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Sensitivity test results on blood cultures of suspected patients with typhoid fever and bacterial analysis of the similarity relations: A study on patients at several hospitals in the city of Jayapura

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ABSTRACT

Bacteremia-septicemia is a complication that causes typhoid fever bacteria in the blood. Based on fenetics numerical analysis using the Vitek 2 Compact and test sensitivity to 14 kinds of antibiotics, bacteria are bacteria obtained diversity of Gram-negative bacilli as much as 3 isolates, Escherecia coli, Pseodomonas aeruginosa, Pseudomonas maltophila and 4 isolates of gram-positive cocci such as Staphylococcus aureus, Staphylococcus epidermidis, Staphylococcus hominis and Staphylococcus saprophyticus. Dendogram structure of fenetics numerical analysis results, obtained 6 clusters with similarity between 56.6% -97.0% using reference strains of S. aureus subspecies aureus ATCC 25923 and P. aeruginosa ATCC 27853. Strain of fenetics numerical analysis based SSM (Simple Matching Coefficient) and UPGMA algorithm (Unweighted Pair Group Method with Arithmetic Average) obtained clusters on the structure dendogram of fenetics numerical analysis results, the first cluster consisted of 2 isolates of coagulase positive Staphylococcus (S. aureus strains subspecies aureus ATCC 25923 and isolate 049III15/Node 2) 97.0% similarity. Cluster II consists of three isolates of the genus Pseudomonas (P. aeruginosa ATCC 27853 strain, isolates and isolates 095III15 054II15/Node 3) similarity of 90.9% - 93.9%. Cluster III consists of 3 isolates of coagulase negative Staphylococcus that Node 3 and isolates similarity 114IV15 87.9% -93.9%. Cluster IV consists of five isolates of Staphylococcus genus, namely Node 2 and Node 5 with 71.7% similarity. Cluster V consists of four isolates of Node 4 and isolates of E. coli (Isolate 035IV15) 70.7% similarity. Claster VI consists of nodes Node 7 and node 6 consists of nine isolates with 56.6% similarity.

Keywords: sensitivity test, suspected typhoid fever, algorithms, similarity, and city of Jayapura.

INTRODUCTION

Determination of typhoid fever diagnosis is carried out through the stages of diagnosis of suspected typhoid fever and typhoid fever are supported by the results of clinical laboratory diagnosis such as diagnosis by serological methods of rapid microbiological diagnosis Widal test and blood cultures [1]. When Salmonella typhi (S. typhi) is found in blood specimens (or feces, urine and bone marrow) in cultured then the patient is definitely suffering from typhoid fever. Typhoid fever is a health problem worldwide [2-3]. Typhoid fever in Indonesia ranked 3rd in the top 10 diseases in all hospitals the number of patients died 274 people.

Bacteria that can be found in the blood is a gram-negative bacteria including *E. coli, Klebsiella spp, Enterobacter spp, S. typhi, Salmonella spp*. other than *S. typhi* and *P. aeruginosa* and gram-positive bacteria include *S. aureus, S. epidermidis, Streptococcus anhaemolyticus* and *Clostridium perfringens* [4]. Based on phenotypic characters found diversity of species of gram-negative bacteria and are divided into four clusters (*S. typhi* included in the first cluster) and gram-positive cocci bacteria are grouped into 6 clusters with the bacterial species highly variable [5].

This study using blood material on suspected cases or in cases of typhoid fever have proved the existence of the diversity of species of bacteria and antibiotic resistance patterns, through phenotypic characterization and testing sensitivity to antibiotics in many countries, including some of the provinces in Indonesia.

MATERIALS AND METHODS

RESEARCH METHODS

This study is an experimental research laboratory with cross sectional approach to determine the diversity of bacteria in blood cultures of suspected patients with typhoid fever based on fenetics numerical analysis and response to antibiotics in Jayapura as well as the analysis of patterns of similarity.

