IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals
ISSN 2349-7203
HUMAN

Human Journals
Research Article
April 2016 Vol.:6, Issue:1

Phytochemical and Haemolytic Activity on Leaves of *Cassia obtusifolia* Linn.

IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals
ISSN 2349-7203
HUMAN

Sufiyan Ahmad[1]*, Rashid Akthar[2], Md. Imran[2], Amit Sinhal[1], Nandvishal Deore[1]

1. *Department of Pharmacognosy, Gangamai college of Pharmacy, Nagaon, Dist. Dhule (M.S.), India.*
2. *Department of Pharmacognosy, KBHSS Trust's college of Pharmacy, Malegaon, Dist. Nashik (M.S.), India.*

Submission: 1 April 2016
Accepted: 7 April 2016
Published: 25 April 2016

Keywords: *Cassia obtusifolia* Linn., Leaves and aqueous extract

ABSTRACT

The objective of present studies deals with the preliminary phytochemical studies on the aqueous extract of leaves of *Cassia obtusifolia* Linn. and phytochemical test for the identification of active constituents. Saponin was identified by foam test. Since the most characteristic properties of saponin is their ability to cause haemolysis, when added to suspension of blood saponin produced changes in erythrocyte membranes, causes haemoglobin to diffuse into surrounding medium. The aqueous extract of *Cassia obtusifolia* Linn. produce haemolysis in the test tube no.11 (i.e. 0.90ml of extract or 0.009gm/ml of extract) and it was calculated by using formula given in the procedure. The haemolytic activity was found to be 722 with reference to standard saponin R i.e. 1000 unit. The result of haemolytic activity is shown in fig. 1.

HUMAN JOURNALS

www.ijppr.humanjournals.com

INTRODUCTION

Cassia obtusifolia Linn. (Fabaceae) is an annual herb or under herb, 0.5-2.0 m in height, Leaves are 7-15cm long, with a conical gland between the lowest pair of leaflets. The tender leaves, twigs and young pods are cooked and eaten as potherb. The leaves are gently aperients. Distributed from Jammu Kashmir and Himachal Pradesh to West Bengal and in Orissa, Madhya Pradesh, Rajasthan, Gujarat and Maharashatra, up to an altitude of 1,200 m. It has been also reported from Andhra Pradesh, Karnataka and Tamilnadu.

It is well known as Marathi (Tarwatta) Rajasthan (Pandia, Pumaria) Uttar Pradesh (Banarh, Chakwar, Rosangi) Mundari (Cakonda) Its leaves is used as ringworm, skin diseases, cough, cold, asthma, mild purgative, in liver complaints, vomiting, stomachache, headache and also applied to foul ulcers[1]. The plant contains anthraquinons, sterol, triterpenoids[2] and anthraquinons glycosides[3]. The objective of present study is to focus on Phytochemical characteristics and haemolytic activity of leaves of *Cassia obtusifolia* Linn.

MATERIALS AND METHODS

Plant material

The plant specimens for the proposed study were collected from Dhule dist. care was taken to select healthy plants and for normal organs. The plant was authenticated by Dr. P.G. Diwakar (joint director) of BSI Pune (M.S.) Ref. No. BSI/WRC/TECH./2011.

Preliminary Phytochemical Parameters

Preliminary phytochemical test of *Cassia obtusifolia* Linn. performed and the chemical constituents were detected[4,5] .

Haemolytic Activity

Many medicinal plant materials, especially those derived from the families Caryophyllaceae, Araliaceae, Sapindaceae, and dioscoreacae contain Saponins. The most characteristic property of Saponins is their ability to cause haemolysis: when added to a suspension of blood. Saponins

produce changes in erythrocyte membranes, causes haemoglobin to diffuse into the surrounding medium.

The haemolytic activity of plant materials, or a preparation containing saponins, is determined by comparison with that of a reference material, Saponins R, which has haemolytic activity of 1000 units per gm. A suspension of erythrocytes is mixed with equal volumes of serial dilution of the plant material extract. The lowest concentration of effect complete haemolysis is determined after allowing the mixtures to stand for given period of time. A similar test is carried out simultaneously with Saponins R.

Procedures proposed for the determination of haemolytic activity of saponaceous medicinal plant material are based on the same principle although the details may vary, e.g. the source of erythrocytes, methods for the preparation of the erythrocytes suspension and the plant material extract, the defined haemolytic activity of the reference material of saponin, and the experimental method. In order to obtain reliable result, it is essential to standardize the experimental conditions, and especially to determine the haemolytic activity by comparison with that of saponin R.

Preparation of erythrocytes suspension

To prepared erythrocyte suspension filled a glass-stoppered flask to one-tenth of its volume with sodium citrate (36.5g/I) TS, swirling to ensure that the inside of the flask is thoroughly moistened. Introduced a sufficient volume of the of blood freshly collected from the healthy ox and shake immediately.

Preparation of reference solution

About 10mg of Methylene salicylate, accurately weighed, to a volumetric flask and add sufficient phosphate buffer pH 7.4 TS to make 100ml. This solution should be freshly prepared.

Preparation of test solution

1gm of aqueous extract of leaves of *Cassia obtusifolia* Linn. was dissolved in 100ml of water to produce a concentration of 0.01 gm/ml.

Procedure

Prepared serial dilution of the plant material extract, with phosphate buffer pH 7.4 TS and blood suspension (2%) using 13 test tubes as shown in table 1.

