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EDITORIAL (Version française)

Chers lecteurs et lectrices, Chers auteurs et autrices ! La deuxième publication de votre revue trimestrielle est disponible dans les délais grâce à l'engagement des pairs évaluateurs, de l'éditeur (les Presses Universitaires de Dakar), des comités scientifique et éditorial et du Secrétariat ayant permis de diligenter dans les délais prévus et dans la rigueur scientifique, le processus d'évaluation et de publication.

Ce volume 2, numéro 1 comporte huit articles scientifiques soumis par de jeunes auteurs mais aussi des séniors aguerris qui contribuent à la diffusion transparente et équitable de la science en accès libre.

Ces articles originaux indexés dans Scopus abordent les plantes médicinales et alimentaires, l'optimisation et la validation de méthodes analytiques de contrôle de médicaments, la qualité du lait en poudre vendu à Dakar, l'accessibilité et la qualité de l'eau à Mboro (Sénégal), la qualité des miels de Casamance (Sénégal), la formation continue du pharmacien d'officine notamment.

Sous le format d'une Lettre à l'Éditeur, nous publions une contribution de la Pharmacopée américaine à travers l'équipe PQM+ (Promoting the Quality of Medicines) qui met la lumière sur la problématique de l'assurance qualité des médicaments en Afrique au Sud du Sahara et les niveaux de performances/reconnaissance des compétences des structures officielles en charge du contrôle de la qualité des médicaments et autres produits de santé. Les principaux résultats de cet exemple de coopération pratique Nord-Sud sont partagés et apportent un brin d'espoir dans la lutte contre les produits médicaux de qualité inférieure/falsifiés pour une meilleure protection de la santé des populations à travers i) une infrastructure de contrôle de qualité performante et ii) l'accessibilité à des produits médicaux de qualité, sûrs et efficaces. Ce présent numéro poursuit l'objectif de cette revue qui offre une opportunité de faire résonner davantage la plume scientifique de l'Afrique dans un monde globalisé dans lequel les sciences analytiques multidisciplinaires et transversales sont au cœur de tout processus de compétitivité, de régulation et de développement.

Nous remercions toute l'équipe éditoriale et les pairs évaluateurs pour leur collaboration. A vos manuscrits et excellente lecture !

EDITORIAL (English version)

Dear readers, Dear authors !

The second publication of your quarterly journal is available on time thanks to the commitment of the peer reviewers, the publisher (Presses Universitaires de Dakar), the scientific and editorial committees and the Secretariat which made it possible to proceed within the planned deadlines and in scientific rigor, the evaluation and publication process.

This volume 2, number 1 includes eight scientific articles submitted by young authors but also seasoned seniors which contributes to the transparent and equitable dissemination of science in open access.

These original articles indexed in Scopus address medicinal and food plants, the optimization and validation of analytical methods for drug control, the quality of powdered milk sold in Dakar, the accessibility and quality of water in Mboro (Senegal), the quality of honeys from Casamance (Senegal), the continuing training of community pharmacists in particular.

In the format of a Letter to the Editor, we are publishing a contribution from the American Pharmacopoeia through the PQM+ (Promoting the Quality of Medicines) team which sheds light on the problem of quality assurance of medicines in Africa South of the Sahara and the levels of performance/recognition of skills of official structures responsible for controlling the quality of medicines and other health products. The main results of this example of practical North-South cooperation are shared and bring a bit of hope in the fight against substandard/falsified medical products for better protection of population health through i) an efficient quality control infrastructure and ii) accessibility to quality, safe and effective medical products.

This present issue pursues the objective of this journal which offers an opportunity to make Africa's scientific writing resonate more in a globalized world in which multidisciplinary and transversal analytical sciences are at the heart of any process of competitiveness, regulation and development.

We thank the entire editorial team and peer reviewers for their collaboration.

To your manuscripts and excellent reading !

