

## RESEARCH PAPER

### A STUDY ON THE EFFICACY OF EXTRACTS OF *BOERHAVIA DIFFUSA* L ON BACTERIAL ISOLATES OF FINGER TIP INFECTIONS (WHITLOW)

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## ABSTRACT

This research was conducted to determine the efficacy of crude extract of *Boerhavia diffusa* L (BHD L) on bacterial isolates of fingertip infection (Whitlow). Seventeen patients with whitlow were studied within 18 months and swabs of active fingertip infections were taken from the patients at Crossing-Kachia in Kaduna State for analysis at the microbiology laboratory of Federal University of Technology Minna, Niger State Nigeria. Associated bacteria were isolated and identified using standard microbiological and biochemical tests. The isolates were tested against extracts of BHD L and commercially available antibiotics using the Kirby Bauer agar well diffusion method. Phytochemical analysis was also conducted in order to determine the bioactive compounds in BHD L that may be responsible for its effectiveness in treatment. The results showed that more males (76.5%) were affected while the mean age of people affected was 28.6years. The predominant causative agent was *Staphylococcus epidermidis* (89.5%) while *Staphylococcus epidermidis* confirmed its resistance to commercial antibiotics, hence difficulty of treatment of whitlow with orthodox medicines. Similarly, extracts of BHD L had no antibacterial activity against *Staphylococcus epidermidis*. Thus, the efficacy of BHD L on the isolates of fingertip infections may be due to some other reasons yet unknown.

**Keywords** - Antibacterial activity, Antibiotics, Efficacy, Photochemical, Whitlow.

## INTRODUCTION

Fingertip infections otherwise referred to as whitlow, are various forms of painful finger infections such as herpetic whitlow (Plate 1) which affects the finger or toes (Nester, Anderson, Roberts, & Nester, 2007). Finger tip infections such as whitlow are uncommon infections; meaning they occur sparingly. When the infection occurs, it results in a lot of pain and discomfort that can be sometimes traumatic. This underscores the need for accessible instant treatment.

Except herpetic whitlow, caused by a virus, most of finger infections are caused bacteria species. The uniqueness of each infection depends on how the infection starts and where it is particularly located. Usually, the initial event is some form of trauma; may be a cut, animal bite, or puncture wound.

Specifically, *Herpetic whitlow* is a caused by the Herpes Simplex Virus types I or II. This is the same virus that causes oral or genital herpes infections. Herpetic whitlow is an occupational hazard because people in certain occupations are more at risk (*see* Plate 1A). These include dentists, hygienists, physicians, nurses, or any other



person that may have contact with saliva or body fluids containing the virus. People with oral or genital herpes may also infect their own fingers (Avitzur and Amir, 2002; Lewis, 2004).

The causative organisms for *Paronychia* are bacteria and usually, staphylococcal or streptococcal organisms. Fungus rarely causes whitlow and it characteristically begins as a hangnail. Oftentimes, the affected individual would attempt to bite off the piece of nail at the corner of the fingertip. This results in an open wound that would allow bacteria on the skin or in the mouth to infect the wound (Plate 1B). The infection can then spread to the surrounding tissue next to the nail and cuticle (Rigopoulos, Larios, Gregoriou and Alevizos, 2008). Interestingly, the infection of the finger pad by same organisms causing paronychia leads to *Felons*, which are usually secondary to puncture wounds allowing the introduction of bacteria deep into the finger pad. The fingertip has multiple compartments with which infections can spread (Plate 1C). Other related infections include *Flexor tenosynovitis* caused by bacteria introduced by a penetrating trauma into the deep structures and tendon sheaths and subsequently spreading along the tendon and associated sheaths (Anderson & Erik, 1987); and *Deep space infections* caused by bacteria that get into the deep tissues through puncture wounds or deep cuts.



