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Original Research Article

Qualitative Determination of Sildenafil and Tadalafil Adulterants in Selected Herbal Aphrodisiac Products

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Abstract

The increasing global demand for herbal approdisiac products has raised concerns regarding their safety and quality due to potential adulteration with synthetic or conventional pharmaceutical active ingredients. This study investigates the presence of sildenafil and tadalafil in selected herbal mixtures marketed for sexual enhancement available in Nigeria, including Mai Sulhu, Mai Rahusa, Mai Sasangi, Jaolin, Saigari Jawaye, and Manisa. A combination of preliminary phytochemical screening, Thin-Layer Chromatography (TLC), and Fourier Transform Infrared Spectroscopy (FTIR) analysis was used to analyze the samples. Phytochemical screening using Dragendorff's reagent indicated the presence of alkaloids in five of the samples, except Jaolin, suggesting the inclusion of bioactive compounds. TLC analysis revealed that four herbal mixtures had Retention Factor (Rf) values matching those of tadalafil, 0.60 (Mai Salhu and Mai Rahusa), and sildenafil citrate, 0.47 (Saigari yawaye and Manisa), indicating possible adulteration. FTIR spectroscopy further confirmed the presence of these synthetic phosphodiesterase-5 (PDE-5) inhibitors, as spectral peaks in some samples closely correlate with those of sildenafil and tadalafil. The detection of these undeclared pharmaceuticals poses significant health risks, including cardiovascular complications and hazardous drug interactions, as well as potential erectile dysfunction. These findings emphasize the need for stricter regulatory oversight, improved quality control measures, and public awareness regarding the risks associated with adulterated herbal aphrodisiacs. Routine screening using advanced analytical techniques such as High-Performance Liquid Chromatography and Mass Spectrometry is recommended to enhance detection accuracy and precision.

Keywords: Aphrodisiacs, sildenafil, tadalafil, Dragendorff, phytochemical screening.

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INTRODUCTION

The demand for aphrodisiac products has increased significantly worldwide, with consumers seeking alternatives to conventional pharmaceutical treatments for sexual dysfunction. Herbal and liquid aphrodisiac preparations are particularly popular due to their perceived natural composition, affordability, and easy accessibility (Shamloul & Ghanem, 2013). These products are widely marketed for their ability to enhance libido, improve erectile function, and boost overall sexual performance. However, growing evidence suggests that some of these products may be adulterated with synthetic phosphodiesterase type 5 (PDE5) inhibitors such as sildenafil and tadalafil, which are commonly used in the medical treatment of erectile dysfunction (Tay et al., 2020). Sildenafil and tadalafil function by inhibiting PDE5, an enzyme responsible for

the breakdown of cyclic guanosine monophosphate (cGMP), leading to prolonged vasodilation and increased blood flow to the penile region (Kloner et al., 2018). While these compounds are effective when prescribed under medical supervision, their unauthorized inclusion in over-the-counter aphrodisiacs poses significant health risks. Undisclosed sildenafil and tadalafil can cause serious adverse effects, including hypotension, priapism (prolonged erection), cardiovascular complications, and dangerous interactions with nitrates and antihypertensive drugs (Liu et al., 2019). The adulteration of herbal and liquid aphrodisiacs has been documented in multiple studies worldwide. A study in China found that nearly 81% of tested herbal aphrodisiac products contained undeclared synthetic PDE5 inhibitors, exposing consumers to unknown health risks (Zhang et al., 2021). Similarly, a study in Nigeria reported the presence of sildenafil in several locally available aphrodisiac drinks, raising concerns about regulatory oversight and public health safety (Akinyemi *et al.*, 2022). The widespread occurrence of adulteration highlights the need for robust analytical methods to detect and quantify these compounds in liquid aphrodisiac preparations.

The deliberate addition of these pharmaceuticals into herbal products without disclosure poses significant health risks, including severe

cardiovascular complications, hypotension, and dangerous drug interactions, especially for individuals with pre-existing medical conditions (Almalki *et al.*, 2022). The unauthorized inclusion of tadalafil in herbal sexual enhancement supplements is an emerging public health concern. Many unregulated products falsely marketed as "natural" contain undisclosed tadalafil, exposing consumers to serious health risks (Venhuis *et al.*, 2021).