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Original paper

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Antioxidant and antimicrobial activities of *Momordica charantia* L. *leaves*

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Abstract
Momordica charantia L. is used as a broad-spectrum antibacterial agent to fight infections. This work aimed at evaluating antioxidant and antimicrobial activities of hydroethanolic extract of *Momordica charantia* leaves. The extract was obtained by maceration of crude leaf powder (hydroethanolic 30:70). The phytochemical screening was focused on the detection of major chemical groups. The total flavonoid contents were studied using the aluminum chloride colorimetric method. The antioxidant capacity was carried out by the phosphomolybdate reduction method and by FRAP method. The antimicrobial activity of the extract was carried out by the diffusion method and micro dilution.
Our extract revealed the presence of alkaloids, phenol compounds, saponins, triterpenes, flavonoids. The total flavonoids contents of our extract is 143.65 ± 2.51 mg RE/g. The antioxidant activity by the phosphomolybdate reduction method and by the FRAP method are respectively 65.42 ± 6.24 mg AAE/g and 165.5 ± 17.55mg FSE/g. The extract of *Momordica charantia* has bactericidal action on *Staphylococcus aureus* ATCC 29213, *Staphylococcus aureus* SARM, *Pseudomonas aeruginosa* ATCC 27853 and *Cutibacterium acnes* ATCC 6919 and fungistatic action on *Candida albicans* ATCC 10231.
Momordica charantia could be a new source of antioxidant and antimicrobial agent. A fractionation and identification of biomolecules will be investigated.

Keywords: *Momordica charantia*, Antioxidant, Antimicrobial, Bactericidal, Fungistatic

1. INTRODUCTION

Since the dawn of time, plants have been humanity's main medicinal resource. Traditional medicine uses plant extracts and continues to provide health coverage to more than 80% of the world's population, especially in developing countries (WHO, 2002). Before an alarming resurgence of new and re-emerging infectious diseases, it is becoming crucial to discover new antimicrobial compounds with diverse chemical properties and mechanisms of action. In addition, microorganisms have developed resistance to available antimicrobials in recent decades, generating a considerable global public health problem (Andersson, 2003).
Several plants are used against microbial in order to reduce resistance and the cost of treatment. One of the most widely used herbs in the treatment of microbial diseases is *Momordica charantia.* Margose (*Momordica charantia* L.), also called bitter melon, bitter cucumber, momordic, African zucchini,

balsamic pear or wonder pear, is a kind of cucumber from Asia (India and China), especially tropical regions since it is a chilly plant that needs heat. Margose is an annual climbing plant that can grow up to 5m tall with rigid stems that cling to each other with tendrils. The leaves, 10 to 15 cm long, are cut into irregularly shaped lobes (Jia et *al.,* 2017). Several bioactive compounds of *M. charantia* have been recorded in the literature. They are categorized into carbohydrates, proteins, fats, and more. *M. charantia* contains triterpenoids, saponins, polypeptides, flavonoids, alkaloids and sterols (Jia et *al.,* 2017). Previous phytochemical studies have shown the bioactive components and their associated functions (Jia et *al.,* 2017). Margose is a plant widely used for the treatment of many diseases. In many African countries, the fruits are used as a purgative and dewormer (Shan et *al.,* 2012) while the leaves are macerated in water and used for diarrhoea

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and dysentery. The leaves are well known especially as a remedy for diabetes mellitus, simply by regular consumption as a vegetable (Johnson et *al*., 2016). Several biological properties of *M. charantia* have been studied, including hypoglycemic activities, antibacterial, antiviral, antitumor, immunomodulatory, antioxidant, antidiabetic, antimutagenic, anthelmintic, antilipolytic, antifertility, hepatoprotective and anti-inflammatory activities, such as anti-ulcerative, antioxidant and immunomodulatory activities (Bao et *al*., 2013; Beloin et *al*., 2005; Braca et *al*., 2008). *In vitro* studies, *M. charantia* proteins (α- and β-momorcharin) have an inhibitory effect against the human immunodeficiency virus (HIV). Its extract can also be used as a broad-spectrum antibacterial agent to fight infections (Saeed and Tariq, 2005).
This study aimed at determining the antibacterial and antifungal activity of *Momordica charantia* for a more reliable recommendation of the latter.

2. MATERIAL AND METHODS

2.1. Plant material

The plant material used was the leaves of *Momordica charantia*. The plant organs were collected in may 2022 at Hahotoe in the maritime region of Togo. The sample was identified at the Laboratory of Botany and Plant Ecology of the Faculty of Sciences of the University of Lomé where a voucher specimen was deposited in the herbarium under the number TOGO02802.

Figure 1: *Momordica charantia* L.