Confirmation testing and identification of bacteria

Colonies of bacteria was elected for rods, gram-negative enteric cultured on media MacConkey Agar (MC, Oxoid) incubated 37 °C for 24 h. Gram-positive cocci bacteria colonies tested coagulase and catalase. Identification of bacteria is a process of determining whether the bacterial strain studied identical to strains of bacteria that have been found previously. The process of identification of bacteria carried out by fenetics numerical analysis based on phenotypic characters include colony morphology, bacterial cell morphology, properties against gram dye, biochemical properties using Vitec 2 systems as well as test sensitivity to 14 antibiotics (suspension turbidity levels according to the standard Mac Farland 0,50- 0.60 using the densiter tool).

Data analysis

Data analysis using univariate analysis design to describe the characteristics of each of the study variables. The data that has been processed using PFE program analyzed using MVSP program (*Multi Variate Statistical Package*). To determine the relationship of similarity to determine the relationship of similarity between the strains and other strains of the bacterial diversity found in blood cultures suspected typhoid fever is based on numerical analysis fenetik used SSM (*Simple Matching Coefficients*). Clustering carried out using algorithms UPGMA (*unweighted pair group method with averages*) are presented in the form dendogram program *Paint Shop Pro* and edited with photoshop Adhobe program. Similarity matrix based SSM (Simple Matching Coefficient), Cluster based algorithm UPGMA (*Unweighted Pair Group Method with Arithmetic Average*) and dendogram. and narrated descriptively.

RESULTS AND DISCUSSION

Based on patient referral, the percentage of the highest obtained in the study came from a personal request or the request itself (APS) 54%, followed by the Bayangkara Hospitals 22%, General Hospital of Jayapura 19%, Doctor Practice 4% and General Hospital of Abepura 1 %. Meanwhile, according to age group, patients with suspected typhoid fever, highest 57.1% in the age group 31-45 years and 0-15 years as many as 42.9%. Distribution of age groups at intervals of 15 years based on patients with positive blood cultures.

The relationship of similarity and clustering.

Based on the analysis of SSM (Simple Matching Coefficient) and algorithms UPGMA (Unweighted Pair Group Method with Arithmetic Average) obtained by the relationship of similarity between the 7 isolates were found and consists of 6 clusters formed in the structure dendogram as shown in the figure below (Fig. 1).

Dendogram results indicate a relationship of similarity between isolates isolates were found with reference strains of *P. aeruginosa* ATCC 27853 for gram-negative isolates and reference strains of *S. aureus* ATCC 25923 for the subspecies aureus isolates were gram positive.

Cluster I consisted of 2 isolates of coagulase positive *Staphylococcus* (*S. aureus* strains subspecies aureus ATCC 25923 and isolate 049III15) with 97.0% similarity. Cluster II consists of three isolates of the genus *Pseudomonas* (*P. aeruginosa* ATCC 27853 strain, isolates and isolates 095III15 054II15) with similarity 90.9% - 93.9%. Cluster III consists of 3 isolates of coagulase negative *Staphylococcus* that Node 3 (034II15 isolates, isolates 005I15) and isolates similarity 114IV15 with 87.9% - 93.9%. Cluster IV consists of five isolates of *Staphylococcus* genus consisting of Node 2 (strain *S. aureus* ATCC 25923 and subspecies aureus isolates 049III15) and Node 5 (034II15 isolates, 005I15 isolates and isolates 114IV15) with 71.7% similarity. Cluster V consists of 4 isolates consisting of Node 4 (strains of *P. aeruginosa* ATCC 27853, 095III15 isolates and isolates 054II15) and isolates of *E. coli* (Isolate 035IV15) has 70.7% similarity. Cluster VI consists of node 7 and nodes 6 consists of nine isolates with 56.6% similarity.

