Effect of a herbal extract (Winniecure) on HIV-1 plasma viral load

J. J. ABDULLAHI[1], D. E. AGBONLAHOR[2], J. E. NJAB [1*], MC P. I. J. OKOYE[1], L. C. ONDOBO[1], O. R. OGIEVA[1], G. O. NWOBU[2], Y. NDEJI[4], M. A. MOMOH[5], O. O. LAMIDI[1], A. J. ONOJA[3]

1.Winners Medical Diagnostic and Research Institute, Plot 1391 Ladongor close P.O.Box 9502, Garki II, Abuja, Nigeria.
2.College of Medicine, Ambrose Alli University, P.M.B. 14, Ekpoma, Edo State, Nigeria.
3.Institute of Human Virology, University of Maryland, Baltimore, U.S.A.
4. Federal Medical Centre, Minna, Niger State, Nigeria
5. Garki General Hospital, Area 3, Abuja, Nigeria

ABSTRACT

Plasma HIV-1 RNA load of 103 volunteers were estimated before and after therapy in a randomized double blind placebo controlled clinical trial, to assess the clinical efficacy of a herbal extract *(Winniecure)* used in Nigeria for the treatment of HIV/AIDS. Volunteers (44 males and 59 females), aged 18-45, were divided into two groups (X and Y). Group X (n=50) was placed on a Placebo (Echinacea), while group Y (N=53) received the extract *(Winniecure)*. The therapeutic period was 8 weeks.Results obtained from the subjects group Y revealed that 46 (86.8%) volunteers had a significant reduction in their viral loads, with 18.9%undetectable, after therapy. On the other hand, no significant reduction was observed in the control group X. Rather, viral RNA significantly increased in 48 (96%) of volunteers. These findings suggest that the herbal extract *(Winniecure)* is a potent antiviral agent that would appear to be a viable candidate for the management of HIV/AIDS.

Key words: Winniecure, viral load, HIV/AIDS management

RESUME

Dans le but d'évaluer l'efficacité d'un extrait végétal (*Winniecure*),utilisé au Nigeria pour le traitement du VIH/SIDA, la charge virale de 103 volontaires séropositifs, a été détermine au cours d'un essai clinique de 8 semaines . Les volontaires (44 males et 59 femelles) agés de 18 a' 45 ans, étaient divises en deux groupes(X et Y).le groupe X s'est fait administrer un échantillon témoin *(Echinacea)*, pendant que le groupe Y recevait l'extrait en étude *(Winniecure)*, tous pour une période de 8 semaines. L'analyse des résultats dans le groupe Y a notamment révèlé une réduction significative ($p<0.05$) chez 46(86.8%) des volontaires, parmi lesquels 18.9% se sont retrouvés avec une charge virale non détectable au terme des 8 semaines d'étude. Dans le groupe X par contre, une augmentation significative ($p<.005$) s'est plutôt fait constater chez 48 (96%) des volontaires. Cette étude suggère pertinemment que l'extrait végétal *(Winniecure)* a des propriétés antivirales dont l'application marquerait un pas décisif vers le contrôle effectif du VIH/SIDA.

Mots clés : Winnicure, charge virale, contrôle de VIH/SIDA

***Corresponding author.**
NJAB JEAN EMILE.
Winners Medical Diagnostic/ Research Institute P.O.Box 9502,
Abuja-NIGERIA. - E-mail: jeannjab@yahoo.com

INTRODUCTION

The Human Immunodeficiency Virus (HIV) and the Acquired ImmunoDeficiency Syndrome (AIDS) have become a Socio economic threat to the development of the African continent (Piot et al, 1993). The most recent epidemiological data reveal that 28.1 million (70.3%) of the 40 million people globally infected, are in sub-Saharan Africa (AIDS EPIDEMIC, 2002). The multisectorial prevention campaigns and the Global Partnership Initiatives (GPI) are yet to significantly stop the spread of the dreaded virus. It therefore appears that HIV/AIDS is primarily a challenge to biomedical scientists. Until a vaccine is found to effective prevent the spread of the virus, we may still have to sustain, prolong and improve the quality of life through effective, available and affordable therapy. In a phase 1 study, the safety and the preliminary phytochemical screening of a herbal extract (Winniecure) were established (unpublished data). Retrospective and prospective studies revealed that, the extract tremendously improve the clinical status of volunteers. The present study was designed to evaluate the effect of the extract on the most relevant virological marker (Cohen et al, 1994) in the clinical management of HIV/AIDS.

MATERIALS AND METHODS

Study design

The protocol design for this study was an Independent Double Blind Randomnised Clinical Trial.

Ethical approval

The ethics committee of Winners Medical Diagnostic and Research Institute, Abuja, Nigeria granted approval.

