

RESEARCH ARTICLE

Efficacy of *Senna occidentalis* Leaf Extracts in the Treatment of Bacterial Gastroenteritis

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Submitted: 20th January 2026 | Accepted: 23rd April 2026 | Published Online: 26th August 2026

ABSTRACT

Bacterial gastroenteritis remains a significant global health challenge due to the rising issue of antibiotic resistance and the high cost of conventional drugs especially in low-resource settings. Addressing this gap, this study evaluated the efficacy of *Senna occidentalis* leaf extracts in treating bacterial gastroenteritis through an in vitro laboratory-based experimental design. Before the commencement of the study, an Ethical clearance certificate and a permit to conduct the study was sought from Kabarak University Research Ethics Committee (KUREC) and National Commission for Science, Technology and Innovation (NACOSTI) respectively. Mature *S. occidentalis* leaves were collected from Kilifi County, authenticated, and extracted using methanol, petroleum ether, and distilled water. Phytochemical screening was carried out using standard qualitative methods. Antibacterial activity against *Salmonella paratyphi* was assessed using the disk diffusion method and Minimum Inhibitory Concentration (MIC) tests, with ciprofloxacin as the positive control and DMSO as the negative control. Methanol extraction produced the highest yield (29.15%), followed by aqueous (13.78%) and petroleum ether (4.32%). Phytochemical analysis revealed high levels of alkaloids, tannins, coumarins, flavonoids, anthraquinones, and cardiac glycosides, with saponins present in low amounts. All extracts showed measurable antibacterial activity, with petroleum ether extracts demonstrating relatively stronger inhibition at lower concentrations and aqueous extracts performing better at higher concentrations. Ciprofloxacin produced significantly larger inhibition zones than all extracts. Data were analyzed using descriptive and non-parametric statistical tests (Kruskal-Wallis and Spearman correlation) in SPSS Version 27 at 95% confidence. The study successfully evaluated the efficacy of *Senna occidentalis* leaf extracts against *Salmonella paratyphi*, confirming its antibacterial potential. However, the modest inhibition zones compared to ciprofloxacin underscore the need for bioassay-guided fractionation, in vivo testing, and toxicity evaluation to isolate and enhance active constituents.

Keywords: Gastroenteritis, *Senna occidentalis*, *Salmonella paratyphi*, phytochemicals, antibacterial activity

How to Cite this Article: GEORGE NYADZUA, E., EVANDER OCHIENG, O., KEITH WAKASIKA, L., FAITH BERYL, U., JOSHUA, K., MOSES KIPLIMO, R., ... WALEKHWA, M. (2026). Efficacy of *Senna occidentalis* Leaf Extracts in Treatment of Bacterial Gastroenteritis. *African Journal of Pharmacy and Alternative Medicine*, 5(02), 306–318. <https://doi.org/10.58460/ajpam.v5i02.199>



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INTRODUCTION

Gastroenteritis is a prevalent and significant health concern globally that is associated with high morbidity and mortality rates among children below 5 years of age (Troeger et al., 2018; Sultan & Hassan 2018) as a result of severe dehydration (Zaidi & Smith-Morris 2015). It is also a significant cause of financial constraint in households and healthcare systems due to loss of wages and reduced productivity (Kankeu et al., 2013). Worldwide, children in this age group are estimated to have an annual prevalence of 1.7 billion episodes of diarrhea, which leads to 124 million clinical visits and 9 million hospitalizations (WHO, 2017). These cases account for 2.2 million deaths annually in children younger than 5 years (Humphries & Linscott, 2015). This means that more than 2000 children of this age group die every day. Generally, it is the cause of 15% of all child deaths with 98% of these deaths occurring in developing countries (WHO, 2013a; Zaidi & Smith-Morris 2015) in which Kenya is a member.

In 2015, diarrheal diseases were estimated to have caused over 9.5 billion episodes and a mortality rate of about 500,000 children younger than 5 years (Collaborators GBD, 2017). Africa and South Asia accounted for 800,000 (10%) of the deaths of children under 5 years globally (Alam et al., 2015; Liu et al., 2012). South Asia demonstrated 32% annual mortality out of an annual 7.6 million deaths worldwide because of diarrhea in children (Sohail & Neupane, 2019). The highest rates of under five children deaths among the 18 countries of the Eastern Mediterranean region were demonstrated by Pakistan, accounting for 464,886 annual deaths (Alam et al., 2015). In developed countries, 1 in 25 children below 5 years of age is diagnosed with acute gastroenteritis (Freedman et al., 2013); for instance, in Canada, more than 5 million cases of acute gastroenteritis are diagnosed each year (Albrecht et al., 2017).

The prevalence of diarrhea in Sub-Saharan Africa is 15.3% and this is coupled with the burden of other comorbidities and poverty (Demissie et al., 2021). In a study by Owusu et al., 2024 the prevalence of diarrhea in West Africa generally was 13.7%. The high burden of diarrheal disease among children under five years of age in West Africa is due to factors like limited access to clean water, inadequate hygiene practices, malnutrition, low vaccination coverage, poverty and limited access to healthcare services which makes it hard for the affected children to receive the required treatment on time (Simen-Kapeu et al., 2021). In Kenya diarrheal infections are estimated to have caused 4471 deaths, 8781 hospitalizations and 1,443,883 hospital visits in children below 5 years of age (Osano et al., 2011). The prevalence of gastroenteritis

in Siaya county in Kenya was 4-11% and this was due to limited access to adequate sanitation amenities (Omore et al., 2013). The condition is primarily caused by bacteria, viral as well as parasitic pathogens (Tam et al., 2012) resulting to severe symptoms like diarrhea, abdominal pain and vomiting. The associated risk factors include poor hygiene and sanitation, inability to access safe drinking water and contaminated food (Elliot, 2007). This explains why diarrhea due to infection is widespread throughout low income countries.

The current treatment of gastroenteritis primarily relies on supportive care including rehydration therapy (Rouhani et al., 2011) to manage dehydration and the use of antibiotics in bacterial infections (Guarino et al., 2018). However, the misuse and overuse of antibiotics have led to a significant rise in antibiotic-resistant strains of bacterial therefore complicating treatment protocols and compromising the efficacy of standard antibiotic therapy (Abd-Elmeged et al., 2015; Rhee et al., 2019; Muloi et al., 2019). The side effects from antibiotics also pose a limitation on these medications. For instance, quinolones like Ciprofloxacin are associated with risks of arthropathy (O'Ryan et al., 2014).

Senna occidentalis commonly known as coffee senna is a plant that belongs to Fabaceae family (Musa et al., 2017). It is widely distributed in tropical and subtropical regions including Asia, Africa, North America, South America, Central America, the Caribbean and India and has been used in traditional medicine for its purported health benefits. Various parts of the plant have been utilized for their potential therapeutic benefits. The leaves, roots and seeds of this plant have been used in traditional medicine for the treatment of various ailments like diabetes mellitus, fever, Typhoid and dysentery (Evans et al., 2002). Local healers among the Mijikenda from the Kenyan coast have reported that the decoction of the leaves of *S. occidentalis* can be used for relief of stomach pain and diarrhea which may also manifest in gastroenteritis. Preliminary studies have indicated that *Senna occidentalis* contains a range of phytochemicals including anthraquinones, flavonoids and tannins which contribute to its medicinal effects (Sadiq et al., 2012).

