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In vitro Antimicrobial Activity of the Flower Buds of Eugenia caryophyllata

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Authors' contributions

This whole work was carried out in collaboration among all authors. Authors FCMR, GA and WW initiated the study, designed the protocol, carried out the test and analyzed the data. Author WW drafted the first manuscript. All authors made significant contributions to the development of the manuscript. The work is part of MSc thesis of author GA.

Original Research Article

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ABSTRACT

Aims: This study investigated the antimicrobial effect of aqueous and ethanol extracts of the flower buds of *Eugenia caryophyllata* (Myrtaeae) against a wide range of bacteria and yeasts cells isolated clinically from patients.

Methodology: The agar diffusion method was used to establish the antimicrobial activity and the zones of inhibition caused by the extracts. The antimicrobial effects of 16% and 32% aqueous and ethanol extracts of *Eugenia caryophyllata* were investigated against 111 pathogenic bacteria and yeasts cells. The microbes used consisted of 11 *Proteus mirabilis,* 20 *Salmonella typhi,* 15 *Pseudomonas aeruginosa,* 18 *Escherichia coli,* 19 *Staphylococcus aureus,* 12 *Klebsiella pneumoniae,* and 16 *Candida albicans* species.

Results: The ethanol extracts inhibited the growth of all the microbes employed in the study with inhibition zones ranging from 8.00 ± 0.00 mm to 24.00 ± 0.00 mm. The aqueous extracts however exhibited different degrees of antimicrobial activity with zones of inhibition ranging from 6.00 ± 0.00 mm to 13.33 ± 0.29 mm.

Conclusion: Our study concludes that the aqueous and ethanol extracts of the flower

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buds of *Eugenia caryophyllata* have relatively good antimicrobial activity against a wide range of medically important pathogenic bacteria and *Candida albicans in vitro*.

Keywords: In vitro; Eugenia caryophyllata; antimicrobial.

1. INTRODUCTION

Infectious diseases remain a public health challenge globally. The situation is however worse in developing countries where the burden of infectious diseases is high. The discovery of antimicrobial agents has contributed greatly to ameliorating the health burden posed by infectious agents. However, this has been met with a new challenge of resistance by these agents [1-3].

The battle against microbial resistance to antimicrobial agents has persisted for decades. Among the factors contributing to microbial resistance are indiscriminate use of antimicrobial agents by both healthcare professionals and patients, and the production of substandard antimicrobials [4-5]. This public health challenge has initiated a global revolution into the search for new antimicrobials and the discovery of alternate ways of developing antimicrobial agents. A critical area targeted for the hunt for new antimicrobials is the search into natural products, particularly botany [6-9].

The importance of medicinal plants in the history of man cannot be over emphasized, particularly in the science of traditional medicine. Over 2600 plants have been archived out of which approximately 700 have known medical use [10-11]. There are reports on the curative potentials or abilities of medicinal plants and their products in the treatment of a wide range of infectious ailments such as urinary tract, gastrointestinal tract, respiratory tract and wound infections [12-14]. Even in the era of modern science, traditional medicine of which plant medicine is an integral part remains the sole source of medical intervention for millions of people living in remote regions of the globe. The demand for medicinal plants has seen a gradual increase in recent years following claims that drugs derived from natural products such as plants are safer and less harmful.

One of such plants is *Eugenia caryophyllata* (Myrtaeae), a small evergreen plant found in Ghana, Madagascar, Moluccas and Nigeria. The flower bud of the plant is reported to be efficacious in the treatment of diarrhoea, dyspepsia, pulmonary and urinary tract infections, sores and infected wounds as well as toothaches [15]. Following these claims, we deemed it important to investigate *in vitro* the effectiveness of the aqueous and ethanol extracts of the flower buds of *Eugenia caryophyllata* against a wide range of clinically isolated bacteria and yeast which have the potential of being implicated in the diseases the rural folks in the Eastern region of Ghana claim to treat with the plant extract.

2. METHODOLOGY

2.1 Experimental Materials

A Taxonomist at the Plant Development Department of the Centre for Scientific Research into Plant Medicine (CSRPM) at Mampong-Akwapim in Ghana aided in identification of the flower buds of *Eugenia caryophyllata*. Voucher specimen (#CSRPM No. 0706) was stored in the herbarium of CSRPM.

