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UTILIZATION, PHYTOCHEMICAL PROFILES, AND TOXICOLOGICAL EVALUATIONS OF COMPLEMENTARY AND ALTERNATIVE MEDICINE (CAM) AMONG PEOPLE LIVING WITH HIV/AIDS

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ABSTRACT

Background: *The concurrent use of complementary and alternative medicine (CAM) alongside conventional Antiretroviral Therapy (ART) is a common but frequently unmapped behavior among people living with HIV/AIDS (PLWHA) in sub-Saharan Africa. While patients often use herbal remedies to manage medication side effects, unregulated CAM use poses risks of herb-drug interactions, systematic tissue toxicity, and altered immunological trajectories.* ***Objective:*** *To investigate the prevalence and drivers of CAM use among PLWHA, analyze the phytochemical profiles of frequently utilized herbal preparations, and evaluate their toxicological profiles using an in-vivo Drosophila melanogaster model and clinical biomarkers.* ***Materials and Methods:*** *This study utilized a multi-phase, biobehavioral, and experimental research design. The behavioral phase surveyed 325 PLWHA and interviewed 10 traditional healers (THs) to identify the primary plants used for HIV management in Ekiti State. The laboratory phase evaluated an aqueous extract of a localized herbal mixture containing Ageratum conyzoides, Aristolochia ringens, African uvaria, Artemisia annua, and Medical cannabis. This mixture was tested across varying concentrations in a Drosophila melanogaster model to monitor survival rates, total protein, total thiol levels, glutathione-S-transferase (GST) activity, and catalase activity. Clinical blood profiles were also analyzed across patient groups to evaluate changes in white blood cell (WBC) counts, packed cell volume (PCV), and liver enzyme levels.* ***Results:*** *Behavioral findings indicated that* ***57.1%*** *of CAM users utilized herbal remedies specifically to counter the physical side effects of conventional ART.*

Laboratory analysis of the plant mixture confirmed the presence of bioactive compounds, including tannins, flavonoids, saponins, and alkaloids. In the Drosophila model, exposure to higher concentrations of the herbal mixture led to a statistically significant decrease in survival rates and climbing capacity, accompanied by a reduction in total thiol levels and an increase in GST and catalase activity, indicating systemic oxidative stress. Clinical evaluations corroborated these physiological risks: patients using both ART and CAM concurrently demonstrated significantly lower mean white blood cell counts (3.89 pm 1.84 µl) and lower packed cell volumes (31.10 pm 7.67%), alongside elevated liver transaminases compared to patients on ART alone. ***Conclusion:*** *Unregulated, concurrent use of CAM and ART presents clear physiological risks, including potential bone marrow suppression and hepatotoxicity. These findings underscore the need for institutional guidelines and targeted patient education to manage herb-drug interactions safely.*

Keywords: Complementary and Alternative Medicine (CAM), HIV/AIDS, Toxicity, Drosophila melanogaster, Phytochemicals, Hepatotoxicity.

Introduction

The introduction of conventional Antiretroviral Therapy (Rx) has successfully transformed HIV/AIDS into a manageable chronic condition. To maintain viral suppression, patients require lifelong adherence to complex drug regimens, which can often induce challenging side effects such as gastrointestinal distress, chronic fatigue, and metabolic changes (Ogregade & Major, 2022). In search of relief, many patients turn to complementary and alternative medicine (CAM), particularly traditional herbal remedies (Kavakli et al., 2019). In Nigeria and across much of West Africa, traditional medicine is deeply rooted in local culture, readily accessible, and highly trusted (Safdar et al., 2021).

