PRODUCTION AND CHARACTERIZATION OF BLOOD TONIC CONSTITUENTS FROM LOCAL HERBS

Jimoh, A. (Ph.D)
Department of Chemical Engineering,
Federal University of Technology Minna, Niger state, Nigeria

Abstract

This paper investigates the presence of blood tonic constituents in local herbs. Phytochemical analysis were carried out on each extract obtained from the barks of theobroma cacao, khaya senegalensis, treculia africana, magnifer indica; pod of parkia filicodea (African locust bean); chaff of sorghum bicolor and spectrophotometric analysis of the mixture extract of these herbs were also carried out. Phytochemical analysis shows that the above named herbs extract contain compounds of alkaloids, saponins, and tannin sesquiterpenes in different proportions. The spectrophotometric analysis shows that the mixture extract contains moderate quantities of vitamin B1 (thiamine), Vitamin B6 (pyridoxine), Vitamin B2-5 phosphate and low amount of ferrous gluconate. Through the characterization, it could be deduced that the above mentioned herb sample has important pharmacological effects and components that are suitable for blood tonic production.

Introduction

Medicinal plants are known to provide a rich source of raw materials for traditional medicine in Africa and other parts of the developing world. There is hardly any need to stress that about 85% of Africans living on the continent are forced to resort to traditional medicine and other practitioners for the continued preservation of their health and also to alleviate their diverse sufferings (Usman, 1990).

It is an established fact that many diseases were treated and still being treated in traditional medical practice with great success. From literature and oral tradition, the diseases which have been or claimed to have been treated with success include malaria, epilepsy, anemia, infertile convulsion, diarrhea, dysentery, bacterial and fungi infections, mental illness, asthma, diabetes, warm infestation, pains, colic and ulcers etc (Akinde, 1980). In addition, medicinal plants have been synthesizing a large variety of chemical substances since their first day of life on earth. These substances include, in addition to the basic metabolites, phenolic compounds, terpenes, steroids, alkaloids, glycosides and a host of other chemical substances referred to as secondary metabolites which are of no importance to the plant's own life but possess important therapeutic properties in the treatment and cure of human diseases (Sofowora, 1982).

In recent time, our traditional medical practice has been subjected to scientific studies. These studies have been mainly on the herbal substances used in the practice, in an attempt to

validate the efficiency of such substances. The studies have been on the pharmacology and toxicology of extractives from the plants, isolation and chemical characterization of the active principles, and to a limit extent, formulation of the substances as medicine (Okpaniyi, 1997). The scientist however who has the task of compiling and evaluating so many common used medicinal plants must establish their botanical profiles and their pharmaceutical potentials including where possible their dosage and uses (Balami, 1984). The extraction of the active constituents such as alkaloids, tannins, phylobatanins, sesquiterpenes etc. was accomplished by the use of phytochemical analysis (Trease & Evans, 1978). Medicinal plant parts had been used in recent times in the preparation of blood tonic mixture by boiling the herbs to a certain temperature to obtain the mixture (Usman, 1990).

Blood tonic mixture is a liquid mixture that effectively corrects deficient hematopoiesis. The vital constituents contained in blood tonic mixture includes; vitamins B - complexes, folate and iron compounds. Literature show that series of works had been done on extraction of important chemicals from parts of plants but little has bee done so far on the preparation of blood tonic mixture using deduction of local herbs. In this investigation, the extract of each of the herb samples was considered for phytochemical analysis to determine the active constituents present and their medicinal values. Also, spectrophotometric method of analysis was performed on specified volume of the prepared mixture in order to determine their stability for blood tonic production.

Methodology

Collection and treatment of samples

Certain portion of each of the herb samples considered was weighed and dried in an oven at about 60°c, a temperature at which no overheating occurred and which is known to be high enough to put an immediate check on enzymic and respiratory action. Drying was completed within 24 hrs.

Hot water extraction

Hot distilled water was employed as solvent. 250ml of distilled water was heated with 30g of each herb sample in a 500ml beaker by placing it on a heater at a temperature of 60°c with the use of a thermometer. The mixture were allowed to settle down and left over night in the refrigerator. The extracts were filtered through filter papers (No. 1, Whatman, U. K.), and the filtrate were used for further analysis.

Phytochemical analysis

Extraction of alkaloids

2ml of each extract was treated with l0ml of 1% HCL in a water bath for 30mins. The solution was then treated with a few drop of Dragendorff's reagent. A whitish substance was precipitated which is then allowed to settle and the solution drained off (Trease & Evans, 1978).

Extraction of tannins

20ml of each extract was mixed with 20ml of distilled water, then few drops of Iron III chloride was added to the mixture. A blue-green substance precipitated, it was allowed to settle and the solution was drained off (Trease & Evans, 1978).

Extraction of sesquiterpenes

0.5ml of each extract was mixed with 0.1ml of methanol by shaking several times. 0.4ml of 5% H_2SO_4 containing 0.5% ferric chloride solution was added to the mixture. The mixture was boiled on a water bath for a minute. A brownish coagulated substance precipitated and the solution drained off (Trease & Evans, 1978).

