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Identification of *Escherichia coli* in Patients with Diabetes Mellitus Suspected of Urinary Tract Infection Complications at the Imbanagara Health Center, Ciamis Regency

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ABSTRACT

Background & Objectives: Diabetes Mellitus (DM) is a metabolic condition characterized by hyperglycemia due to irregular insulin production and action. Increased levels of glucose in the blood cause people with diabetes mellitus to experience glycosuria and neutrophil dysfunction, causing the risk of being susceptible to infectious diseases. One disease that can cause complications is urinary tract infection. This study aims to determine the presence or absence of *Escherichia coli* bacteria in the urinary tract of patients with diabetes mellitus suspected of complications of urinary tract infection. **Methods:** The method used in this study uses a descriptive method, namely the researcher only identifies *Escherichia coli* in patients with diabetes mellitus suspected of complications of urinary tract infection at the Imbanagara Health Center, Ciamis Regency **Results:** Based on the results of the research that has been done, it can be concluded that of the 12 samples examined, 5 samples were indicated positive for *Escherichia coli* bacteria, while 7 other samples found *Enterobacter* bacteria. **Conclusion:** Based on the results of the research that has been carried out, it can be concluded that of the 12 samples examined, 5 samples indicated positive for *Escherichia coli* bacteria, while 7 other samples found *Enterobacter* bacteria.

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Introduction

Diabetes Mellitus (DM) is a metabolic condition characterized by hyperglycemia due to irregular insulin production and action. Diabetes mellitus is not contagious, although it is a major public health problem in many developing countries including Indonesia (Nur, Mardhia, and Mahyarudin 2022).

According to the International Diabetes Federation (IDF), there are 537 million cases worldwide in 2021 who suffer from diabetes mellitus (Euis Inayah 2022). Based on Riskesdas data in 2018, the national prevalence of diabetes mellitus reached 10.09% or around 20.4 million cases in Indonesia (Indonesian Ministry of Health 2020). Based on data obtained from the Medical Records of the Imbanagara Ciamis Health Center, there were 119 patients with diabetes from January to October 2023 who were outpatients (Health Office 2020).

Decreased immune dysfunction and increased blood glucose levels cause people with diabetes mellitus to experience glycosuria, increasing the number of bacteria in normal urine. High glucose levels can also cause damage to neutrophils so that they are at risk of being susceptible to infection with diseases including urinary tract infections (Geerlings et al. 2016).

Chronic hyperglycemia in diabetes mellitus is associated with abnormalities in several parts of the body including the eyes, nerves, heart, skin, kidneys and blood vessels. In addition to these organs, the genitourinary tract that experiences inflammation is at risk of urinary tract infection. The genitourinary tract consists of the urinary system and reproductive system in both men and women (Ni Made Susilawati, Marni Tangkelangi, and Dorotia Masi Daen 2022). Sepsis, vesicourethal reflux (RVU), urinary tract obstruction, or the use of a new urethral device all contribute to an increase in urinary tract infections. Women are at higher risk of suffering from urinary tract infections than men (Sugireng, Syarif, and Amelia 2022). Urinary tract infection is a condition caused by microbes such as *Escherichia coli* bacteria that attack the urinary system.

Escherichia coli bacteria are gram-negative rod-shaped bacteria that can survive as normal intestinal flora in the human body around the anus, perineal skin, vaginal introitus and penile prepuce. *Escherichia coli* bacteria can be pathogenic to the human body in the form of the onset of various infections such as urinary tract infections caused by increased bacterial colonization and inflammation in the urinary tract causing infection (Sri Rahayu 2023). Efforts to prevent infections caused by *Escherichia coli* bacteria by treatment as early as possible require diagnosis by performing bacterial culture to determine the morphological characteristics of the bacterial colonies that cause the infection (Hutasoit 2020).

Bacterial culture is the process of breeding bacteria in the laboratory using media containing nutrients as a carbon source and culture consists of water, energy sources, nitrogen, sulfur, phosphate, hydrogen, and other components. Amino acids, vitamins, or nutritional nucleotides are examples of growth factors that can be added to the basic media material. Types of bacterial cultures include EMB, MCA, Sugar-Sugar Test and Biochemical Test (Intan Camilia Febyayuningrum, Rudina Azimata Rosyidah, and Resmi Aini 2021).