Consequently, many patients use herbal preparations concurrently with their prescribed antiretrovirals, often without disclosing this to their healthcare providers (Ogbe et al., 2020). While these plants contain valuable bioactive compounds with therapeutic potential, their unregulated use introduces significant clinical risks. Unstandardized dosages, unknown mechanisms of action, and potential herb-drug interactions can cause systemic toxicity and compromise the efficacy of conventional ART (Adeloye et al., 2017). This study addresses these risks by combining behavioral data on CAM use with experimental evaluations of commonly used local plant mixtures, using an in-vivo *Drosophila melanogaster* model and clinical hematological and hepatic biomarkers.

Materials and Methods

Identification of Herbal Formulations

Through individual semi-structured interviews with 30 PLWHA and 10 practicing traditional healers (THs) across Ekiti State, a specific 5-plant herbal formulation was identified as the most common remedy for HIV symptoms. The formulation consists of: *Ageratum conyzoides* (white weed), *Aristolochia ringens*, *African uvaria*, *Artemisia annua*, and *Medical cannabis*.

Phytochemical Screening and Aqueous Extraction

Fresh samples of the five plants were collected, verified, and dried. An aqueous extract of the combined herbal mixture (AEHM) was prepared to replicate local traditional preparation methods. Standard qualitative and quantitative screening protocols were used to profile core bioactive compound groups, including alkaloids, saponins, tannins, flavonoids, and glycosides.

Experimental In-Vivo Design (*Drosophila melanogaster*)

Wild-type *Drosophila melanogaster* (Harwich strain) were used as the in-vivo model. Equal cohorts of flies were exposed to varying concentrations of AEHM (control, 1mg/g, 5mg/g, and 10mg/g of diet) over a set period. Key outcomes evaluated included:

- Overall survival curves and mortality rates.
- Locomotor function, measured via standardized negative geotaxis (climbing) assays.
- Tissue homogenate markers for oxidative stress, including total protein concentration, total thiol levels, glutathione-S-transferase (GST) activity, and catalase activity.

Human Biomarker Triangulation

To validate the laboratory findings clinically, a comparative cross-sectional analysis of blood profiles was conducted across four distinct patient groups (n=40 per arm) at participating clinics: (1) treatment-naïve HIV patients, (2) patients on ART using CAM concurrently, (3) patients using ART alone, and (4) healthy, HIV-negative controls. Standard laboratory assays quantified white blood cell (WBC) counts, packed cell volume (PCV), and liver transaminases (AST and ALT).

Statistical Analysis

Data were analyzed using SPSS Version 26. In-vivo metrics and human biomarkers were compared across groups using One-Way Analysis of Variance (ANOVA) followed by Tukey's post-hoc tests, with statistical significance defined as $p < 0.05$.

Results

Behavioural Findings and Phytochemical Profile

The survey data showed that 57.1% of CAM-using participants relied on traditional remedies primarily to manage the physical side effects of conventional antiretrovirals. Phytochemical analysis of the multi-plant extract confirmed high concentrations of tannins, flavonoids, saponins, phenolics, and alkaloids.

In-Vivo Oxidative Stress and Locomotor Safety Profiles

Testing in the *Drosophila melanogaster* model showed that higher concentrations of the herbal mixture introduced clear physiological strain, as detailed below:

Table 1: Biochemical and Behavioural Markers in *Drosophila melanogaster* Exposed to AEHM

Experimental Groups (Dietary Dose)	7-Day Survival Rate (%)	Climbing Performance Index (%)	Total Thiol Level (µmol/g protein)	GST Enzyme Activity (µmol/min/mg protein)
Control Group (0 mg/g)	98.5 pm 1.1	94.2 pm 2.1	54.2 pm 3.1	0.12 pm 0.02

Low Dose (1 mg/g)	95.1 pm 1.8	89.4 pm 2.6	51.0 pm 2.8	0.14 pm 0.03
Mid Dose (5 mg/g)	82.4 pm 2.9*	71.1 pm 3.4*	38.3 pm 2.2*	0.26 pm 0.05*
High Dose (10 mg/g)	64.0 pm 4.2dagger	48.3 pm 4.1dagger	21.4 pm 1.9dagger	0.41 pm 0.07dagger

Note: All values are expressed as Mean pm SD. Symbols (, dagger) indicate statistically significant differences from the control group ($p < 0.05$).*

As shown in table 1, higher doses of the extract led to a significant increase in GST and catalase activity, alongside a sharp drop in total thiol levels, confirming that the formulation induces dose-dependent oxidative stress.

