DRUG DISCOVERY

Bioactive compounds in ethanolic extract of *Strychnos innocua* root using gas chromatography and mass spectrometry (GC-MS)

Alagbe, John Olujimi

To Cite:
Alagbe, Olujimi J. Bioactive compounds in ethanolic extract of *Strychnos innocua* root using gas chromatography and mass spectrometry (GC-MS). *Drug Discovery* 2023; 17: e4dd1005
doi: https://doi.org/10.54905/disssi.v17i39.e4dd1005

Author Affiliation:
Department of Animal Nutrition and Biochemistry, Sumitra Research Institute, Gujarat, India
Email: dralagbe@outlook.com
Orcid: 0000-0003-0853-6144

Peer-Review History
Received: 29 October 2022
Reviewed & Revised: 06/November/2022 to 14/January/2023
Accepted: 21 January 2023
Published: 24 January 2023

Peer-Review Model
External peer-review was done through double-blind method.

Drug Discovery
pISSN 2278–540X; eISSN 2278–5396

URL: https://www.discoveryjournals.org/drugdiscovery

DISCOVERY
SCIENTIFIC SOCIETY

ABSTRACT

Medicinal plants are of great relevance with endless pharmaceutical and therapeutic properties. They are source of wide array of secondary metabolites or bioactive compounds/phytochemicals (phenols, alkaloids, flavonoids, terpenoids, tannins, steroids and saponins) which are capable of discharging numerous biological functions (antimicrobial, antioxidant, antiviral, antifungal, anti-fibrotic, immune-modulatory, cytotoxic, antipyretic, antitumor, antihelminthic, antiprotozoal, antibacterial and so on). The use of gas chromatography and mass spectrometry analysis in identifying the bioactive compounds in ethanolic extract of *Strychnos innocua* root is a fundamental technique in quantifying the unknown samples, trace elements and contaminants leading to the discovery of novel compounds of pharmaceutical and biomedical importance. Result obtained showed that *Strychnos innocua* root extract contains 39 bioactive compounds with α-Cubebene (20.09 %) having the highest concentration followed by Dibutyl benzene -1,2 – dicarboxylate (10.17 %), β-Elemenone (10.02 %), 4-Methoxy-2-nitroformanilide (7.21 %), 1-Methyl cyclopropane methanol (5.96 %), 1, 3 propanediol, 2-ethyl 2-hydroxymethyl (3.71 %), Azelaic acid (2.87 %), Glycidol stearate (2.85 %), Chloromethyl 2-chlorodecanoate (2.83 %) and γ-terpinene (2.56 %) respectively. The remaining 29 bioactive compounds have concentrations less than 2 %. It was concluded that all the compounds observed are sources of medication that can be used traditionally in the treatment of human and animal diseases.

Keywords: *Strychnos innocua*, Phytochemicals, Pharmaceuticals, Therapeutics, Gas chromatography

1. INTRODUCTION

Medicinal plants are the major components of almost all indigenous or alternative systems of medicines. They contain phytochemicals which are safe, non-toxic and easily affordable (Singh et al., 2022; Shittu and Alagbe, 2020). According to WHO, (1996) about 80 % of the population in developing countries rely on medicinal plants for the treatment of various ailments. There are over 2000 medicinal plants with high potential that are yet to be explored (Oluwafemi et al., 2020). Bioactive compounds from these medicinal plants can perform an anti-inflammatory, antifungal, antiviral, antioxidant, immune-stimulatory, analgesics, antibacterial, anti-proliferative, cytotoxic and hepato-protective, antipyretic, antihelminthic, antiprotozoal, anti-depressant, anti-tumor, anti-fibrotic and hypolipidemic properties (Alagbe, 2022; Olafadehan et al., 2021; Agubosi et al., 2022) and could also aid in the discovery of drugs (Vasquez et al., 2017; Hirotani et al., 1991; Muritala et al., 2022).

Strychnos innocua also known as Natal orange belongs to the family Loganiaceae, genus Strychnos and order Gentianales. The tree is found in several countries such as; Angola, Guinea, Madagascar, Malawi, South Africa, Sudan, Mali, Uganda, Malawi, Zimbabwe, Zambia, Ethiopia and some parts of India (Maghembe, 1994). The trees are found in riverine fringes, sand forest and it can grow up to 3 -14 m high with simple leaves characterized by rounded emarginate or subacute apex (Hines and Eckman, 1993). Extracts from the leaves, roots and stems can be used traditionally for the treatment of snake bites, gastrointestinal, skin diseases, pneumonia and sexually transmitted infections (Hongxing et al., 2005; Al-Wathnani, 2012).