Figure 1: Structural features of Sildenafil (A) and Tadalafil (B)

PDE5 inhibitors are a class of drugs widely used for the treatment of erectile dysfunction (ED), pulmonary arterial hypertension (PAH), and benign prostatic hyperplasia (BPH) (Andersson, 2021). PDE5 inhibitors exert their effects by selectively inhibiting the PDE5 enzyme, which is responsible for the degradation of cyclic guanosine monophosphate (cGMP) in vascular smooth muscle cells (Boolell et al., 2021). During sexual stimulation, nitric oxide (NO) is released from endothelial cells, activating guanylate cyclase, which increases cGMP levels. Elevated cGMP leads to relaxation of smooth muscle and increased blood flow, particularly in the corpus cavernosum of the penis, facilitating erection (Gacci et al., 2020). This mechanism also underlies the use of PDE5 inhibitors in PAH, where they promote vasodilation in pulmonary arteries (Humbert et al., 2021).

Sildenafil citrate, a PDE5 inhibitor, is used in the management of erectile dysfunction (ED) and pulmonary arterial hypertension (PAH) (Kloner, 2000). By blocking PDE5, it enhances cGMP activity, improving blood flow for erections and reducing pulmonary pressure (Nehra et al., 2001). Approved in 1998 (Goldstein et al., 2000), it has high efficacy (60-85% success in ED) and is metabolized by CYP3A4, with a 3 - 5-hour half-life (Goldstein et al., 2016). For PAH, it improves exercise capacity, as shown in the SUPER-1 trial (Galie et al., 2005). However, illegal adulteration of herbal supplements with sildenafil poses health risks due to unregulated dosing and potential drug interactions (Venhuis et al., 2011). Despite its safety profile, misuse remains a public health concern (Gryniewicz et al., 2013). On the other hand, Tadalafil, a long-acting PDE5 inhibitor, is also used in the management of ED, benign prostatic hyperplasia (BPH),

and PAH (Peixoto & Gomes, 2015). It offers extended efficacy (up to 36 hours) due to its ~17.5-hour half-life, enabling once-daily dosing (Gacci *et al.*, 2020; Gomelsky *et al.*, 2021). By inhibiting PDE5, it enhances cGMP-mediated vasodilation, improving blood flow for ED and reducing pulmonary pressure in PAH (Andersson & Mulhall, 2021; Boolell *et al.*, 2021). For BPH, it relaxes smooth muscle, alleviating urinary symptoms (Cantrell *et al.*, 2013). Unlike sildenafil, its absorption is unaffected by food, and it is metabolized by CYP3A4 (Gacci *et al.*, 2020; Andersson & Mulhall, 2021). Approved in 2003, tadalafil provides flexibility for on-demand or daily use, improving sexual function and quality of life (Gacci *et al.*, 2020).

Analytical techniques such as Fourier Transform Infrared Spectroscopy (FTIR) and Thin Layer Chromatography (TLC) are widely used to detect active pharmaceutical ingredients and natural products chemical constituents (Awala et al., 2019; Bunu et al., 2020; 2020a; Bunu et al., 2022; Ebeshi et al., 2022; Bunu et al., 2023; 2023a; Ebeshi et al., 2023). TLC is a versatile analytical technique used to assess drug purity, detect adulteration, and evaluate formulation stability (Blau & Halket, 2020). The food industry uses it for quality control and detecting adulterants (Reich & Schibli, 2007), while herbal medicine research relies on TLC for phytochemical identification and product authentication (Kowalska & Sajewicz, 2022). FTIR is another essential analytical tool used in identifying active ingredients, excipients, and impurities of pharmaceuticals (Pavia et al., 2020).

