ANTIBIOTICS SUSCEPTIBILITY AND MOLECULAR CHARACTERIZATION OF CLINICAL ISOLATES OF *E. coli* FROM AHMADU BELLO UNIVERSITY TEACHING HOSPITAL SHIKA, ZARIA

BY

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M.SC/ PHARM. SCI/ 4967/ 2010-2011

A THESIS SUBMITTED TO THE SCHOOL OF POST-GRADUATE STUDIES AHMADU BELLO UNIVERSITY IN PARTIAL FULFILLMENT OF THE REQUIREMENT FOR THE AWARD OF MASTER DEGREE (M.Sc.) IN PHARMACEUTICAL MICROBIOLOGY

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DECEMBER, 2011

DECLARATION

I declare that the work reported in this thesis entitled "Antibiotics Susceptibility and Molecular Characterization of clinical isolates of *E. coli* from Ahmadu Bello University Teaching Hospital Shika, Zaria" was carried out by me in the department of pharmaceutics and Pharmaceutical Microbiology, Faculty of Pharmaceutical sciences, Ahmadu Bello University, Zaria, Nigeria under the supervision of Prof. J. A. Onaolapo and Prof. J. O. Ehinmidu. All information derived from the literature was acknowledged and referred to accordingly. I declare that no part of this thesis has been submitted elsewhere for a degree or diploma.

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CERTIFICATION

This thesis entitled "Antibiotics Susceptibility and Molecular Characterization of clinical isolates of *E. coli* from Ahmadu Bello University Teaching Hospital Shika, Zaria" by James Chibueze, IGWE meets the regulations governing the award of the degree of Master of Science (Pharmaceutical Microbiology) of the Ahmadu Bello University, Zaria, and is approved for the scholarly contribution to knowledge and literary presentation.

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This work is dedicated to God Almighty and my beloved family.

ACKNOWLEDGEMENT

From the depth of my heart I wish to sincerely express my profound gratitude to you Sir, Prof. J. A. Onaolapo (Major Supervisor) for your support, advise, contribution and thorough guidance during the course of this work. I say God bless you and your family. Also I wish to appreciate the constructive supervision and contribution of Prof. J. O. Ehinmidu (Co-supervisor) that led to the actualization of this dreamed work. I say thank you sir for your support.

I also appreciate the effort of the H.O.D, my departmental lecturers, laboratory technicians and the entire staff in the Faculty of Pharmaceutical Sciences, A.B.U Zaria. Who in one way or the other showed interest in my work. God bless you all.

Words are not enough to thank you ma, Mrs. M. I. Okeke and Mrs. Olajumoke, Rebecca Bolaji of the Department of Pharmaceutical Microbiology and Dr. (Mrs.) Afolabi-Balogun, Nusrah Bolatito of the Centre for Biotechnology Research and Training, A.B.U Zaria, thanks for your relentless effort, encouragement and motherly companion.

My sincere gratitude to Prof. Alex U. Akpa and all the staff of the Department of Medical Biotechnology, National Biotechnology Development Agency (NABDA), Abuja, for their support that challenged and encouraged me in the course of my study.

Also, my heartfelt gratitude to my dearly belovely and caring sweet mother Mrs. Igwe Mary, my elder brother Capt. J. O. Igwe, my noble uncle, Mr. Ajauwa B. and my lovely and careering elder sister, Mrs. Racheal Kelechukwu who have always encouraged me to further my academic pursuit and live a responsible life.

Finally, I wish to acknowledge my beloved friends and colleagues: Okorie, Chinenye Felicia; Okpala, Chinedu Jude; Mukosolu, U. Nzekwe; Ugwuneji, Nnaemeka V; Ogwu, Nkechi; Yakubu, Michael Nosano; Mari, Mshelia; Olatunji, Aremu, Kayode; Odeh, Ruth; Aboh, Mercy; Paul, Sunday; Abudullahi Musa; Salihu, Musa; Mrs. Jane David; Onyeka, Ifeanyi Peter; Okeke Izunna S and Udoh, Faith; I say thank you all for your encouragement, prayers and support.