2.2 Test Organisms Used

The test organisms used in this study were clinical isolates consisting 11 *Proteus mirabilis*, 20 *Salmonella typhi*, 15 *Pseudomonas aeruginosa*, 18 *Escherichia coli*, 19 *Staphylococcus aureus*, 12 *Klebsiella pneumoniae*, and16 *Candida albicans* species.

2.3 Aqueous Extract Preparation

One kilogram (1kg) of dried pulverized flower buds of *Eugenia caryophyllata* was soaked in 1L of distilled water for 30 minutes. The mixture was subsequently boiled for 30 minutes and then allowed to simmer under reduced heat. The filtrate was lyophilized and the freeze dried sample reconstituted 16% and 32% using sterile distilled water. The reconstituted samples were stored in a refrigerator at 4°C until needed.

2.4 Ethanol Extract Preparation

One kilogram (1kg) of dried pulverized flower buds of *Eugenia caryophyllata* was macerated in 1L of ethanol for two days (48 hours). The resultant extract was filtered and concentrated using rotary evaporator and later freeze dried. The freeze dried sample was reconstituted 16% and 32% using sterile 20% dimethyl sulphoxide (DMSO). These were stored in a refrigerator at 4°C until needed.

2.5 Antimicrobial Activity of the Reconstituted Formulations

The agar diffusion method was used to investigate the antibacterial properties of each of the reconstituted four extracts (16% aqueous, 32% aqueous, 16% ethanol and 32% ethanol extracts) of Eugenia caryophyllata, as described by the National Committee for Clinical Laboratory Standards and the National Center for Infectious Disease, Center for Disease Control and Prevention [16-17]. The entire surface of Mueller-Hinton agar plate was seeded with the test organism within 15 minutes by the swabbing technique after adjusting the turbidity of the inoculum suspension equivalent to 0.5 McFarland standard. A sterile cock borer of an internal diameter of 4 mm was used to punch five wells in the medium, and the plant extracts aseptically dispensed into the respective labeled wells. Disc of Standard drug chloramphenicol 30 µg/disc used as positive control was overlayed on the Mueller-Hinton agar plates, while 20% v/v DMSO was used as negative control. Triplicates of each plate were made and the procedure repeated for all the isolates. The plates were kept in the refrigerator at 4°C for 4 hours to ensure complete diffusion of the extract before incubating at 37°C for 3 days (for bacteria). The same procedure was applied for C. albicans but on Sabouraud dextrose agar plates and incubated at 28°C for 7 days. After the incubation period, the diameter of each zone of inhibition was measured in millimeters (mm) with a standard ruler.

3. RESULTS

The 32% aqueous extract and the two ethanol extracts (16% and 32%) exhibited 100% inhibitory activity against all the *C. albicans* isolates (Table 1). Also 100% inhibitory activity was observed among *S. aureus* isolates by all the four different extracts (Table 2). Three out of 18 *E. coli* isolates were not susceptible to the 16% aqueous extract while the remaining extracts showed 100% inhibitory activity (Table 5). All the four extracts also registered 100% inhibitory activity against all 20 *S. typhi* isolates (Table 7) as well as all the 11 *P. mirabilis*

isolates (Table 6). All the 15 *P. aeruginosa* isolates were inhibited by the ethanol extracts. Three (3) out of 15 and 4 out of 15 isolates were however not susceptible to the 16% and 32% aqueous extracts respectively (Table 4). Five (5) out of 12 and 1 out of 12 *K. pneumoniae* isolates were not susceptible to the 16% and 32% aqueous extracts respectively. The ethanol extracts however inhibited all the 12 isolates (Table 3).

4. DISCUSSION

Scientists quest to discover alternate ways of developing new antimicrobials for therapeutic purposes keep increasing as reports on microbial resistance to existing antimicrobials continue to soar [18-20]. Amidst other interventions, most scientists believe employing modern scientific research methods to investigate historic plants of medicinal importance could contain the key to unravel novel antimicrobial compounds [21-22]. It is this same believe that motivated us to investigate the antimicrobial efficacy of *Eugenia caryophyllata* against clinically isolated human pathogens consisting 11 *Proteus mirabilis*, 20 *Salmonella typhi*, 15 *Pseudomonas aeruginosa*, 18 *Escherichia coli*, 19 *Staphylococcus aureus*, 12 *Klebsiella pneumoniae*, and 16 *Candida albicans*. The present study revealed varying degrees of antimicrobial activity by both the aqueous and ethanol extracts used.