**Plate 1 (A, B and C): Herpetic whitlow in a young child (A); Active clinical infection (B) (paronychia); and Active clinical infection (C) (Felon)**

Although there are standard clinically procedures for the treatment of whitlow infections, suggestions are rife that extracts of *Boerhavia diffusa* L can effectively be used for the treatment of whitlow and the result is always good. The finger is usually healed and saved from permanent damage (see Plates 4 – 9 below). Nevertheless, it is well known that improper treatment of whitlow infections may result in permanent deformity of an affected finger (Nester et al., 2007). *Boerhavia diffusa* plant has been seen to be used for treatment of whitlow infections in some parts of northern Nigeria. This brought about the need to research into its efficacy and possible improvement. *Boerhavia diffusa* L (BHD L) is a species of flowering plant in the four o'clock family, commonly known as tar vine, “punarnava” meaning ‘that which rejuvenates’ or ‘renews the body’, or ‘red spiderling’. Its common name is Hogweed or common pigweed. Its vernacular names include Babba-juji in Hausa language. It grows as a perennial herb at about 50- 100cm high. The stem is semi woody and glabrous. It also bears tubers like cassava. The roots have been seen to be effective in the treatment of whitlow infection. It is pan tropical and grow wild and as a weed in farms (Mann, Gbate, & Ndaumar, 2003). The plant is presently seen or available at Crossing-Kachia in Kaduna State and around FUT Minna, Bosso ampus. The roots are actually the useful part in therapy. During treatment, the roots are dried, pounded or mashed with little water to make a poultice and placed on the site of infection i.e. the fingertip. The infection gets cured in 24hours-72hours.

Experience has shown that most people come for this treatment when conventional antibiotics fail. Unfortunately, the practice of treating whitlow infections with herbs has been going on for several years without any scientific validation; hence the need for this study. This study therefore, was designed to determine the efficacy of crude extract of *Boerhavia diffusa* L (BHD L) on Whitlow infections with the following as specific objectives: to isolate and identify causative organisms involved in localized whitlow infections; to establish the susceptibility profile of



causative organisms to crude extract of herb; to determine the minimum inhibitory concentration (MIC.), minimum bacteriocidal concentration (MBC.) and to carry out phytochemical screening of roots of herbs; and to compare the efficacy of crude extracts of *Boerhavia diffusa* L and commercially available antibiotics. We practically relied on the hypothesis that there is no significant difference between the effect of herbal extracts on isolates of fingertip infection and those of conventional antibiotics.

## MATERIALS AND METHODS

**Study Area:** Kaduna State is in the Northwest region of Nigeria and it shares borders with Niger State which is in the North central Geopolitical region of northern Nigeria. Kachia and Minna are both located within these Geopolitical Zones.

**Collection of samples:** Link was established with a traditional healer/herbal practitioner known to treat cases of fingertip infections using local herbs. Fingertips of subjects were swabbed using sterile swab sticks (Evepon brand®) at Crossing-Kachia, Kaduna State and taken to the Microbiology laboratory of Federal University of Technology Minna, Niger State Nigeria within 24hours in an ice pack for processing. People of all ages who were suffering from whitlow infection were sampled. The ages of respondents were ascertained through oral interview with the herbal practitioner. Samples collected were analyzed statistically using simple percentages and inferential statistics such as chi square and correlation coefficient.

**Isolation of organisms from whitlow infections:** Swab from active infection was inoculated into 15ml sterile Nutrient broth and incubated at 37°C for 24hrs in accordance with the method described by Sharma (2007). This was later sub-cultured onto Nutrient agar (NA), Blood agar (BA) and Saboraud dextrose agar (SDA). The use of Nutrient agar was to provide cultures for storage for future use. The blood agar was to enhance growth of large spectrum of organism in order to guarantee proper isolation without missing any important organism. The use of Saboraud dextrose agar was to afford the growth of fungi.

**Identification of isolates:** The morphological characteristics of the isolates were determined using the Gram staining technique as described by Sharma (2007). Biochemical tests were also conducted such as: growing isolates on Manitol Salt Agar (MSA), Sheep blood agar, Catalase test, Coagulase test, Urease test, Phosphatase test, and  $\beta$ -galactosidase test.

**Collection of plant material:** Plant was collected from surrounding environment of Federal University of Technology, Minna. The plant was taken to the Department of Chemistry, Federal University of Technology Minna, for identification and authentication by a team of plant specialists/phytochemists led by Dr. Abdullahi Mann. The plant was identified as *Boerhavia diffusa* L.