2.2. Chemicals

Phosphomolybdate, rutting, aluminum chloride, ascorbic acid. sulphuric acid, 2,4,6-tripyridyl-s-triazine (TPTZ).

2.3. Microbial strains Tested

The microbial strains used are wild-type reference strains (confirmed by susceptibility testing) that were obtained from the

American Type Culture Collection (ATCC). They are *Staphylococcus aureus* (ATCC29213); *Staphylococcus aureus* MRSA; *Pseudomonas aeruginosa* (ATCC27853)*; Cutibacterium acnes* (ATCC6919) and *Candida albicans* (ATCC 10231). They were provided to us by the microbiology laboratory of the National Institute of Hygiene (INH).

2.4. Plant extract preparation

The extraction was carried out following the previous work of Hoekou (2016). The hydroethanolic extract of *Momordica charantia* leaves was obtained by maceration under continuous agitation of 200 g of vegetable material powder in 2000 mL of ethanol-water mixture (70 : 30) at room temperature (25 - 30°C) for 48 hours. The maceration was filtered with Whatman N°1 paper. The filtrate was evaporated with rotavapor under vacuum at 40°C and then freeze-dried. The resulting solid was weighed to determine the extraction yield Y and then stored in the refrigerator in tubes at 4°C, protected from light, until use. Extraction yield is determined by the following formula (*Equation 1*):

Y = 100 (Mass of dry residue of evaporated extract) / Mass of dry plant material powder (Equation 1)

2.5. Phytochemical Screening

Phytochemical tests focused on the detection of major chemical groups (alkaloids, flavonoids, phenolic compounds, saponins, sterols and triterpenes, reducing compounds) by tube reactions. Using standard procedures as described by Harborne (1998). The dry extract was dissolved in distilled water at a concentration of 1mg/ml for phytochemical testing.

2.6. Determination of total flavonoids

The total flavonoids contents of hydroethanolic extract of plant material was studied using the aluminum chloride colorimetry method described by Okselni et *al*. (2018). The volume 1.5 mL dry extract was dissolved in distilled water (1 mg/ml), 1.5 mL of 2% aluminum chloride was added and the optical density was measured at a wavelength of 415 nm with a spectrophotometer after 30 minutes at laboratory temperature. This was repeated three times. Flavonoids levels were obtained from the rutting calibration curve and expressed as rutting equivalents per gram of dry matter (mg RE/g).

2.7. Antioxidant activity

2.7.1. The Phosphomolybdate Reduction Method

The reduction of phosphomolybdate was carried out according to the method described by Ouadja *et al*. (2018). To the volume 1 mL of 1 mg/mL extract, 9 mL of the working reagent was added. The whole thing was placed in a water bath at 95°C for 90 minutes and then the optical density was measured at a wavelength of 695 nm with a spectrophotometer. The working reagent consists of 90 mL of 0.6M sulphuric acid, 5 ml of 0.1% sodium hydrogen phosphate and 5 mL of 1% ammonium molybdate. This has been repeated three times. Ascorbic acid was used as a standard antioxidant under the same experimental conditions. Results were expressed in milligrams of ascorbic acid equivalent per gram of dry extract (mg AAE/g).

2.7.2. The FRAP Method

The ability to reduce ferric ions was measured using the method described by Kantati *et al*. (2022). To the volume 3 mL of 1 mg/mL extract, 3 mL of FRAP reagent was added [pH acid buffer = 3.5 (50 mL), 2,4,6-tripyridyl-s-triazine (TPTZ) solution (5 mL) and iron III chloride solution (5 mL)] and the optical density was measured at a wavelength of 695 nm on the spectrophotometer after 10 minutes. This has been repeated three times. A calibration line with ferrous sulphate ($FeSO_4$) as the reference molecule was used for the determination of concentrations. The values obtained are expressed as mg equivalent of ferrous sulphate per gram of dry matter (mg FSE/g).

2.8. Antimicrobial activity

Antimicrobial tests were performed using the liquid micro dilution method coupled with appropriate solid media spreading (Anani et *al*., 2015).

2.8.1. Preparation of the extract

The crude hydroethanolic extract from the leaves of *Momordica charantia* L. has been used. The dry extract was dissolved in distilled water to prepare 100 mg/mL solutions and then filtered on a 0.45 µm millipore membrane. The sterility of the extract was verified by inoculating a 100 µL aliquot of the extract onto Muller Hinton (MH), chocolate agar (GC) and Sabouraud chloramphenicol agar.