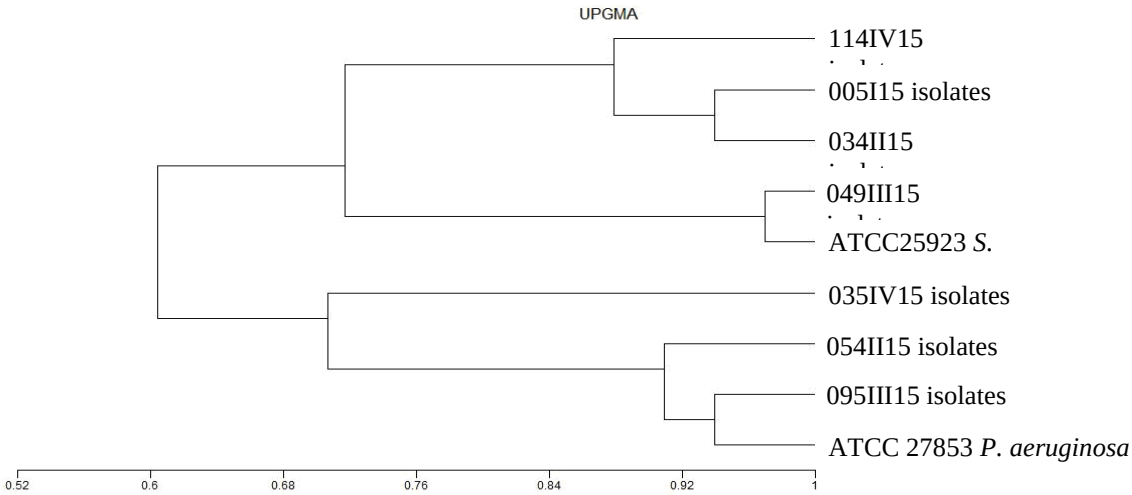

Fig 1. The relationship of similarity between the results of blood culture isolates of patients with suspected typhoid fever in Jayapura with reference strains of *P. aeruginosa* ATCC 27853 for gram-negative isolates and reference strains of *S. aureus* ATCC 25923 for the subspecies aureus isolates were gram positive

Table 1. The similarity of isolates blood culture results of suspected patients with typhoid fever in Jayapura city with reference strains based on the analysis of the Simple Matching Coefficient (SSM) and the algorithm UPGMA

No	Group 1	Group 2	Similarity (%)	Amount Group member
1	*Strain S. aureus subsp Aureus* ATCC 25923	049III15 isolates	97.0	2
2	*Strain P. aeruginosa* ATCC 27853	095III15 isolates	93.9	2
3	Isolat 034II15	005I15 isolates	93.9	2
4	Node 2	054II15 isolates	90.9	3
5	Node 3	114IV15 isolates	87.9	3
6	Node 1	Node 5	71.7	5
7	Node 4	035IV15 isolates	70.7	4
8	Node 7	Node 6	60.9	9

Sensitivity test results on blood cultures of suspected patients with typhoid fever in the city of Jayapura

Based on research results from blood cultures of suspected patients with typhoid fever in the city of Jayapura, obtained 14 kinds of antibiotics with varying responses against 7 isolates discovered. The sensitivity of each of these antibiotics can be seen in the table below (Table 2).

Table 2. Response isolates of gram-positive and gram-negative bacilli Coccus to antibiotics

No	Types of antibiotics	Antibiotic sensitivity	
		Sensitive (%)	Resistant (%)
	Gram-Positive and Gram-Negative		
1	*Trim-sulfamethoxazole*	42.9	57.1
2	*Ciprofloxacin*	71.4	28.6
3	*Amikacin*	50.0	50.0
4	*Ampicillin-sulbactam*	66.7	33.3
5	*Nitrofurantoin*	100.0	0.0
6	*Piperacillin-tazobactam*	60.0	40.0
7	*Gentamycin*	100.0	75.0
8	*Oxacillin*	75.0	25.0
9	*Cefepime*	50.0	50.0
10	*Meropenem*	75.0	25.0
	Options for gram negative		
11	*Ceftazidime*	33.3	66.7
	Options for gram positive		
12	*Clindamycin*	100.0	0.0
13	*Linezolit*	66.7	33.3
14	*Vancomycin*	66.7	33.3