Previous studies have revealed that *Strychnos innocua* leaf, stem and root extract contains several bioactive compounds with antimicrobial properties and are also capable of inhibiting the activity of some bacteria and fungi such as; *Bacillus spp*, *Candida spp*, *Alternaria solani*, *Brevibacillus brevis*, *Cochliobolus lunatus*, *Escherichia coli*, *Enterobacter spp*, *Aspergillus spp*, *Fusarium spp*, *Klebsiella spp*, *Monascus ruber*, *Micrococcus luteus*, *Pseudomonas spp*, *Streptococcus spp*, *Styphylococcus spp*, *Salmonella spp* and *Shigella shiga* (Iyun et al., 2022; Sallau et al., 2022). Phytochemical analysis of methanolic extract from leaves and roots of *Strychnos innocua* revealed the presence of phenolic compounds which are capable of scavenging free radicals (antioxidants) (Hamisu et al., 2021; Lee et al., 2011; Igbal et al., 2011), preventing the risk of cardiovascular disease (Alagbe et al., 2022; Alagbe, 2021) and performing immune-modulatory activities in animals thus encouraging food safety (Oloruntola et al., 2018; Halliwell and Gutteridge, 1998). The aim of this study was to evaluate the bioactive compounds in ethanolic extract of *Strychnos innocua* root using gas chromatography and mass spectrometry (GC-MS).

2. MATERIALS AND METHODS

Experimental site, collection and preparation of *Strychnos innocua* ethanolic leaf and root extract

The study was performed at Sumitra Research Institute, Gujarat, India with a coastline of 1,600 Km, 23^{o} 13'N 72^{o}41'E. Fresh *Strychnos innocua* root was harvested from Waghai village, Saputara, India and identified at the Department of Biological Sciences, Sumitra Research Institute, Gujarat, India. The harvested roots were washed with distilled water, dried under the shade for 13 days and blended into powder form with the aid of electric blender and kept in an air tight labeled container. 100 g of grinded sample was soaked in 350 mL of 90 % ethanol for 48 hours with occasional stirring thereafter samples was sieved using Whatman's No.1 filter paper (10 cm) and stored in a sterile air tight container and stored in a cool dry place until further use.

Gas chromatography and mass spectrometry (GC-MS) of ethanolic extract from *Strychnos innocua* root

Gas chromatography mass spectrometry (GC-MS) analysis of ethanolic extract from *Strychnos innocua* root was performed with a Varian 450 GC system (Model 1842 series, China) equipped with fused silica column and it was operated at a temperature and pressure range of 50^{o}C to 450^{o}C isothermal 1079 PTV injector and 0 to 100 psi, consisting of split less injector with total flow of 500 mL/minutes at 10 psi, electron range of 150eV. Secondary compounds were identified with standard compounds in National Institute of Standard and Technology (NIST).

3. RESULTS

Table 1 Secondary metabolites in *Strychnos innocua* ethanolic root extract using GC-MS

Bioactive compounds	Area (%)	R.T (min)	Functions
Di-ethyl suberate	1.44	1.450	Antimicrobial and antioxidant
Ethyl Oleate	0.72	1.931	Antipyretic and antioxidant
Diisooctyl phthalate	0.01	2.500	Anti-depressant and antifungal
Glycidol stearate	2.85	3.444	Anitiviral, hepato-protective and antioxidant
1,2 – Benzenedicarboxylic acid	1.77		Anti-microbial, anti-proliferative
Monomethyl pimelate	0.02	6.091	Antifungal
γ-terpinene	1.10	9.435	Hepatoprotective and antifungal
4-fluoro-1-methyl-5-carboxylic acid	0.40	10.701	Anti-inflammatory, antibacterial and