This study aims to assess the presence of sildenafil and tadalafil in selected herbal mixtures and evaluate their authenticity and safety using

phytochemical screening, TLC, and FTIR, and assess the potential health risks associated with adulteration, particularly regarding consumer safety and drug interactions

METHODS

Drug Samples and Preparation of Reagents

The drug samples, tadalafil and sildenafil, were graciously supplied by Tuyil Pharmaceutical Industry, Nigeria, and their identities of tadalafil and sildenafil were confirmed using FTIR, ensuring authenticity and compliance with regulatory standards. Additionally, the herbal mixtures were sourced from various local markets and herbal stores across different locations in Bayelsa State. These sources were selected to capture a diverse range of herbal formulations commonly used by the local population. All reagents were prepared according to standard laboratory protocols.

The preparation of Dragendorff's reagent was carried out in two stages. First, Solution A was prepared by dissolving 1.7 g of bismuth subnitrate in 20 mL of glacial acetic acid, after which the mixture was diluted with 80 mL of distilled water while being stirred to ensure complete dissolution. Next, Solution B was prepared by dissolving 40 g of potassium iodide in 100 mL of distilled water, ensuring that all the potassium iodide crystals were fully dissolved. Once both solutions were ready, they were mixed in equal volumes to obtain the final Dragendorff's reagent. The prepared reagent was then stored in an ambercolored bottle to prevent light-induced degradation.

Qualitative Phytochemical Screening

The herbal mixture samples tested include Mai Sulhu, Mai Rahusa, Mai Sasangi, Jaolin, Saigari Jawaye, and Manisa herbal mixtures. The samples were initially subjected to qualitative phytochemical screening to detect the presence of alkaloids, using Dragendorff's reagent as a specific alkaloid-detecting reagent. This preliminary test was conducted because sildenafil citrate is classified as a semi-synthetic alkaloidal drug, meaning it is derived from alkaloidal precursors through chemical modifications. Dragendorff's reagent test was chosen due to its high specificity for alkaloids, which form an orange or reddish-brown precipitate in their presence. This step was essential to confirm whether the herbal samples contained alkaloid-like substances, including potential adulterants such as sildenafil citrate or other synthetic alkaloids. The results of this test provided an initial indication of possible adulteration before proceeding with TLC analysis for further identification and comparison.

Thin Layer Chromatography

TLC was performed on sildenafil, tadalafil, and the herbal preparations using a mobile phase composed of ethyl acetate, methanol, and ammonia (8:2:0.2). The developed TLC plate was examined under a UV lamp at a wavelength of 254 nm to detect any fluorescent or UVabsorbing spots. The plate was then sprayed with Dragendorff's reagent to visualize alkaloids, and the retention factors (Rf values) were calculated to analyze the separation and identification of the compounds. The samples were evaporated to near dryness using a water bath and subsequently reconstituted with methanol. The reconstituted samples were then filtered, and the resulting filtrate was concentrated by evaporation. Each of the six samples was co-spotted with sildenafil citrate and tadalafil on a TLC plate. After development, the plate was examined under a UV lamp at a wavelength of 254 nm to detect UV-absorbing compounds. The plate was then sprayed with Dragendorff's reagent to visualize alkaloids. The results were analyzed to determine the presence of adulterants in the herbal samples by comparing their Rf values with those of sildenafil citrate and tadalafil.

Fourier Transform Infrared Spectroscopy

Finally, sildenafil citrate, tadalafil, and the six herbal samples were subjected to FTIR analysis using an Agilent Technologies **FTIR** spectrometer. Exactly 0.1g of each powdered herbal sample was weighed and transferred into a beaker. This analysis was performed to determine the infrared (IR) absorption spectrum of each sample, which provides valuable information about the functional groups present in the compounds. FTIR spectroscopy works by measuring the absorption of infrared radiation at different wavelengths, producing a spectrum that serves as a chemical fingerprint of the sample. The resulting spectra were then analyzed and compared to identify similarities or differences between the herbal samples and the reference compounds (sildenafil citrate and tadalafil), aiding in the detection of potential adulterants or unknown chemical constituents within the herbal formulations.

