TO STUDY DIURETIC ACTIVITY OF *PASSIFLORA INCARNATA* LEAVES

Suvarna P.Ingale*, Harshad Kute [a], Nitin Patil [a] and Sanjay Kastute [#]

*****Assistant Professor, Dept. of Pharmacology, Indira College of Pharmacy, Tathawade, Pune-411033
[a] Student, Indira College of Pharmacy, Tathawade, Pune-411033
Pinnacle Biomedical Research Institute, Bhopal.
Email address for correspondence: suvarnaingale@gmail.com

Summary

Diuretics play an important role in situations of fluid overload like acute and chronic renal failure and cirrhosis of liver. Passionflowers have been used in folk medicine for ages and finding an increasingly important place in modern medicine. The literature review revealed the reports on anxiolytic, sedative-hypnotic, anti convulsant, anti inflammatory, antitussive, anti asthmatic and aphrodisiac activity of *Passiflora incarnata* Linn. However, there is no report on its diuretic potential. Hence, present study was designed to study diuretic activity of buanol and ethyl acetate extracts of *Passiflora incarnata* (BEPI and EAEPI). The method of Lipschitz *et al* was employed for the assessment of diuretic activity. In present study, BEPI and EAEPI in a dose of 150 and 300 mg/ kg, ip increased urine volume, urinary sodium, potassium and chloride excretion significantly compared to vehicle treated animals. Thus, the results of present investigation conclude that the BEPI and EAEPI posses' significant diuretic activity which is due to presence of flavonoid content of extract.

Introduction

Diuretics play an important role in conditions of fluid overload like acute and chronic renal failure and cirrhosis of liver. A number of diuretics like thiazides, furosemide, and ethacrynic acid are used in practice. Drug-induced diuresis is beneficial in many life-threatening disease conditions such as congestive heart failure, nephritic syndrome, cirrhosis, renal failure, hypertension, and pregnancy toxemia. (1)

Passionflowers have been used in folk medicine for ages and finding an increasingly important place in modern medicine. Though many species are used in rural areas, *Passiflora incarnata* is the most economically important medicinal species on the international market. In their native habitats, many Passionflower species have been used in folk medicine for a very long time to combat various diseases. Throughout Central America, an infusion of leaves from various Passiflora species is used as a diuretic. In many parts of

South America the leaves of *Passiflora edulis* and other species are used as diuretic. (2)
The literature review revealed the reports on anxiolytic (3), sedative-hypnotic (4), anti asthmatic (5), aphrodisiac (6) anti convulsant, antitussive activity (7) of *Passiflora incarnata* Linn. However, there is no report on its diuretic potential. Hence, present study was designed to study diuretic activity of *Passiflora incarnata*.

Materials and Methods

Animals:
Male Wistar rats weighing 150-200 gm were used for the study. Animals were kept for 1 week to acclimatize to laboratory conditions before starting the experiment; they were given free access to water and standard rat feed except during experimentation.

Plant material:
The fresh leaves of *Passiflora incarnata* were collected in the month of June, July and August from local nursery in Pune India. The plant was identified and authenticated (Voucher Specimen No: PASSIN 3) by Botanical Survey of India, Pune, India. Shade dried leaves (1000gm), powdered and macerated with ethanol for 48 hrs. The extract was evaporated to dryness. The leaf extract was suspended in water and extracted successively with hexane, chloroform, ethyl acetate and n-butanol to obtain hexane (HEPI), chloroform (CEPI), ethyl acetate (EAEPI) and n- butanol (BEPI) extracts of Passiflora *incarnata.* The yield was 11.7% w/w, 1.1% w/w, 0.9% w/w and 21.1% w/w for hexane, chloroform, ethyl acetate and n- butanol extracts, respectively. Qualitative chemical tests were conducted for above extracts of Passiflora *incarnata* to identify the various phytoconstituents (8). The results of preliminary phytochemical investigation are shown in [Table 1].

Diuretic Activity (9):
The method of Lipschitz *et al*. was employed for the assessment of diuretic activity. The animals were divided into six groups each six animals. They were fasted and deprived of food and water for 18 hours prior to experiment. The first group received only saline (25 ml/kg, ip). The second group served as the standard group, received the standard drug furosemide (20 mg/kg, ip.). Rest of the four groups received each of EAEPI (150 and 300 mg/kg, ip.) and BEPI (150 and 300 mg/kg, ip.) After drug administration, each animal were placed in an individual metabolic cages specially designed to separate feces and urine at room temperature. Urine excretion is recorded after 24 h. Na^+ and K^+ concentrations were measured by Flame photometer and Cl^- concentration was estimated by titration with silver nitrate solution

(N/50) using three drops of 5% potassium chromate as an indicator. The concentration of the electrolytes in urine is expressed in terms of Meq/L.

Statistical Analysis:
All values are expressed as mean ± SEM. The values obtained for the above parameters in case of the extracts were compared with control group by using Analysis of Variance (ANOVA) followed by Dunnett's test. $P<0.01$ is considered as significant.