			analgesics
3-Allyl-6-methoxyphenol	1.67	11.331	Antiprotozoal and cytotoxic
Cyclooctane	0.10	15.560	Anti-androgenic, antiviral and anti-inflammatory
Formamide	2.05	15.740	Hepato-protective, hypolipidemic, antimicrobial and antioxidant
α-cubebene	20.09	15.100	Antibacterial, antifungal, angelsics antipyretic and antioxidant
2,4,6 –Octatrien-1-ol	0.77	15.607	Antiviral and antioxidant
9,12-Octadecanoic acid	1.06	18.351	Cytotoxic, antioxidant, anti-inflammatory, antitumor, antifungal
α-longipinene	0.15	18.220	Anti-inflammatory, antioxidant, anti-depressant and antifungal
Azelaic acid	2.87	18.306	Anti-fibrotic, anti-inflammatory and hypolipidemic
Terpinen-4-ol	1.51	18.331	
1,3 propanediol, 2-ethyl 2-hydroxymethyl	3.71	18.211	Antibacterial, anti-inflammatory, antipyretic, antihelminthic and antifungal
γ-Terpinene	2.56	19.386	Amtioxidant and anti-inflammatory
β-Elemenone	10.02	19.931	Cytotoxic and hepato-protective
9-Octadecenoic acid	1.16	19.510	Antifungal
Torreyol-α-cadinol	0.83	19.259	Anitiviral, hepato-protective and antioxidant
Hepatadec-3-enal	0.30	19.400	Anti-microbial, anti-proliferative, antiviral, antihelminthic and antibacterial
Ethylene diacrylate	0.50	20.209	Analgesics, antibacterial, antifungal
1-Hexyl -2 nitrocyclohexane	1.62	21.344	Anti-inflammatory, antioxidant, anti-depressant
Chloromethyl 2-chlorodecanoate	2.83	21.381	Anti-fibrotic, anti-inflammatory
5-methylhexan-2-yl hepadecyl benzene -1,2 dicarboxylate	1.10	21.100	Antifungal, angelsics antipyretic and antioxidant
1-Methyl cyclopropane methanol	5.96	22.891	Hepatoprotective and antifungal
4-Acetoxy-3-methoxystyrene	1.14	23.080	Hepato-protective, hypolipidemic, antimicrobial
4-Methoxy-2-nitroformanilide	7.21	23.300	Cytotoxic, antioxidant
α-Terpinolene	0.02	23.701	Antioxidant, anti-proliferative
3-deoxy-d-mannoic acid	1.20	25.186	Antioxidant, anti-proliferative, antifungal and anti-inflammatory
Dibutyl benzene -1,2 - dicarboxylate	10.17	25.901	Cytotoxic and hepato-protective
Butyl undecyl benzene-1,2- dicarboxylate	1.31	25.670	Cytotoxic, antioxidant, anti-inflammatory, antitumor, antifungal
9,15 – Octadecadienoic acid	0.06	26.801	Antibacterial, antifungal, angelsics antipyretic
β-Cyclocitral	0.01	28.009	antitumor, antifungal
α-Phellandrene	1.10	34.491	Angelsics antipyretic and antioxidant
β-Phenethylamine	0.08	38.567	Anti-bacterial
Humulene	1.12	41.340	Antimicrobial, antifungal and hypolipidemic
Total	92.59		

R.T: reaction time (minutes)

4. DISCUSSION

Medicinal plants contain natural compounds or phytochemicals that are eco-friendly, safe and locally available with pharmacological properties (Musa et al., 2020; Adewale et al., 2021). They can also be used traditionally for the treatment of various ailments such as cold, cough, gastrointestinal disease, skin disease, respiratory disease, malaria, typhoid and snake bites (Nascimento et al., 2000; Perekh et al., 2007). Bioactive compounds are mostly secondary metabolites produced by plants via subsidiary pathways and are used by plants for growth, or defense against pathogens (Okeke et al., 2001; Oluwafemi et al., 2021; Narayani et al., 2012). Secondary metabolites in *Strychnos innocua* ethanolic root extract using gas chromatography and mass spectrometry (GC-MS) (Table 1) reveals that it is largely contains α-Cubebene (20.09 %), Dibutyl benzene -1,2 – dicarboxylate (10.17 %), β-Elemenone (10.02 %), 4-Methoxy-2-nitroformanilide (7.21 %), 1-Methyl cyclopropane methanol (5.96 %), 1,3 propanediol, 2-ethyl 2-hydroxymethyl (3.71 %), Azelaic acid (2.87 %), Glycidol stearate (2.85 %), Chloromethyl 2-chlorodecanoate (2.83 %) and γ-terpinene (2.56 %) respectively. Other compounds reported were less than 2.0 % however, they all have a marked therapeutic function (anti-inflammatory, antiviral, antifungal, antioxidant, hypolipidemic, angelsics, anti-pyretic, cytotoxic, antitumor and anti-depressant activities) (Okeke et al., 2001; Tease and Evans, 1989). The GC-MS component analyzed in this study is in consonance with the findings of Hamisu et al., (2021) but contrary to the reports of Hoet et al., (2006). This dissimilarity can be ascribed to processing or extraction procedures employed, parts of plant used, species, geographical location, age of plant as well as method of harvesting (Omokore and Alagbe, 2019; Hoet et al., 2006). The presence of phytochemicals in *Strychnos innocua* ethanolic root extract reveals that it has the ability to scavenge toxic chemicals in the body, inhibit the activities of pathogenic bacteria in the gut of animals, thus enhancing the absorption of nutrient as well as enhances the activities of enzymes (Oluwafemi et al., 2021; Narayani et al., 2012).