Despite the traditional use of *Senna occidentalis* in treating gastrointestinal ailments, there is lack of comprehensive scientific evidence to support its efficacy particularly in the context of treating gastroenteritis.

Therefore, against this background, this study aimed to systematically investigate the phytochemicals present in the leaves of *Senna occidentalis* and determine the efficacy of the leaf extracts in the treatment of gastroenteritis through in vitro studies.

METHODOLOGY

Research Design

This was a laboratory based experimental study according to Garton et al., (2005). Initially the leaves were collected, dried, and subjected to solvent extraction to obtain the crude extracts. *Salmonella paratyphi*, commonly associated with gastroenteritis was cultured under controlled laboratory conditions. The bacterial cultures were then exposed to different concentrations of the *S. occidentalis* extracts. The antibacterial activity was assessed by measuring the diameters of the zones of inhibition. The results were then compared with standard antibiotics to evaluate the relative efficacy of the plant extract.

Study Location

S. occidentalis leaves were obtained from Mazeras in Kilifi County while the bacterial strains were obtained from Kenya Medical Research Institute (KEMRI). The experimental work of the study was first conducted at the pharmacognosy laboratory where extraction took place. This was then followed by antibacterial assays at the Microbiology laboratory in the School of Pharmacy (Kabarak University).

Study Population

Salmonella paratyphi, *Senna occidentalis* leaf extract, Ciprofloxacin, Dimethyl sulfoxide (DMSO) as negative control.

Inclusion and Exclusion Criteria

Inclusion Criteria

Fresh, mature, and disease-free *Senna occidentalis* leaves collected from Mazeras, Kilifi County, and authenticated by a qualified botanist. Leaves harvested from plants growing in their natural habitat, free from pesticide or chemical exposure. Bacterial strain: *Salmonella paratyphi* obtained from a reputable reference source (Kenya Medical Research Institute - KEMRI). Only solvents of analytical grade (methanol, petroleum ether, and distilled water) and Culture media and laboratory materials sterilized before use to prevent contamination.

Exclusion Criteria

Immature, insect-damaged, or mold-contaminated *S. occidentalis* leaves, plant materials exposed to chemical treatment or harvested from polluted environments (e.g., roadsides or waste disposal areas), bacterial isolates other than *Salmonella paratyphi* and solvents, reagents, or culture media

that were expired, contaminated, or not meeting analytical-grade standards were excluded.

Sampling

Sampling Technique

Purposive sampling technique was used in this study as outlined by Rai & Thapa (2015). This is a non-probability sampling technique where the researcher selects samples based on specific characteristics and the objective of the study. It is particularly useful in studies where specific types of subjects or materials are required. Purposive sampling ensured that the collected samples were specifically relevant to the study's objectives, focusing on the plant material and bacterial strains pertinent to gastroenteritis. For instance, in the identification of plant materials, mature and healthy *Senna occidentalis* plants were selected during collection to prevent potential variation in phytochemical composition due to environmental factors. A qualified botanist ensured the authenticity of the plant specimen. Leaves were also collected during the same season to maintain consistency in the phytochemical profile. Only common gastroenteritis-causing bacterial strains (*Salmonella paratyphi*) were chosen for the study.

Sample Collection

Senna occidentalis leaves from mature plants were collected from Mazeras in Kilifi County. The collection was done in the morning to ensure maximum potency of the phytochemicals. The collected leaves were then rinsed thoroughly with water to remove any soil or debris. This was then followed by air drying the leaves in a shaded, well ventilated area for 10 days to prevent degradation of sensitive phytochemicals. The dried leaves were then transported to Nakuru County in a carton. The plant material was then authenticated by a botanist to ensure correct identification and then a voucher specimen was deposited at the Laboratory Herbarium. The leaves were then grinded into fine powder using a laboratory grinder. The powdered leaves were then stored in airtight containers at room temperature waiting for extraction. The *Salmonella paratyphi* was obtained from Kenya Medical Research Institute (KEMRI). Biochemical tests were performed to confirm the identity of the bacterium. The sample was then sub-cultured onto nutrient broth for storage. Short term storage was done at 4 degrees.

Laboratory Analysis

Extraction Process

The powdered leaves were subjected to solvent extraction using different solvents to obtain various extracts. Solvents such as petroleum ether, water and methanol were used for extraction.

For the methanolic extract, 100 grams of the powdered leaves was macerated in 500 ml of methanol for 48 hours with frequent agitation. The mixture was then filtered and the filtrate was concentrated under reduced pressure using a rotary evaporator. The same procedure as methanolic extraction was followed using petroleum ether as the solvent to obtain the petroleum ether extract. For aqueous extraction, 100 grams of the powdered leaves were macerated in 500 ml of distilled water for 48 hours with frequent agitation. Before filtration, the sample was placed in a water bath for 30 minutes, cooled and filtered and the filtrate was lyophilized to obtain a dry extract. The concentrated extracts were stored in airtight containers at 4 degrees Celsius.

Phytochemical Screening

The extracts were analyzed for the presence of alkaloids, tannins, saponins, flavonoids, glycosides, phenols, anthraquinones, cardiac glycosides, steroids and coumarins using standard qualitative methods.

Disk Diffusion Method

The antibacterial activity of *Senna occidentalis* leaf extracts was evaluated using the Kirby-Bauer disk diffusion method (Humphries et al., 2018). *Salmonella paratyphi* was inoculated into 2ml of normal saline and the turbidity of the suspension was compared with that of a 0.5 McFarland standard reagent whose turbidity is equivalent to $1-2 \times 10^8$ Colony Forming Units (CFU)/ml. Sterile Mueller-Hinton Agar (MHA) plates were prepared according to the instructions on the label of the container and inoculated with the bacterial suspensions via streaking method using a sterile swab to ensure even distribution. Small round disks with a diameter of 6mm were soaked into different concentrations of the extracts (made by dissolving in 1 ml of DMSO) and then left to dry for 10-15 minutes. These disks impregnated with the extracts, together with the disks containing the selected standard antibiotics were placed on the inoculated culture plates. Disks soaked in the plain DMSO were used as negative controls. The plates were then incubated at 37 degrees for 24 hours and afterwards the zones of inhibition around each disk were measured in millimeters using a Vernier calipers to assess the antimicrobial activity. The antibacterial activity of *Senna occidentalis* leaf extracts and effectiveness was assessed relative to antibiotics such as ciprofloxacin by analyzing the diameter of the zones of inhibition (Charles, 2009).

