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Evaluation of antibacterial and antioxidant activity from fruit extract of *Cassia fistula L.*

HARISH GUPTA[1,2], KISHLAY KUMAR[2] and R.C. AGRAWAL[1]*

[1]Department of Research, Jawaharlal Nehru Cancer Hospital & Research Centre, Idgah Hills, Bhopal (India).
[2]RKDF College of Pharmacy, Bhopal (India).

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ABSTRACT

The in vitro antibacterial and antioxidant properties of 50% methanolic extract of fruit from *Cassia fistula L.* (Indian Laburnum) was investigated. In this study the antibacterial activity of *Cassia fistula* against different gram positive and gram negative bacteria by disc diffusion method and Antioxidant activity by Fenton reaction was undertaken. *Cassia fistula* has shown effective against Gram positive bacteria and Gram negative bacteria and the MIC value was measured. In another experiment antioxidant activity was observed and IC_{50} was shown at 1200 µg. concentration. These results are important because this plant is widely used for medicinal and ornamental purposes..

Key words: *Cassia fistula*: antibacterial: antioxidant.

INTRODUCTION

Cassia fistula Linn is known as Amaltas, in Hindi which is a medium size tree belongs to family Caesalpiniodeae and is widely cultivated throughout India, It is widely used for its medicinal properties. The main property which is reported are mild laxative[1], cure of skin disease[2], wound healing[3], Hypoglycemic[4], antibacterial[5], antifungal[6], hypocholestremic[7] hepatoprotective [8,9], antitumour [10], antioxidant[11.12], antifertility [13]. This plant is widely used in traditional medicine, in the present we are reporting antibacterial and antioxidant potential of 50% methanolic extract of the fruit from *Cassia fistula.*

MATERIAL AND METHODS

Plant Material

The fresh fruit of *Cassia fistula* linn was collected from shops of Bhopal, India and identified by N.K.Pandey of Central Research Institute, Gwalior, India. A voucher specimen (field book No. 10630) has kept in our laboratory for future reference. The fruit were dried under shed and dried powdered was kept in a well closed container for further experiment.

Preparation of extract

Powder of *Cassia fistula* was treated with petroleum ether for defatiation for 30 minutes then extracted with 50% methanol until the colourless solution was obtained using separating funnel. The upper layer was collected and then extract was kept at water bath for drying thus the powder obtained which was used for the further experiment.

Test organism

The following gram negative and gram positive bacteria i.e. *Pseudomonas aeroginosae, Staphylococcus aureus, Staphylococcus epidermidis, Shigella flexineri, Bacillus subtilis & Escherichia coli* were used for antibacterial activity which were received from stock culture of our laboratory.

Methodology

Antibacterial activity

Antibacterial activity of 50% methanolic, extract from fruit of *Cassia fistula* were investigated using disk diffusion method[14]. 20 ml of sterile Nutrient agar was added in the test tube after that Petri plate were prepared and cultures were swabbed on the top of the solidified media and allow to dry . The test were conducted at four different concentration of the crude extract 25%, 50%, 75% & 100% (100 µl per disk), the disk which was soaked with extract were placed on the surface of the medium the plate and were incubated for 12-18 h at 37^{0}c and the zone > 8mm were considered not active against microorganism as reported in the literature[15].

