International Journal of Life Sciences, Biotechnology and Pharma Research Vol. 13, No. 10, October 2024 Online ISSN: 2250-3137
Print ISSN: 2977-0122

DOI: 10.69605/ijlbpr_13.10.2024.131

ORIGINAL RESEARCH

Urinary Microbiota Associated with Bladder Cancer-A cross sectional study

[1]Shanthi Swaroop B, [2]Thiruvasagamani B, [3]Mangaiyarkarasi S, [4]Rajakumari S N, [5]Subhakanesh, [6]Saravanaraja

[1]Resident, [2]Professor, [3]Associate Professor, [4,5,6]Assistant Professor, Department of Urology, TVMCH, PMSSY Hospital, Tirunelveli, Tamilnadu, India

Corresponding author
Dr. Shanthi Swaroop B
Resident, Department of Urology, TVMCH, PMSSY Hospital, Tirunelveli, Tamilnadu, India

Received Date: 27 August, 2024 Accepted Date: 30 September, 2024

ABSTRACT

Background: Several biological factors such as bacterial infections and immunological status are implicated in predisposing individuals to bladder cancer. Bacterial infection of urinary tract has been related to increase the risk of bladder cancer. **Patients and Methods:** On CLED agar, urine culture will be carried out. At 24 and 48 hours, all plates will be read after being incubated at 37 degree Celsius. **Results:** Bacterial growth was observed in 15 urine samples of those patients represent (27.2 %) while, 40(72.72%) yielded no growth (negative results). It is obvious that E. coli is the most predominant organisms followed by K. pneumoniae and Ps. Aeruginosa. **Conclusion:** There is predominance of gram negative, facultative rod shaped anaerobes in the urine of carcinoma bladder cases included in the study

Keywords: Bladder cancer, Bacterial infection.

INTRODUCTION

Several biological factors such as bacterial infections and immunological status are implicated in predisposing individuals to bladder cancer (1, 2). Bladder cancer (BC) carcinogenesis is probably related to bacterial and viral infections, commonly associated with bilharzial infection rather than the parasite itself (3). Bacterial infection of urinary tract has been reported to increase the risk of BC (4). Significant etiological factors have not been identified, but chronic inflammation caused by infectious agent appears to playing a role in this disease (5). Bladder cancer in particular Squamous Cell Carcinoma (SCC) also may be caused by chronic cystitis (bladder inflammation) due to long term urinary tract infection (UTI)(6). Chronic urinary tract infections are thought to contribute to bladder carcinogenesis by several mechanisms. Repeated chronic irritation can lead to metaplastic changes, then dysplasia, and finally carcinoma. Glandular metaplasia may be seen in cases of calculus, chronic bacterial infections particularly due to Escherichia coli, schistosomiasis and in extrophy of the bladder (7) The best-documented relationship between bacterial infection and malignancy is Helicobacter pylori and gastric carcinoma. In early childhood, H. pylori alter the gastric mucosa at the cellular level resulting in chronic inflammation Mycoplasma-like organisms have been suggested to be associated with Hodgkin's disease(12). The present investigation is a trial to isolate bacteria from urine of patients with carcinoma bladder.

MATERIALS AND METHODS

Procedure

All experiments will be performed in accordance with relevant guidelines and regulations and participants need to be given written informed consent for urine collection and analysis for research purposes. Clean catch, midstream urine is collected from all participants.

Samples processing and culturing

The urine was mixed thoroughly and centrifuged before inoculation and the top of the container was removed. The calibrated loop was inserted vertically into the urine in a cup.

The following prepared culture media (Blood agar, Nutrient agar and MacConkey agar) were inoculated and streaked to obtain isolated colonies. Urine culture was incubated over-night at 37C˚ for 18-24 hours to detect uropathogens.

The isolated colonies were studied using the standard cultural characteristics to describe and identify the bacterial isolates.

On CLED agar, urine culture will be carried out. At 24 and 48 hours, all plates will be read after being incubated at 37 degree celsius

DOI: 10.69605/ijlbpr_13.10.2024.131

Inclusion Criteria

Only individuals those who were not taking antibiotics for any reason (urinary or non-urinary) for one month prior to urine collection, were included into our study.

Exclusion criteria

Those with positive history of sexually transmitted or recent urinary infections, diabetes and obesity.

RESULTS

An attempt to isolate pathogens associated with bladder cancer tumors, particularly bacterial pathogens was done. Urine sample was obtained under sterile conditions. Bacterial growth was observed in 15 urine samples of those patients represent (27.27%) while, 40(72.72%) yielded no growth (negative results).

From the above results, it is obvious that E. coli is the most predominant organisms followed by K. pneumoniae and Ps. aeruginosa. The other uropathogens isolated more or less of equal distribution.