Results

Table no. 2 depicts Urinary output and electrolyte excretion of control and treated groups. The results of present investigation showed that the standard diuretic furosemide and extracts (BEPI and EAEPI) in all doses (150 and 300 mg/kg, ip) caused significant ($p < 0.01$) increase in urine volume and electrolyte excretion as compared to vehicle control group. In BEPI treated groups (150 and 300 mg/kg, ip), the urine volume is increased to 1.02±0.03 and 1.12±0.03 respectively from 0.54±0.06 in vehicle treated group. Similarly, In EAEPI treated groups (150 and 300 mg/kg, ip), the urine volume is increased to 0.98±0.03 and 1.02±0.05 respectively.
The butanol extract of P. incarnata (BEPI) in both the dose (150 and 300 mg/kg, ip) increased the Na^+, K^+ and Cl^- excretion compared to vehicle treated group. But, BEPI in a dose of 300 mg/kg, ip increased the Na^+, K^+ and Cl^- excretion significantly ($p < 0.01$) compared to vehicle treated group. Similarly, ethyl acetate extract of P. incarnata (EAEPI) in both the dose (150 and 300 mg/kg, ip) increased the Na^+, K^+ and Cl^- excretion compared to vehicle treated group and EAEPI in a dose of 300 mg/kg, ip increased the Na^+, and Cl^- excretion significantly compared to vehicle treated group.
It was observed that the ratio of sodium to potassium ions increased after treatment with BEPI and EAEPI. Na^+/ K^+ ratios in BEPI and EAEPI treated groups (2.09 and 2.27) in a dose of 300 mg/ kg were very much similar to that of Frusemide treated group (2.28)

Table 1: Preliminary Phytochemical Investigation

Sr no	Extract	Phytoconstituent present
1	n- butanol extract of Passiflora incarnata leaves(BEPI)	Tannins, Glycosides, Alkaloids, Flavonoids
2	Ethyl acetate extract of Passiflora incarnata leaves(EAEPI)	Tannins, Lipids
3	Chloroform extract of Passiflora incarnata leaves (CEPI)	Lipids
4	Hexane extract of Passiflora incarnata leaves(HEPI)	Lipids

Table 2: Diuretic activity of BEPI and EAEPI by Lipschitz method

Treatment	Dose	Urine Volume (ml)	Urinary Electrolyte concentration (24 hr)			Na^+/ K^+ Ratio
			Na^+(mEq/l)	K^+ (mEq/l)	Cl^- (mEq/l)	
Control	25 ml/kg	0.54±0.06	63.08±2.33	35.98±3.49	58.43±5.72	1.75
Frusemide	20 mg/kg	1.14±0.05*	144.20±8.86*	63.14±2.26*	89.02±4.16*	2.28
BEPI	150 mg/kg	1.02±0.03*	94.54±3.67	50.72±3.04	86.03±2.95*	1.86
BEPI	300 mg/kg	1.12±0.03*	110.54±8.19*	52.72±3.40*	95.03±3.68*	2.09
EAEPI	150 mg/kg	0.98±0.03*	96.52±7.83	51.93±3.22*	82.26±2.06*	1.85
EAEPI	300 mg/kg	1.02±0.05*	104.52±11.96*	45.93±4.33	88.66±2.28*	2.27

Values are mean ± SEM, n=6 in each group
*P<0.01 compared to control (ANOVA followed by Dunnett test)

Discussion

The objective of the present study is to evaluate the diuretic activity of butanol and ethyl acetate extracts of *P. incarnata* leaves. Diuretics play an important role in various cardiovascular disorders like hypertension, congestive heart failute, etc. The control of sodium ion concentration is important for regulation of blood volume and pressure and the potassium ion concentration maintains proper function of cardiac and skeletal muscles. The Na^+/ K^+ balance is also intimately related to acid- base balance of body (9).

The results of the present study revealed that the BEPI and EAEPI possess a potent diuretic activity which is expressed as increase in urine volume and urinary electrolyte excretion. The diuretic potency was comparable to that of the standard drug frusemide. BEPI and EAEPI increases the Na^+ and K^+ excretion, which may be acting like a loop diuretic, frusemide.

The active phytoconstituents like saponins, triterpinoids and flavonoids are known to possess diuretic potential (9). The preliminary phytochemical analysis of BEPI and EAEPI revealed that Tannins, Glycosides, Alkaloids, Flavonoids are present in butanol extract of P. incarnata leaves. The ethyl acetate extract contains Tannins and Lipids. Thus, these phytoconstituents might be acting synergistically or individually to produce diuretic effect. In conclusion, the present studies support traditional use of *P. incarnata* leaves for its diuretic effect.

Acknowledgement

The author is thankful to the Principal and Management of SCES's Indira College of Pharmacy, Pune-411033 for providing necessary facilities for carrying out this research work.

References

1. Koti BC, Ashok P. Diuretic activity of extracts of *Mimusops elengi* Linn. Bark. Int J Green Pharm 2010; 4:90-2.
2. Torsten Ulmer and John M. MacDougal, Passiflora: Passisonflowers of the World, Timber Press Inc., Portland, USA, 2004, 66-67.
3. Dhawan K, Kumar S, Sharma A. Anxiolytic activity of aerial and underground parts of *Passiflora incarnata*. Fitoterapia 2001; 72: 921–925.
4. Capasso A, Sorrentinob L.Pharmacological studies on the sedative and hypnotic effect of Kava kava and Passiflora extracts combination Phytomedicine 2005; 12:39–45.
5. Dhawan, K, Kumar S, Sharma A. Anti-asthmatic activity evaluation of methanol extract of leaves of *P. incarnata*. Phytotherapy Research 2003; 17: 821–822.
6. Dhawan K, Kumar S, Sharma A. Aphrodisiac activity of methanol extract of leaves of *P. incarnata* in mice. Phytotherapy Research 2002; 17:401–403.
7. Dhawan K, Sharma A. Antitussive activity of the methanol extract of leaves of *P. incarnata*. Fitoterapia 2002;73:399–401.
8. Kokate CK, Purohirt AR, Gokhale CB. Pharmacognosy. 27 th ed. Nirali Prakashan; 2004: 344.
9. Jain DL, Baheti AM, Parakh SR, Ingale SP, Ingale PL. Study of antacid and diuretic activity of ash and extracts of *Musa sapientum* L. fruit peel. Pharmacog Magazine 2007;3:116-9.