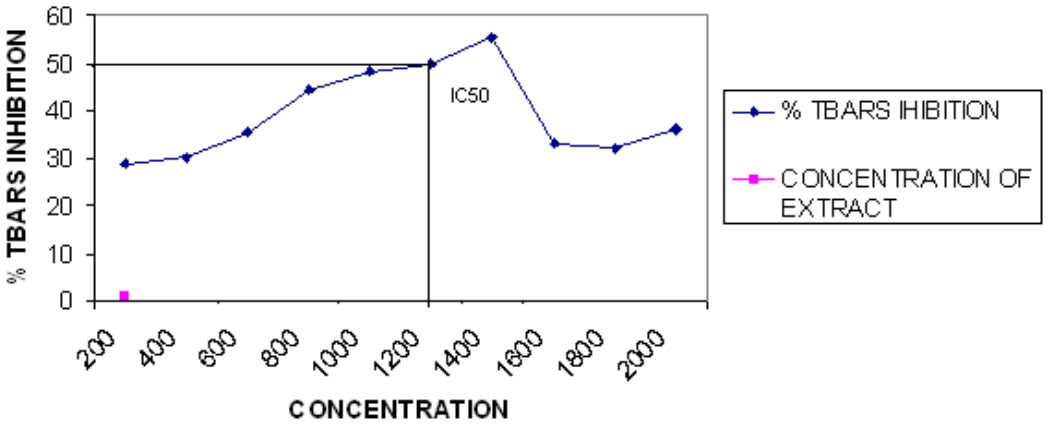
Antioxidant Protocol

The hydroxyl radical attacked deoxyribose and initiated a series of reaction that eventually resulted in the formation of Thiobarbituric acid reactive substance (TBARS). The measurement of TBARS thus given an index of free radical scavenging activity. The reaction mixture consisted of a deoxyribose (3mM, 100µl) Ferric chloride (Fe^{3+} 0.2 mM 50µl) EDTA (0.1mM, 50 µl) ascorbic acid (0.1 mM 100µl) and H_2O_2 (1mM, 100µl) stock solution of *Cassia fistula* extract 2mg/ml were prepared from which 200-2000 µl were added in Reaction mixture . The final volume was made up to 1 ml by adding adequate quantity of phosphate buffer saline (p^H 7.4) and incubated for 1h at 37^{0}c. The Reaction was stopped by adding 0.5ml of 5% TCA and 0.5ml of 1% TBA the mixture was than incubated for 20 min in a boiling water bath. The absorbance was measured at 532 nm. Ascorbic acid was used as the positive control. The results are expressed as the percentage inhibition of TBARS as reported.[16]

RESULTS AND DISCUSSION

Antibacterial activity

50% methanolic extract of *Cassia fistula* at the different concentration i.e. 25%, 50%, 75%, 100% exhibited antibacterial against *Bacillus subtilis* (9-13 mm), *Staphylococcus epidermidis* (10-12mm), *Staphylococcus aureus* (9-12mm) but the lower activity was observed against *Shigella flexineri* (9 mm at 100%) *E. coli* (9 mm at 100%) and *Pseudomonas aeroginosae* (9 mm at 100%) the Minimum inhibitory concentration (MIC) of *Cassia fistula* against gram positive bacteria i.e. *Bacillus subtilis, Staphylococcus epidermidis, Staphylococcus aureus* was 25% but against gram negative bacteria Zone of inhibition was observed only in 100% extract . Other concentration i.e. 25%,50 % and 75 % has not shown any zone of inhibition (table No.-1.) Result showed *Cassia fistula* was effective against gram positive bacteria but it was very less effective against gram negative bacteria. Cassia fistula was also reported significant antibacterial activity against the some bacteria.[5] Our findings confirm the traditional therapeutic claims of this herb.



Graph 1: % inhibition of TBARS by different concentration of *Cassia fistulaextract*

Table 1: **Antibacterial activity of *Cassia fistula* against different bacterial strains**

Name of Bacteria	Zone of Inhibition (mm) Concentration of drug			
	25%	**50%**	**75%**	**100%**
(+ve)				
Bacillus subtilis	10	9	11	13
Staphylococcus epidermidis	10	10	11	12
Staphylococcs aureus	9	10	12	11
(-ve)				
Shigella flexineri	-	-	-	9
Escherichia Coli	-	-	-	9
Pseudomonas aeroginosae	-	-	-	9

Table 2: In vitro antioxidant activity of 50% methanolic fruit extract from *Cassia fistula* extract

S. No.	Concentration of ascorbic acid (µg)	% TBARS inhibition ± SEM	Concentration of *cassia fistula* (µg)	% TBARS inhibition ± SEM
1	50	43.42±0.63	200	28.80±0.21
2	100	44.17±2.11	400	30.29±0.42
3	150	40.88±0.00	600	35.4±0.42
4	200	50±0.84	800	44.16±0.21
5	25	54.01±1.26	1000	48.17±1.48
6	300	58.03±1.26	1200	50±00*
7	350	75.70±1.83	1400	55.47±0.84
8	400	54.75±0.64	1600	32.97±2.00
9	450	62.78±2.11	1800	32.11±0.21
10	500	80.77±3.16	2000	36.13±2.11

* denoted IC50 value of cassia extract

Antioxidant activity

The extract of *Casia fistula* showed good antioxidant property in Fenton reaction model. The test drug was compared with a low concentration of ascorbic acid. Table-2 showed the % TBARS inhibition of methanolic extract of *Cassia fistula* and ascorbic acid. Crude 50% methanolic extract of *Cassia fistula* showed IC_{50} at 1200 ìg and the ascorbic acid showed IC_{50} at 200 ìg. Aqueous extract of *Cassia fistula (Linn.)* flowers (ACF) was reported for its antioxidant effect in alloxan induced diabetic rats [12] but there are no report about the antioxidant activity of fruit extract of *Cassia fistula* using Fenton reaction.