Objective

This study aims to determine the presence or absence of *Escherichia coli* bacteria in the urinary tract of patients with diabetes mellitus suspected of complications of urinary tract infection.

Method

The method used in this study uses descriptive methods, namely researchers only identify *Escherichia coli* in patients with diabetes mellitus suspected of

complications of Urinary Tract Infection at the Imbanagara Health Center, Ciamis Regency with sampling techniques using incidental sampling techniques conducted from January to June 2024. Respondents in this study amounted to 12 people with diabetes mellitus suspected of complications of urinary tract infection at the Imbanagara Health Center, Ciamis Regency. Sampling was carried out at the Imbanagara Health Center and the examination was carried out at the STIKes Muhammadiyah Ciamis Bacteriology Laboratory. The measurement methods used were gram staining, culture on media, sugar test, and biochemical test. The instruments used were microscope, TSB media, MCA media, EMB, sucrose, lactose, glucose, TSIA, SIM, SC, MR, and VP. The research procedure consists of Pre analytical, analytical, and Post analytical. The pre-analytic stage consists of: Use of PPE, Sterilization of tools, Preparation of media, Recording patient identity and sampling. The analytic stage consists of: TSB media cultivation, gram staining, MCA media cultivation, MCA media cultivation, sugar test, and biochemical test. Post analytical stage consists of: After practicing the work table is cleaned using disinfectants to avoid viruses, bacteria and others, Observation of bacterial growth on TSB media, microscopic gram staining, EMB media, MCA media, sugar test, and biochemical tests.

Results

Media quality tests are carried out before examination of the sample, the finished media is treated the same as the sample to be tested. The media to be planted is declared suitable for use if there are no colonies growing on each media.

First Day

Samples were planted on fertilizer media, namely TSB media, from 12 samples planted, it was found that turbidity occurred in each media. The results of planting TSB media samples are shown in table 1.

TABLE 1. Gram stain observation results

Sample code	Gram staining result			
	Color	Shape	Arrangement	Properties
Control	-	-	-	-
A01	Red	Bacilli	Monobacilli	Gram Negative
A02	Red	Bacilli	Monobacilli	Gram Negative
A03	Red	Bacilli	Monobacilli	Gram Negative
A04	Red	Bacilli	Monobacilli	Gram Negative
A05	Red	Bacilli	Monobacilli	Gram Negative
A06	Red	Bacilli	Monobacilli	Gram Negative
A07	Red	Bacilli	Monobacilli	Gram Negative
A08	Red	Bacilli	Monobacilli	Gram Negative
A09	Red	Bacilli	Monobacilli	Gram Negative
A10	Red	Bacilli	Monobacilli	Gram Negative
A11	Red	Bacilli	Monobacilli	Gram Negative
A12	Red	Bacilli	Monobacilli	Gram Negative

Second Day

Turbid samples were planted on selective media, namely MCA and EMB media and then incubated at 37 °C for 24 hours. Observe the growth of colonies on the media and note in table 2 and table 3.

TABLE 2 Macroscopic Observation Results on MCA Media

Sample Code	Color	Shape	Elevation	Properties
Control	-	-	-	-
A01	Back	Round	Convex	Non Lactose Fermenter
A02	Back	Round	Convex	Non Lactose Fermenter
A03	Back	Round	Convex	Non Lactose Fermenter
A04	Back	Round	Convex	Non Lactose Fermenter
A05	Back	Round	Convex	Non Lactose Fermenter
A06	Light red	Round	Convex	Lactose Fermenter
A07	Back	Round	Convex	Non Lactose Fermenter
A08	Light red	Round	Convex	Lactose Fermenter
A09	Light red	Round	Convex	Lactose Fermenter
A10	Light red	Round	Convex	Lactose Fermenter
A11	Light red	Round	Convex	Lactose Fermenter
A12	Back	Round	Convex	Non Lactose Fermenter