**Preparation of plant material:** The roots were collected from the plant and washed thoroughly with clean water and sun-dried for 3-5days in order to replicate how the herbal practitioners prepare the plants. After drying, the plant was pounded with mortar and pestle into a semi-powder form and stored in air-tight bottles for further action. The semi-powdered plant material was extracted using 70% ethanol, hexane, chloroform and water; using soxhlet extractor. Twenty grams of root sample was placed in each extraction chamber wrapped in a folded paper to avoid spilling and placed inside a thimble. Two hundred milliliters of each solvent placed inside the flat bottom flask was heated at 95°C for ethanol, 50°C for hexane, 60°C for chloroform, 100°C for water; and kept running in succession from ethanol to hexane to chloroform, and water for 4hrs each in order to ensure proper extraction. The extracts were later heated at corresponding temperatures to dryness using rotary evaporator and stored with a drop of toluene for future use (Sofowora, 1994).

About 0.4g of each of the extract was weighed and mixed with 100ml distilled water to produce a 0.4mg concentration of the extract. Similarly, 0.8mg and 1.0mg concentrations were prepared by weighing 0.8g and 1.0g respectively of the extracts and dissolving in 100ml of distilled water each. This was used in the test of antimicrobial activity. Test of antimicrobial activity of extract.



**Phytochemical Analysis:** The phytochemical analysis of plant extract was carried out in order to determine active ingredients responsible for antimicrobial efficacy, using standard phytochemical methods as described by Sofowora (1993). The following constituents were tested for: alkaloids, flavonoids, saponins, terpenoids, total phenols, cardiac glycosides, tannins, phlobatanins, anthraquinones and cardenolides. The procedures were both qualitative and quantitative

**Sensitivity Test:** Different concentrations (0.4mg, 0.8mg and 1.0mg), of the extract obtained above were used against the microbial isolates obtained from infections, on nutrient agar plates. The sensitivity of isolate to extract was tested using agar diffusion method (Bonjar and Nik, 2004; Zhang, Nagao, Tanaka, Yang, Okabe & Kouno, 2004; Deore & Khadabadi, 2009). The agar diffusion media were Nutrient agar plates with agar wells of 6mm diameter. Single direct colony of a pure culture of *Staphylococcus epidermidis* was aseptically transferred using sterile wire loop into nutrient broth and incubated for about 6hrs. Serial dilutions were made in tubes containing sterile normal saline (0.85% NaCl), and was adjusted to a turbidity that matches that of 0.5 McFarland standards. The corresponding suspension was then uniformly inoculated on the surface of sterile Nutrient agar plates using sterile cotton swab to form a lawn culture. A sterile 6 mm diameter cork borer was used to make holes (wells) into the set of inoculated agar media. The wells were filled with different concentrations of the extracts and the plates were incubated at 37°C for 24hrs in triplicates. The mean zones of inhibition were taken. The same procedure was conducted on similar culture plates as in the main experiment, using commercially available antibiotic disks. The zones of inhibition were compared, and inferences made in accordance with the standard (Coyle, 2005).

Bacterial isolates from wound infection (*Staphylococcus aureus*, *Streptococcus pyogenes*, *Pseudomonas aeruginosa*) were passed through same procedure above using plant extract and commercial antibiotics, and the zones of inhibition observed. It should be noted that the antibiotic used in comparison with wound isolates was Ampiclox due to its wide use as drug of choice in the treatment of whitlow infection in the hospitals.

## RESULTS

On age and sex distribution of patients sampled, the results showed that the whitlow infection cuts across all ages irrespective of the sex (Table I). Majority of people affected were males 13(80%) and within the age group of 20 – 29yrs (7; 41.2%), while the least affected were those within the age groups of 0-9yrs, 40-49yrs, 50-59yrs and 60-69yrs. (1; 1.5%). Age group 10-19yrs and 30-39yrs had 3(17.6%) people affected respectively. Overall, the age specific prevalence was not statistically significant but the mean age was 28.6years with a standard deviation of 14.6. The results from biochemical reactions of the isolates (Table 2) showed that two organisms were associated with whitlow infections. The most predominant was staphylococci which appeared in 17(89.5%) cases. Bacilli cells were also observed in 2 (10.5%) samples. The presence of bacilli cells was mostly that of contamination rather than association with whitlow infection because none of the supposed cells showed any biochemical reaction as typical of microbial cells. The presence of staphylococci in all samples processed was likely that of association; hence its predominance.