Volunteers

Volunteers in this study were all HIV-1 seroprositive apparently asymptomatic individuals, who consented in writing to partake in the study. The aim and objectives of the study were communicated to them, as well as the responsibilities and duties of the investigators. Those on placebo were administered the herbal extract (Winniecure) at the end of the study. Inclusion and exclusion criteria were as follows:

Inclusion

- HIV seropositive
- Age 18-50
- Sex males and females
- Normal complete blood count
- Negative Hepatitis B surface antigen
- Volunteers' readiness to be followed up for a period of 8 weeks.

Exclusion

- Age below 18 or above 50
- Pregnant and Lactating females
- History and/or evidence of previous or current antiretroviral drug(s) use.
- Presence of any other immunesuppressive disorder or im munologic deficiency syndromes such as systemic lupus erythematosus, polyartritis nodosa, isolated IgA defi ciency, Digeorge syndrome.
- Repeated uncontrollable vomiting and diarrhea
- Need for parenteral treatment
- Addicted Alcoholics/ or Smokers
- Severe Anaemia.
- Medical or psychiatric condition or occupational responsibilities which prectude subject compliance with the study.
- History of suicide attempts
- Presence of suicidal ideation
- Past/present requirement for antipsy chotic medication.
- Uncontrolled diabetes mellitus and hy pertension.

Volunteers were divided into two groups X and Y using a table of random numbers for purpose of drug placement. The subject group Y, was on the herbal extract and the control group X on a placebo *(Echinacea).*

The herbal extract (Winniecure)

Winniecure is the trademark of an aqueous herbal combination of *Ficus exaspirata, Ficus asperifolia, Ficus sur and Sida corymbosa.* The physicochemical properties of the extract reveal a dark brown crystalline powder, with a strong pungent leafy smell, bland taste and a fine granular texture. Spectrophotometric analysis identified a single peak at 302nm against a 0.1% w/v solution of the extract. The extract is strongly hydrophilic with a moisture content of 0.1 % and a true density of 1.936+- 0.2.Phytochemical screening only indicated the presence of tannins, saponins and complex carbohydrates. Preliminary laboratory studies in animals and humans demonstrated that the extract was safe in humans (Agbonlahor et al, 2003).

Placebo

The placebo chosen for the purpose of this study was *Echinacia*, which is a herbal extract reported to be an

"effective immunomodulator" (KATZUNG.B.G, 2001)

Specimen collection and handling

Ten milliliters (10mls) of venous blood were collected from each volunteer, into lithium heparin vacutainer, before and after therapy. Whole blood was stabilized for 20 minutes on an electrical blood mixer (RM-500, HOSPITEX), then processed in duplicate into plasma and stored at-10°C.

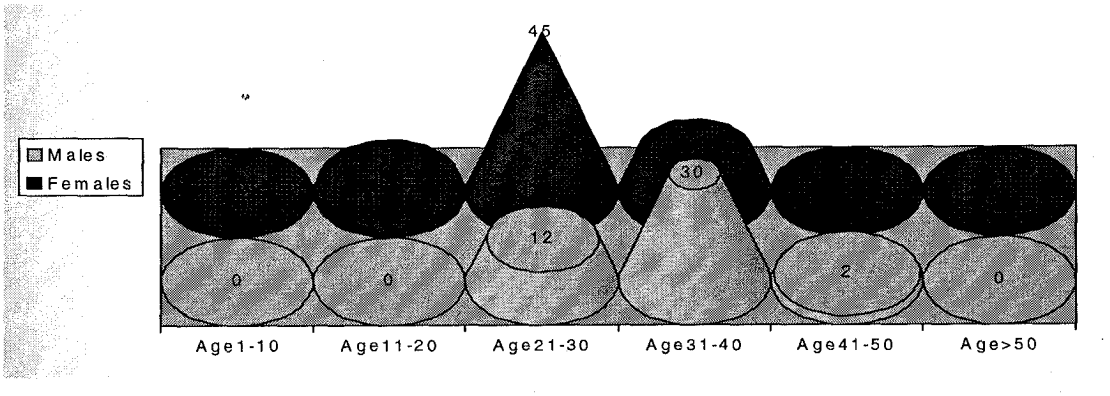
HIV-1 plasma RNA assay

Samples were run in batches of 10 for a period of three weeks. Calibrators provided by Organon Teknika were co-extracted with samples using the boom extraction method (Brusse and Vanderbiezen, 2001), co-amplified employing the Nucleic Acid Sequence Based Amplification (NASBA) at 41°C for 90 minutes. Hybridization was carried out at 41°C for 30 minutes. Detection was performed at room temperature (26°C) using the version 1.3r1 Nuclisens Reader of Organon Teknika (Dehaan and Vanderbiezen, 2000).