Table 1:

Extract Yield Based on Extraction Solvent Used

Extraction solvent	Initial powder weight (g)	Extract weight (g)
Methanol	100	29.146g
Petroleum ether	100	4.321g
Distilled water	100	13.783g

Minimum Inhibitory Concentration

Nutrient broth was prepared according to the manufacturer's instructions on the container's label and 2ml of the media was placed into six sterile test tubes. Then 1ml of the bacterial suspension was added into each test tube containing the media. Then 6 other sterile test tubes were labelled 1,2,3,4,5 and 6. Test tubes 2,3,4,5 and 6 contained 0.5ml of dimethyl sulfoxide (DMSO). In test tube 1, 1000mg of the extract/1ml of DMSO was added. 0.5ml from test tube 1 was transferred to test tube 2 and 0.5ml from test tube 2 was transferred to test tube 3. This process was continued for the rest of the test tubes and 0.5ml from the last test tube was discarded. This formed concentrations ranging from 31.25mg/ml to 1000mg/ml. The nutrient broth containing the bacterial suspension was transferred into the test tubes containing the different concentrations of the extracts and incubation was done. The MIC was to be the lowest concentration of the extracts that showed no turbidity in the test tube after incubation.

Data Analysis

Data were analyzed using descriptive and non-parametric statistical tests (Kruskal-Wallis and Spearman correlation) in SPSS Version 27 at 95% confidence.

Ethical Considerations

Prior to the commencement of the study, a letter from the School of Pharmacy management to carry out the study was sought. Ethical clearance was obtained from the Kabarak University Research Ethics Committee (KUREC), under reference number **KABU01/KUREC/001/18/05/25**. Additionally, a research permit was secured from the National Commission for Science, Technology and Innovation (NACOSTI) with license number: **NACOSTI/P/25/4174535**.

RESULTS

Percentage Yield of Extracts

Three solvents, namely: methanol, petroleum ether and distilled water were used each for extraction. The findings in table 1 below show that methanol-based extraction gave the highest yield weight (29.146g) followed by distilled water (13.783g) and finally petroleum ether-based extraction (4.321g).

Figure 1 below shows a comparative analysis of the percentage yield based on different solvents used for extraction. As noted previously, methanol solvent had the highest percent yield (29.15%) followed by distilled water (13.78%) and finally petroleum ether (4.32%).

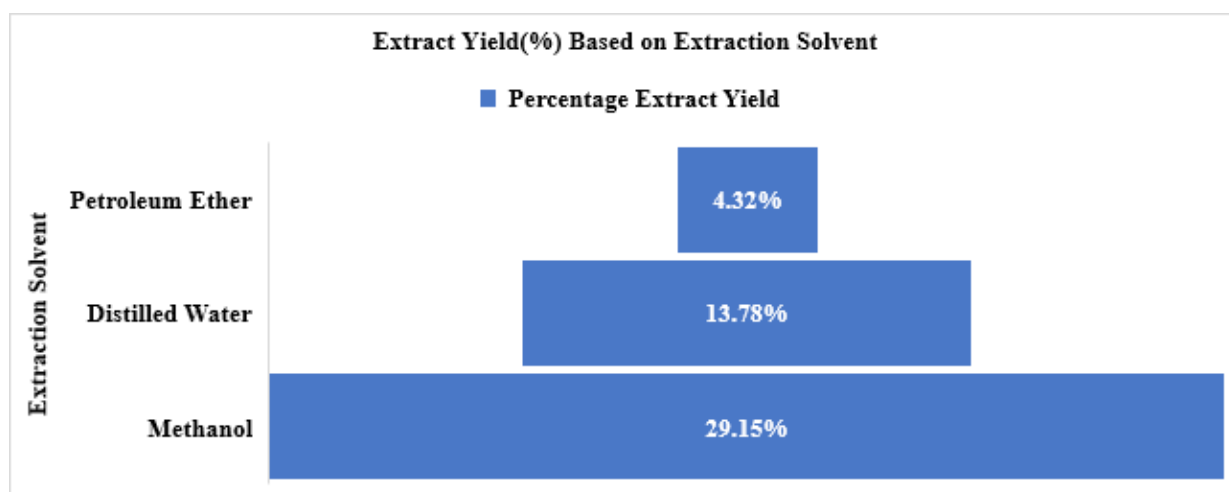


Figure 1: Comparative Analysis of Extract Yield Based on Extraction Solvent Used

Phytochemical Screening

Phytochemical screening of *Senna occidentalis* leaf extract was also conducted. The findings in table 2 show that the extract had high amounts of alkaloids, tannins, coumarins, cardiac glycosides, anthraquinone & anthrones, flavonoids and phytosterol. Saponins were present in low amounts while phenolic compounds, terpenoids, cardenolides and dianthones were found to be absent.

Table 2:

Results of the Phytochemical Screening of the Extract and Fractions of *Senna occidentalis* Leaves

Phytochemical Test	Expected Color Changes	Intensity	Inference
ALKALOID			
Picric acid test	Formation of a yellow precipitate	Observed(+++)	Alkaloid present in high amount
Dragendroff's Test	Formation of an orange precipitate	Observed(+++)	Alkaloid present in high amount
Wagner's Test	Formation of a brown precipitate	Observed(+++)	Alkaloid present in high amount
Mayer's Test	Formation of a creamy white precipitate	Observed(+++)	Alkaloid present in high amount
SAPONINS			
Foam Test	The foam formed, it persisted for 15 minutes but was less than 1.5cm	Observed(+)	Saponin present in small amounts
TANNINS			
5% Ferric Chloride Test	Formation of a brown precipitate	Observed(+++)	Tannin present in high amount
10% Lead acetate Test	Formation of a cream white-yellow precipitate	Observed (++++)	Tannin present in high amount
Potassium Dichromate Test	Formation of a brown color	Observed (+)	Tannin present in small amount
COUMARINS			
Ferric chloride Test	Formation of a blue-black color	Observed(+++)	Coumarins present in high amount
FLAVONOIDS			
5% Ferric Chloride Test	Formation of a yellow precipitate	Not observed (-)	Absence (phenolic compound)

10%Lead Acetate Test	Formation of a yellow precipitation	Observe(+++)	Flavonoid present in high amount
SodiumHydroxide Test	Formation of an intense yellow color that disappears after adding 10% sulphuric acid	Observed(+++)	Flavonoids present in high amount
Shinoda Test	Formation of an orange reddish color	Observed(+++)	Flavonoids present in high amount
Conc Sulphuric Acid Test	Formation of a deep red color	Observed(+++)	Flavanes present in high amount
PHYTOSTEROLS			
Salkowski Test	No formation of a reddish color	Not observed(-)	Absence (Terpenoids)
Liebermann's Test	Formation of a Bluish-green layer	Observed(+++)	Phytosterol present in high amount
CARDIAC GLYCOSIDES			
Baljet's Test	Formation of an orange color	Observed(+++)	Cardiac glycosides present in high amount
Kedde's Reagent	No purple color formed	Not observed(-)	Absence (Cardenolides)
Keller-Killiani Test	Formation of upper bluish green layer and reddish-brown lower layer	Observed(+++)	Cardiac glycosides present in high amount
Liebermann's Test	Formation of bluish green color	Observed(+++)	Cardiacglycosides present in high amount
ANTHRAQUINONE GYLCSIDES			
Borntrager's Test	Formation of a pink red color	Observed(+++)	anthraquinone & anthrones present in high amount
Modified Borntrager's Test	Formation of an orange color	Not observed (-)	Dianthrone absent

(-) Absent; (+) Present in Small Amounts; (++) Present in Moderate Amounts; (+++) Present in High Amounts

In terms of abundance, figure 2 shows that alkaloids (1.0), coumarins (1.0), flavonoids (0.8), tannins (0.78) and cardiac glycosides (0.75) were highly abundant while phytosterols (0.5) and anthraquinone glycosides (0.5) were moderately abundance in the leaf extract. Saponins were found to be present in small quantities (0.3). Phytochemical abundance was calculated by coding '+++' as 3 to mean highly abundant, '++' as 2 to mean moderately abundant, '+' as 1 to mean low amounts and '-' as 0 to represent phytochemical absence. The percentages were computed as average abundances for each phytochemical.