The 16% aqueous extract of *Eugenia caryophyllata* inhibited the growth of 14 out of 16 *Candida albicans* isolates while the 32% inhibited the growth of all the isolates (100%) with zones of inhibition ranging from 6.83 ± 0.76 mm to 13.33 ± 0.29 mm. The 16% as well as the 32% ethanol extracts inhibited the growth of all the *C. albicans* isolates used in the study with zones of inhibition ranging from 8.67 ± 0.29 mm to 21.50 ± 0.50 mm (for the 16% extract) and 8.83 ± 0.29 mm to 23.00 ± 1.00 mm (for the 32% extract) Table 1. It has been reported that essential oils from *Eugenia caryophyllata* possess strong antifungal activity against a wide range of fungi as well as *C. albicans* [23]. The potential of the plant extract in the treatment of mouth candidiasis as well as toothaches as claimed by traditional folks could be justified, following reports that its essential oils are used in dentistry as anodyne [24-26].

Both the aqueous and ethanol extracts inhibited the growth of all the 19 S. *aureus* isolates employed in the investigation. The aqueous extract however exhibited inhibition zones ranging from 6.83 ± 0.29 mm to 10.33 ± 0.29 mm (for the 16% extract) and 6.67 ± 0.29 mm to 11.17 ± 0.29 mm (for the 32% extract) while the ethanol extract showed inhibition zones ranging from 7.83 ± 0.29 mm to 16.83 ± 0.76 (for the 16% extract) and 9.83 ± 0.29 mm to 19.33 ± 0.58 mm (for the 32% extract) Table 2. The outcome of the present study commensurates with similar works, which reported on Gram positive bacteria, particularly S. *aureus* [27-28]. Most plants extracts are efficacious against Gram positive bacteria and this has been attributed to the morphological differences in cell wall existing between Gram positive and Gram negative bacteria [29-30]. Claims that the plant extract is used by traditional healer to manage various forms of abscesses could be justified.

The 16% aqueous extract inhibited the growth of 7 out of 12 *K. pneumoniae* isolates with zones of inhibition ranging from 6.20 ± 0.35 mm to 9.33 ± 0.29 mm while the 32% extract inhibited 11 out of 12 with inhibition zones ranging from 6.83 ± 0.29 mm to 8.00 ± 0.00 mm. Both the 16% and 32% ethanol extracts inhibited the growth of all the 12 *K. pneumoniae* isolates employed in the study, with zones of inhibition ranging from 8.00 ± 0.00 mm to 9.33 ± 0.29 mm and 8.17 ± 0.29 mm to 10.17 ± 0.29 mm for the 16% and 32% extract respectively, (Table 3). The present study seems to disagree with Negero et al. [23] who reported in their work the resistance of *K. pneumoniae* and *E. coli*. Even though capsulated bacteria are mostly resistant against antimicrobials, our study seems to suggest the

presence of potent compound(s) in the extracts which is/are active against usually capsulated bacteria such as *K. pneumoniae*.

Isolate code	Zone of inhibition (mm)			
	16% Aq extract	32% Aq. extract	16% EtOH extract	32% EtOH extract
H/2029	10.33±0.29	13.33±0.29	14.50±0.50	16.00±1.00
H/2030	6.83±0.76	9.83±0.29	8.83±0.29	10.00±0.00
H/2031	7.83±0.29	8.33±0.29	9.33±0.58	12.17±0.29
H/2051	8.17±0.76	8.83±0.29	9.17±0.29	10.00±0.00
H/2052	8.50±0.50	13.17±0.29	10.67±0.29	16.00±0.00
H/2054	13.33±0.38	9.50±1.00	14.67±0.29	15.67±0.58
H/2057	8.00±0.00	7.67±0.58	17.33±0.58	15.33±0.58
H/2058	10.50±0.50	10.50±0.50	21.50±0.50	23.00±1.00
H/2062	8.33±0.29	9.33±0.29	10.00±0.00	10.17±0.29
H/2072	8.00±0.10	11.17±0.29	8.83±0.29	9.00±0.00
H/2074	7.33±0.29	8.33±0.29	9.00±0.50	8.83±0.29
H/2085	7.83±0.29	10.83±0.29	9.00±0.00	11.83±0.29
H/2087	N/S	7.83±0.29	8.83±0.29	9.17±0.29
H/2088	N/S	11.00±0.00	8.67±0.58	9.00±0.00
H/2094	N/S	8.50±0.00	8.67±0.29	25.33±0.58
H/2102	9.00±0.00	9.17±0.29	11.17±0.29	15.50±0.00