TABLE 3. Macroscopic Observation Results on EMB Media

Sample Code	Color	Shape	Elevation	Properties
Control	-	-	-	-
A01	No color	Round	Convex	Non Lactose Fermenter
A02	No color	Round	Convex	Non Lactose Fermenter
A03	No color	Round	Convex	Non Lactose Fermenter
A04	No color	Round	Convex	Non Lactose Fermenter
A05	No color	Round	Convex	Non Lactose Fermenter
A06	Green Metallic	Round	Convex	Lactose Fermenter
A07	No color	Round	Convex	Non Lactose Fermenter
A08	Green Metallic	Round	Convex	Lactose Fermenter
A09	Green Metallic	Round	Convex	Lactose Fermenter
A10	Green Metallic	Round	Convex	Lactose Fermenter
A11	Green Metallic	Round	Convex	Lactose Fermenter
A12	No color	Round	Convex	Non Lactose Fermenter

Day Three

Samples that have grown on MCA and EMB media are then confirmed with sugar tests such as glucose, sucrose and lactose. The test results can be seen in table 4.4 below.

TABLE 4. Observation Results on the Sugar-Sugar Test

Sample Code	Glucose	Sucrose	Lactose
A01	(-)	(-)	(-)
A02	(-)	(-)	(-)
A03	(-)	(-)	(-)
A04	(-)	(-)	(-)
A05	(-)	(-)	(-)
A06	(+)	(+)	(+)
A07	(-)	(-)	(-)
A08	(+)	(+)	(+)
A09	(+)	(+)	(+)
A10	(+)	(+)	(+)
A11	(+)	(+)	(+)

A12	(-)	(-)	(-)

Description: Positive (+): Color change from purple to yellow
Negative (-): No change in color remains purple

For biochemical testing such as TSIA, SIM, Simon citrate and MR-VP tests. The test results can be shown in table 5 below.

TABLE 5. Observation Results on Biochemical Tests

Sample Code	TSIA			SIM	Simon Citrat	MR	VP
	Slope/Base	H2S	Gas				
A01	Y/R	(-)	(-)	Sulfur (-) Indol (-) Motility (-)	(+)	(-)	(+)
A02	Y/R	(-)	(-)	Sulfur (-) Indol (-) Motility (-)	(+)	(-)	(+)
A03	Y/R	(-)	(-)	Sulfur (-) Indol (-) Motility (-)	(+)	(-)	(+)
A04	Y/R	(-)	(-)	Sulfur (-) Indol (-) Motility (-)	(+)	(-)	(+)
A05	Y/R	(-)	(-)	Sulfur (-) Indol (-) Motility (-)	(+)	(-)	(+)
A06	Y/Y	(-)	(+)	Sulfur (-) Indol (+) Motility (+)	(-)	(+)	(-)
A07	Y/R	(-)	(-)	Sulfur (-) Indol (-) Motility (-)	(+)	(-)	(+)
A08	Y/Y	(-)	(+)	Sulfur (-) Indol (+) Motility (+)	(-)	(+)	(-)
A09	Y/Y	(-)	(+)	Sulfur (-) Indol (-) Motility (+)	(-)	(+)	(-)
A10	Y/Y	(-)	(+)	Sulfur (-) Indol (+) Motility (+)	(-)	(+)	(-)
A11	Y/Y	(-)	(+)	Sulfur (-) Indol (+) Motility (+)	(-)	(+)	(-)
A12	Y/R	(-)	(+)	Sulfur (-) Indol (-) Motility (-)	(+)	(-)	(+)

Description:
TSIA: Y/Y yellow slope yellow base
H2S (-) no black color is formed on the media

Gas (+) there is gas in the media
Gas (-) there is no gas in the media
SIM: Positive (+) indole, formed red color ring
Negative (-) indole, no red ring formed
Negative (-) sulfur, does not produce a black hue on the media
Negative (-) motility, lack of movement, white hue at the puncture point
Positive (+) motility, there is white movement near the puncture site
SC: Positive (+) media changes to blue color
Negative (-) media remains green
MR: Positive (+) red colored solution
Negative (-) the solution remains white
VP: Positive (+) red colored solution
Negative (-) yellow colored solution

Discussion

In table 1 the samples were grown on TSB media as a fertilizer medium. Of the 12 samples grown on TSB media, the turbidity caused by light deflection due to the presence of bacteria. Samples that are positive for turbidity in TSB media are gram stained to determine the shape, composition, and nature of the bacteria. Bacteria were found in 12 samples with red color, bacillus shape, and gram-negative. The most dominating positive result is *Escherichia coli* with bacillus shape, monobacillus arrangement and red color.