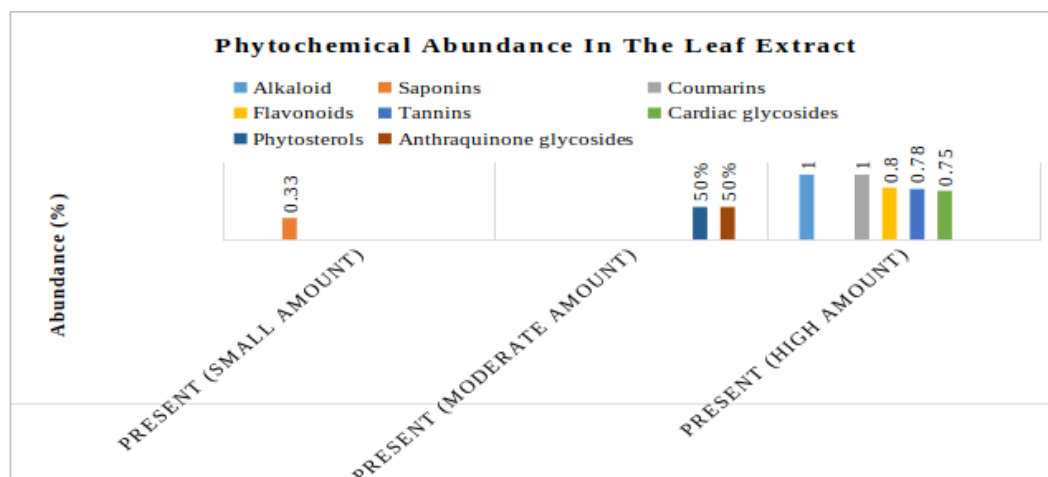


Figure 2: Abundance of Phytochemicals in *Senna occidentalis* Leaf Extract

Antibacterial Activity of *Senna occidentalis* Leaf Extract

The disk diffusion method was used to investigate the antibacterial activity of *Senna occidentalis* leaf extract, at different concentrations against *Salmonella paratyphi* - a common causative agent for gastroenteritis. For each concentration, two replicates for the zone of inhibition were included.

Table 3 below outlines the findings of diameters for zones of inhibition for each extract at each concentration of the plant extract. Notably, with increase in concentration of extract based on each of the three extraction solvents, there was subtle change in the diameter for the zones of inhibition.

Table 3:

Diameter for Zones of Inhibitions of Leaf Extract at Different Concentrations

Concentration (g/ml)	Plate 1 Zone (mm)	Plate2 Zone (mm)	Mean Zone of Inhibition (mm)
METHANOLIC EXTRACTION			
0.1 g/ml	6mm	6mm	6mm
0.2g/ml	6mm	6mm	6mm
0.3g/ml	6mm	6mm	6mm
0.6g/ml	8mm	9mm	8.5mm
0.8g/ml	6mm	6mm	6mm
1 g/ml	9mm	8mm	8.5mm
PETROLEUM ETHER EXTRACTION			
0.1 g/ml	9 mm	9 mm	9 mm
0.2 g/ml	6 mm	6 mm	6 mm
0.3 g/ml	8 mm	9 mm	8.5 mm
0.5 g/ml	9 mm	9 mm	9 mm
0.6 g/ml	10 mm	9 mm	9.5 mm
1 g/ml	6 mm	6 mm	6 mm
AQUEOUS EXTRACTION			
0.1 g/ml	7 mm	8 mm	7.5 mm
0.2 g/ml	6 mm	6 mm	6 mm
0.3 g/ml	6 mm	6 mm	6 mm
0.6 g/ml	6 mm	6 mm	6 mm
0.8 g/ml	6 mm	6 mm	6 mm
1 g/ml	8 mm	10 mm	9 mm
CIPROFLOXACIN DISK			
5 mcg/disk	32 mm	35 mm	33.5 mm

Figure 3 below shows a comparison of the mean zones of inhibition at each concentration, for the different extraction methods. Notably, extract from petroleum ether-based extraction had the highest mean zone of inhibition diameter in most of the different extract concentration levels examined.

This was followed secondly by methanol-based extract and finally aqueous based-extract. To note, at the very low concentration of lcompounds that possess gastro-protective activity in *Phyllanthus discoideus* leaves is recommended.

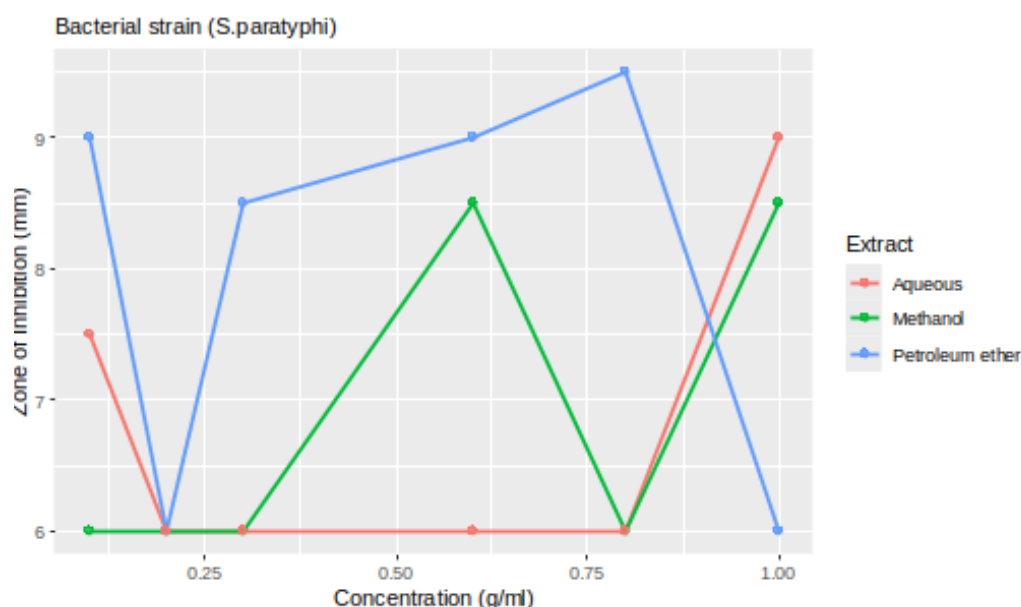


Figure 3: Leaf Extract-Mean Zone of Inhibition Against Salmonella Type at Different Extract Concentration

Comparison of Antibacterial Activity Between Extracts

To compare the antibacterial activity between extracts, we first checked if the data is normally distributed using Kolmogorov-Smirnov and Shapiro-Wilk tests. The test indicated that extraction solvent and concentration groups deviated significantly from a normal distribution ($p < 0.05$). Thus, non-parametric tests were used for inferential analysis. We used the Kruskal-Wallis test to check the effect of concentration on size of zone of inhibition.