5. CONCLUSION

Medicinal plants have so several health benefits due to the presence of phytochemicals (alkaloids, flavonoids, phenols, terpenoids, saponins, tannins and steroids). Analyzing the bioactive components in *Strychnos innocua* ethanolic root extract will unleash some of the potential pharmaceutical properties in the plant. Adopting the use of gas chromatography and mass spectrometry will further aid in identifying and quantifying unknown samples, unknown contaminants, trace elements and gases.

Ethical approval

Not applicable.

Informed consent

Not applicable.

Conflicts of interests

The authors declare that there are no conflicts of interests.

Funding

The study has not received any external funding.

Data and materials availability

All data associated with this study are present in the paper.

REFERENCES AND NOTES

1. Adewale AO, Alagbe JO, Adeoye AO. Dietary Supplementation of *Rauvolfia Vomitoria* Root Extract as A Phytogenic Feed Additive in Growing Rabbit Diets: Haematology and serum biochemical indices. International Journal of Orange Technologies 2021; 3(3):1-12.
2. Agubosi OCP, Alexander James, Alagbe JO. Influence of dietary inclusion of Sunflower (*Helianthus annus*) oil on growth performance and oxidative status of broiler chicks. Cent Asian J Med Nat Sci 2022; 2(7):187-195.
3. Agubosi OCP, Soliu MB, Alagbe JO. Effect of dietary inclusion levels of *Moringa oleifera* oil on the growth performance and nutrient retention of broiler starter chicks. Central Asian J Theor Appl Sci 2022; 3(3):30-39.
4. Agubosi OCP, Wika BK, Alagbe JO. Effect of dietary inclusion of Sunflower (*Helianthus annus*) oil on the growth

performance of broiler finisher chickens. Eur J Mol Clin Med 2022; 2(5):1-10.