All isolates (3 isolates of Gram-negative bacilli and 4 isolates of gram-positive cocci) found in this study indicates that the sensitivity varies sequentially from the highest response nitrofurantoin, gentamycin and clindamycin (especially isolates of gram positive) 100%; oxacillin and meropenem 75.0%; ciprofloxacin 71.4%; vancomycin and

linezolid (in particular gram-positive isolates), 66.7% ampicillin-sulbactam and piperacillin-Tazobactam 60%. The highest resistance to ceftazidime (particularly gram-negative isolates) 75% and trimethoprim-sulfamethoxazole 57.1%. Moderate sensitivity found in amicacin and cefepime is 50.0%.

Pseudomonas aeruginosa showed a response to 8 (8/14) type of antibiotic resistance results 100% to trimethoprim-sulfamethoxazole, ciprofloxacin, amikacin, ampicillin-sulbactam, piperacillin-Tazobactam, cefepime, meropenem and ceftazidime. There are 6 types of antibiotics used in this study does not provide the sensitivity results are nitrofurantoin, gentamycin, oxacillin, clindamycin, and vancomycin linezolit. Instead Pseudomonas matophilia respond 100% sensitive 10 (10/14) antibiotics are trimethoprim-sulfamethoxazole, ciprofloxacin, amikacin, ampicillin-sulbactam, nitrofurantoin, gentamycin, oxacillin, cefepime, meropenem and ceftazidime. There are 4 types of antibiotics used in this study does not provide results of sensitivity that piperacillin-Tazobactam, clindamycin, and vancomycin linezolit.

Escherichia coli provide a response that had a variation of 11 (11/14) antibiotics. Response sensitive to amikacin, nitrofurantoin, gentamycin, meropenem and resistant to trimethoprim-sulfamethoxazole, ciprofloxacin, ampicillin-sulbactam, piperacillin-Tazobactam, oxacillin, cefepime and ceftazidime. Staphylococcus aureus showed a 100% response sensitive to 12 (12/14) antibiotics are trimethoprim-sulfamethoxazole, ciprofloxacin, amikacin, ampicillin-sulbactam, nitrofurantoin, piperacillin-Tazobactam, gentamycin, cefepime, meropenem, clindamycin, and vancomycin linezolit.

Staphylococcus epidermidis and *Staphylococcus hominis* demonstrated varying response to the 10 (10/14) antibiotics. Sensitive response shown in ciprofloxacin, ampicillin-sulbactam, nitrofurantoin, piperacillin-Tazobactam, oxacillin and clindamycin. The response of resistance shown by trimethoprim-sulfamethoxazole and amikacin. Differences can be seen in the two isolates sensitive response to *Staphylococcus epidermidis* is linezolit and resistant to vancomycin. In contrast to the response *Staphylococcus hominis* sensitive to vancomycin and resistant to linezolit. *Staphylococcus saprophyticus* showed 100% response sensitive to 4 (10/14) antibiotics are trimethoprim-sulfamethoxazole, ciprofloxacin, nitrofurantoin, and gentamycin.

The results of similarity analysis based SSM and UPGMA algorithm, displayed in a dendrogram, obtained 6 clusters composed of 7 nodes. Clusters I, consisting of two isolates of coagulase positive *Staphylococcus* reference strains of *S. aureus* that subspecies aureus ATCC 25923 and isolate 049III15 with 97.0% similarity. In addition to phenotypic characterization colony morphology, cell morphology with gram staining, both isolates similarities can also be seen based on biochemical properties that produce the enzyme coagulase so-called coagulase positive *Staphylococcus*.

Coagulase enzymes could crumple plasma oxalate or citrate plasma reactive for coagulase factor in the serum so that the test results positive coagulase formed agglutination. Coagulase enzyme reacts with coagulase factor in serum produce an esterase to form fibrin clots to occur deposit on the surface of bacteria that can inhibit phagocytosis [6-11].