Notably, the test showed a statistically significant difference in the sizes of zone of inhibition between difference concentrations as suggested in table 4 below. I.e., at least one concentration group differed significantly from the others. The Kruskal-Wallis test also indicated significant differences among the extraction solvents used denoting that solvent type significantly influences size of zone of inhibition.

Table 4:

Findings of the Independent-Samples Kruskal-Wallis Test

Independent-Samples Kruskal-Wallis Test		
Null hypothesis	Sig.	Decision
The distribution of zone of inhibition is the same across different extract concentrations	.009	Reject null hypothesis
The distribution of zone of inhibition is the same across leaf extract based on solvent used for extraction	.007	Reject null hypothesis

Correlation Between Extract Concentration and Antibacterial Effect

Since the data was not normally distributed, we used Spearman correlation analysis to correlate concentration with size of inhibition zone. The analysis showed no significant monotonic relation

between concentration and zone size ($p > 0.05$), i.e., higher or lower concentrations were not correlated with bigger or smaller sizes of zones of inhibition; as shown in figure 4 below.

Correlations			concentration_g/ml	zone_mm
Spearman's rho	concentration_g/ml	Correlation Coefficient	1.000	-.056
		Sig. (2-tailed)	.	.739
		N	38	38
	zone_mm	Correlation Coefficient	-.056	1.000
		Sig. (2-tailed)	.739	.
		N	38	38

Figure 4: Spearman Correlation Between Extract Concentration and Antibacterial Effect

Figure 5 below depicts a scatterplot showing relationship between zone of inhibition and concentration. Notably, there is a positive relationship between extract concentration and their corresponding zones of inhibition for the aqueous and methanol extract. Conversely, for the petroleum ether extract, the best line of fit for the scatter plot shows that as extract concentration increases, there is a slight negative correlation with the corresponding zone of inhibition.

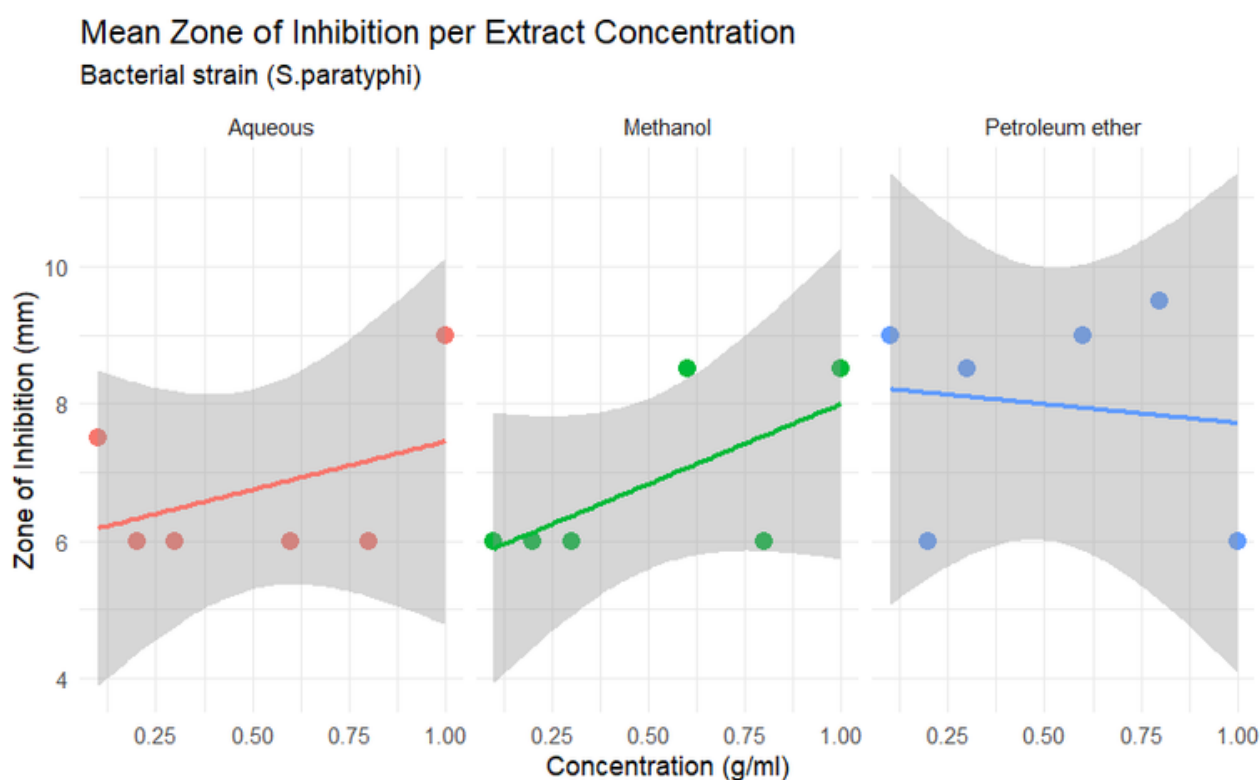


Figure 5: Scatter Plot Showing Relationship Between Extract Concentration and Zones of Inhibition of the Three Extracts

DISCUSSION

The study revealed that methanol extraction yielded the highest proportion of crude extract (29.15%), followed by water (13.78%) and petroleum ether (4.32%). This finding is consistent with previous studies where polar solvents such as methanol have been shown to yield higher

quantities of extractable compounds compared to non-polar solvents (Sadiq et al., 2012; Mohammed et al., 2012). The polarity of methanol enhances its ability to solubilize a wide spectrum of phytochemicals, including alkaloids, glycosides, and flavonoids (Musa et al., 2018). Conversely, the low yield from petroleum

ether extraction is attributable to its non-polar nature, which restricts the extraction mainly to lipophilic compounds, a finding echoed in similar extraction studies (El-Mahmood & Doughari, 2008). Critically, although methanol gave the highest yield, high yield does not necessarily correlate with high biological activity. Non-polar solvents such as petroleum ether may yield fewer compounds, yet those compounds can be more biologically potent. This highlights the need for bioassay-guided fractionation to distinguish between quantity and therapeutic relevance of extracts.

Phytochemical screening of *Senna occidentalis* leaves demonstrated the presence of alkaloids, tannins, coumarins, flavonoids, cardiac glycosides, anthraquinones, and phytosterols in varying quantities, with saponins present in low amounts, while phenolic compounds and terpenoids were absent. These results are consistent with findings by Sadiq et al. (2012), Idu et al. (2007) and Mohammed et al. (2012), who also identified alkaloids, anthraquinones, tannins, flavonoids, and glycosides in *S. occidentalis* extracts. Similarly, Adamu et al. (2018) reported that methanolic extracts were rich in alkaloids, anthraquinones, sterols, and flavonoids. However, some studies such as Evans et al. (2002) and Tamasi et al. (2021) have reported phenolic compounds in moderate amounts, indicating that phytochemical composition may vary depending on geographical location, environmental conditions, harvesting time, and solvent used (Faruq et al., 2006).