ABSTRACT

Antimicrobial drug resistance is a global challenge with the emergence of resistant bacterial strains worldwide. This study was conducted to determine the incidence of *E. coli* in ABUTH Shika, Zaria (from March 2011 to February 2012), their antibiotics susceptibility pattern, molecular characterization and possible presence of *E. coli* serotype O157:H7. Clinical isolates of *E. coli* from inpatients and outpatients were collected and cultured on eosin methylene blue to obtain pure cultures. A retrospective analysis of records of the Medical Microbiology unit for the same period (March 2011 to February 2012) was carried out. The incidence of *E. coli* isolates in clinical samples was found to be 50% in stool, 26.7% in urine, 13.3% in blood, 6.6% in urogenital and 3.3% in wounds. The retrospective analysis of the prevalence of *E. coli* associated infections in ABUTH Shika, showed that out of the 751 patients bio-data evaluated, female patients were mostly infected 62.3%(468) compared with male patients 37.4%(281). Of the 751 patients bio-data evaluated, 150 isolates were collected and 60 isolates were confirmed as *E. coli.* The antibiotic susceptibility profile of clinical isolates of *E. coli* to fourteen (14) commonly prescribed antibiotics in the treatment of *E. coli* associated infections showed that 76.7% of the isolates were resistant to ceftazidime, 78.3% to amoxicillin-clavulanic acid, 70% to ampicillin-sulbactam, 56.7% to tetracycline, 43.3% to cefalexin, 41.7% to nalidixic acid, 36.7% to amoxicillin and 30% to cefuroxime. The isolates were also found to be sensitive to ceftriaxone (88.3%), gentamicin (78.3%), chloramphenicol (78.3%), ciprofloxacin (71.7%), nitrofurantoin (78.4%), ofloxacin (73.3%). Higher percentage (80%) of the isolates were multidrug resistant (MDR), 11.7% were extensively drug resistant (XDR). Statistical analysis at p value < 0.05, showed a significant difference in the level of resistance expressed by *E. coli* from different clinical samples. At MARI ≥ 0.3, 71.6% of the patients showed a frequent use of the antibiotics

usually prescribed in the hospital for *E. coli* infections. The high minimum inhibitory concentration (μg/ml) of amoxicillin–clavulanic acid and ceftriaxone to the transconjugants of the multidrug resistant *E. coli* showed that the resistance exhibited was plasmid encoded. The extended spectrum β-lactamase (ESBLs) using double disc diffusion method showed that of the 10 isolates tested, all were resistant to amoxicillin-clavulanic acid, 7(70%) to ceftazidime, 5(50%) to ceftriazone and cefpodoxime. Seventy percent 7(70%) of the 10 selected multidrug resistant clinical isolates were ESBLs positive; a ≥ 5mm increase in zone diameter for either antibiotics compared to its zone when tested alone, while 30% (3) of the selected isolates showed production of AmpC gene. *E. coli* serotype O157:H7 was isolated in 11(18.3%) of the 60 confirmed *E. coli* isolates (that is, 36.7% (11) of the stool isolates (30) were *E. coli* serotype O157:H7, while other serotypes in stool isolates amounted to 63.3%). Hetero-resistant isolates of *E. coli* (small colonies variant, found within the diameter of the zone of inhibition of cefoxitin) showed a far higher resistance to some tested antibiotics than that of their parental clinical isolates. Using molecular characterization by polymerase chain reaction (PCR), the ESBLs encoding genes, TEM, SHV and OXA and plasmid bands were detected in the multidrug resistant isolates, and in line with documented works, this suggests that these genes were plasmid encoded. *E. coli* expressing ESBLs and AmpC enzyme are present in *E. coli* isolated from ABUTH, Shika, Zaria. These suggest that patients with infections associated with *E. coli* producing ESBLs and AmpC enzyme may have complication in therapy and limited treatment options, which will lead to higher mortality rate, high economic burden and longer hospital stays.

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