2.8.2. Preparation of Microbial Suspensions

The microbial strains tested were successively transplanted into Muller-Hinton broths for bacteria and Sabouraud broth for yeast and then onto agar (Muller-Hinton and GC for bacteria; Sabouraud chloramphenicol agar for yeast). A 24-hour colony (48 hours for *Cutibacterium acnes* (ATCC 6919) of each strain was collected using a sterile loop and inoculated in 10 mL of suitable broth (Muller-Hinton

broth for bacteria and Sabouraud broth for yeast). From this suspension, dilutions to the thousandth (10^{-3}) were made. 100 µL of these dilutions were spread over agar milieu (Muller-Hinton agar for bacteria, Sabouraud chloramphenicol agar for yeast and chocolate agar for *Cutibacterium acnes* (ATCC 6919) to assess the microbial load of the suspensions before the tests were performed.

2.8.3. Determination of antimicrobial potency

The microdilution technique in 96-well microplates was used to determine the Minimum Inhibitory Concentrations (MIC) and the Minimum Bactericidal Concentrations (MBC) or Fungicidal (MFC) in order to derive the antimicrobial potency (AP) of the extracts (Anani et *al*., 2015). A 100 µL aliquot of Muller-Hinton broth was deposited in all but the first wells of the microplate. 100 µL of the stock solution of the extracts (100 mg/mL) were deposited in these first and second wells. The mixture of the contents of the second wells was homogenized and then half dilutions were carried out by taking 100 µL of the solution each time. At the end of the dilutions, the concentrations of the extract obtained are : 100 ; 50 ; 25 ; 12.5 ; 6.25 and 3.125 mg/mL. Then 100 µL of microbial suspension was added to the contents of each well. The trials were carried out in triplicata. The micro plates were incubated at 37 °C for 24 hours (48 hours for *Cutibacterium acnes* (ATCC 6919). Macroscopic observations of the various wells were made to determine the MIC. The MIC of the extract is the smallest of the concentrations of the extract that does not show visible growth of the microorganism tested by the naked eye. The Minimum Bactericidal Concentration (BMC) or Fungicide (MFC) was determined by spreading the contents of all wells with an extract concentration greater than or equal to the MIC on agar milieu (Muller-Hinton agar for bacteria, Sabouraud chloramphenicol agar for yeast and chocolate agar for *Cutibacterium acnes* (ATCC 6919). Colonies were counted after incubation of the milieu at 37°C for 24 hours (72 hours for *Cutibacterium acnes* (ATCC 6919) and the specified MBC. The lowest concentration of the contents of the well without culture after spreading corresponds to the minimum bactericidal concentration (MBC) or minimum fungicidal concentration (MFC) (99.99% inhibition of the starting inoculum). The MBC/MIC (or MFC/MIC) report has made it possible to specify the modality of action of the extracts. If the MBC/MIC ratio is less than or equal to 2, the substance is said to be bactericidal or fungicidal. On the other hand, if it is greater than 2, the substance is said to be bacteriostatic or fungistatic (Fauchère, 2002).

2.9. Data analysis

The data collected were analyzed (calculation of percentages, means and standard deviations) with Microsoft's Excel spreadsheet, version 2019. Differences between results were considered significant at the 5% threshold (p-value < 0.05).

3. RESULTS

3.1. Extract yield

The hydroethanolic extract of *Momordica charantia* leaves has a greenish color; its pH is 6.75 and 7% of yield.

3.2. Phytochemical Screening

The hydroethanolic extract of the leaves of *Momordica charantia L*. revealed the presence of alkaloids, phenolic compounds, saponins, triterpenes, flavonoids but it revealed the absence of reducing compounds (*Table 1*).

Table 1: Phytochemical screening of *Momordica charantia* leaves extract

Chemical groups	***M. charantia* extract**
Favonoids	+
Phenolics compounds	+
Saponins	+
Alkaloids	+
Reducing compounds	-

Legend: +: presence; -: absence

3.3. Total flavonoids contents

The total flavonoids contents of *Momordica charantia* leaves hydroethanolic extract is 143.65 ± 2.51 mg RE/g.

3.4. Antioxidant activity

The antioxidant activity of the extract by the phosphomolybdate reduction (PR) method and by the FRAP method are presented below (*Table 2*).

