provide empirical data that intensive, disciplined and regular treatment can produce relatively fast recovery, even faster than 2 months (eight weeks.). The Indonesian Ministry of Health, 2018 states that good TB treatment is where 2 months after treatment there is a conversion or change from positive smear to negative smear on microscopic examination of sputum preparations. So it is possible that within 2 months there will be a change in the morphology of the Mycobacterium tuberculosis bacteria before it finally dies and becomes smear negative.

Bacterial measurements were carried out by measuring the length and width of the Mycobacterium tuberculosis bacteria using a trinocular microscope expressed in m, while the calculation of the percentage of damage to the Mycobacterium tuberculosis bacteria was also carried out simultaneously with the measurement of Mycobacterium tuberculosis bacteria, so that the researchers used the same field of view to measure and to calculate the percentage of bacteria found damaged. The field of view used is 20 fields of view that are in the area of the longest horizontal line in the sputum preparation.

The data showed that the size of the bacteria continued to decrease in size with increasing treatment time each week. Its size is reduced from 0.19 m – 0.49 m. Cell size decreased by an average of 46% with the smallest decrease at week 2 of 4% and the largest decrease at week six by 61% before finally being undetectable in size again at week eight. Cell size every two weeks from suspect to two months of treatment in the length range 1.07 m – 1.40 m and width range 0.19 m – 0.35 m. TB patients have undergone conversion or change from smear positive to smear positive to negative after two months of intensive treatment.

In this study, the sputum specimens used were sputum in patients with smear positive, then taking the intensive stage of anti-TB drugs. The drug is a RHZE tablet consisting of Rifampicin (R), Isoniazid (H), Pyrazinamide (Z) and Ethambutol (E) to be taken for 2 months.

Mycobacterium tuberculosis bacteria when experiencing a slight disturbance will be cut off into short rods or cocci (Soemarno, 2000). Mycobacterium tuberculosis bacteria will also have difficulty surviving if the patient gets anti-tuberculosis drugs (Velayati, 2012).

Mycobacterium tuberculosis bacteria have an inner compartment consisting of peptidoglycan (PG), arabinogalactan (AG), and mycolic acid (MA) which together form covalent bonds. These bonding strands are the main agents that function to maintain the shape of the bacteria (Velayati, 2012). One of the anti-TB drugs consumed by patients is Isoniazid. Isoniazid has the action of inhibiting the formation of mycolic acid, a component of the cell envelope (envelope) in mycobacteria (Nugroho, 2012). As we already know that mycolic acid is one of the components in the main agent that functions to maintain the shape of bacteria. If the formation of mycolic acid is inhibited, the Mycobacterium tuberculosis bacteria will be easily damaged because there is nothing to maintain its shape.

The granules that appear in the Mycobacterium tuberculosis bacteria in the sputum preparations of patients with positive smear are also used as parameters in determining bacterial damage. As we have

seen in the description above, most TB drugs destroy the Mycobacterium tuberculosis bacteria by attacking the bacterial cell wall. If the cell wall is damaged, the cell wall will peel off and bacterial granules will appear. Visible bacterial granules indicate that the bacteria have damaged the cell wall and what can be seen with a light microscope are the bacterial granules.

Conclusions

Morphological profile of Mycobacterium tuberculosis bacteria microscopically decreased the size of bacterial cells continuously starting from week 2 measuring 0.79 m to week 6 (0.19 m). The increase in bacterial cell damage averaged 29% with the level of damage in the 0 to 2 week period of 53%. In the end, at the eighth week it was 100% damaged and negative AFB.Routine and disciplined and intensive treatment are the main capital to achieve healing for TB patients while maintaining the consumption of nutritious food, so that TB patients and their families should continue to be educated to always be disciplined at the intensive treatment stage.

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