

**ANTIDIARRHOEAGENIC POTENTIALS OF SYNERGISTIC
ACTIVITIES OF WATER EXTRACTS OF *ALOE VERA* AND *HYPTIS
SUAVEOLENS* AGAINST *GIARDIA LAMBLIA* AND *SALMONELLA*
SPECIES AMONG 0-5 YEARS IN BAUCHI STATE, NIGERIA.**

ABSTRACT: Antidiarrhoeal activity was determined in the water extracts obtained from *Aloe vera* and *Hyptis suaveolens* plants against co-infections of *Giardia lamblia* and *Salmonella* species in 0-5years, antidiarrhoeal activity of *Salmonella* species was carried out by Kirby-Bauer methods and *in-vitro* susceptibility assays methods of *Giardia lamblia*, parasites mortality was determined using hemocytometer counter. Zone of inhibitions, parasites mortality as produced from the efficacies of these extracts were expressed as mean \pm SEM and the differences between means were analyzed and compared. The phytochemical constituents in these extracts, revealed the presence of saponins, tannins, alkaloids, flavonoids, terpenoids, alkaloids and phenols. The activity of water extracts of these plants singly and in combinations on *Salmonella* species, revealed antibacterial activity with good zone of inhibitions of *Aloe vera* extracts (0.302 \pm 18.00), *Hyptis suaveolens* extracts (0.315 \pm 19.67) and the combined *Aloe vera* and *Hyptis suaveolens* extracts (0.413 \pm 30.00). Also, on *Giardia lamblia*, revealed anti-giardial activity of *Aloe vera* extracts (0.002 \pm 0.505), *Hyptis suaveolens* extracts (0.002 \pm 0.478) and of the combined *Aloe vera* and *Hyptis suaveolens* extracts (0.002 \pm 0.643), as in all the cases, the plants, concentrations and time were determinants factors for the anti-giardial and antibacterial activity.

Keywords: Plant materials, *Giardia lamblia*, *Salmonella* species and Water extractions

INTRODUCTION

Plants remains one of the potential sources of effective agents against microbes including the deadly infections like HIV/AIDS, tuberculosis (caused by *Mycobacterium tuberculosis*), syphilis (caused by *Treponema pallidum*), gonorrhoea (*Neisseria gonorrhoea*), skin and wound infections (*Staphylococcus aureus*), diarrhoea (*Escherichia coli*), typhoid fever (*Salmonella typhus*) and *Pseudomonas aeruginosa* which directly infects the urinary tract, pulmonary tract, wounds, burns and also causes other blood infections [12]. The genus *Aloe* belongs to *Aloaceae* (Liliaceae) family which has around 360 to 400 different species [14]. *Aloe vera* leaf components have been credited for its antibacterial, antifungal, antiviral and anti-helminthic medicinal properties [14]. The plant *Hyptis suaveolens* (L.) commonly known as bush tea belonging to the family Lamiaceae which is a potent medicinal herb [2]. Pharmacological property of *Hyptis suaveolens* includes antioxidant, antimicrobial, antibacterial, anticancer, anti-inflammatory, antiseptic and anti-malarial [12]. Diarrhoea remains a major cause of mortality and morbidity among children under five years of age especially in developing countries [8]. The prevalence rate of acute diarrhoea in Nigeria is about 18.8%, one of the worst in sub-Saharan Africa [9]. Medicinal plants are widely used to treat different diseases in different parts of the world [1]. Giardiasis is a protozoan infections caused by *Giardia lamblia*; a flagellate protozoan, an infection principally of the upper small intestine and remains largely asymptomatic bringing on acute self-limited diarrhea [3]; [5]. Its occurrence is world-wide. Children are frequently more infected than adults. Prevalence is higher in area of poor sanitation in institutions with overcrowded human conditions and areas of children not toilet trained [7]. *Salmonella* is a genus

46 of enteric pathogens that consists of two species, *Salmonella enterica* and *Salmonella bongori*
47 [13]; [6], broad host range *Salmonella typhimurium* cause gastroenteritis in human and other
48 mammals [17].

49 This study is aim at evaluating the antimicrobial potentials of the water extracts of *Aloe vera* and
50 *Hyptis suaveolens* singly and in combinations against infections caused by *Salmonella* species
51 and *Giardia lamblia* in under five children and to determining their qualitative phytochemical
52 compositions.