Extraction of phylobatanns

2ml of each extract was boiled with 5ml of 5% HCL. A red precipitate was observed and allowed to settle after which the solution was drained off (Trease & Evans, 1978).

Spectrophotometric analysis of mixture extract

Spectrophotometric analysis was performed on the mixture extract as follow:

Mixture extract preparation

The blood tonic herbs were considered and mixed in the proportion shown below;

(i) 220g sorghum bicolor chaff
(ii) 240g magnifera indica bark
(iii) 240g theobroma cacao bark
(iv) 200g parkia filicoidea pod
(v) 240g khaya senegalensis bark
(vi) 20g potash
(vii) 240g Treculia Africana bark

The herbs were boiled together for 30 minutes. The mixture was allowed to cool and then decanted.

Assay: ferrous gluconate

Test: 5ml of the mixture was transferred into 500ml volumetric flask, 20ml of 1M H_2SO_4 was added and made up to mark with water.

Standard: 200mg ferrous gluconate was weighed into another 50ml volumetric flask. Little quantity of 1M H_2S0_4 was added, and made up to mark with water. 1Oml of both test and standard was transferred into 50ml flask each. 4ml of 20% citric acid and 5 drops of thioglicolic acid were added and shaken. The mixture was then made up to mark with dilute ammonia solution. Then the absorbance was read at 550nm.

Assay: vitamin B2-5 phosphate

Test: 10ml of test was transferred into 100ml volumetric flask and made up to mark with water.10ml of the standard was also transferred into 100ml volumetric flask and made up to mark with water. 10ml of both standard and test was transfer into 50ml flask each. 2ml each of 20% citric acid and of 45% $KMN0_4$ solution were added to the mixture and shaken; the mixture was then allowed to stand for 2 minutes. 1ml of H_2O_2 solution was subsequently added and the solution was allowed to stand for 5 minutes and the absorbency was read at 450nm.

Standard: 300mg of B2-5 phosphate was weighed into 1000ml flask and made up to volume.

Blank: 10ml of the mixture was used without B2-5 phosphate and treated as above.

Assay: vitamin B1 (thiamine)

Bromothymol Blue Solution: 62.44mg of Bromothymol blue was weighed into a 250ml volumetric flask. It was dissolved and made up to volume with chloroform.

Standard: 40mg of Vitamin B1 was weighed into a 100ml flask, dissolved and made up to volume with water. 10ml of the resulting solution was pipette into a 250ml volumetric flask and made up to volume with chloroform.

Test: 100ml of the sample was pipette into a repeating funnel and treated as the above standard. The absorbency of both the standard and test was read at 420nm using chloroform as the blank.

Assay: vitamin B6 (pyridoxine)

Standard: 100mg of pyridoxine previously dried was weighed accurately into a 1000ml flask. 10ml of 0.1N HCL was added and shaken to dissolve; it was then diluted to 1000ml with water.

10ml from the resulting solution was pipette into a 100ml flask and made up to mark with 0.1N HCL.

Colour development: This was performed in a dry test tube and the following reagents were added in succession mixing after the other. 200ml of the test was measured into 200ml volumetric flask.

Table 1: different reagents used during the experiment

	Standard blank	Test	Test blank	Standard
Test	-	2ml	2ml	-
Isopropyl	4ml	4ml	4ml	4ml
Ammonium buffer 10.9	2ml	2ml	2ml	2ml
Boric acid	1ml	-	1ml	-
Water	1ml	-	1ml	-
Standard	2ml	-	-	2ml

Results and discussion

Table 2: Phytochemical analysis of the extract

	Extracts					
AC	MTB	MATB	SC	LBP	CTB	ABTB
Alkaloids	-	-	-	-	++	-
Anthraquinones	-	-	-	-	-	-
Cardial glycosides	+	-	-	++	-	-
Saponins	+	++	-	-	-	+
Tannin	+	-	-	++	-	-
Sesquiterpenes	+	-	-	++	-	-
Phylobatannins	+	-	++	+++	-	-

Active constituents = AC; Mango tree bark = MTB; Mahogany tree bark = MATB; Sorghum chaff = SC; Locust bean pod = LBP; Cocoa tree bark = CTB; African breadfruit tree bark = ABTB.

+++ Abundantly present.

++ Moderately present.

+ Sparing present

- Absent.

Table 3: Spectrophotometric analysis of the mixture extract

Mixture extract			
Assay	Absorbance test	Absorbance standard	Potency (mg)
Vitamin B1 (thiamine)	0.039	0.040	1.95
Vitamin B2-5 phosphate	0.061	0.082	1.04
Vitamin B6 (pyridoxine)	0.049	0.030	2.0
Ferrous gluconate	0.024	0.576	8.33

Discussion

Phytochemical analysis is the basis for the extraction of active constituents present in local herbs. Generally, the result of the phytochemical analysis of sample extracts obtained in this study (table 2) is close to those obtained in the literature (Oliver, 1960).