The high abundance of alkaloids, tannins, and flavonoids in this study is significant given their established antibacterial and anti-inflammatory properties (Edoga et al., 2005; Evans, 2009). Alkaloids act by disrupting bacterial DNA and protein synthesis, while flavonoids exert effects by inhibiting nucleic acid synthesis and disrupting cell membranes. Tannins, on the other hand, are known to exert antimicrobial effects by precipitating microbial proteins and forming complexes with bacterial cell walls, thereby inhibiting growth (Sallau et al., 2005; Shimada, 2006). Coumarins which were also abundant, have similarly been linked with antimicrobial activities via the inhibition of bacterial enzymes and interference with cell wall synthesis further supporting the therapeutic potential of *S. occidentalis* (Musa et al., 2017). The presence of these metabolites provides a pharmacological basis for the traditional use of *S. occidentalis* in managing diarrhea and related gastrointestinal disorders. From a critical standpoint, the absence of phenolics and terpenoids in this study could either reflect true absence or methodological limitations in extraction and detection. Since

phenolics are reported in other studies, it is possible that differences in extraction solvents or sensitivity of the screening methods influenced the results. This suggests that complementary analytical methods, such as HPLC or LC-MS, should be employed in future studies to provide a more accurate phytochemical profile.

The antibacterial activity of *S. occidentalis* extracts against *Salmonella paratyphi* revealed that petroleum ether extracts showed relatively stronger inhibition at lower concentrations, while aqueous extracts became more effective at higher concentrations (9mm at 1g/ml). Methanol extracts demonstrated moderate inhibition across concentrations (8.5mm at 0.6g/ml and 1g/ml). These findings correspond with those of Mohammed et al. (2012), who reported that petroleum ether and aqueous extracts displayed dose-dependent antibacterial activity against *E. coli* and *S. aureus*. Similarly, Musa et al. (2018) observed that ethanolic and petroleum ether extracts of *S. occidentalis* showed notable inhibitory effects against Gram-negative bacteria. On the other hand, these findings contrast with Adamu et al. (2018), who reported considerably higher inhibition zones (7–23 mm) against *S. typhi* and *S. paratyphi* A/B using methanolic and ethanolic extracts, with the methanolic extract giving the highest activity (23 mm). The mean inhibition zones recorded in this study (6–10 mm) were also lower than those reported by Sadiq et al. (2012), who found inhibition zones ranging between 12–18 mm against *Salmonella typhi* and *Shigella* spp. The weaker activity in the present study could be attributed to differences in extract concentration and extraction procedures, solvent polarity, possible loss of active compounds during preparation or bacterial strain variability. Notably, ciprofloxacin, the positive control, produced much larger inhibition zones (33.5 mm), underscoring the fact that while *S. occidentalis* has antibacterial potential, its crude extracts are less potent compared to standard antibiotics. The lack of a strong dose-response correlation, as revealed by Spearman's test is concerning because dose-dependence is a hallmark of reliable pharmacological activity. There was no significant monotonic relationship between concentration and inhibition zone, which suggests that activity may not increase proportionally with concentration, possibly due to compound saturation effects or antagonistic interactions between phytochemicals at certain doses. Importantly, the use of crude extracts in this study further illustrated the findings by Faruq et al. (2006), that fractionation and purification of *S. occidentalis* extracts can enhance antibacterial activity by isolating the most active constituents.

A key limitation of this study is that only crude extracts were evaluated, without successfully determining the Minimum Inhibitory Concentration (MIC) or Minimum Bactericidal Concentration (MBC), unlike Adamu et al. (2018) who reported MIC values as low as 62.5 µg/ml. In this study the extracts were extremely dark in color and hence turbidity was not visible while conducting MIC. Secondly, variations in solvent extraction efficiencies could have masked the true potential of the plant. Additionally, the antibacterial assays were performed in vitro and may not directly translate to in vivo efficacy, since factors such as metabolism, bioavailability, and toxicity could alter the activity of the phytochemicals in a living organism. Lastly, the study was limited to *Salmonella paratyphi*, and the results may not be generalizable to other causative agents of gastroenteritis. Future studies should incorporate fractionation, bioassay-guided isolation, and toxicity testing to better evaluate efficacy and safety. Additionally, advanced analytical techniques should be employed to enable visualization of turbidity in darker extracts especially when carrying out MIC. Despite these limitations, the findings provide a scientific basis for the traditional use of *S. occidentalis* in managing gastroenteritis. In regions with limited access to conventional drugs, herbal remedies such as *S. occidentalis* could offer a cost-effective complementary approach, provided further pharmacological and toxicological studies confirm their safety and efficacy.

The results support traditional claims of using *S. occidentalis* leaves for treating gastrointestinal disorders such as diarrhea and dysentery (Musa et al., 2017; Evans et al., 2002). Similar studies by Sadiq et al. (2012) and Mohammed et al. (2012) demonstrated antibacterial effects of ethanolic and aqueous extracts of *S. occidentalis* against *E. coli*, *Shigella*, *Staphylococcus aureus*, and *Salmonella typhi*. Our results therefore contribute further evidence of its broad-spectrum antibacterial potential, although the modest zones of inhibition observed here suggest that the crude extracts may be less potent compared to purified compounds or standard antibiotics.

Conclusion

The study successfully evaluated the efficacy of *Senna occidentalis* leaf extracts against *Salmonella paratyphi*, confirming its antibacterial potential. Phytochemical screening revealed the presence of alkaloids, tannins, flavonoids, coumarins, cardiac glycosides, and anthraquinones, compounds known for antimicrobial activity. Although the crude extracts exhibited measurable inhibition, their

potency was considerably lower compared to ciprofloxacin. These findings support the ethnomedicinal use of *S. occidentalis* in managing bacterial gastroenteritis and highlight its potential as a source of lead compounds for developing novel antibacterial agents.

Recommendations

From our findings, we recommend the following:

1. Bioassay-guided fractionation should be conducted to isolate and identify the specific active compounds responsible for antibacterial activity.
2. Perform in vivo studies to evaluate pharmacological efficacy, toxicity, and safety profiles.
3. Synergistic effects between *S. occidentalis* extracts and standard antibiotics to combat resistant bacterial strains should be investigated.
4. Advanced analytical techniques to be employed for detailed phytochemical characterization and visualization of turbidity in darker extracts for the case of MIC.

Conflict of Interest

The authors declare no conflict of interest.

Funding Sources

The study was privately funded by the members of the research team.

Authors' Contribution

G.E.N conceptualized the study, G.E.N, L.K.W and O.E.O handled data collection, cleaning and data analysis was performed by F.O. G.E.N wrote the first draft of the manuscript and all authors together with the supervisors M.N.W & T.S provided creative inputs during manuscript drafting and revisions. All authors read and approved the final manuscript.