Carried out the dilutions and evaluation as mentioned in following table and observed the results after 24 hours. Calculate the amount of medicinal plant material in g, or of the preparation in g or ml, that produces the total haemolysis. To eliminate the effect of individual's variations in resistance of the erythrocyte suspension to saponin solutions, prepare serial dilutions of saponin R in the same manner as described above for the plant material extract. Calculate the quantity of saponin R that produces total haemolysis (Table 1)[6].

Table 1: Haemolytic activity determination procedure

	1	2	3	4	5	6	7	8	9	10	11	12	13
Plant material extract (diluted if necessary) (ml)	0.40	0.45	0.50	0.55	0.60	0.65	0.70	0.75	0.80	0.85	0.90	0.95	1.00
Phosphate buffer PH 7.4 TS (ml)	0.60	0.55	0.50	0.45	0.40	0.35	0.30	0.25	0.20	0.15	0.10	0.5	--
Blood suspension (2%) (ml)	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0

RESULTS AND DISCUSSION

Preliminary Phytochemical Studies

Aqueous extract of leaves of *Cassia obtusifolia* Linn. showed the presence of various Phytoconstituents such as glycosides, saponins, triterpenoids, tannins, flavanoid, alkaloids and carbohydrates. Saponin was identified by foam test. since the most characteristic properties of saponin is their ability to cause haemolysis, when added to a suspension of blood saponin

produced changes in erythrocyte membranes, causes haemoglobin to diffuse into surrounding medium.

The aqueous extract of leaves of *Cassia obtusifolia* Linn. produce haemolysis in the test tube no.11(i.e. 0.90ml of extract or 0.009gm/ml of extract) and it was calculated by using formula given in the procedure. The haemolytic activity was found to be 722 with reference to standard saponin R i.e. 1000 unit. The result of haemolytic activity is shown in fig. 1.

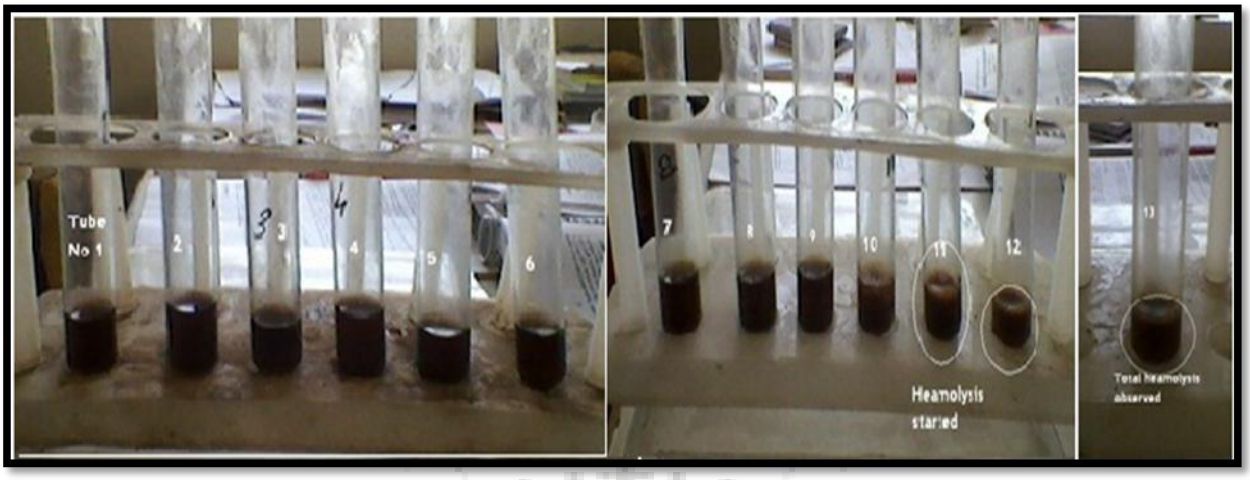

Fig. 1: Result of haemolytic activity

Calculate the haemolytic activity of the medicinal plant material using the following formula:

$$1000 * a/b$$

Where 1000= the defined haemolytic activity of saponin R relation to OX blood,

a= quantity of saponin R that produces total haemolysis (g),

b= quantity of plant material that produces total haemolysis (g).

= 1000*0.65/0.90

= 722

CONCLUSION

The present phytochemical studies on leaves of *Cassia obtusifolia* Linn. might be useful to supplement assumed significantly in the way of acceptability of herbal drugs in present scenario that lacks regulatory laws to control the quality of herbal drugs.

ACKNOWLEDGEMENTS

Authors thank Nagaon Education Society's Gangamai College of Pharmacy, Nagaon Dist. Dhule, for their kind support for doing this research work.

REFERENCES

1. Y. R. Chandra, the wealth of India, publication and information directorate, New Delhi, 1976, vol. Sp – w, 349.
2. Sylvain valere T. Sob, *et al*, Biochemistry systematic & ecology 2010, 38(3), 342-345.
3. Li Ying Tang e*t al*, Chinese chemical letters, 2008, 19 (9), 1083-1085.
4. C.K. Kokate, Practical Pharmacognosy, Vallabh prakashan, 1994, 107-111.
5. Brain K and Turner, T.D., 'The Practical evaluation of phytopharmacueticals, Wright Scienteania Bristol, 1983, 103-106.
6. WHO guidelines, Quality control method for medicinal plant material, published by world health organization, genera. A.I.T.B.S publisher & distributors Delhi-51, 41-43.