The results on the incidence of bacteria in whitlow infections revealed can be classified as ‘pure’ and ‘mixed’ infections (Table 3). Mixed infections were 2(21.1%) containing staphylococci and bacilli, while pure isolates of *Staphylococcus epidermidis* was predominant 15 (78.9%).

On the qualitative and quantitative analyses of the phytochemicals in *Boerhavia diffusa* L (Table 4), our results revealed the presence of alkaloid, flavonoid, phenols, tannins, and saponins whereas, cardiac glycosides, phlobatannins, terpenoids and cardenolides were absent. The Quantitative phytochemical analysis showed that *Boerhavia diffusa* L contained higher amounts of alkaloids ( $6.67 \pm 0.418\text{g}/100\text{g}$ ), flavonoids( $16.66 \pm 0.700\text{g}/100\text{g}$ ), and tannins( $14.09 \pm 0.959\text{g}/100\text{g}$ ). Other phytochemicals include saponins ( $1.73 \pm 0.212\text{g}/100\text{g}$ ) and phenols ( $0.95 \pm 0.000\text{g}/100\text{g}$ ).



**Table 1: Age and Sex distribution of patients with finger tip infections collected from Kachia and Minna**

Age (years)	Number examined		Total (%)
	Male	Female	
0 – 9	0	1	1(5.9%)
10 -19	2	1	3(17.6%)
20 -29	5	2	7(41.2%)
30 – 39	3	0	3(17.6%)
40 – 49	1	0	1(5.9%)
50 -59	1	0	1(5.9%)
60 -69	1	0	1(5.9%)
Total	13(76.5%)	4(23.5%)	17(100%)

**Table 2: Biochemical reactions and identification of *Staphylococcus epidermidis***

Test	1	2
GRAM REACTION	Positive cocci	Positive bacilli
CATALASE	+	-
COAGULASE	-	-
UREASE	+	-
ALKALINE PHOSPHATASE	+	-
β- GALACTOSIDASE	-	-
ORGANISM IDENTIFIED	<i>S. epidermidis</i>	<i>Bacilli</i>
FREQUENCY /(% OCCURRENCE	17(89.5%)	2(10.5%)

The results of susceptibility test showed no zone of inhibition for all the different extracts to the whitlow isolate. Other organisms from wound infection also showed no traces of susceptibility to extracts of *Boerhavia diffusa* L (Table 5). However, standard antibiotics used as control showed high zones of inhibition in wound isolates. *S. aureus*: 22.0 ±2.00mm, *S. pyogenes*: 20.6 ±0.503mm, and *P. aeruginosa*: 23.6 ±0.421mm average zones of inhibition (Table 5). The susceptibility of bacterial isolates to commercial antibiotics (Table 6) showed that *Staphylococcus epidermidis* was highly resistant to commercially available antibiotics as there was no zone of inhibition. Other isolates presented various forms of sensitivity from intermediate to susceptible zones of inhibition ranging from 16mm to 25mm.

**Table 3: Occurrence of bacteria in patients with finger tip infections**

Category	Isolates	Frequency	% occurrence
Mixed infections	Staphylococci and Baccilli	2(2)	21.1
Pure isolates	<i>Staphylococcus epidermidis</i>	15	78.9
Total		19	100



**Table 4: Qualitative and Quantitative analyses of phytochemicals in Boerhavia diffusa L**

Phytochemical	Qualitative	Quantitative(g/100g)
Alkaloids	+	6.67 ± 0.418
Flavonoids	+	16.66 ± 0.7000
Steroids	-	NA
Terpenoids	-	NA
Saponins	+	1.73 ± 0.212
Cardiac glycosides	-	NA
Tannins	+	14.09 ± 0.959
Anthraquinones	-	NA
Phenols	+	0.95 ± 0.000
Phlobatanins	-	NA
Cardenolides	-	NA