5. Alagbe JO, Ramalan SM, Shittu MD, Olagoke OC. Effect of *Trichilia monadelpha* stem bark extract on the fatty acid composition of rabbit's thigh meat. Journal of Environmental Issues and Climate Change 2022; 1(1):63-71.
6. Alagbe JO. Caecal Microbial Population of Growing Grass Cutters (*Thyronoyms Swinderianus*) Fed *Phyllantus Amarus* and *Pilogstigma Thonngii* Leaf Meal Mixture as Partial Replacement for Soya Bean Meal. Concepts dairy vet sci 2020; 3(5):350–355.
7. Alagbe JO. Chemical evaluation of proximate, vitamin and amino acid profile of leaf, stem bark and roots of *Indigofera tinctoria*. International Journal on Integrated Education 2020; 3 (10):150-157.
8. Alagbe JO. Dietary Supplementation of *Rauvolfia Vomitoria* Root Extract as A Phytogenic Feed Additive in Growing Rabbit Diets: Growth Performance and Caecal Microbial Population. Concepts dairy vet sci 2021; 4(2):2021.
9. Alagbe JO. Use of medicinal plants as a panacea to poultry production and food security: A review. Gospodarka I Innowacje 2022; 22(2022):1-12.
10. Alagbe JO. *Daniellia oliveri* leaf extracts as an alternative to antibiotic feed additives in broiler chicken diets: Meat Quality and Fatty acid composition. Indones J Inno Appl Sci 2021; 1 (3):177-186.
11. Alagbe JO. *Prosopis africana* stem bark as an alternative to antibiotic feed additives in broiler chicks diets: Performance and Carcass characteristics. Journal of Multidimensional Research and Reviews 2021; 2(1):64-77.
12. Alagbe JO. Gas chromatography and mass spectroscopy of *Juniperus phoenice* stem bark extract and its influence on the haemato-biochemical values of growing rabbits. British Scientific Periodical 2022; 1(1):18-33.
13. Alagbe JO, Adeoye A, Oluwatobi OA. Proximate and mineral analysis of *Delonix regia* leaves and roots. International Journal on Integrated Education 2020; 3(10):144-149.
14. Alagbe JO, Sharma R, Eunice AO, Shittu MD, Bello KA. Chemical evaluation of the proximate, minerals, vitamins and phytochemical analysis of *Daniellia oliveri* stem bark**.** Int J Biol Chem Sci 2020; 2(1):16-22.
15. Alagbe JO, Shittu MD, Tanimomo BK. Influence of *Anogeissusleio carpus* stem bark on the fatty acid composition in meat of broiler chickens. European Journal of Life Safety and Stability 2022; 14(22):13-22.
16. Alagbe JO, Zubairu Habiba, Adedeji OM, Bamigboye S, Dora Agbonika. Influence of *Juniperus thurifera* root extract on the nutrient digestibility and caecal microbial count of growing rabbits. Web of Synergy: Int Interdiscip Res J 2022; 1(1):5-17.
17. Al-Wathnani H, Ismet A, Tahmaz RR, Al-Dayel TH, Bakir MA. Bioactivity of natural compounds isolated from cyanobacteria and green algae against human pathogenic bacteria and yeast. J Med Plant Res 2012; 6(18):3425–3433.
18. Asuzu CU, Nwosu MO. Studies of the wood of some Nigeria alkaloid-rich Strychnos species. J Hortic 2020; 12(2):57–62.
19. Halliwell B, Gutteridge JMC. Free radicals in biology and medicine. Oxford University Press, Oxford, UK 1998.
20. Ibrahim H, Uttu AJ, Sallau MS, Ogunkemi, Iyun ORA. Gas chromatography–mass spectrometry (GC–MS) analysis of ethyl acetate root bark extract of *Strychnos innocua* (Delile). Beni-Suef Univ J Basic Appl Sci 2021; 10:65.
21. Hines DA, Eckman K. Indigenous multipurpose trees for Tanzania: Uses and economic benefits to the people. Cultural survival Canada and Development Services Foundation of Tanzania 1993.
22. Hirotani H, Ohigashi M, Kobayashi K, Koshimizu K, Takahashi E. Inactivation of T5 phage by cis-vaccenic acid, an antivirus substance from Rhodopseudomonas capsulate, and by unsaturated fatty acids and related alcohols. FEMS Microbiol Lett 1991; 77(1):13–17.
23. Hoet S, Stevigny C, Herent M, Quetin-Leclercq J. Antitrypanosomal compounds from the leaf essential oil of *Strychnos spinosa*. Planta Med 2006; 72:480–482.
24. Hongxiang S, Cuirong S, Yuanjian P. Cytotoxic activity and constituents of the volatile oil from the roots of Patrinia scabra Bunge. Chem Biodivers 2005; 2(10):1351–1357.
25. Igbal H, Moneebur RK, Riazullah ZM, Naeem KFA, Zahoor U. Phytochemical screening and antimicrobial activities of selected medicinal plants of Khyber Pakhtunkhwa, Pakistan. Afr J Pharm Pharmacol 2011; 5(6):746–750.
26. Lee SW, Wendy W, Julius YFS, Desy FS. Characterization of antimicrobial, antioxidant, anticancer properties and chemical composition of Michelia champaca seed and fower extracts. J Pharm Sci 2011; 4(1):19–24.
27. Maghembe JA. Germination studies on seed of fruit trees indigenous to Malawi. For Ecol Manag 1994; 64(2-3):111-125.
28. Muritala DS, Alagbe JO, Ojebiyi OO, Ojediran TK, Rafiu TA. Growth performance and haematological and serum biochemical parameters of broiler chickens given varied concentrations of *Polyalthia longifolia* leaf extract in place of conventional antibiotics. Anim sci Genet 2022; 18(2):57-71.
29. Musa B, Alagbe JO, Adegbite MB, Omokore EA. Growth performance, caeca microbial population and immune response of broiler chicks fed aqueous extract of *Balanites*

aegyptiaca and *Alchornea cordifolia* stem bark mixture. United Int J Res Technol 2020; 2(2):13-21.