In table 2 on MCA Media from the observation of 5 samples found pink colonies, round spots, smooth surface. In 7 samples found black colonies, round spots, smooth surface. MCA media is used as a growth medium for various bacteria and contains crystal violet and bile salts which have the ability to stop the development of gram-positive bacteria. Selective media aims to identify gram-negative bacteria because not all bacteria can grow well on it. On MCA media, bacteria that are unable to ferment lactose will develop into black colonies, while bacteria that can ferment lactose will multiply into acids such as *Escherichia coli*.

In table 3 on EMB media from the observation of 5 samples found metallic green colonies, round spots and smooth surfaces. EMB media is used for the growth of gram-negative bacteria and is more specific for *Escherichia coli* bacteria because EMB is a selective media. In addition to *Escherichia coli* bacteria that can grow on EMB media, other gram-negative bacteria can also grow such as coliform. Bacteria that can ferment strong lactose and produce acid on EMB media will grow metallic green colonies on EMB media such as *Escherichia coli* bacteria. There is no colony growth on EMB media because bacteria cannot ferment lactose, namely other bacteria.

In table 4 in the sugar test from the observation of 5 samples on glucose, sucrose, and lactose positively changed color from purple to yellow. In 7 samples on glucose, sucrose, and negative lactose remained purple. To determine microorganisms that can ferment carbohydrates, use the sugar test. The color of the Durham tube changes from purple to yellow during the fermentation test, and gas bubbles begin to form. In the biochemical test from the observation of 5 samples in the TSIA K/K test, the slope and base of the yellow color, H2S is negative and there is positive gas. In the SIM test sulfur is negative, positive indole produces a red color ring and negative motilty. Simon citrate test was negative. In the MR test the results are positive in red and in the VP test the results are negative. Of the 7 samples in the TSIA K / M test. slope and red

color base. In the SIM test negative, in the Simon Citrat test positive turns into blue color. The MR test was negative and the positive VP test was red.

The TSIA test measures the ability of bacteria to ferment carbohydrates. TSIA medium contains three types of carbohydrates: glucose, lactose, and sucrose. The indication is that phenol red changes color from red-orange to yellow under acidic conditions. Lactose and sucrose are found on the slopes, while glucose is near the bottom of the medium.

The SIM test assesses the capacity of an organism to accomplish various tasks, including breaking down sulfur, producing indole, and developing motility. Sulfur can be converted to hydrogen sulfide (H2S) through the desulfurase enzyme system or the amino acid catabolism system during anaerobic respiration. If hydrogen sulfide is generated, the medium turns black; otherwise, the medium will remain yellow. Indole is used to identify the capacity of bacteria to use tryptophanase enzymes. Some bacteria oxidize tryptophan, producing indole, pyruvic acid, and ammonia. The white cloud that grows on the surface of SIM media helps bacteria to migrate.

The Simon Citrate test evaluates an organism's ability to use citrate as a primary source of carbon and energy. If the bacteria can use citrate as a carbon source, the pH increases and the medium turns blue. The MR test is designed to assess an organism's capacity to create and maintain a stable acidic end product from glucose fermentation. The VP test identifies acetonin in a liquid culture of bacteria by using a red fixed pH indicator. In this experiment, alpha naphthol is used. Red represents a positive finding, but yellow, brown, and colorless represent negative results.