Compounds of alkaloids, tannins, cardiac glycosides, saponins, Sesquiterpenes and phylobatannins were found to be present in the sample extract (table 2) in different proportions and are soluble in water. While Anthraquinones were totally absent (although they are insoluble in water). The herbs samples collected contain some pharmacological components which are useful in medicine due to the presence of compounds of alkaloids and saponins. Alkaloids have a number of pharmacological properties which includes: central nervous system (CNS) and respiratory simulations, skeletal muscle stimulation, diuresis, cardiac stimulation and smooth muscle relaxation (Margaret & Brian, 1990). Saponins have low oral toxicity with potentially useful nutritive value. It reduces blood cholesterol levels in humans under conditions, which could be expected to induce high levels of blood cholesterol (Macrae, Robinson & Sadler, 1997).

The spectrophotometric method of analysis is used in determining the various vitamins and iron contents contained in mixture extract. The result obtained from the analysis of the mixture extract (table 3) shows that Vitamin B1 (thiamine), Vitamin B2-5 phosphate, Vitamin B6 (Pyridoxine) and ferrous gluconate were present in the extract. The Potency analysis of the mixture extract shows that Vitamin B1 (thiamine) is present in a moderate quantity with a value

of 1.95mg, which falls within the specified range in the literature (1.8 - 2.4) mg (Macrae, Robinson & Sadler, 1997). The moderate value of Vitamin B1 in the mixture could be attributed to the labile nature of the nutrient. The Vitamin loss noticed could be as a result of unprotected boiling that may be avoided through boiling in plastic bags. Thus, this shows that preparation technique does influence the loss (Bender, 1992). The potency of Vitamin B2-*5* phosphate is 1.04mg, this is in agreement with the ranges 1.3 - 1.7mg reported in the literature (Macrae, Robinson & Sadler, 1997). 8.33mg potency value obtained for Ferrous gluconate is very insignificant when compare to the specified value of 190 – 210mg reported (Macrae, Robinson & Sadler, 1997). The low value of ferrous gluconate in the mixture extract may be attributed to the unstable property of iron at a pH greater than 3.5 as earlier reported (Macrae, Robinson & Sadler, 1997) compared to that of the mixture extract that was read at a pH 7.0. Vitamin B6 (pyridoxine) could be considered as present in a moderate quantity with potency value of 2:0mg that falls within the range of 1.8 - 2.4 mg reported (Macrae, Robinson, & Sadler, 1997). The various Vitamins and Iron compounds have very useful pharmacological effects in blood tonic production. The presence of the various vitamins is necessary for normal cellular metabolism and the co-ordination of many vital intermediate chemical reactions and enzymes (Olaniyi, 1980).

Conclusion

The phytochemical analysis of the mixture extract from local herbs shows the presence of active constituents such as compounds of alkaloids, saponins etc. that has very useful pharmacological effects in blood tonic. It was also deduced that water is the best solvent that can extract almost all the active constituents extracted from the local herbs. The various vitamins and iron compounds contained in the mixture extract also proves the reliability of the local herbs combination for blood tonic production.

References

Akinde, B. E. (1980). *Phytochemical and microbiological evaluation of the oils from the leaves of a Calypha willcesiana mull.* Arg. OAU press, University of Ile-Ife, Nigeria. Pp. 41-42.

Balami, A. H. (1984). *Screening of some local medicinal plant, B.Pharm.* Unpublished Thesis, Ahrnadu Bello University, Zaria, Nigeria, Pp 10-25.

Bender, D. A. (1992). *Nutritional Biochemistry of the vitamins.* 3rd edition, Cambridge University press, Cambridge. Britain. Pp. 267-269.

Culei, I. (1982). *Methodology for analysis of vegetable drugs: Practice manual on industrial utilization of medicinal and aromatic plants.* Bucharest of the joint UNIDO, Romania. Pp. 2-67.

Macrae, R., Robinson, R. K., & Sadler, M. J. (1997). *Encyclopedia of food science, food technology and nutrition*, Vol. 6. Academic press limited. London. Pp. 3998- 3999.

Margaret, L.V. & Brian, V. (1990). *Plant products of tropical Africa.* Macrnillan College Editions. Macrnillan press Ltd. London. Pp. 71-74.

Okpaniyi, S. N. (1997). *Perspectives in medicinal plant research today.* Local bulletin of Dept. of Research Unit (DRU), Ile-Ife, Nigeria. Pp. 89

Oliver, B. (1960). *Medicinal plants in Nigeria*. Private Edition. Pp. 24-26.

Olaniyi, A.A. (1980). *Nigeria Journal of Pharmacy, Vol.11, pp.133.*

Sofowora, A. (1982). *Medicinal plants and traditional medicine in Africa.* John Wiley and Sons Ltd., New York. USA. Pp.1-20.

Trease, G. E. & Evans, W. C. (1978). *Textbook of Pharmacognosy.* (11th Edition). Baillere Tindall, London. Britain. Pp. 60-75.

Usman, A. (1990). *The practice of traditional medicine in the treatment of Anemia.* Unpublished B. Pharm. Thesis, Ahmadu Bello University, Zaria, Nigeria.