( + = Positive, - = Negative, NA=Not applicable )

Further test of *Staphylococcus epidermidis* on aqueous extracts using the agar well diffusion of 6mm at different concentrations of 0.4mg, 0.8mg, and 1.0mg showed no sensitivity at different concentrations and at longer times (Table 7). The susceptibility of *Staphylococcus epidermidis* to aqueous plant extract using broth method The broth or tube method also showed no susceptibility at different concentrations of 0.4mg, 0.8mg and 1.0mg and at different times from 6hours to 48hours (Table 8).

**Table 5: Susceptibility of bacterial isolates from finger tip and wound infections to extracts of Boerhavia diffusa L**

Isolates	Zone of inhibition(mm)				CONTROL Standard antibiotics( Ampiclox) average zone of inhibition (mm)
	AE	HE	CE	EE	
<i>S. epidermidis</i>	NIL	NIL	NIL	NIL	0.0mm
<i>S. aureus</i>	"	"	"	"	22.0mm
<i>S. pyogenes</i>	"	"	"	"	20.6mm
<i>P. Aeruginosa</i>	"	"	"	"	23.6mm

( AE = aqueous, HE = hexane, CE = chloroform, EE = ethanol )



**Table 6: Susceptibility of bacterial isolates from fingertip and wound infections to commercially available antibiotics**

Antibiotic( Conc.)	Mean Zone of inhibition (mm)			
	<i>S. epidermidis</i>	<i>S. aureus</i>	<i>S. pyogenes</i>	<i>P. eruginosa</i>
Ciproxin 1.0mg	0	20±1.00	22±1.76	24±2.54
Norfloxacin 1.0mg	0	18±1.00	20±0.92	20±1.23
Chloramphenicol 3.0mg	0	17±1.33	19±0.77	18±0.76
Streptomycin 3.0mg	0	23±3.05	18±1.00	21±1.00
Amoxycilin 2.0mg	0	20±0.89	19±0.63	18±1.20
Ampiclox 2.0mg	0	22±1.57	23±1.61	21±0.99
Erythromycin 3.0mg	0	17±1.55	17±1.21	18±1.00
Gentamicin 1.0mg	0	16±1.00	20±1.62	18±0.78
Rifampicin 2.0mg	0	22±2.00	18±0.73	16±0.65
Levofloxacin 2.0mg	0	25±1.93	20±0.50	22±1.00
Control without antibiotic	0	0	0	0

**Table 7: Susceptibility of *Staphylococcus epidermidis* to aqueous plant extract at different concentrations**

Plant extract	Zones of inhibition( mm)		
	at 12hours	at 24hours	at 48hours
0.4mg	0.0	0.0	0.0
0.8mg	0.0	0.0	0.0
1.0mg	0.0	0.0	0.0

**Table 8: Susceptibility of *Staphylococcus epidermidis* to aqueous plant extract at different concentrations using broth or tube method.**

Extract	Turbidity			
	+ Isolate	After 6hours	After 12hours	After 24hours
0.4mg	+	+	++	++
0.8mg	+	+	++	++
1.0mg	+	+	++	++

+ = cloudy, ++ = more cloudy



A breakdown of healing processes which begins from presentation of infection through application of herbal extract to actual healing is shown in Plates 2 (A, B, C, D, E and F). The process takes from 2days to 8days in some cases. Plates 3 (A and B) and Plate 4 (A and B) shows permanently damaged fingers previously infected with whitlow and fingers healed as a result of treatment with extract of *Boerhavia diffusa* L.



**PLATE 2 (A, B, C, C, D and F):** Showing healing processes following treatment with extract of *Boerhavia diffusa* L. Note finger at presentation with swollen pad (A); poultice of plant applied and bandaged immediately (B); addition of more extracts after three days with healing in progress (C); infected finger five days later (D); finger healing with scars seven days later (E); and finger completely healed after eight days.