Escherichia coli bacterial infection in the urinary tract of patients with diabetes mellitus due to predisposing factors, such as lack of personal hygiene (personal hygine), the habit of holding urine, drinking less water, often wearing tight clothes and irregular sexual intercourse so that it becomes a trigger factor for bacteria to easily develop and the onset of infection. Patients with diabetes mellitus are susceptible to infection due to decreased immunity due to autonomic neuropathy resulting in incomplete emptying of the bladder which makes urine remaining in the bladder in an increase in the number of bacteria that cause infection and become complications in patients if the patient does not maintain personal hygiene. There are several other factors that diabetes mellitus patients cause complications of urinary tract infections, namely gender and age. UTIs are more at risk in women than men in patients with diabetes mellitus because women have a short urethral structure, absence of prostate secretion and pericium contamination of the urinary tract with fecal flora which allows microorganisms to more easily reach the bladder resulting in bacterial growth. In this study, 9 respondents were female and 3 were male. The characteristics of each sample or respondent, for example, in sample code 1,2,3,4,5,7,12 have in common, namely the habit of urinating frequently and having symptoms of pain when urinating, while in sample code 6,8,9,10,11 have in common the habit of holding urine, drinking insufficient water, having symptoms of pain in the lower abdomen and pain when urinating. Symptoms of pain when urinating are caused by bacteria entering the urinary tract through the urethra, causing infection and pain. The habit of holding urine is caused by a buildup of bacteria around the urethra or anus, causing pain and discomfort. *Escherichia coli* is a harmful bacteria that can cause infections in the urinary tract because the urine of people with diabetes mellitus has a high sugar content so that these bacteria are easier to develop in the urinary tract and bladder and also cause an invasion disease through the mechanism of toxin release.

Conclusion

Based on the results of the research conducted, it can be concluded that of the 12 samples examined, 5 samples were positive for *Escherichia coli* bacteria, while 7 other samples were found to have *Enterobacter* bacteria.

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References

1. Euis Inayah, Ika Juita Giyaningtyas. 2022. "Hubungan Dukungan Keluarga Dengan Kejadian Harga Diri Rendah Pada Pasien Diabetes Mellitus Di Klinik Dwi Cipta 1 Cibatu Cikarang Selatan Kabupaten Bekasi Tahun 2022." : 89. http://www.nber.org/papers/w16019.
2. Geerlings, Suzanne et al. 2016. "Genital and Urinary Tract Infections in Diabetes: Impact of Pharmacologically-Induced Glucosuria." Diabetes Research and Clinical Practice 103(3): 373–81. http://dx.doi.org/10.1016/j.diabres.2013.12.052.
3. Hutasoit, Dion Pardameian. 2020. "Pengaruh Sanitasi Makanan Dan Kontaminasi Bakteri Escherichia Coli Terhadap Penyakit Diare." Jurnal Ilmiah Kesehatan Sandi Husada 12(2): 779–86.
4. Intan Camilia Febyayuningrum, Rudina Azimata Rosyidah, and Resmi Aini. 2021. "Kontaminasi Bakteri Alat Stetoskop Dengan Media Bap Dan Mca Di Ruang Penyadapan Darah Udd Pmi Kabupaten Sleman Diy." Jurnal Ilmu Kedokteran dan Kesehatan Indonesia 1(3): 106–16.
5. Kementerian Kesehatan RI. 2020. "Peraturan Kemenkes Tahun 2020 Tentang Diabetes Mellitus." 2507(February): 1–9.
6. Ni Made Susilawati, Marni Tangkelangi, and Dorotia Masi Daen. 2022. "Prevalensi Infeksi Saluran Kemih Pada Penderita Diabetes Melitus Di Rsud Prof. Dr. W. Z Johannes Kupang Tahun 2021." Jurnal Kesehatan dan Kedokteran 1(3): 19–23.
7. Nur, Zaitin, Mardhia Mardhia, and Mahyarudin. 2022. "Gambaran Bakteri Penyebab Infeksi Saluran Kemih Pada Urin Penderita Diabetes Melitus Tipe 2." Majalah Kedokteran Andalas 45(2): 173–84. http://jurnalmka.fk.unand.ac.id.
8. Sri Rahayu, Risma. 2023. "Identifikasi Bakteri Escherichia Coli Pada Urine Pasien Penderita Diabetes Mellitus Tipe 2 Di RSU Bahteramas Provinsi Sulawesi Tenggara." NBER Working Papers (Dm): 89. http://www.nber.org/papers/w16019.
9. Sugireng, Satriani Syarif, and Imelda Amelia. 2022. "Karakterisasi Bakteri Penyebab Infeksi Saluran Kemih (ISK) Pada Pasien Diabetes Mellitus Tipe 2 Secara Fisiologi Dan Biokimia." 6(2).