RESULTS

Histogram 1 and 2 illustrate the age and sex distribution frequency of the volunteers, as well as their marital status. Out of the 103 volunteers, 57.3% were females while 42.7% were males. Their age range was 18 to 45 and the age bracket 21 to 41 was the most represented. Summaries of statistics and t-test of significance (P=0.05) are found in table 1 and 2, for group Y and X respectively. Table 1 show that there was a significant reduction in viral load in the subject group. Table 2 reveals that there was a significant increase in viral load in the control group X. Virologic responses after therapy are summarized in table 3 and 4, for Y and X respectively. In the subject group table 3 illustrates that 46 volunteers (86.8%) has significant (P<0.05) viral load reduction, of which 10 (21.2%) had undetectable viral load at the end of the 8 weeks therapy. Seven (13.2%) did not achieve any significant viral load reduction. In table 4, 48 volunteers (96%) had significant viral load increase, while only 2 volunteers (4%) recorded significant viral load reduction.

Histogram 1: Age Distribution of Patients



Histogram 2: Group and Sex Distribution of Patients

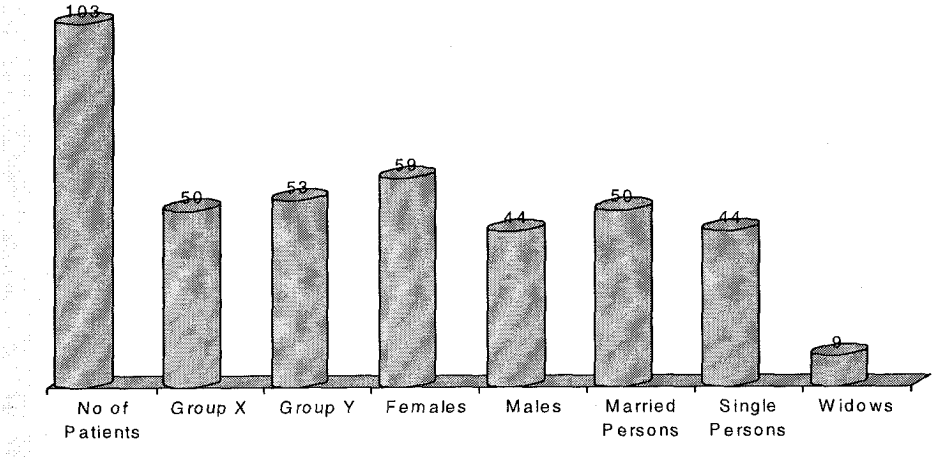


Table 1: Effect of Winniecure on the treatment group Y.

	Total viral load (cp/ml)	Mean viral load (cp/ml)	Standard Error of Mean	Standard Deviation	Calculated-t value	Tabulated-t
BEFORE THERAPY	1,549,350	29,233	9,696	70,585		
AFTER THERAPY	851,430	16,065	4,873	35,474	2.3	2.0

Table 2: Effect of placebo on the control group X.

	Total viral load (cp/ml)	Mean viral load (cp/ml)	Standard Error of Mean	Standard Deviation	Calculated-t value	Tabulated-t
BEFORE THERAPY	881,690	17,634	6,677	47,215		
AFTER THERAPY	1,429,860	28,597	6,902	48,807	4.1	2.0

Table 3: Summary of the virologic response in the treatment group Y

NUMBER OF VOLUNTEERS	PERCENTAGE
Viral RNA load reduction after therapy (46)	**86.8%**
Undetectable viral RNA load (10)	**18.9%**
More than 50% viral RNA load reduction (30)	**56.6%**
Less than 50% viral RNA load reduction (6)	**11%**
Virologic failure (7)	**13.2%**

Table 4: Summary of the virologic response in the control group X.

NUMBER OF VOLUNTEERS	PERCENTAGE
Viral RNA load increase after therapy (48)	**96%**
Viral RNA load reduction (2)	**4%**
More than 50% viral RNA load reduction (1)	**2%**
Less than 50% viral RNA load reduction (1)	**2%**

DISCUSSION

In every drug development, assessment of drug efficacy is indeed an indispensable exercise. The goal in therapeutics is the eradication of an etiological agent and resolving the clinical presentations brought about by such agent(s). In HIV infection therefore, the current objectives are to achieve the greatest possible reduction in viral load, and to reconstitute the qualitative and quantitative architecture of the immune system (HOPKINS J., 2001).

This study has clearly demonstrated that the herbal extract *(Winniecure)* can be used to achieve the first objective in HIV/AIDS treatment, as is evident in the marked and significant reduction in the HIV-1 RNA copies of subjects relative to that of the control after herbal therapy. While in the study group, the mean viral RNA copies was reduced from 29,333 copies/ml before herbal therapy to 16,065 copies/ml after therapy, that of individuals on the placebo instead increased from a baseline mean value of 17,634 copies/ml to 28,597 copies/ml after 8 weeks of study.