REFERENCES

- Abd-Elmeged, G.M., Khairy, R.M., Abo-Eloyoon, S.M., & Abdelwahab, S.F. (2015). Changing patterns of drug-resistant *Shigella* isolates in Egypt. *Microbial Drug Resistance*, 21(3), 286-291. <https://doi.org/10.1089/mdr.2014.0187>
- Adamu, U., Abdulrazak, M. H., Yusha'u, M., & aslisu, B. (2018). Antibacterial potentials And toxicity study of *Cassia occidentalis* leaf extracts against clinical isolates of *Salmonella* sp. *Science World Journal*, 13(1), 77-81. <https://www.scienceworldjournal.org/article/view/18375>

- Alam, M. T., Karim, M. M., & Khan, S. N. (2009). Antibacterial Activity of Different Organic Extracts of *Achyranthes Aspera* and *Cassia Alata*. *Journal of Scientific Research*, 1(2), 393–398.
<https://doi.org/10.3329/jsr.v1i2.2298>
- Albrecht, L., Hartling, L., & Scott, S.D. (2017). Pediatric acute gastroenteritis: understanding caregivers' experiences and information needs. *Canadian Journal of Emergency Medicine*, 19(3), 198–206.
<https://doi.org/10.1017/cem.2016.363>
- Charles, K.M. (2009). Antimicrobial analysis of leaf extract *Cassia fistula*. *Journal of Ethnopharmacology*, 3(2):11–13
<https://doi.org/10.2174/0122150838270781231006184609>
- Collaborators GBD (2017). Estimates of global, regional & national morbidity, mortality and aetiologies of diarrheal diseases: a systemic analysis of Global Burden of Disease study 2015.
[https://doi.org/10.1016/S1473-3099\(18\)30362-1](https://doi.org/10.1016/S1473-3099(18)30362-1)
- Demissie, G.D., Yeshaw, Y., Alemine, W., & Akalu, Y. (2021). Diarrhea and associated factors among under five children in sub-Saharan Africa: evidence from demographic and health surveys of 34 sub-Saharan countries. *Plos one*, 16 (9), e0257522.
<https://doi.org/10.1371/journal.pone.0257522>
- Edeoga, H. O., Okwu, D. E., & Mbaebie, B. O. (2005). Phytochemical constituents of some Nigerian medicinal plants. *African Journal of Biotechnology*, 4(7), 685–688.
<https://doi.org/10.4314/ajb.v4i7.15167>
- Elliot, E.J., (2007). Acute gastroenteritis in children. *Bmj*, 334 (7583), 35–40.
<https://doi.org/10.1136/bmj.39036.406169.80>
- El-Mahmood, A.M., & Doughari, J.H. (2008). Phytochemical screening and antibacterial evaluation of the leaf and root extracts of *Cassia alata* Linn. *African Journal of Pharmacy and Pharmacology*, 2(7), 124–129.
<https://academicjournals.org/article/article1380819596>
- Evans, C.E., Banso, A., & Samuel, O.A. (2002). Efficacy of some nupe medicinal plants against *Salmonella typhi*: an in vitro study. *Journal of Ethnopharmacology*, 80 (1), 21–24.
[https://doi.org/10.1016/S0378-8741\(01\)00378-6](https://doi.org/10.1016/S0378-8741(01)00378-6)
- Evans, W.C. (2009). *Trease and Evans' pharmacognosy*. Elsevier Health Sciences.
- Faruq, U.Z., Hassan, L.G., Manga, B.S., Uba, A., Olatunji, M.K., Tal, M.T., & Isiak, F.S. (2006). Anti-bacterial activity of *Cassia occidentalis* in *Salmonella typhi*. In *Proceedings of the 28th Annual International Conference Chemicals Society of Nigeria* (pp 177–181).
[doi: 10.1111/j.1467-9280.2009.02454.x](https://doi.org/10.1111/j.1467-9280.2009.02454.x) Epub 2
- Freedman, S.B., Ali, S., Oleszczuk, M., Gouin, S., & Hartling, L. (2013). Treatment of Acute gastroenteritis in children: an overview of systematic reviews of interventions commonly used in developed countries. *Evidence- Based Child Health: A Cochrane Review Journal*, 8 (4), 1123–1137.
<https://doi.org/10.1002/ebch.1932>
- Garton, E.O., Ratti, J.T., & Giudice, J.H. (2005). Research and experimental design. Techniques for wildlife investigations and management. Sixth edition. The Wildlife Society, Bethesda, Maryland, USA, 43–71.
<https://cir.nii.ac.jp/crid/1573950399913040384>
- Guarino, A., Lo Vecchio, A., Dias, J.A., Berkley, J.A., Boey, C., Bruzzese, D., ... & Shimizu, T. (2018). Universal recommendations for the management of acute diarrhea in non-malnourished children. *Journal of pediatric gastroenterology and nutrition*, 67(5), 586–593.
<https://doi.org/10.1097/MPG.00000000000002053>
- Humphries, R.M., & Linscott, A.J. (2015). Practical guidance for clinical microbiology laboratories: diagnosis of bacterial gastroenteritis. *Clinical microbiology reviews*, 28(1), 3–31.
<https://doi.org/10.1128/cmr.00073-14>
- Humphries, R. M., Amber, J., Mitchell, S.L., Castanheira, M., Dingle, T., Hindler, J., ... & Sei, K. (2018). CLSI methods development and standardization working group best practices for evaluation of antimicrobial susceptibility tests. *Journal of clinical microbiology*, 56 (4), 10–1128.
<https://doi.org/10.1128/jcm.01934-17>
- Idu, M., Omonigho, S.E., & Igeleke, C.L. (2007). Preliminary investigation on the phytochemistry and antimicrobial activity of *Senna alata* L. flower. *Pakistan journal of biological sciences: PJBS*, 10(5), 806–809.
<https://doi.org/10.3923/pjbs.2007.806.809>