RESULT

Phytochemical Screening

The presence of alkaloids in the herbal samples was determined using Dragendorff's reagent, which produces an orange or reddish-brown precipitate in the presence of alkaloids.

Table 1: Alkaloid Screening with Dragendorff's

Reagent		
Test Sample	Result	
Mai Sulhu	+	
Mai Rahusa	+	
Mai Sasangi	-	
Jaolin	+	
Saigari Yawaye	+	
Manisa	+	

Legend: + = **Present,** - = **Absent**

Thin-layer chromatography

To examine the presence of sildenafil citrate and tadalafil in the herbal samples, TLC analysis was conducted by co-spotting the six herbal mixtures with the reference standards (sildenafil citrate

and tadalafil). The retention factor (Rf value) was calculated for each standard compound and herbal formulation, and compared to determine potential adulteration.

Table 2: TLC Retention Factors (Rf)

Test Sample	Rf value	Adulterant detected
Sildenafil citrate	0.47	=
Tadalafil	0.60	=
Mai Salhu	0.60	Tadalafil
Mai Rahusa	0.60	Tadalafil
Mai Sasangi	-	None
Jaolin	-	None
Saigari yawaye	0.47	Sildenafil citrate
Manisa	0.47	Sildenafil citrate

Fourier Transform Infrared Spectroscopy Fourier Transform Infrared Spectroscopy

The Fourier Transform Infrared Spectrum of standard compounds and some test herbal formulations.

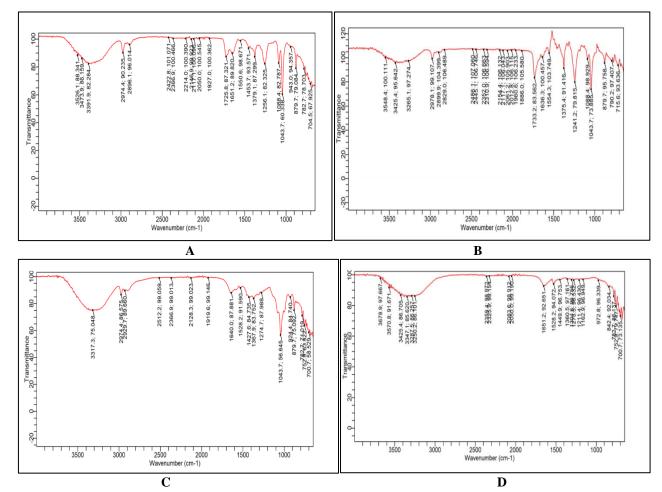
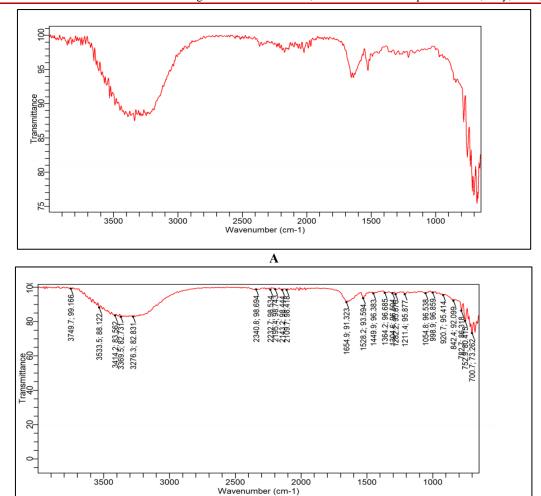


Figure 2: Fourier Transform Infrared Spectrum of (A) Sildenafil citrate, (B) Tadalafil, (C) Saigari Yawaye, and (D) Mai Rahusa herbal mixtures



B
Figure 3: Fourier Transform Infrared Spectrum of (A) Manisa and (B) Mai Salhu herbal mixture