Table 1. Susceptibility of Candida albicans to Eugenia caryophellata extracts

Key: N/S= Not susceptible, Aq= Aqueous, EtOH =Ethanol

	Table 2. Susceptibility of S	Staphvlococcus aureus to	Eugenia caryophellata extracts
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Isolate code	Zone of inhibition (mm)			
	16% Aq extract	32% Aq. extract	16% EtOH extract	32% EtOH extract
B/10400	6.83±0.29	8.33±0.29	7.83±0.29	11.00±0.00
B/10800	7.83±0.29	8.17±0.29	9.83±0.29	12.33±0.58
B/10820	10.17±0.29	10.33±0.29	10.00±0.00	11.00±0.00
B/10824	7.17±0.29	8.00±0.50	13.17±1.04	13.17±0.29
B/10824	6.83±0.29	8.00±0.50	13.00±0.00	14.17±1.04
B/10870	10.33±0.29	11.17±0.29	9.67±0.58	17.33±1.04
B/10956	8.83±0.29	9.50±0.50	13.00±0.00	10.17±0.29
B/8055	8.33±0.29	9.67±0.76	9.83±0.29	12.17±0.29
B/8074	8.33±0.29	7.67±0.76	10.33±0.58	9.83±0.29
B/8180	8.00±0.50	8.33±0.29	9.83±0.29	11.67±0.58
B/8183	10.00±0.50	9.33±0.29	12.67±0.58	13.00±0.00
B/9055	9.67±0.58	9.33±0.29	12.83±0.76	10.00±0.00
B/9829	9.00±0.50	6.67±0.29	14.17±0.29	9.67±0.58
B9763	10.17±0.29	9.00±0.50	16.83±0.76	11.67±0.58
U/2004	7.33±0.29	7.17±0.58	10.00±0.00	11.67±0.58
U/2006	8.17±0.29	8.00±0.50	10.83±0.29	10.33±0.58
W/2005	7.33±0.29	10.50±0.00	10.00±1.00	10.17±0.29
W/2036	9.50±0.00	10.33±0.29	11.83±0.29	15.00±0.00
W/2055	9.00±0.00	9.50±0.00	10.17±0.29	19.33±0.58

Key: Aq= Aqueous, EtOH =Ethanol

Isolate code	Zone of inhibition (mm)			
	16% Aq extract	32% Aq. extract	16% EtOH extract	32% EtOH extract
B/10034	8.33±1.04	7.83±0.29	8.00±0.00	9.17±0.29
B/10036	6.20±0.35	7.83±0.29	8.33±0.29	9.50±0.00
B/9082	8.00±0.00	6.83±0.29	9.33±0.29	8.33±0.29
B/9970	9.33±0.29	8.00±0.00	9.17±0.29	10.17±0.29
U/1190	7.50±0.50	7.00±0.50	8.33±0.29	9.00±0.00
U/2043	N/S	7.00±0.00	8.17±0.29	9.17±0.29
U/2058	N/S	6.83±0.29	9.00±0.00	9.17±0.29
U/2473	8.00±0.50	7.50±0.00	9.00±0.00	8.17±0.29
U/2667	N/S	7.00±0.50	9.33±0.29	8.33±0.29
U/3324	9.00±0.50	7.50±0.50	9.17±0.29	8.17±0.29
U/3443	N/S	N/S	8.17±0.29	10.17±0.29
W/2046	8.33±0.29	7.00±0.50	8.00±0.00	8.50±0.00

Table 3. Susceptibility of Klebsiella pneumoniae to Eugenia caryophellata extracts