Total no. of patients	Type of culture isolates	No	percentage	Bacterial isolates	No.	percentage
55	No growth	40	72.72	o		
	Pure bacterial isolates	5		1.E.coli	3	60%
				2.Klebsiella	1	20%
				3.Pseudomonas aeruginosa	1	20%
	Mixed bacterial isolates	10		1.E.coli and Klebsiella	6	60%
				2.E.coli, pseudomonas	2	20%
				3.Klebsiella, pseudomonas	1	10%
				4.Staph aureus, E.coli		
					1	10%
	Bacterial growth combined	15	27.27			

DISCUSSION

Bacterial infection has not traditionally been considered as a major causes of cancer. Several epidemiological studies have suggested that infection or inflammation of the urinary tract may be a risk factor for cancer of bladder. When phagocytes (neurophils, eosinophils, monocytes, macrophages) are exposed to an inflammatory stimulus (e.g. bacteria), they become activated and begin to generate large quantities of reactive oxygen and nitrogen intermediates that could lead to direct DNA damage . Reactive oxygen intermediates, also generically referred to as oxidants, are derivatives of molecular oxygen such as superoxide, hydrogen peroxide, hypochlorous acid, singlet oxygen, and the hydroxyl radical. Under normal circumstances, phagocyte-derived oxidants serve a protective function by killing invading bacteria and parasites. However, they can also have detrimental effects causing tissue damage and contributing to the development or progression of numerous diseases including cancer. Furthermore, E. coli isolates that seems to be the major bacterial species was found in urine culture in 3 patients as a single isolates. E. coli is the most common organism causing UTIs and predominates strongly at most ages . In additional to virulence factors like O- antigen, K antigens, haemolysin production, haemagglutination of human RBCs, adhesions, aerobactin and colicin production then resistance to bactericidal effect of human serum.

On the other hand, K. pneumoniae observed in 1(20%) of the cases as a single isolate and mixed isolate only in two cases with Ps. aeruginosa represent (20%) and S. aureus (10%). The incidence of this bacteria may be related to virulence properties. It was contributed that this bacterium can adhere to the epithelial cells and colonize the infected tissue and may be ascribe to their opportunistic ability particularly when there was some lowered tissue resistance or other predisposing factors like production of cytotoxins, enterotoxins and hemolysin.

Ps. Aeruginosa is another bacterial isolate found in urine culture of the same patients present as a single infection or dual infections with E. coli or S. aureus . Most of the isolated strains from clinical specimens produce large number of virulence-associated exoproducts. These include exoenzyme S, the protolytic enzymes alkaline proteases, elastases, collagenases, two hemolysins: a heat-labile phosphatase C and heat-stable glycolipid, and exotoxin, which iscomparable in action to diphtherial toxin(20). Notably, existence of mixed infections in urine cultures may support bacterial ability to cause progressive lesion(25).

Whereas, the existence of S. aureus in urine of carcinoma bladder patient may be related to their

DOI: 10.69605/ijlbpr_13.10.2024.131

ability to multiply and spread widely in tissues through their production many extracellular substance. Some of these include enzymes such as catalase, coagulase, hyaluronidase, staphylokinase, while others are considered to be toxins such as haemolysin(26). Other explanation for relating bacteria with such malignancy, in general, bacteria are able to produce a wide range of carcinogens, mutagens or tumor promoters from a wide range of substance such as Tryptophan metabolites, Volatile phenols, and N-nitroso compounds.

Many bacteria present in the urine reduce diet-derived nitrate to nitrite, which under mildly acidic or neutral conditions becomes a potent nitrosating agent. The production of N-nitrosamines by the nitrosation of amine precursors was detected in the urine of bacterially infected rats . Therefore, in addition to the N-nitrosamine exposure originating from the external environment, individuals with bacterial cystitis are potentially more exposed to nitrate and/or nitrite, which would then greatly increase the risk of in situ formation of carcinogenic alkylating agents, e.g., N-nitrosamine. Nitrosamines are capable of inducing bladder cancer in animal model(28) Gram negative – bacteria were able to produce nitosoamine compounds as a potent carcinogenic agent, in the urine of patients with bladder cancer.

CONCLUSION

Many bacteria present in the urine reduce diet-derived nitrate to nitrite, which under mildly acidic or neutral conditions becomes a potent nitrosating agent.

Gram negative – bacteria were able to produce nitosoamine compounds as a potent carcinogenic agent, in the urine of patients with bladder cancer **E.Col**i is the predominant organism isolated in the urine of patients enrolled in the study.