Table 3: Fourier Transform Infrared Spectrum Specific Absorbance

Sample	Wave number	Interpretation
_	(cm^{-1})	
	704 - 879	Aromatic C-H bending
Sildenafil citrate	943 - 1256	C-N stretching (alkyl amines)
	1379 - 1550	N-H bending (Amines), C=C stretch (Aromatic ring)
	1651	C = O stretching (amide, urea)
	2896 - 3526	O-H/N-H stretching (hydroxyl and amine groups
	715 - 879	Aromatic C-H bending
Tadalafil	1043 - 1088	C-N stretching (amines)
	1375 - 1554	N-H bending (Amines), C=C stretch (Aromatic ring)
	1636-1733	C = O stretching (amide)
	2899 - 3548	O-H/N-H stretching (hydroxyl and amine groups
Saigari Yawaye	700 - 879	Aromatic C-H bending
	1043 - 1274	C-N stretching (alkyl amines)
	1640	C = O stretching (amide, similar to sildenafil)
	2896 - 3526	O-H/N-H stretching (hydroxyl and amine groups
Mai Rahusa	704 - 879	Aromatic C-H bending
	943 - 1256	C-N stretching (alkyl amines)
	1379 - 1550	N-H bending (Amines), C=C stretch (Aromatic ring)
	1651	C = O stretching (amide, urea)
	2896 - 3526	O-H/N-H stretching (hydroxyl and amine groups

DISCUSSION

The presence of alkaloids in the herbal mixtures was determined using Dragendorff's reagent. Alkaloids were detected in five out of the six samples, suggesting that these herbal formulations contain bioactive compounds with potential pharmacological activities. Alkaloids are known for their wide range of effects, including analgesic, stimulant, and vasodilatory properties (Cicero *et al.*, 2020). The absence of alkaloids in the Mai Sasangi herbal mixture suggests that its composition may differ significantly from the others.

TLC analysis revealed that two herbal mixtures (Mai Sulhu and Mai Rahusa) exhibited an Rf value of 0.60, matching tadalafil, while two others (Saigari Yawaye and Manisa Herbal Mixture) had an Rf value of 0.47, matching sildenafil citrate. These findings indicate that four out of the six herbal samples were adulterated with synthetic phosphodiesterase-5 (PDE-5) inhibitors.

The presence of these adulterants in herbal products is concerning as sildenafil and tadalafil are prescription drugs used to treat erectile dysfunction and have significant side effects, including cardiovascular risks (Sharma *et al.*, 2018). The absence of adulterants in Jaolin and Mai Sasangi suggests that these formulations are not contaminated with synthetic PDE-5 inhibitors.

FTIR spectroscopy further supported the presence of sildenafil and tadalafil in specific herbal mixtures. The spectral peaks in the Saigari Yawaye sample closely resembled those of sildenafil citrate, particularly the C=O stretching at 1640 cm⁻¹ and N-H bending at 1379-1550 cm⁻¹. Similarly, Mai Rahusa displayed peaks characteristic of tadalafil, including C=O stretching at 1651 cm⁻¹. This confirms the presence of these synthetic drugs, further validating the TLC results.

The findings of this study point towards stronger regulatory enforcement and pharmacovigilance interventions (Miediegha & Bunu, 2022) to monitor and control the adulteration of herbal products. National drug regulatory agencies should implement routine screening of herbal formulations to ensure consumer safety (WHO, 2021). Consumers should be educated about the risks associated with adulterated herbal supplements. Awareness programs can help individuals make informed decisions regarding the safety and efficacy of these products. Manufacturers should be required to adhere to Good Manufacturing Practices (GMP) and undergo stringent quality control tests to prevent contamination with pharmaceutical substances (Zhang et al., 2020). The use of more sensitive and reliable techniques, such as HPLC and Mass Spectrometry (MS), should be encouraged for the detection of adulterants in herbal products. Legal measures should be taken against manufacturers and distributors found guilty of adulteration to deter such practices and protect public health. Additionally, herbal formulations should be

rigorously tested for efficacy and safety to ensure they meet therapeutic standards without the need for synthetic drug adulteration.

CONCLUSION

The study confirms that a significant number of herbal mixtures tested contained undeclared sildenafil citrate and tadalafil, posing serious health risks to consumers. These findings highlight the urgent need for stricter regulation, better quality control, and public education to prevent the risks associated with adulterated herbal products.

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