Key: N/S= Not susceptible, Ag= Aqueous, EtOH =Ethanol

All the 15 *P. aeruginosa* were inhibited in growth by both the 16% and 32% ethanol extracts with zones of inhibition ranging from 8.17 ± 0.29 mm to 24.00 ± 0.00 mm. Greater inhibitions were however associated with the 32% extract. The 16% aqueous extract inhibited 3 out of 15 *P. aeruginosa* isolates used with zones of inhibition ranging from 7.00 ± 0.50 mm to 8.00 ± 0.50 mm, while the 32% extract inhibited 11 out of 15 with inhibition zones ranging from 7.00 ± 0.50 mm to 10.00 ± 0.50 mm, (Table 4). *P. aeruginosa*, an Estended Spectrum Beta-Lactamase (ESBL) producer found not susceptible to the aqueous extract but susceptible to the ethanol extract affirms similar work by Sabahat and Perween who reported that *K. ozaenae*, *K. pneumoniae*, *S. marcescens*, *S. typhi*, *S. dysentriae* and *V. cholera* were resistant to aqueous infusion and decoction while essential oil showed strong antibacterial activity against all bacterial isolates tested [31]. Probably the ethanol is capable of extracting more of such oils from the plant, hence the relatively high antimicrobial activity observed.

Three out of 18 *E. coli* isolates employed in the study were non-susceptible to the 16% aqueous extract. Fifteen (15) out of 18 were however susceptible, exhibiting inhibition zones ranging from 6.33 ± 0.29 mm to 9.00 ± 0.50 mm. On the contrary, the 32% aqueous extract inhibited the growth of all the 18 *E. coli* isolates with inhibition zones ranging from 6.00 ± 0.00 mm to 9.50 ± 1.00 mm. Both the 16% and 32% ethanol extracts also exhibited 100% activity against all the 18 *E. coli* isolates with inhibition zones ranging from 8.17 ± 0.58 mm to 12.33 ± 0.29 mm, (Table 5). The susceptibility of *E. coli* (the commonest cause of urinary tract infection) [28-29,32] to the extracts probably confirms the use of the plant in the treatment of some infections of the urinary tract. The plant has also been proven to be potent against diarrhoea causing bacteria such as *E. coli* [32-33].

Both the aqueous and ethanol extracts exhibited 100% activities against all the 11 *P. mirabilis* isolates. The aqueous extract however showed narrow range of inhibition zones ranging from 6.67 ± 0.29 mm to 10.33 ± 0.76 mm while the ethanol extract showed inhibition zones ranging from 9.67 ± 0.27 mm to 16.67 ± 0.76 mm, Table 6. *P. mirabilis* is among bacteria which cause urinary tract infection as well as wound infection [32]. It is therefore convincing if tradition/herbal healers used the plant in treating such infections as revealed by the present study.

Isolate code	Zone of inhibition (mm)			
	16% Aq extract	32% Aq. extract	16% EtOH extract	32% EtOH extract
B/7875	N/S	N/S	15.17±0.29	18.50±0.87
B/8228	N/S	8.33±0.29	14.00±0.00	13.00±0.00
B/8425	7.50±0.50	9.50±0.00	15.33±0.58	17.67±0.58
B/8518	N/S	7.33±0.29	17.17±0.29	19.83±0.29
B/9496	N/S	N/S	14.50±0.50	17.33±0.29
B/9510	N/S	7.83±0.29	15.83±0.76	18.17±0.29
U/1950	7.00±0.50	7.00±0.50	8.17±0.29	8.83±0.29
U/2038	N/S	N/S	16.50±0.00	18.00±0.00
U/2448	8.00±0.50	10.00±0.50	15.33±0.29	15.67±0.58
U/2660	N/S	7.50±0.50	11.83±0.29	13.17±0.58
U/3483	N/S	N/S	12.17±0.29	16.00±0.00
W/1950	N/S	7.83±0.29	14.33±0.58	15.00±1.00
W/1951	N/S	7.17±0.27	16.00±0.00	20.17±0.29
W/2007	8.00±0.50	9.00±0.50	18.00±0.00	23.33±0.58
W/2246	N/S	9.17±0.58	17.17±0.29	24.00±0.00

Table 4. Susceptibility of Pseudomonas aeruginosa to Eugenia caryophellata extracts