Both isolates produced catalase enzyme that breaks down hydrogen peroxide (H_2O_2) into water and oxygen (O_2) so that visible bubbles - air bubbles in the colony after added a solution of 3% H_2O_2. Catalase enzyme is an enzyme of the oxidase class that describes hydrogen peroxide (H_2O_2) into water and oxygen (O_2). This enzyme include non-toxic metabolites that are owned by *Staphylococcus* catalase test is used to differentiate *Staphylococcus* (catalase positive) with *Streptococcus* (catalase negative).

Similarities cluster II is based on the ability to break down carbohydrates to produce acid isolates in aerobic atmosphere by fermenting glucose, maltose, sucrose, mannitol, trehalose, but not ferment xylose. Strains of S. aureus reference difference subspecies aureus ATCC 25923 with 049III15 isolates can be seen in the results of biochemical tests of VITEC 2 systems which isolates 049III15 found not ferment lactose and do not produce the enzyme urease. According to the results of previous studies, some strains of *S. aureus* do not produce acid from lactose such as *Staphylococcus aureus* subspecies *Anaerobius* [11].

Urease activity was discovered more than 200 species of gram-positive and gram-negative, but the isolates 049III15 not found urease activity and is unable to break down urea to form ammonia molecules.

Staphylococcus aureus; Gram-positive, does not move, not the spores, could produce a capsule, measuring 0.5 - 1.0 mm, shaped cocci in pairs and in groups, could also be seen single. Colony morphology diameter >5 mm, smooth, convex, shiny, transparent. Pigmentation varies gray to yellow orange, facultative anaerobes, but can grow in aerobic conditions. Grows well in medium containing 10% NaCl, less fertile in 15% NaCl. The growth temperature of about 15-45 °C; optimum 30-37 °C. Thecanoat acid containing containing ribitol and N-acetylglucosamine.

Produces acid in aerobic atmosphere by fermenting fructosa, galactosa, glucose, lactose, maltose, mannitol, mannose, ribose, sucrose, trehalose, and does not ferment adonitol, arabitol, arabinose, cellobiose, dextrin, dulcitol, sorbitol, xylitol and xylose. Coagulase positive reaction, catalase, urease, hemolysis and negative reaction in the oxidase, β-galactosidase, β-glucuronidase.

Staphylococcus aureus is a bacterium that is pathogenic because this bacterium is able to coagulate plasma, capable of hemolysis blood, produces a variety of extracellular enzymes and toxins, that is what distinguishes *Staphylococcus aureus* and species Staphylococcus others, can lead to infection, food poisoning and toxic shock syndrome [11].

Cluster II consists of three isolates of the genus *Pseudomonas* (*P. aeruginosa* ATCC 27853 strain, 095III15 isolates and isolates 054II15) with similarity 90.9% - 93.9%. Similarity of strain ATCC 27853 and *P. aeruginosa* isolates 095III15 93.9%. In this study is based on in addition to the morphological characterization of the colony and cell morphology (Gram negative rods form red), obtained by biochemical tests of 66 characters to have similar results in the ability to produce enziim oxidase, urease and catalase, ability to break down carbohydrates to produce acid isolates without any gas in an aerobic atmosphere by fermenting glucose, maltose, mannitol and acid formed in trehalose and lactose (nonfermented lactose). The difference is based on the ability of isolates 095III15 ferment mannose, sucrose and xylose whereas *P. aeruginosa* ATCC 27853 strain is not capable of fermenting mannose, sucrose and xylose.

Note 2 and isolates 054II15 microscopically including gram-negative rod-shaped bacterium slightly curved, have flagella, no spores, not encapsulated, gray white colonies were grown at a temperature of 42 °C, do not ferment lactose and decarbokxylase ornithine negative reaction. Node 2 and isolates 054II15 distinguished by the presence of the enzyme oxidase which isolates 054II15 not have the enzyme is different from other *Pseudomonas* isolates. Additionally isolates 054II15 able to ferment lactose.