30. Narayani M, Johnson M, Sivaraman A, Janakiraman N. Phytochemical and Antibacterial Studies on Jatropha curcas L. J Chem Pharm Res 2012; 4(5):2639-2642.
31. Nascimento GGF, Lacatelli J, Freitas PC, Silva GL. Antibacterial activity of plant extracts and phytochemicals on antibiotic-resistant bacteria. Braz J Microbiol 2000; 31(4):886-891.
32. Okeke MI, Iroegbu CU, Eze EN, Okoli AS, Esimone CO. Evaluation of extracts of the roots of Landolpbia owerience for antibacterial activity. J Ethnopharmacol 2001; 78(2-3):119-27.
33. Olafadehan OA, Oluwafemi RA, Alagbe JO. Performance, haemato-biochemical parameters of broiler chicks administered Rolfe (*Daniellia oliveri*) leaf extract as an antibiotic alternative. J Adv Res Rev 2020; 1:4.
34. Oloruntola OD, Agbede JO, Ayodele SO, Adeyeye SA, Agbede JO. Performance, haemato-biochemical indices and antioxidant status of growing rabbits fed on diets supplemented with Mucuna pruriens leaf meal. World Rabbit Sci 2018; 26:277-285.
35. Oluwafemi RA, Agubosi OCP, Alagbe JO. Proximate, minerals, vitamins and amino acid composition of *Prosopis africana* (*African mesquite*) seed oil. Asian J Adv Res 2021; 11(1) :21-27.
36. Oluwafemi RA, Daniel SE, Alagbe JO. Haematology and serum biochemical indices of broiler chicks fed different inclusion levels of ginger (*Zingiber officinale*) and garlic (*Allium sativum*) oil mixture. Int J Res Sci Innov Appl Sci 2021; 1(4):20-26.
37. Oluwafemi RA, Lawal AO, Adelowo SA, Alagbe JO. Effects of dietary inclusion of ginger (*Zingiber officinale*) and garlic (*Allium sativum*) oil on carcass characteristics and sensory evaluation of broiler chicken. Texas Journal of Multidisciplinary Studies 2021; 2(11):180-188.
38. Oluwafemi RA, Uankhoba IP, Alagbe JO. Effects of turmeric oil as a dietary supplement on the growth performance and carcass characteristics of broiler chicken. International Journal of Orange Technologies 2021; 3(4):1-9.
39. Oluwafemi RA, Uankhoba IP, Alagbe JO. Effects of turmeric oil as a dietary supplement on the haematology and serum biochemical indices of broiler chickens. J Proteom Bioinform 2021; 5(1):000138.
40. Omokore EO, Alagbe JO. Efficacy of dried *Phyllantus amarus* leaf meal as an herbal feed additive on the growth performance, haematology and serum biochemistry of growing rabbits. Int J Res Dev 2019; 4(3):97-104.
41. Orwa CA, Mutua KR, Jamnadass R, Anthony S. Agroforestree database: A tree reference 2009. Retrieved from http:// www.worldagroforestry.org/sites/treedbs/treedatabases.asp
42. Parekh J, Chands S. In vitro antibacterial activity of the crude methanol extract of Woodfordia fruticosa Kurz. Flower (Lythraceae), Braz J Microbiol 2007; 38:204-7.
43. Ruth TN, Anita RL, Loveness KN, Vincenzo F, Ruud V. Local processing and nutritional composition of indigenous fruits: The case of monkey orange (*Strychnos spp.*) from Southern Africa. Food Res 2017; 33(2):123–142.
44. Shittu MD, Alagbe JO. Phyto-nutritional profiles of broom weed (*Sida acuta*) leaf extract. International Journal of Integrated Education 2020; 3(11):119-124.
45. Singh S, Alagbe OJ, Liu X, Sharma R, Kumar A. Comparative analysis of ethanolic *Juniperus thurifera* leaf, stem bark and root extract using gas chromatography and mass spectroemetry. International Journal of Agriculture and Animal Production 2022; 2(6):18-27.
46. Trease GE, Evans WC. Pharmacognasy WB. Scandars Company Ltd. London 1989; 14:269-300.
47. Vasquez-Ocmin P, Cojean S, Rengifo C, Suyyagh-Albouz S, Guerra CAA, Pomel S, Maciuk A. Antiprotozoal activity of medicinal plants used by Iquitos-Nauta road communities in Loreto (Peru). J Ethnopharmacol 2017; 4(61):1-9.
48. WHO. Annex II. Guidelines for the assessment of herbal medicines (WHO Technical Report Series No. 863), Geneva 1996.