Genotypic detection of the Virulence Factors of Uropathogenic
*Escherichia coli* Isolated from Diarrheic and Urinary Tract Infected
Patients in Khartoum State

اﻟﻜﺸﻒ اﻟﺠﺰﺋﻲ ﻋﻦ ﻋﻮاﻣﻞ اﻟﻀﺮاوة ﻓﻲ ﺑﻜﺘﺮﯿﺎ اﻻﺷﺮﯾﻜﯿﺔ اﻟﻘﻮﻟﻮﻧﯿﺔ ﻟﻌﺪوى اﻟﻤﺴﺎﻟﻚ البولیة و الاسﮭﺎﻻت ﻓﻲ مرضى من ولاية الخرطوم

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بسم الله الرحمن الرحيم

قال تعالى:

آمن الرسول بما أنزل إلينا من ربه والمؤمنون كل آمن بالله والمائدة وكتبه ورسله
لا نفرق بين أحد من رسوله وقالوا سمعنا وأطعنا غفرانك ربنا وإليك المصير

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Dedication

I dedicate this work:

To whom breastfed me the love and compassion to the symbol of love, and healing balms to pure whiteness heart.

(My mother)

To who spend His life working to give me a drop of love and happiness to that who pave my way to science to the big heart?

(My father)

To the pure hearts and innocent souls to my life basil's.

(My brothers and sister)

Now open the sails and raise the anchor for the ship to start running in the dark sea, the sea of life there is no light in this darkness except candles of distant memory, to those I loved them and loved me.

(My friends)

For all world nation that fight for life, for all Islamic nations and our lovely home.

Best wishes…
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I am grateful to my patients for their co-operation and consent to collect the clinical specimens used in this project.
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ABSTRACT

The aim of this study was to determine the virulence factor of Uropathogenic *E. coli* isolated from diarrheic and urinary tract infected patients in Khartoum State by multiplex PCR assay. A total of 100 clinical specimens (50 urine, 50 diarrhea) were collected in this study. Urine samples were culture on CLED agar, while diarrhea samples were culture on MacConky agar, identification scheme was done by conventional method. Modified Kirby-bauer method was performed using the following antibiotic discs; Gentamicin, Amikacin, Ciprofloxacin and Co-trimoxazole. Fifty five percent of samples were found sensitive to Gentamicin, 96% were sensitive to Amikacin, 57% were sensitive to Ciprofloxacin and 63% were sensitive to Co-trimoxazole. Boiling method was adopted for DNA extraction. Finally Multiplex PCR was done for the detection of *E. coli* virulent genes (*pap, fim, sfa, aer and hly)*.

Most study population were females 57(57%); 42 of them suffering from UTIs and 15 suffering from diarrhea, while males were 43(43%); 8 of them were suffering from UTIs and 35 of them were suffering from diarrhea. Among enrolled subjects, 82 were positive for one or more Uropathogenic *E. coli* virulent genes. While 18 isolates were negative for all genes. The results of multiplex PCR searching for different virulent factors revealed the following: Thirty tow (n=32) diarrheal samples appear as *aer* gene positive while the remaining fourteen (n=14) urine samples appear as *aer* gene positive. Thirty three (n=33) urine samples appear as *fim* gene positive while the remaining eight (n=8) diarrheal samples appear as *fim* gene positive. Twenty four (n=24) urine samples appear as *pap* gene positive while the remaining nine (n=9) diarrheal samples appear as
pap gene positive. Fourteen (n=14) urine samples appear as hly gene positive while the remaining three (n=3) diarrheal samples appear as hly gene positive. Fifteen (n=15) urine samples appear as sfa gene positive while in diarrheal samples was not detected. The study concluded that fim gene was highly prevalent among UTIs patients, aer gene was high prevalent among diarrhea patients and Amikacin is the most effective antibiotic
المستخلص

هدفت هذه الدراسة إلى تحديد عوامل الضراوة في البكتيريا الاشريكية القولونية بنسبة لعدد المسالك البولية المعزولة من مرضى الإنسال ومرضى المسالك البولية في ولاية الخرطوم، حيث تم الكشف عنها عن طريق فحص تفاعل البلمرة المتعدد المحتوي على عدسة بادئة. تم جمع 100 عينة (50 عينة بول، 50 عينة إسهال) في هذه الدراسة. ثم تم تزريع عينات البول على وسط MacConkey وعينات الإسهال على وسط CLED وتم التعرف على البكتريا بالطرق التقليدية. تم استخدام طريقة Kirby-bauer المحورية لإجراء اختبار الحساسية باستخدام أقراص المضادات الحيوية التالية: جنتاميسين، أميكاسين، سيبروفلوكساسين والكوريموكسازول وجديدا أن 55% من العينات حساسة للجناميسين، 96% حساسة للأميكاسين، 57% حساسة للسيبروفلوكساسين و 63% حساسة للكوريموكسازول. تم استخدام طريقة الغليان لاستخراج الحمض النووي أخيرا تم إجراء اختبار تفاعل البلمرة المتعدد المحتوي على عدة بادئة للكشف عن عوامل الضراوة في بكتريا الاشريكية القولونية.

وكان معظم المشاركين في الدراسة من الإناث 57 (75%); 42 منهم يعانون من عددى المسالك البولية و 15 منهم يعانون من الإنسال، في حين بلغ عدد الذكور 43 (43%); 8 منهم يعانون من التهاب المسالك البولية و 35 منهم يعانون من الإنسال. من عينات الدراسة، كانت 82 (82%) عينة إيجابية لواحدة أو أكثر منجينات الضراوة، بينما كانت 18 (18%) عينة سلبية لجميع الجينات. نتيجة اختبار تفاعل البلمرة المتعدد المحتوي على عدة بادئة للكشف عن عوامل
الضرأوة كانت كالالتالي: اثنين و ثلاثون (ن=32) عينة إسهال ظهرت موجبة للجين aer و المتباقي
اربعة عشر (ن=14) عينة بول ظهرت موجبة للجين fim و المتباقي ثمانية (ن=8) عينات إسهال ظهرت موجبة للجين.
اربعة و عشرون (ن=24) عينة بول ظهرت موجبة للجين pap و المتباقي تسعة (ن=9) عينات إسهال ظهرت
اربعة عشر (ن=14) عينة بول ظهرت موجبة للجين hly و المتباقي ثلاثة.
خمسة عشر (ن=15) عينة بول ظهرت موجبة hly.
لم يظهر نفس الجين في أي عينة إسهال. وخلصت الدراسة إلى أن الجين fim للجين fim
منتشرًا بكثرة بين مرضى المسالك البولية، وكان الجين hab عالي الانتشار بين مرضى
الإسهال ووجدنا المضاد الحيوي الأميكاسين هو المضاد الحيوي الأكثر فعالية.
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