

Review Form 1.6

Journal Name:	South Asian Journal of Research in Microbiology
Manuscript Number:	Ms_SAJRM_84658
Title of the Manuscript:	Virulence response of Escherichia coli to different concentrations of combined herbal drugs
Type of the Article	Original Research Article

General guideline for Peer Review process:

This journal's peer review policy states that **NO** manuscript should be rejected only on the basis of '**lack of Novelty**', provided the manuscript is scientifically robust and technically sound. To know the complete guideline for Peer Review process, reviewers are requested to visit this link:

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PART 1: Review Comments

	Reviewer's comment	Author's comment (if agreed with reviewer, correct the manuscript and highlight that part in the manuscript. It is mandatory that authors should write his/her feedback here)
Compulsory REVISION comments	<p>1-The authors showed that; The used herbal drugs were purchased from Mile 3 Market. How can the authors use the products from the markets without confirmation about the purity of the product? Are their any other constituent like preservatives have antibacterial effect? What about the concentration of the active constituents n the product ???</p> <p>2 The authors showed that; <i>Escherichia coli</i> strain used in this research was purchased from Lahor Research Laboratory, Benin City, Edo State.</p> <p>What about the detailed characteristics of the used strain of E.coli?? Is it sensitive to antimicrobials, is it multidrug resistant , Pan drug resistant or what?????</p> <p>3- Why the authors don't test the herbal effect on other multidrug resistant organisms???</p> <p>4- The manuscript requires English editing to correct the grammar and flow of sentences.</p> <p>5- The authors did not mention the ethical approval statement in the manuscript.</p> <p>6- The title "Virulence response of <i>Escherichia coli</i> to different concentrations of combined herbal drugs" is broad and don't describe the work. It should be changed to be more precise.</p> <p>Suggested title is " Antimicrobial activity of some combined herbal drugs against E.coli "</p>	<p>1. The purity of herbal products were confirmed by culturing the herbal concoction on nutrient agar to investigate for purity. The products used were confirmed to have no bacterial growth.</p> <p>2. The strain of the bacterial used was <i>E. coli</i> ATCC 252922. It was sensitive to the herbal drugs used. In the work the negative control is the pure isolate of the bacteria</p> <p>3. This was done using a continuous broth culture method after 48 hr which are shown in figure 1. Some level of efficacy was observed overtime</p> <p>4. Grammarly software has been used to edit the English of the work.</p> <p>5. The authors worked with a category 1 microorganism.</p>
Minor REVISION comments	<p>1. INTRODUCTION</p> <p>Authors showed write about the used combined herbal drugs; chemical structure, suggested mechanisms of action against microbes.</p> <p>2. materials and methods</p> <p>2.1 Herbal Drugs</p> <p>The herbal drugs used were Beta herbal mixture (B), Deep root herbal mixture (D), Goko cleanser (G). These drugs were purchased from Mile 3 Market, Porthacourt,</p> <p>How can the authors use the products from the markets without confirmation about the purity of the product? Is there any other constituent like preservatives have antibacterial effect? What about the concentration of the active constituents in the product??? Purification??</p>	<p>The chemical structure of the herbal drugs used have not been profiled chemically.</p> <p>The purity of herbal products were confirmed by culturing the herbal concoction on nutrient agar to investigate for purity. The products used were confirmed to have no bacterial growth.</p>

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	<p>What are these herbs? What is the active constituent that have the antibacterial effect ??</p> <p>The reference for the manipulation with these products must be added</p> <p>2.2 Test Organisms</p> <p><i>Escherichia coli</i> strain used in this research was purchased from Lahor Research Laboratory, Benin City, Edo State. The identity of the microorganism was subjected to confirmatory testing such as MacConkey agar, indole and Grams stains according to Chesbrough [10]</p> <p>What about the detailed characteristics of the used strain of <i>E. coli</i>?? Is it sensitive to antimicrobials, is it multidrug resistant, Pan drug resistant or what?????</p> <p>Why the authors don't test the herbal effect on other multidrug resistant organisms like <i>P. aeuroginosa</i>???</p> <p>Also the effect on Gram positive organisms like MRSA, VRSA and fungi like <i>Candida speies</i> should be tested,</p> <p>2.4 Growth Response to the Herbal Drugs</p> <p>The growth response was carried out by using the spectrophotometric method. In the spectrophotometric method, overnight inoculum was serially diluted to 10^8 and treated with different concentrations of BG and BD serially diluted in different concentration of the overnight inoculums, and incubated for 18 hrs at 37 °C. The optical densities of the various concentrations were obtained using spectrophotometer at a wavelength of 600 nm.</p> <p>The unit of 10^8 must be mentioned The reference of the method should be added How can you standardize the test</p> <p>different concentrations of BG and BD serially diluted in different concentration of the overnight inoculums?????? How can you select these concentrations, where is the MIC determination of these used herbal products</p> <p>2.5 Antimicrobial Susceptibility Testing of the Herbal Drug-Treated <i>E. coli</i></p> <p>What about the reference for this test?? Why the author select these antibiotics only ? what about other classes of antibiotics?? How can you use the same strain of the E.coil as control group?? Is the test is according to the guidelines CLISI ??? Mention</p> <p>3. Results; are weak</p> <p>References:</p> <p>NO RECENT REFERENCES ARE PRESENT</p>	<p>These drugs are yet to be verified in terms of their actual potency, hence, the reference for these manipulation are difficult to get.</p> <p>The test organism used was <i>E. coli</i> ATCC 252922.</p> <p>The focus of this work was on <i>E. coli</i> hence there was no need including <i>Pseudomonas</i> as they are of different class of bacteria. However, further work can assess these bacteria</p> <p>The 10^8 was a dilution factor. The actual number of bacteria in this dilution was 10^3 cfu/ml which has been indicated in the work.</p> <p>The work used <i>E. coli</i> that has not been exposed to the herbal drug has control.</p> <p>The authors have added more recent references.</p>
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Optional/General comments	The manuscript requires English editing to correct the grammar and flow of sentences.	
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PART 2:

	Reviewer's comment	Author's comment (if agreed with reviewer, correct the manuscript and highlight that part in the manuscript. It is mandatory that authors should write his/her feedback here)
Are there ethical issues in this manuscript?	(If yes, Kindly please write down the ethical issues here in details)	