Pseudomonas maltophilia (*Xanthomonas maltophilia* or *Stenotrophomonas maltophilia*); Gram negative, moving (single polar flagellum), no spores, does not have a capsule, measuring 0.5-1.0 mm, slightly curved rod-shaped straight, mostly in pairs or single, short-chain sometimes. Colonies smooth, shiny, pale white pigment - could turn yellow and brown, obligate aerobic, optimum growth temperature of 35 °C. Produces acid in an aerobic atmosphere in lactose, glucose, maltose, trehalose. The positive reaction on catalase and oxidase negative reaction and urease. *P. maltophilia* has been isolated from a number of water sources including rivers, wells, lakes hypereutrofics, bottled water and sewage. Iizuka and Komagata reported the isolation of S. maltophilia from the brine, oil and other related materials from oil fields in Japan.

P. aeruginosa is found in soil, water, ± 10% of the normal flora of the colon (large intestine), the skin and the outer ear, and *P. maltophilia* found in soil and plants but can infect humans who have immune system is decreased. Infects through food, beverages, water, contaminated hands, handling and equipment - equipment that is not sterile in the hospital, the air flow (Nosocomial), Moreover, animals could also pass (flies, mosquitoes, and others) that have been contaminated. Enzymes-*Psedomonas* extracellular enzymes such as elastase and proteases have the effect histotoxic and facilitate the invasion of the organism into the bloodstream.

Cluster III consists of 3 isolates of coagulase negative *Staphylococcus* that Node 3 (005I15 isolates, 034II15 isolates) and isolates similarity 114IV15 with 87.9% - 93.9%. Similarities owned cluster III is the inability to produce the enzyme coagulase isolates so-called coagulase negative Staphylococcus. Third isolates produce enzymes catalase, producing urease enzyme capable of breaking down urea to form two molecules of ammonia, has the ability to break down carbohydrates to produce acid in aerobic atmosphere by fermenting lactose, maltose, sucrose, trehalose, but not ferment xylose.

Isolates 034II15 and isolates 005I15 (Node 3) have similarities 93.9% can be seen by 66 characters biochemicals of them in addition to producing the enzyme catalase, urease and ferment lactose, maltose, sucrose, trehalose, both isolates have the ability to break down carbohydrates to produce acid by fermenting galactose, but not ferment mannose and coagulase reaction and oxidase negative, the typical difference between the two isolates in this study where isolates 034II15 besides not ferment mannose also do not ferment mannitol and obtained a negative reaction on opthochin novobiocin resistant and resistant. Instead Isolates 005I15 ferment mannitol, a positive reaction on opthochin resistant and novobiocin resistant.

Staphylococcus epidermidis; Gram-positive, not moving, not the spores, does not move, measuring 0.8 - 1.0 mm, shaped cocci in pairs in groups, can also be seen single. Colony diameter of 2.5 - 4.0 mm, smooth, convex, lustrous, usually not pigmented or pigment - gray or grayish white, may be slimy and facultative anaerobes, but can grow in

aerobic conditions. The growth temperature of about 15-45 ° C; optimum 30-37 °C. Teikhoat acid containing glycerol, N-acetylglucosamine, and glucose. β-Galactose and lactose are metabolized through the D-Tagatose-6-phosphate. Produces acid in an aerobic atmosphere with fermenting fructose, glucose, maltose, mannose, sucrose and do not ferment mannitol, trehalose, xylitol or xylose. The positive reaction on catalase and urease reaction and oxidase enzymes coagulase negative.

Staphylococcus epidermidis (*Staphylococcus albus*), one species of coagulase negative the normal flora of the skin and mucous membranes of humans, mouth, respiratory tract, gastrointestinal tract, urinary tract, and in the air. *S. epidermidis* can be found in humans from the age of neonates, but these bacteria can also cause infection if it is in an unfamiliar location, in large quantities, and if there are factors - predisposing factors.