The increase observed in the placebo group is not unexpected, as the placebo has no known antiretroviral properties, the viruses replicated unabated in the system, hence the marked increase in viral load at the end of the study. On the other hand, individuals in the

study group had their viral loads drastically reduced because they were placed on an agent, which either suppressed or inhibited the active viral replication of the HIV in their peripheral blood and this agent is clearly the Winniecure they were administered.

The degree of viral RNA reduction is of paramount interest in antiretroviral therapy and this was also analysed in this study. As can be seen in table 3, 46 of the 53 individuals (86.8%) in the study group, showed a significant reduction in their viral load, and 10 of these (18.9%) had undetectable viral RNA copies/ml after 8 weeks of herbal treatment. Where as in the placebo group no such reduction was observed, except in 2 individuals (4%) who showed reduction in their viral load, one of them with reduction > 50%. This may be attributed to reasons we cannot explain, but not due to the placebo.

Though it may be difficult to make direct comparisons, it is worthy to note that a review of 17 clinical trials (HILL C., 1999) revealed that the mean percent achieving "virologic success" was 81% by "as-treated" analyses (<500 copies/ml therapeutic goal) and 52% by "intent-to-treat" analyses. Against this background, the 18.9% virologic success (undetectable HIV-1 RNA copies/ml) obtained in this study may appear very low. However, the findings mentioned above were observed in studies that lasted a minimum of 16 weeks. In addition, the probability of achieving the goal of <20-50 copies/ml with Highly Active Antiretroviral Therapy (HAART) in treatment of naïve patients is estimated at 16-30 weeks (HOPKINS J., 2001). The time required to reach undetectable viral loads depends largely on the baseline plasma HIV-1 RNA copies/ml.

Prolonging the duration of treatment with *Winniecure* and matching baseline HIV-1 RNA copies to values similar to those of individuals in studies mentioned above, may therefore make for a better comparison and this calls for a further evaluation. It's also pertinent to note that virologic failure is not uncommon even in the use of HAART. This has often been ascribed to lack of adherence, reduced potency of regimen, pharmacological failure due to reduced drug delivery to site of infection and resistance (HOPKINS J., 2001). These factors may also be responsible for the virologic failure observed in this study in the 7 individuals (13.2%) who were administered the herbal extract. Though the first two reasons are unlikely, as volunteers in this study complied and cooperated fully all through the duration of the study, this in any case calls for a further research. More so, the pharmacokinetics of the herbal extract used in this study are yet to be established. Further more, it was difficult in this study, to ensure that every volunteer was a naïve patient, in which case primary resistance may have played a part in the very small, but important failure rate observed in this study.

CONCLUSION

This study strongly suggests that the herbal extract *Winniecure* is a potent antiviral agent that would appear to be a viable candidate for the management of HIV/AIDS, especially in sub-Saharan Africa where synthetic regimens are not always available, let alone affordable.

REFERENCES

AGBONLAHOR D.E., ABDULLAHI J.J., NWOBU G.O., NJAB J.E., ONDOBO C., OGIEVA O.R., OKOYE M.I.J., ONOJA A.J. (2003). The effects of a herbal extract (Winniecure) on CD+4 T-Lymphocyte counts of asymptomatic HIV Seropositive individuals. *Journal of Applied and Basic Sciences, vol* 1,3-7.

BRUSSE R., VANDERBIEZEN J.I.M. (2000). Training manual of *Organon teknika*(DOC code:8419/TM/09).

COHEN P.T., SANDE M.A., VOLBERDING P.A. (1994). The AIDS Knowledge Base, 2nd ed. Little, Brown; 2.4-5.

DEHAAN C.F., VANDERBIEZEN J.I.M. (2000). Training reference manual for nuclisens. *Organon teknika*, Boxtel; 10.

HILL C. (1999). Clinical trial: Results and Interpretations, *6th CROI*, 111:394.

HOPKINS J. (2001). AIDS service and medical management (ed). 10-13.

Joint united programme on HIV/AIDS (UNAIDS), world health organization (WHO). (2001). *AIDS update*. 20-25.

KATZUNG B.G. (2001). *Basic and clinical pharmacology* (8th ed). Mc Gram Hill. 1089.

PIOT P., KAPITA B.M., NGUGI E.N., MANN J.M., COLEBUNDERS R., WABITSCH R. (1993). AIDS in Africa: A manual for physician. *WHO publication*. 2-3.

Received: 23/02/2004
Accepted: 28/07/2004