53 MATERIALS AND METHODS

54 The plants were randomly collected in Jos, Plateau State, authenticated by the plant curator of
55 Federal College of Forestry, Jos, Nigeria. The design of the study is community and hospital
56 based which allows for the collections, laboratory isolations, identifications and culturing of
57 *Giardia lamblia* and *Salmonella* sp. occurring in both symptomatic and asymptomatic infections
58 among 0-5 years and the antimicrobial potentials of the crude extracts of *Aloe vera* and *Hyptis*
59 *suavolens* against them in Bauchi Metropolis. The air dried leaves of *Hyptis suaveolens* L. was
60 grounded into powder soaked in water for 72 hours, placed in Gallenkamp shaker rotating at 65
61 revolutions per minute, the contents were then homogenized and filtered using Whatman filter
62 paper no.1. The filtrate were poured into a round bottom flask and concentrated using a Buchi
63 Rotavapor R-200 to yield *Hyptis suaveolens* in required concentrates and also, the grounded
64 powder *Aloe vera* soaked in water in conical flasks and left to stand for 3days as reported by
65 [15]. Stool samples collected were placed in a clean disposable plastic tubes with tight fittings,
66 microscopically examined for *Giardia lamblia* cysts and trophozoites presence, positively
67 detected 50 mg of stool was inoculated immediately in an axenic medium for culture of *Giardia*
68 *lamblia* trophozoites. Also, *Salmonella* species, stool samples collected were inoculated within
69 two hours of collections onto selective and differential media: MacConkey (MAC) agar,
70 *Salmonella-Shigella* (SS) agar, and xylose lysine deoxycholate (XLD) agar, using a calibrated
71 inoculating loop in the spread plate method. The media were then incubated aerobically at 35°C
72 for 18 to 24 hours as described by [18] and [10], biochemical test was carried out according to
73 the methods described by [4] and [16]. The combined effect was determined by using same
74 solvent extractions of *Aloe vera* and *Hyptis suaveolens* making a combination in the ratio of 1:1.

75 RESULTS AND DISCUSSIONS

76 The results in table 1, shows the phytochemical constituents of water extracts of *Aloe vera* and
77 *Hyptis suaveolens*. All the plant extracts were found to contain saponins, tannins, alkaloids,
78 flavonoids, terpenoids, alkaloids, phenols.

79 **Table 1: Phytochemical Constituents of *Aloe vera* and *Hyptis suaveolens***

Name of Test	<i>Aloe vera</i> Extractions	<i>Hyptis suaveolens</i>
Extractions	Water	Water
Saponins	+	+
Tannins	+	+
Flavonoids	-	-
Terpenoids	-	-
Steroids	-	-
Cardiac glycosides	-	-
Anthraquinones	-	-
Alkaloid	-	-
(Wagner's test)		
Alkaloid	-	-
(Mayer's test)		

Phenolics

+

+

80 **Key:** (+) present, (-) absent

81 The results as presented in table 2, shows the mean efficacy of treatments and time of *Aloe vera*
 82 and *Hyptis suaveolens* on cultured *Giardia lamblia* trophozoite produced after 48 hours which
 83 was significantly (P=0.05) different after 48 hours reveals the highest mean value treatments and
 84 time, efficacy were found with water extractions of *Aloe vera* (0.002±0.505) and time
 85 (0.002±0.507) when compared with positive control (0.002±0.641), *Hyptis suaveolens*
 86 (0.002±0.478) and time (0.002±0.563) when compared with positive control (0.002±0.563).

87 **Table 2: Standard Error and Mean Efficacy of Treatments (*Aloe vera*) and (*Hyptis*
 88 *suaveolens*) and Time on Cultured *Giardia lamblia* Trophozoite**

89

S.E ± Mean Effects after 48 hours			
EXTRACTIONS		<i>Aloe vera</i>	<i>Hyptis suaveolens</i>
		Water	Water
Treatment	80mg	0.002±0.505 ^b	0.002±0.478 ^b
	70mg	0.002±0.430 ^c	0.002±0.445 ^c
	60mg	0.002±0.357 ^d	0.002±0.386 ^d
	50mg	0.002±0.260 ^e	0.002±0.263 ^e
	40mg	0.002±0.041 ^f	0.002±0.058 ^f
	-ve Ctrl	0.002±0.004 ^g	0.002±0.009 ^g
	+ve Ctrl	0.002±0.641 ^a	0.002±0.638 ^a
Time (Hours)	48	0.002±0.507 ^a	0.002±0.563 ^a
	40	0.002±0.456 ^b	0.002±0.452 ^b
	32	0.002±0.378 ^c	0.002±0.360 ^c
	24	0.002±0.314 ^d	0.002±0.300 ^d
	16	0.002±0.169 ^e	0.002±0.177 ^e
	8	0.002±0.093 ^f	0.002±0.101 ^f

90 Each value is a mean of ± standard error of three replicates. Mean followed by the same superscripts in a column are not significantly different from each other.