CONCLUSION

From the above result it may be concluded that 50% methanolic fruit extract of Cassia *fistula* has antibacterial activity against gram positive bacteria but less activity in gram negative bacteria. The antioxidant activity of this extract was also observed. These results are important because this plant is widely used for medicinal and ornamental purposes.

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REFERENCES

1. Akanmu, M A., Iwalewa, E.O., Elujoba, A.A., Adelusola, K.A. " Toxicity potential of *Cassia fistula* fruits as laxative with reference to *Senna*". *African J. of Biomedical Research* **7**(1): 23-26 (2004).
2. Chopra, R.N., Nayar, S.L., Chopra I.C. "Glossary of Indian Medicinal Plant" Directorate of CSIR, New-Delhi 54 (1992).
3. Bhakta, T., Mukharjee, P.K., Saha, K., Pal, K., Saha, B.P. Hypoglycemic activity of *Cassia fistula linn* leaf methanolic extract in alloxon induce diabetic rat. *J. of Ethanobotany* **9**: 35-38 (1997(b)).
4. Bhakta, T., Mukharjee, P.K., Pal, K., Saha, B.P. "Studies on In-Vivo wound healing activity of *Cassia fistula Linn* leaves in rat" *Natural product science* **4**(2): 84-87 (1997(a)).
5. Samy, PR., Ignacimutha, S., Seh, A. "Screening of 34 Indian medicinal plant for antibacterial properties". *J. of Ethanopharmacology* **62**: 176-182 (1998).
6. Souwalak, P., Nongyao, P., vatcharin, R., " Antifungal activity from leaf extract of *Cassia alata L., Cassia fistula L., Cassia tora L.*" Songkalahkarin *J. of Science and Technology* **26**(5): 741-748 (2004).
7. Saadany- El-, S.S., El-massry, R.A., Labib, S.M., Sitohy, M.Z., "Biochemical role and Hypocholestrolemic potential of the legume *Cassia fistula* in Hypocholestrolemic rat" *Nenrung* **35**: 807-815 (1991).
8. Pradeep, K., Chandraskaran, V.R.M., Gobianand, K., Karthikeyan, S., "Effect of *Cassia fistula linn.* Leaf extract on diethylnitrosamine induced hepatic injury in Rat" Chemico- *Biological intraction* **167**: 12-18 (2007).
9. Bhakta, T., Pulok, K., Mukharjee, P.K. Banerjee, S., Subhash, C.M., Maity, T.K., Pal, M., saha, B.P. Evaluation of hepatoprotective activity of *Cassia fistula* leaf extract *J. of Ethanopharmacology* **66**: 277-282 (1999).
10. Gupta, M., Mazumdar, U.K., Rath, N., Mukhopandhyay, D.K. "Antitumour activity of methanolic extract of *Cassia fistula L.*, Seeds against Erich ascites carcinoma *J. of Etharnopharmacology* **72**: 151-156 (2000).
11. Siddhuraja, P., Mohan, P.S., Becker, k., Studies on Antioxidant activity of Indian laburnum (*Cassia fistula L.*): a Preliminary assessment of and extract from stem bark, leaves, flowers and fruit pulp. *J. of food chemistry* **79**:61-67 (2002).
12. Manomani, G., Bhavpriya, V., Kalpana,S., Govindsamy., S., Apparaanatham, T. Antioxidant activity of *Cassia fistula (Linn)* flower in alloxon induced. Diabetic rats *J. of Ethanopharmacology* **97**: 39-42 (2005).
13. Yadav, R., Jain, G.C. Antifertility effect and hormonal profile of petroleum ether extract of seeds of *Cassia fistula* in female Rats *International J. of Pharma Tech Research* **1**(3): 438-444 (2009).
14. Bauer, A.W., Kirby WMM, Sherries, J.C., Turck, M., Antibiotic susceptibility testing by standardized single disc method *American J. of clinical pathology* **36**: 493-96 (1966).
15. Nehru, S.S., Zuraini, Z., Sasidharan, S., Suryani, S., Antimicrobial activities of *Cassia surattensis and Cassia fistula J. of molecular Biology and Biotechnology* **1**: 1-4 (2008).
16. Uma Devi, P., Vrinda, B., "radiation protection of human Limphocyte chromosomes in vitra by Orientin and Vicenin" *Mutation research* **498**: 39-46 (2001).