Human Blood Profile and Tissue Function Markers

Clinical blood tests across the patient groups supported the laboratory findings, showing distinct variations in hematological and hepatic markers:

Table 2: Clinical Biomarkers across Cross-Sectional Patient Cohorts

Patient Treatment Group Assessed	Total White Blood Cell Count (µl)	Packed Cell Volume (PCV %)	Alanine Aminotransferase (ALT U/L)	Aspartate Aminotransferase (AST U/L)
Group 1: Naïve HIV Patients	4.12 pm 1.20	34.20 pm 5.10	32.40 pm 6.20	38.10 pm 7.40
Group 2: Concurrent ART + CAM	3.89 pm 1.84*	31.10 pm 7.67*	68.40 pm 12.30 dagger	74.20 pm 14.10 dagger
Group 3: ART Treatment Only	5.64 pm 1.50	38.50 pm 4.20	28.10 pm 5.40	31.00 pm 6.10
Group 4: Healthy Controls	6.80 pm 1.10	42.10 pm 3.80	22.30 pm 4.10	24.50 pm 4.80

Note: Asterisks and daggers indicate statistical significance ($p < 0.05$) relative to Group 3 (ART Only).

Patients using both ART and CAM concurrently exhibited the lowest mean white blood cell counts (3.89 pm 1.84 µl) and packed cell volumes (31.10 pm 7.67%), alongside significantly elevated liver transaminases (ALT and AST), pointing to potential bone marrow suppression and hepatotoxicity.

Discussion

The experimental results show that while the studied herbal mixture contains therapeutic bioactive compounds like flavonoids and phenolic, its unstandardized use poses clear health risks. In the *Drosophila melanogaster* model, high doses of the extract caused a significant increase in mortality and impaired climbing capacity. This was accompanied by a sharp drop in total thiol levels and an increase in GST activity, confirming that the formulation triggers significant intracellular oxidative stress at higher concentrations and agrees with the findings of (SB et al., 2021). These laboratory findings are closely mirrored by the clinical data from human patients and agrees with the findings of similar studies (Mohamud et

al., 2024) . The cohort using CAM concurrently with conventional ART showed a significant reduction in white blood cell counts and packed cell volumes (31.10pm 7.67%), alongside elevated liver transaminases (Mitjà et al., 2023). This indicates that unregulated herb-drug combinations can induce systemic tissue strain, including bone marrow suppression and hepatotoxicity. This physiological strain is largely driven by a behavioural factor: 57.1% of patients turn to CAM independently to cope with ART side effects. Related findings also exist in the literature (Wonkam & Makani, 2019) Because patients rarely disclose this to clinic staff, they inadvertently expose themselves to risky herb-drug interactions. These findings highlight the critical need to integrate laboratory data with clinical practice to ensure safer, more transparent patient care.

Conclusion and Recommendations

Unregulated concurrent use of complementary herbal medicine alongside conventional antiretroviral therapy introduces clear physiological risks, including potential bone marrow suppression, tissue oxidative stress, and hepatotoxicity. To improve patient safety, the following institutional interventions are recommended:

1. Health authorities and institutional review boards should establish clear clinical guidelines on CAM use, helping healthcare providers identify and manage potential herb-drug interactions safely.
2. Antiretroviral clinics should integrate routine haematological and liver function testing for patients using alternative remedies to catch and manage potential tissue toxicity early.
3. Hospitals and health departments should create collaborative platforms bringing together traditional healers and medical professionals to improve patient safety, standardize herbal use, and build safer referral networks.

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