91 Table 3, presented the results of the mean efficacy of treatments and time of the combined *Aloe*
 92 *vera* and *Hyptis suaveolens* on cultured *Giardia lamblia* trophozoites produced after 48 hours,
 93 significant (P=0.05) after 48 hours, revealed highest means value treatments and time. Best
 94 efficacy was found (0.002±0.643) and time (0.002±0.719) when compared with the positive
 95 control (0.002±0.621).

96 **Table 3: Standard Error and Mean Efficacy of Treatments (Combined *Aloe vera* and
 97 *Hyptis suaveolens* Extracts) and Time on Cultured *Giardia lamblia* Trophozoites**

98

S.E ± Mean Effects after 48 hours		
EXTRACTIONS		Water
Treatment	80mg	0.002±0.643 ^a
	70mg	0.002±0.582 ^c
	60mg	0.002±0.497 ^d
	50mg	0.002±0.444 ^e

	40mg	0.002±0.309 ^f
	-ve Ctrl	0.002±0.011 ^g
	+ve Ctrl	0.002±0.621 ^b
Time (Hours)	48	0.002±0.719 ^a
	40	0.002±0.607 ^b
	32	0.002±0.495 ^c
	24	0.002±0.420 ^d
	16	0.002±0.272 ^e
	8	0.002±0.150 ^f

99 Each value is a mean of ± standard error of three replicates. Mean followed by the same
100 superscripts in a column are not significantly different from each other.

101
102 The results as tabulated in table 4, shows the mean efficacy of inhibition zones of treatments of
103 *Aloe vera* and *Hyptis suaveolens* on cultured *Salmonella* species, was significantly (P=0.05)
104 different which shows zones of inhibitions of water extractions of *Aloe vera* (0.302±18.00) when
105 compared to control (0.302±29.50) and *Hyptis suaveolens* (0.315±19.67) when compared with
106 positive control (0.315±29.50).

107 **Table 4: Standard Error and Mean Efficacy of Inhibition Zone Diameters of Treatments**
108 **of *Aloe vera* and *Hyptis suaveolens* on Cultured *Salmonella* species**

109 S.E ± Mean Effects after 48 hours

EXTRACTIONS		<i>Aloe vera</i>	<i>Hyptis suaveolens</i>
		Water	Water
Treatment	80mg	0.302±18.00 ^b	0.315±19.67 ^c
	70mg	0.302±15.00 ^c	0.315±17.33 ^c
	60mg	0.302±12.67 ^d	0.315±14.83 ^d
	50mg	0.302±9.167 ^e	0.315±12.00 ^e
	40mg	0.302±7.333 ^f	0.315±9.167 ^f
	-ve Ctrl	0.302±0.667 ^g	0.315±0.000 ^g
	+ve Ctrl	0.302±29.50 ^a	0.315±29.50 ^a

110 Each value is a mean of ± standard error of three replicates. Mean followed by the same
111 superscripts in a column are not significantly different from each other.

112
113 The results tabulated in table 5, shows the mean efficacy of inhibition zones of treatments of the
114 combined *Aloe vera* and *Hyptis suaveolens* on cultured *Salmonella* species, was significantly
115 (P=0.05) different which shows zones of inhibitions of water extractions of the combined *Aloe*
116 *vera* and *Hyptis suaveolens* (0.413±30.00) when compared to control (0.413±30.00).

117 **Table 5: Standard Error and Mean Efficacy of Inhibition Zone Diameters of Treatments**
118 **of the Combined *Aloe vera* and *Hyptis suaveolens* on Cultured *Salmonella* species**

119 S.E ± Mean Effects after 48 hours

EXTRACTIONS		Water
Treatment	80mg	0.413±30.00 ^a
	70mg	0.413±28.00 ^b
	60mg	0.413±24.67 ^c

50mg	0.413±22.50 ^d
40mg	0.413±18.67 ^e
-ve Ctrl	0.413±0.667 ^f
+ve Ctrl	0.413±30.00 ^a

120 Each value is a mean of ± standard error of three replicates. Mean followed by the same
 121 superscripts in a column are not significantly different from each other.

122 CONCLUSION

123 Based on the findings of this research work, water extracts of *Hyptis suaveolens*, *Aloe vera* and
 124 of the combined all exhibited good activity on *Giardia lamblia* and *Salmonella* species. Hence,
 125 water extracts of *Aloe vera* has a good antibacterial activity, *Hyptis suaveolens* has a better
 126 activity and the combined thus shows the best antibacterial activity on *Salmonella* species. Also,
 127 on *Giardia lamblia*, *Aloe vera* has better activity, *Hyptis suaveolens* has good activity and the
 128 combined extracts performed best. Therefore, these plants possess antimicrobial potentials.
 129 There was the presence of phytochemicals in these plant extracts, it is thus concluded that these
 130 plants are promising and are very important antidiarrhoeogenic agents.

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