Key: N/S= Not susceptible, Aq= Aqueous, EtOH =Ethanol

Isolate code	Zone of inhibition (mm)			
	16% Aq extract	32% Aq. extract	16% EtOH extract	32% EtOH extract
B/10125	7.83±0.29	6.83±0.29	9.67±0.76	9.50±0.50
B/10128	6.33±0.29	7.17±0.29	8.00±0.00	9.17±0.58
B/10186	7.00±0.50	7.33±0.29	8.17±0.58	10.50±0.50
B/8082	7.00±0.00	8.00±0.50	9.00±0.00	9.00±0.00
B/9259	6.50±0.50	7.00±0.00	9.00±0.00	9.83±0.29
B/9260	7.00±0.50	7.50±0.50	9.00±0.00	9.83±0.29
B10905	7.17±0.58	6.00±0.00	9.00±0.00	9.00±0.00
U/2671	8.00±0.00	6.17±0.29	9.00±0.00	10.00±0.00
U/2797	N/S	8.83±0.29	8.33±0.29	9.67±0.76
U/2845	N/S	6.50±0.50	8.50±0.50	9.83±0.29
U/3080	9.00±0.50	7.83±0.29	8.17±0.58	9.83±0.29
U/3242	8.83±0.29	9.50±1.00	11.67±0.76	12.33±0.29
U/3372	7.83±0.29	5.83±0.29	10.00±0.00	12.00±0.00
U/3374	7.50±0.50	7.33±0.29	10.00±0.00	10.17±0.58
U/3387	6.83±0.29	7.17±0.76	8.00±0.00	10.17±0.58
U/3392	N/S	9.00±0.50	8.33±0.29	9.00±0.00
U/3393	7.00±0.50	7.17±0.29	8.17±0.58	10.33±0.29
U/6065	8.17±0.29	7.67±0.76	9.83±0.29	11.00±0.00

Key: N/S= Not susceptible, Aq= Aqueous, EtOH =Ethanol

All the 20 S. *typhi* isolates were susceptible to both the aqueous and ethanol extracts. The aqueous extract exhibited inhibition zones ranging from 7.00 ± 0.10 mm to 9.50 ± 0.50 mm, while the ethanol extract showed zones of inhibition ranging from 8.00 ± 1.00 to 11.83 ± 0.76 mm, asshown in Table 7. These results confirm work done by Rahman et al. [34] who reported that extract from *Eugenia caryophyllata* buds screened against some diarrhoea causing bacteria demonstrated strong antimicrobial activity against *S. typhimurium* and

Shigella dysenteriae. Similar report by Sabahat and Perween, and Bishnu et al. also justify the susceptibility of *S. typhi* to *Eugenia caryophyllata* extract as observed in this study [27,31].

Zone of inhibition (mm)			
16% Aq extract	32% Aq. extract	16% EtOH extract	32% EtOH extract
9.50±0.50	10.33±0.76	9.67±0.76	11.67±0.76
8.17±0.76	9.83±0.29	9.67±0.76	9.83±1.26
6.67±0.29	8.00±1.00	9.83±1.26	11.83±0.76
8.83±0.76	7.17±0.76	10.00±0.00	11.00±0.00
9.50±0.50	7.50±0.50	10.83±1.26	12.00±0.00
8.00±0.00	6.00±1.00	11.33±0.76	14.17±0.76
9.00±0.00	8.17±1.04	16.67±0.76	16.17±0.76
8.17±0.76	7.67±0.76	11.17±0.27	15.67±0.76
8.00±1.00	8.33±0.58	11.67±0.76	14.00±1.00
8.83±0.27	7.67±0.58	11.00±0.00	12.17±0.76
7.67±0.58	7.83±1.26	9.83±0.27	10.17±0.76
	$\begin{array}{c} 9.50\pm0.50\\ 8.17\pm0.76\\ 6.67\pm0.29\\ 8.83\pm0.76\\ 9.50\pm0.50\\ 8.00\pm0.00\\ 9.00\pm0.00\\ 8.17\pm0.76\\ 8.00\pm1.00\\ 8.83\pm0.27\\ 7.67\pm0.58\end{array}$	16% Aq extract32% Aq. extract9.50±0.5010.33±0.768.17±0.769.83±0.296.67±0.298.00±1.008.83±0.767.17±0.769.50±0.507.50±0.508.00±0.006.00±1.009.00±0.008.17±1.048.17±0.767.67±0.768.00±1.008.33±0.588.83±0.277.67±0.587.67±0.587.83±1.26	16% Aq extract32% Aq. extract16% EtOH extract 9.50 ± 0.50 10.33 ± 0.76 9.67 ± 0.76 8.17 ± 0.76 9.83 ± 0.29 9.67 ± 0.76 6.67 ± 0.29 8.00 ± 1.00 9.83 ± 1.26 8.83 ± 0.76 7.17 ± 0.76 10.00 ± 0.00 9.50 ± 0.50 7.50 ± 0.50 10.83 ± 1.26 8.00 ± 0.00 6.00 ± 1.00 11.33 ± 0.76 9.00 ± 0.00 8.17 ± 1.04 16.67 ± 0.76 8.17 ± 0.76 7.67 ± 0.76 11.17 ± 0.27 8.00 ± 1.00 8.33 ± 0.58 11.67 ± 0.76 8.83 ± 0.27 7.67 ± 0.58 11.00 ± 0.00