## DISCUSSION

Results in Table VI and Table VII showed no sensitivity of isolate to conventional antibiotics and herbal extracts respectively. This conformed to our null hypothesis hence it was accepted. Measure of relationship between effects of herbal extracts and conventional antibiotics could not be determined because both had no activity.

The results in Table I showed that majority of people affected are males and the average age infected with whitlow was 28.6years. This implies that youths are more vulnerable than children and the elderly people. This may likely be linked to the age of activities where manual or mechanical work is predominant, which often results in bruises or injuries that serve as an opening for onset of whitlow infections. Jacobs (2006) had earlier posited that trauma to the cuticle of the fingers and damage of fingers through work and overenthusiastic manicure are usually responsible for the onset of whitlow infection as both situations allows infections in. These definitely, are youth-related activities and provide explanation and possible reasons for the predominant incidence of whitlow infections within the youthful age bracket.







Plate 3: Permanently damaged fingers previously infected with whitlow



Plate 4: Fingers healed as a result of treatment with extract of *Boerhavia diffusa* L



However, the age specific test of significance gave a chi square result below chi square tabulated at  $df= 6$ . This means that there is no relationship between the ages seen on our distribution and the prevalence of whitlow infection seen from the samples. It shows that age is inconsequential in the manifestation of whitlow infection within the population studied. Relying on the statistical analysis, we can say that whitlow infection can occur at any age without prejudice. It can affect both young and old at any time. But the sex specific test of significance gave a correlation that showed relationship between sex and whitlow incidence. This implies that the result in Table 1 is significant and shows that whitlow affects males more than females within the population studied. This is likely connected to the higher involvement of men in daily endeavors that have to do with fending for the family which are predominantly manual hence, exposes men more to injuries that may predispose to whitlow infection.

From the findings in Tables 2 and 3, it can be inferred that *Staphylococcus epidermidis* was the bacteria mostly associated with whitlow infections in the area of study. *Staphylococcus epidermidis*, is a Gram positive bacteria hence this implies that mostly Gram positive bacteria are the ones associated with whitlow infections. The results also showed clearly that there were no fungi in whitlow infections because no growth was seen on any of the seventeen Sarbourauds Dextrose Agar plates inoculated. This agrees with Rigopoulos *et al.* (2008) who observed that whitlow infection is rarely caused by fungi.

The results in Table 4 showed the presence of high amounts of alkaloids ( $6.6 \pm 0.418\text{g}/100\text{g}$ ), flavonoids ( $16.66 \pm 0.700\text{g}/100\text{g}$ ) and tannins ( $14.09 \pm 0.959\text{g}/100\text{g}$ ) following phytochemical analysis. Some reports have it that tannins, alkaloids and flavonoids possess antimicrobial activities (Aliyu *et al.*, 2008; Sofowora, 1993). Flavonoids and other phenolic compounds are potent water soluble antioxidants and free radical scavengers, which prevent oxidative cell damage, have strong anti cancer activity (Salah, Miller, Paganje, Tijburg, Bolwell, Rice & Evans, 1995; Del-Rio, Obdulio, Casfillo, Marin & Ortuno, 1997; Okwu, 2004). Alkaloids are very important in medicine and constitute most of the valuable drugs. They have marked physiological effect on animals (Edeoga and Eriata, 2001). Tannin content was also high which may agree with the report of Okwu (2004) which says some medicinal plants are used in the treatment of sore throat, diarrhea, hemorrhage, and wound healing due to presence of tannins. These reports give us possible explanation for the therapeutic efficacy of *Boerhavia diffusa* L often seen in the treatment of whitlow infection. However, the use of all extracts of *Boerhavia diffusa* L on *Staphylococcus epidermidis* in the agar diffusion media did not show any antibacterial activity.