Staphylococcus hominis; Gram-positive, does not move, not berspora, measuring 1.0 - 1.5 mm, shaped cocci (*tetracoccus*) small - small, can also be seen single. Colony diameter of 3.0 - 5.0 mm, smooth, convex, opaque, usually not pigmented or pigment yellow - orange, aerobic or facultative anaerobes, but thrives in anaerobic conditions. The growth temperature of about 20-45 °C; optimum 30-40 °C, containing glycerol teikhoat acid and N-acetylglucosamine. Acid production of N-acetylglucosamine, galactose, lactose, mannitol, trehalose. Produces acid in an aerobic atmosphere with fermenting fructose, glucose, maltose, sucrose and do not ferment mannitol, mannose, sorbitol, and xylitol or xylose, a positive reaction on catalase and urease and coagulase and negative oxidase.

Staphylococcus hominis was coagulase negative species that are commensal on human skin and animals, so it is not dangerous but can infect humans are immune compromised and as a cause of nosocomial infection in infants neonatal age. Similarity node 3 and isolates 114IV15 for 87.9% whereas in biochemical tests using Vitec 2 systems similarities with the results obtained positive reactions in opthochin resistant and novobiocin resistant and ferment mannitol. Differences isolates node 3 and isolates 114IV15 based on its ability to break down carbohydrates to produce acid by fermenting mannose, but not ferment galactose.

Staphylococcus saprophyticus; Gram-positive, does not move, not the spores, measuring 0.6 - 1.2 mm. shaped cocci in pairs in groups, can also be seen single. Morfoloogi colony diameter of 4.0 - 9.0 mm, smooth, convex, shiny, opaque, usually not pigmented or pale yellow pigment, facultative anaerobes, but thrives on aerobic conditions. The growth temperature of about 10-40 °C; optimum 28-35 °C, acid-containing ribitol teikhoat, slightly glycerol, and N-acetylglucosamine. Produces acid in an aerobic atmosphere with fermenting fructose, glucose, maltose, trehalose and do not ferment galactose, mannose, ribose, sorbitol, xylitol or xylose, a positive reaction on catalase and urease, coagulase negative reaction and oxidase.

Staphylococcus saprophyticus including coagulase negative *Staphylococcus* species, are commensal on the skin but can cause urinary tract infections, especially young women, and is the cause cystitis other than *Escherichia coli*. A variety of infections caused by coagulase negative Staphylococcus among other nosocomial infections, urinary tract infections, infections due planted tools (such as catheters, infusion, prosthetic valves), osteomyelitis and bacteremia in someone with impaired immune.

Cluster IV consists of five isolates of Staphylococcus sp genus consisting of node 2 (strain *S. aureus* ATCC 25923 and subspecies *aureus* isolates 049III15) and node 5 (034II15 isolates, 005I15 isolates and 114IV15 isolates) with 71.7% similarity. Morphologies colony to-5 isolates genus Staphylococcus sp found in this study was a colony of round, smooth, convex, cell morphology gram-positive, purple, shaped cocci clustered but differ in their ability to lyse blood (hemolytic or anhemolitik), pigment varying (not pigmented, pigment white or gray to orange or golden yellow), there are capsulated strain but generally not encapsulated. Results of biochemical tests on vitec 2 systems using the Vitek 2 Compact referring to Bergey's Manual of Systematic Bacteriology, obtained similarity with probability 92-99% using a strain of reference *S. aureu*s subspecies aureus ATCC 25923 and the main difference is the ability isolates produce enzymes coagulase which isolates node 2 produces enzymes coagulase and isolates node 5 does not produce the enzyme coagulase.

*Staphylococcu*s virulence depends on the ability to form the enzyme coagulase. Coagulase positive *staphylococci* produce a variety of extracellular enzymes and toxins (such as bacteriocins), cause hemolysis, invasive making it easier for bacteria to enter the blood vessels. Coagulase negative *Staphylococcus* otherwise cause hemolysis and are not invasive but by infections that cause local tissue necrosis and abscess formation if fibrin wall around the abscess broken then the bacteria can spread into the blood to occur bacteremia.