Table 6. Susceptibility of Proteus mirabilis to Eugenia caryophellata extracts

Key: Aq= Aqueous, EtOH =Ethanol

Table 7. Susceptibility of Salmonella typhi to Eugenia caryophellata extracts

Isolate code	Zone of inhibition (mm)			
	16% Aq extract	32% Aq. extract	16% EtOH extract	32% EtOH extract
B/3036	7.67±0.76	8.33±0.58	9.83±0.29	10.00±0.00
B/3092	8.00±1.00	7.00±1.00	10.00±0.00	10.17±0.29
B/3700	7.50±0.50	8.00±0.50	10.00±1.00	10.17±0.29
B/3991	7.83±0.29	7.50±0.00	9.83±0.29	11.83±0.76
B/5386	9.50±0.50	8.83±0.29	10.5±0.500	10.17±0.29
B/5504	8.17±0.76	7.83±0.76	9.33±0.58	9.00±0.00
B/5609	7.50±0.50	8.50±0.87	10.17±0.76	9.50±0.50
B/5702	8.17±1.04	8.00±1.00	10.67±0.76	10.17±1.04
B/5881	8.50±0.50	7.00±0.50	10.17±0.76	8.83±0.29
B/6271	7.17±0.29	8.50±0.00	9.17±0.76	9.17±0.29
B/6876	9.50±0.50	7.33±0.29	9.83±0.29	10.00±1.00
B/7052	8.00±0.00	7.50±0.50	11.00±0.00	11.17±0.29
B/7207	7.67±0.29	8.33±0.58	9.67±0.29	11.17±0.29
B/8113	7.00±1.00	7.67±0.29	9.83±0.29	11.00±1.00
B/8195	8.00±1.00	6.50±0.50	8.00±1.00	11.50±0.50
B/8215	9.00±1.00	7.00±0.50	9.83±0.29	10.83±0.76
B/8280	7.33±0.58	8.50±0.50	8.83±0.29	10.83±0.76
B/8285	7.50±0.50	9.00±0.00	10.17±0.76	10.00±0.00
B/8340	9.00±0.00	8.17±0.76	9.00±0.00	9.00±0.00
B/8617	8.17±0.29	7.00±1.00	10.50±0.50	10.00±0.00

Key: Aq= Aqueous, EtOH =Ethanol

Generally the antimicrobial activity exhibited in the study could be attributed to eugenol and sugnyl acetate: antimicrobial compounds present in the flower bud of *Eugenia caryophyllata* [35-36]. The variation in resistance observed among the studied isolates could be genetic or

difference in active compounds in the extracts. There may therefore be present in the ethanol extract certain potent compound which are less or absent in the aqueous phase.

5. CONCLUSION

In conclusion, aqueous and ethanol extracts of *Eugenia caryophyllata* has antimicrobial activity against wide range of common pathogenic bacteria and yeast. The antimicrobial efficacy of the extracts seems to be dose-dependent. Better antimicrobial activity was however observed in the ethanol extracts.

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CONSENT

Not applicable.

ETHICAL APPROVAL

Not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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