- Liu, L., Johnson, H.L., Cousens, S., Perin, J., Scott, S., Lawn, J.E., ... & Black, R.E. (2012). Global, regional, and national causes of child mortality: an updated systematic analysis for 2010 with time trends since 2000. *The Lancet*, 379 (9832), 2151-2161. [https://www.thelancet.com/journals/lancet/article/PIIS0140-6736\(12\)60560-1/abstract](https://www.thelancet.com/journals/lancet/article/PIIS0140-6736(12)60560-1/abstract)
- Mohammed, M., Aboki, M.A., Saidu, H.M., Victor, O., Tawakalitu, A., & Maikano, S.A. (2012). Phytochemical and some antimicrobial activity of *Cassia occidentalis* L. (Caesalpiniaceae). *International Journal of Science and Technology*, 2(4), 200-209. <https://www.semanticscholar.org/paper/Phytochemical-and-Some-Antimicrobial-Activity-of-L./ad2978c9ca61535474b0f03e4c36c417ce9d3d58>
- Muloi D., Fevre, E. M., Bettridge, J., Rono, R., Ong'are, D., Hassell, J. M., ... & Woolhouse, M. (2019). A cross-sectional survey of practices and knowledge among antibiotic retailers in Nairobi, Kenya. *Journal of global health*, 9(2), 020412. <https://doi.org/10.7189/jogh.09.020412>
- Musa, D.D., Aliero, A.A., & Bashir, K.A. (2017). Preliminary allelopathic activity of aqueous leaf extract of *Senna occidentalis* on the germination and early seedling growth of cowpea (*Vigna unguiculata*). *International Journal of Current Science and Studies*, 1(1), 23-25. <https://www.researchgate.net/profile/Abdulaziz-Bashir-kutawa/publication/321073883>
- Musa, D.D., Bashir, K.A., & Hassan, K.Y. (2018). Phytochemical screening and antibacterial activity of leaves extract of *Senna occidentalis* (L.) FUDMA Journal of Sciences, 2 (1), 59-65. <https://fjs.fudutsinma.edu.ng/index.php/fjs/article/view/1260>
- Omoro, R., O'Reilly, C.E., Williamson, J., Moke, F., Were, V., Farag, T.H., ... & Breiman, R.F. (2013). Health care-seeking behavior during childhood diarrheal illness: results of health care utilization and attitudes surveys of caretakers in western Kenya, 2007-2010. *The American journal of tropical medicine and hygiene*, 89(1 Suppl), 29-40. <https://doi.org/10.4269/ajtmh.12-0755>
- O'Ryan G, M., Ashkenazi-Hoffnung, L., O'Ryan-Soriano, M.A., & Ashkenazi, S. (2014). Management of acute infectious diarrhea for children living in resource-limited settings. *Expert Review of Anti-Infective Therapy*, 12 (5), 621-632. <https://doi.org/10.1586/14787210.2014.901168>
- Osano, B.O., Wang'ombe, J.K., Kamenwa, R.W., & Wamalwa, D. (2011). Cost analysis of care for children admitted to Kenyatta national hospital with rotavirus gastroenteritis. *Vaccine*, 29 (23), 4019-4024. <https://doi.org/10.1016/j.vaccine.2011.03.053>
- Owusu, D.N., Duah, H.O., Dwomoh, D., & Alhassan, Y. (2024). Prevalence and determinants of diarrhea and acute respiratory infections among children aged under five years in West Africa: evidence from demographic and health surveys. *International Health*, 97-106. <https://doi.org/10.1093/inthealth/ihad046>
- Rai, N., & Thapa, B. (2015). A study on purposive sampling method in research. *Kathmandu: Kathmandu School of Law*, 5(1), 8-15. <https://dlwqtxts1xzle7.cloudfront.net/48403395>
- Rhee, C., Aol, G., Ouma, A., Audi, A., Muema, S., Auko, J., ... & Verani, J.R. (2019). Inappropriate use of antibiotics for childhood diarrhea case management- Kenya, 2009-2016. *BMC Public Health*, 19, 1-12. <https://doi.org/10.1186/s12889-019-6771-8>
- Rouhani, S., Meloney, L., Ahn, R., Nelson, B.D., & Burke, T.F. (2011). Alternative rehydration methods: a systematic review and lessons for resource-limited care. *Pediatrics*, 127 (3), e748-e757. <https://doi.org/10.1542/peds.2010-0952>
- Sadiq, I., Shuaibu, M., Bello, A., Isah, S.T.A., Izuagie, T., Nasiru, S., & Kamaru, M. (2012). Phytochemistry and antimicrobial activities of *Cassia occidentalis* used for Herbal remedies. *Methods*, 18, 19-28. <https://www.researchgate.net/publication/284034861>

- Sallau A. B., Njoki G. C., Olokisi A. R., Wurochekke A. U., Abdulkadir A. A., Isah S., Abubakar M. S., and Ibrahim S., Effects of *Guiera senegalensis* leaf extracts on some *Echis carinatus* venom enzymes. *Journal of Medicinal Science*, 5: 2880-2883 (2005).
<http://repository.futminna.edu.ng:8080/jspui/handle/123456789/16394>
- Shimada T. 2006. Salivary proteins as a defense against dietary tannins. *J Chem Ecol* 32 (6): 1149-1163.
<https://doi.org/10.1007/s10886-006-9077-0>
- Simen-Kapeu, A., Bogler, L., Weber, A.C., Ntambi, J., Zagre, N.M., Vollmer, S., & Ekpini, R.E. (2021). Prevalence of diarrhea, acute respiratory infections and malaria over time (1995-2017): A regional analysis of 23 countries in West and Central Africa. *Journal of global Health*, 11, 13008.
<https://doi.org/10.7189/jogh.11.13008>
- Sohail, H., & Neupane, S. (2019). Prevalence of and factors associated with under-5 mortality in South Asia. *International Health*, 11(2), 119–127.
<https://doi.org/10.1093/inthealth/ihy065>
- Sultan, M.A., & Hassan, Z. (2018). Assessment of severity of acute gastroenteritis in the paediatric Pakistani population by Modified Vesikari Score. *J Pak Med Assoc*, 68(2), 159-164.
<https://d1wqtxts1xzle7.cloudfront.net/103005051/8547>
- Tam, C.C., O' Brien, S.J., Tompkins, D. S., Bolton, F.J., Berry, L., Dodds J., & ... IID2 Study Executive Committee. (2012). Changes in causes of acute gastroenteritis in the United Kingdom over 15 years: microbiologic findings from 2 prospective, population-based studies of infectious intestinal disease. *Clinical Infectious Diseases*, 54(9), 1275-1286.
<https://doi.org/10.1093/cid/cis028>
- Tamasi, A. A., Shoge, M. O., Adegboyega, T. T., & Chukwuma, E. C. (2021). Phytochemical analysis and in-vitro antimicrobial screening of the leaf extract of *Senna occidentalis* (Fabaceae). *Asian Journal of Natural Product Biochemistry*, 19(2).
<https://doi.org/10.13057/biofar/f190203>
- Troeger, C., Blacker, B.F., Khalil, I. A., Rao, P. C., Cao, S., Zimsen, S.R., ... & Reiner, R.C. (2018). Estimates of the global, regional, and national morbidity, mortality, and aetiologies of diarrhea in 195 countries: a systematic analysis for the Global Burden of Disease Study 2016. *The Lancet Infectious Diseases*, 18(11), 1211-1228.
[https://doi.org/10.1016/S1473-3099\(18\)30362-1](https://doi.org/10.1016/S1473-3099(18)30362-1)
- World Health Organization (WHO), 2013a. Diarrheal disease Fact Sheet. Available at: <http://www.who.int/mediacentre/factsheets/fs330/en>.
- World Health Organization (WHO), (2017). Diarrheal Disease fact sheet. Available at: <https://www.who.int/news-room/fact-sheets/detail/diarrheal-disease>.
- Zaidi, S.H., & Smith-Morris, C. (2015). Diapers in war zones: Ethnomedical factors in Acute childhood gastroenteritis in Peshawar, Pakistan. *PLoS One*, 10 (3), e0119069.
<https://doi.org/10.1371/journal.pone.0119069>