Cluster V consists of 4 isolates consisting of node 4 consists of 3 isolates (strains of *P. aeruginosa* ATCC 27853, 095III15 isolates and 054II15 isolates) and isolates of *E. coli* (isolate 035IV15) has a 70.7% similarity. Similarities properties assessed on the basis of similarity index >70%, it still has a 70.7% similarity based on the similarity of

cell morphology are gram-negative rod-shaped bacteria, not berspora, not encapsulated, moving on to the flagellum. But differ in the ability ferment lactose, node 4 cluster does not ferment lactose and Isolate 035IV15 have the ability to break down carbohydrates to produce acid and gas by fermenting lactose.

Isolate colony morphology 035IV15 smooth rounded, convex, glistening red, straight, short, produce the enzyme catalase, do not produce the enzyme oxidase and urease, have the ability to ferment glucose and produce acid gases, ferment almost all carbs except one of which is xylose. *E. coli* are classified by characteristic virulence properties, and each group to cause disease via a different mechanism. Enteropathogenic *E. coli* (EPEC) is attached to the small intestine mucosal cells causing diarrhea; Colonization factor of enterotoxigenic *E. coli* (ETEC) that are specific to the human cause ETEC adhesion to the epithelial cells of the small intestine; Enteroinvasive *E. coli* (EIEC) cause disease through invasion of epithelial cells of the intestinal mucosa. Outside intestinal E. coli can enter the bloodstream and cause sepsis. *Escherichia coli* is opportunistic bacteria commonly found in the large intestine (colon) as normal flora, but can cause infection in the intestines and other body tissues outside the intestine.

Cluster VI consists of node 7 and node 6 consists of nine isolates with 56.6% similarity. Similarities properties assessed on the basis of similarity index > 70%, then 56.6% similarity no significant similarity. Node 7 are included in the phylum *Firmicutes*, Class *bacilli*, Order Bacillales, Family *Staphylococcaceae*, Genus *Staphylococcus* that is shaped bacteria cocci, clustered, *tetracoccus* or sometimes visible single, gram-positive, purple, and node 6 is Phylum Proteobacteria, Class *Gammaproteobacteria* consisting of 3 order that Order *Xanthomonadales*, Family *Xanthomonadaceae*, genus *Stenotrophomonas maltophilia* (also called *Pseudomonas maltophilia* were rechristened *Xanthomonas maltophilia* and now known *Stenotrophomonas maltophilia*), Order *Pseudomonadales*, Family *Pseudomonadaceae*, genus *Pseudomonas aeruginosa* and Order Enterobacteriales, Family *Enterobacteriaceae*, genus *Escherichia coli* type isolates shaped trunks, sometimes seen single, gram-negative red [12-19].

Under normal circumstances, blood and tissue are sterile. However, the abnormal state of the commensal flora can infect and cause bacteremia. Complications of bacteremia-a major cause of septicemia often the discovery of bacteria in blood culture is gram-negative bacteria including *E. coli* and *P. aeruginosa* and gram-positive bacteria *S. aureus* and *S. epidermidis* [20-25].

CONCLUSION

Fenetics numerical analysis based SSM (*Simple Matching Coefficient*) and algorithms UPGMA (*Unweighted Pair Group Method with Arithmetic Average*) obtained clusters on the structure dendogram fenetics numerical analysis results, the first cluster consisted of 2 isolates of coagulase positive *Staphylococcus* (*S. aureus* strains subspecies aureus ATCC 25923 and isolates 049III15/Node 2) 97.0% similarity. Cluster II consists of three isolates of the genus *Pseudomonas* (*P. aeruginosa* ATCC 27853 strain, 095III15 isolates and 054II15 isolates/Node 3) similarity of 90.9% - 93.9%. Cluster III consists of 3 isolates of coagulase negative *Staphylococcus* that Node 3 and isolates similarity 114IV15 87.9% -93.9%. Cluster IV consists of five isolates of *Staphylococcus* genus, namely Node 2 and Node 5 with 71.7% similarity. Cluster V consists of four isolates of Node 4 and isolates of *E. coli* (Isolate 035IV15) 70.7% similarity. Claster VI consists of nodes Node 7 and 6 consists of nine isolates with 56.6% similarity.

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