Table 2: Results of antioxidant activity by the phosphomolybdate reduction (PR) method and by the FRAP method

	PR (mg AAE/g)	FRAP (mg FSE/g)
Momordica charantia L.	65.42 ± 6.24	165.50 ± 17.55

Legend: mg AAE/g : mg equivalent of ascorbic acid per gram of extract, mg ESF/g : mg equivalent of ferrous sulphate per gram of extract.

3.5. Antimicrobial activity

The results indicate that the hydroethanolic extract of *Momordica charantia* L. inhibits the *in vitro* growth of all tested germs to varying degrees. The results are reported in *Table 3*. The MIC for the crude hydroethanolic extract from the leaves of *Momordica charantia* range from 3.125 mg/mL to 50 mg/mL while the MBC ranges from 6.25 mg/mL to 100 mg/mL (Table 3).

In general, it is noted that the extract of *Momordica charantia* have a bactericidal action on strains of *Staphylococcus aureus* ATCC 29213, *Staphylococcus aureus* SARM, *Pseudomonas aeruginosa* ATCC 27853 and *Cutibacterium acnes* ATCC 6919 (the ratio of MBC/MIC = 2) and a fungistatic action on *Candida albicans* ATCC 10231 (with a MFC/MIC = 4).

Table 3: Action of the crude hydroethanolic extract of the leaves of *Momordica charantia L.* on microorganisms

Microbial strains	MIC (mg/mL)	MBC or MFC (mg/mL)	MBC/MIC or MFC/MIC	Types of activity
Staphylococcus aureus ATCC 29213	50	100	2	Bactericidal
Staphylococcus aureus MRSA	12.5	25	2	Bactericidal
Pseudomonas aeruginosa ATCC 27853	6.25	12.5	2	Bactericidal
Cutibacterium acnes ATCC 6919	3.125	6.25	2	Bactericidal
Candida albicans ATCC 10231	25	100	4	Fungistatic

MIC: Minimum inhibitory concentration in mg/mL; MBC: Minimum bactericidal concentration in mg/mL; MFC: Minimum fungicidal concentration in mg/mL

4. DISCUSSION

The results show that this extract has bactericidal and fungistatic activities on the microorganisms tested. This extract could therefore treat Infections including folliculitis, acne and skin fungus. Antimicrobial activities on Gram-negative bacteria could be beneficial on people with folliculitis. Anti *Cutibacterium acnes* shows promise for treating acne. The antifungal activity shows promise for the treatment of cutaneous fungal infections. The antimicrobial activity obtained from the hydroethanolic extract of *Momordica charantia L.* is in line with the work of Mada *et al*., (2013), Mwambete (2009) and Hsu *et al*., (2012) ; the results of which also revealed inhibitory activities of *Momordica charantia L.* on *Pseudomonas aeruginosa, Candida albicans, Staphylococcus aureus* and *Cutibacterium acnes.*

Significant antioxidant properties have been recorded in phytochemicals that are necessary for the reduction in the occurrence of many diseases caused by these microbes. Many polyphenolic constituents derived from plants are more effective antioxidants. Phytoconstituents employed by plants to protect them against pathogenic insects, bacteria, fungi or protozoa have been of relevance in human medicine. Thus the bactericidal and fungistatic activities of *Momordica charantia* L. could be explained by this hypothese.

The results of the phytochemical screening of *Momordica charantia* L. are in line with other work Mada et *al*. (2013) whose phytochemical screening of the crude extract of the leaves of *Momordica charantia L.* revealed the presence of alkaloids, tannins, saponins and flavonoids. The same applies to the work of Johnson et *al*. (2016). The results of antioxidant activity are similar to the results of Rezaeizadeh et *al*., (2011) whose work revealed that the methanolic extract of the fruits of *Momordica charantia L.* It has a strong antioxidant activity.

The compounds revealed by phytochemical screening are known to be biologically active and therefore contribute to the antimicrobial and antioxidant activities of *M. charantia*. These secondary metabolites exert antimicrobial activity through different mechanisms. Like what tannins have been shown to form irreversible complexes with the proline-rich protein (Shimada, 2006) resulting in the inhibition of cellular protein synthesis. The work of Behidj-Benyounes et *al*., (2013) confirm the high antimicrobial potential