A test of the extract on isolates from wound infection such as *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Streptococcus pyogenes* did not show antibacterial activity either. Looking at the test of hypothesis, one will observe that the null hypothesis passed; hence making the research hypothesis a failure. This on the other hand means that contrary to speculations, the herbal extract is theoretically not better than conventional antibiotics in the treatment of whitlow. This in effect means that extract of *Boerhavia diffusa* L does not have antibacterial activity in-vitro but the plant contains phytochemicals known to have antimicrobial activities. Its efficacy in the treatment of whitlow infection may likely be attributed to antiviral and other possibilities such as the Prontosil scenario in the history of antibiotics (Nester *et al.*, 2007).

Prontosil (a red dye), was dramatically effective in the treatment of streptococcal infections in animals. Surprisingly, Prontosil did not have effect on streptococci growing in test tubes. It was later discovered that enzymes in the blood of the animal split the Prontosil molecule, producing a smaller molecule called sulfanilamide; this break-down product acted against the infecting streptococci. It can be said here that as Prontosil did not show direct antibacterial activity and was discovered later to carry out intermediate enzymatic metabolism which aided eventual cure; this may be a possibility for extracts of *Boerhavia diffusa* L. Also, a trial of standard/commercial antibiotics on *Staphylococcus epidermidis* in a comparison, showed no antibacterial activity also. This may imply outright resistance to commercially available antibiotics tested. This also reveals a possible reason why antibiotics do not cure whitlow infections when patients go to hospital for treatment.

Another possible reason is the fact that the infection may be caused by a virus as reported by Clark (2003) he said whitlow can be caused by herpes simplex virus type 1 or type 2. This report gives an explanation why the herbal extracts did not show sensitivity to the bacterial isolates- the etiologic organisms may actually be HSV-1 or HSV-2.



During presentation and treatment with extracts of *Boerhavia diffusa* L the patient continues to suffer pains, hence by implication this is an indication that the extract does not relieve pain and as such may not have anti-inflammatory properties. As a matter of fact, some patients are given pain killer drugs during treatment while the infection gets cured.

A close observation of patients undergoing therapy with extracts of *Boerhavia diffusa* L showed that from presentation to healing takes an average of 5 days as shown in Plate 2. During infection, some patients remain on orthodox treatment which never give them healing. After a long time, the infection goes on its own leaving the patient with serious damage such as chopped off fingers etc. (Plate 3). On the other hand, those who apply crude extracts of *Boerhavia diffusa* L usually get healing within few days to a week without much complication – only scars of previous infection will be seen (Plate 4).

Invariably, this study has shown that the organism mostly associated with whitlow infection is *Staphylococcus epidermidis* and that crude extract of *Boerhavia diffusa* L does not have direct antibacterial activity against it. The possible reason why it is efficient in treatment may be due to the extract creating a change in growth environment that may be uncondusive for the isolate thereby resulting into its death with time. Such change in environment does not occur in petridish; or that the underlying causative agent may be a virus which is susceptible to extracts of *Boerhavia diffusa* L and dies off with time.

In conclusion, this research has afforded better understanding on the functioning of *Boerhavia diffusa* L in the treatment of whitlow infection, and also added information on the use of the plant which was hitherto not known. *Staphylococcus epidermidis* was found to be mostly associated with whitlow infection rather than being causative. The anti-microbial efficacy or order wise of the plant to *Staphylococcus epidermidis* has also been established for future references, and the principle of action of plant extract in the treatment of whitlow infection now somewhat known. The failure of plant extract to show antibacterial activity somehow gives a clue as to the possibility of viral etiology for whitlow diseases. Some of the objectives of the study were not achieved as a result of the research discovery which turned out to be contrary to initial expectations. We recommend that similar work be conducted in different regions of the country in order to be able to afford a holistic report on findings in Nigeria as a whole, which can be presented to the world. More work should be done on other possible reasons why *Boerhavia diffusa* L is efficacious on *Staphylococcus epidermidis* even though without antibacterial activity. Viral whitlow (herpetic whitlow) and the effect of *Boerhavia diffusa* L should also be studied.

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#### **AUTHORS CONTRIBUTIONS.**

Danboyi and Mawak are microbiologists who saw to the microbiology aspects of the research work while Egwim and Ghaji who are biochemists also gave biochemical guidance to the job. Wunzani is a chemist while Auta is a biologist who assisted in the phytochemistry and identification of plant respectively.