REFERENCES

1. Badawi, A.F., M.H. Mostafa, and P.J. O'Connor. Involvement of alkylating agents in schistosomeassociated bladder cancer: the possible mechanisms of induction. Cancer Lett. 1992; 63: 171-188.
2. World Health Organization. Evaluation of carcinogenic risk to humans. Schistosomes, liver flukes and Helicobacter pylori. IARC Monogr. 1994; 61: 45-119.
3. Shokeir, A-A. (2004): Squamous cell carcinoma of the bladder: Pathology, diagnosis and treatment. BJUInt 2004 Jan; 93(2): 216-20.
4. Mostafa, M.H., S. Helmi, A.F. Badawi, A.R. Tricker, B. Spiegelhalder, and R. Preussman: Nitrate, nitrate and volatile N-nitroso compounds in the urine of Schistosoma mansoni infected patients. Carcinogenesis, 1994; 15: 619-625.
5. Rosin, M.P., S.S. El-Din, A.J. Ward, and W.A. Anwar. Involvement of inflammatory reactions and elevated cell proliferation in the development of bladder cancer in schistosomiasis patients. Mutat. Res. 1994; 305: 283-292.
6. Mostafa, M.H., S.A. Sheweita, and P.J. O'Connor. Relationship between schistosomiasis and bladder cancer. Clin. Microbiol. Rev. 1999; 12: 97-111.
7. Saxena S., Rina B, Mohanty NK, Manoj T. Primary adenocarcinoma of the Urinary Bladder-A case Reported with review of literature. Indian J. Pathol. Microbiol. 1994; 37: 4; 453-458.
8. Sipponen P, Kosunen TU, Valle J, Riihela M, Seppala. Helicobacter pylori infection and chronic gastritis in gastric cancer. J. Clin. Pathol. 1992; 45: 319-323.
9. El-Omar, E.M.; Carrington, M. and Chow, W.H. Interleukin-1 polymorphisms associated with increased risk of gastric cancer. Nature 2000; 404: 398-402.
10. Oshima, O. and Barstch, B. Chronic infection and inflammation causes as a cancer risk factors: possible for nitric oxide in carcinogenesis. Mutation. Res. 1994; 305: 253-264.
11. Alberchet S., Hofstadter S., Artsob H., Chaban O. and from I. Lymphadenosis benign cutis resulting from Borrelia infection. J. Am. Acad. Dermatol. 1991; 24: (Abstract).
12. Sauter, S. Is Hodgkin's disease a human counterpart of bacterially induced crown-gall tumors? Lancet. 1995; 346: 1433-1435.
13. Abrams, J.; Vonderheid, E.; Kolbe, S.; Appelt, D.; Arking, E. And Balin, B. Sezary T-cell activating factor is a Chlamydia pneuminae associated protein. Clin. Diagn. Lab. Immun. 1995; 6: 895-905.
14. Colle, J.G., Miles R.S., and Watt B. Tests for the identification of bacteria. In: Collee J. G., Fraser A.G., Marimo B.P., Simmons A. Mackie and MaCartney. Practical Medical Microbiology, (14th) edition. Churchill Livingstone, New York, 1996; Pp. 148.
15. Appell, R.A.; Flynn, J.T.; Parris, A.M.I.; and Blandy, J.P. Occult bacterial colonization of bladder tumors. J. Urol. 1980; 24: 345.
16. Lattimer, J. and Tannenbaum, MI. Personal communication (1997).
17. Kantor, A.F., P. Hartge, R.N. Hoover, A.S. Naragana, J.W. Sullivan, and J.F. Fraumeni. Urinary tract infection and risk of bladder cancer. Am. J. Epidemiol. 1994; 119: 510-515.
18. Babior BM. Phagocytes and oxidative stress. Am. J. Med. 2000; 109: 33-44.
19. Hassani, H.H. Cytogenetical and Bacteriological studies on Posterior Urethral Tumors. Ph.D. Thesis submitted to the College of Sciences. University of Baghdad, 2003.
20. Jawetz, E.J.; Melnick, L. and Adelberg, E.A. Medical microbiology, 21st (ed.) 1998; Appleton and Lange, Librarie due Libu, Lebanon.
21. Higaki, M., T. Chida, H. Takano, and R. Nakaya. Cytotoxic component(s) of Klebsiella oxytoca on HEp2 cells. Microbiol. Immunol. 1990; 34: 147-151.
22. Minami, J., A. Okabe, J. Shiode, and H. Hayashi. Production of a unique cytotoxin by Klebsiella neumoniae. Microb. Pathog. 1989; 7: 203-211.
23. Guarino, A., S. Guandalini, M. Alessio, F. Gentile, L. Tarallo, G. Capano, M. Migliavacca, and A.

DOI: 10.69605/ijlbpr_13.10.2024.131

Rubino. Characteristics and mechanism of action of a heat-stale enterotoxin produced by Klebsiella pneumoniae from infants with secretory diarrhea. Pediatr. Res. 1989; 25: 514-518.

24. Barberis, L. I., A.J. Eraso, M.C. Pajaro, and I. Albesa. Molecular weight determination and partial characterization of Klebsiella pneumoniae hemolysins. Can. J. Microbiol. 1986; 32: 884-888. 2.
25. A HP Singh, DC Shetty, A Kumar, R Chavan, DD Shori, J Mali. molecular insight into the role of inflammation in the behavior and pathogenesis of odontogenic cysts. Annals of medical and health sciences research 3 (4), 523-528