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Phytochemical analysis, antioxidant and xanthine oxidase inhibitory activity of *Tephrosia purpurea* Linn. root extract

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An attempt has been made to search for xanthine oxidase (XO) inhibitors from the root extracts of *Tephrosia purpurea* Linn. which is traditionally used in folk medicine in India. Root extracts were screened for *in vitro* antioxidant and xanthine oxidase inhibitory activity. Antioxidant activity was measured using ABTS, DPPH, FRAP and ORAC methods. The enzyme inhibitory activity was tested on purified milk xanthine oxidase. The root extracts and phytochemicals, obtained in distilled water, inhibited bovine milk XO in a concentration-dependent manner, with an additional superoxide scavenging capacity, the average antioxidant activity of *T. purpurea* root extract in the concentration range of 100-200 µg/mL. The reacting system revealed significant antioxidant activity, viz. 42.2 (ABTS), 28.7 (DPPH), 36.5 (FRAP) and 25.6 per cent by ORAC assay. Screening of xanthine oxidase inhibitory activity by extract in terms of kinetic parameters revealed noncompetitive mode of inhibition, where the K_m and V_{max} values in presence of (25 to 100 µg/mL) *T. purpurea* root extract is 0.18 µg and 0.040,0.037,0.034 and 0.030 (µg/min) while for control K_m and V_{max} is 0.21 µg and 0.043 (µg/min), respectively. The phytochemical analysis revealed presence of significant amount of polyphenols and flavonoids (90% and 80%, respectively). These findings suggest that *T. purpurea* root extract possess prominent medicinal properties and can be exploited as natural drug to treat the diseases associated with free radical formation, oxidative stress and xanthine oxidase activity.

Keywords: *Tephrosia purpurea,* Antioxidant, Xanthine oxidase, Inhibition, Purple Tephrosia, Phytochemical analysis.

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Introduction

Plant extracts or secondary metabolites have served as antioxidants in phytotherapeutic medicines to protect against various diseases for centuries[1]. *Tephrosia purpurea* Linn. (Fabaceae), is a pantropical, polymorphic, branched, sub-erect, perennial herb popularly known as *Sarapunkha* in Sanskrit, Purple Tephrosia in English and *Unali* in Marathi[2]. The plant is used in folk medicine as an antidiabetic, antipyretic, anticancer and antiulcer agent in addition to its usefulness in treatment of diseases related to oxidative stress and free radicals activity[3]. This phenomenon develops due to the interaction of the free radicals like Reactive Oxygen Species (ROS) and cellular components resulting in cellular damage and tissue injury[4]. Free radicals include reactive species such as hydroxyl radicals, peroxyl radicals, super oxide radicals, hydrogen peroxide, singlet oxygen and various lipid peroxides[5,6]. ROS are also capable of reacting with membrane lipids, proteins, nucleic acids, various metabolic enzymes and small molecules of living systems. They play an important role in the initiation and progression of various diseases such as atherosclerosis, cardiovascular diseases, aging, respiratory diseases, cancer and gout[7,8]. Gout develops due to the deposition of uric acid in the form of urate monohydrate crystals in the synovial joints during purine catabolism by xanthine oxidase[9], which catalyses the conversion of hypoxanthine to xanthine and xanthine to uric acid with concomitant production of hydrogen peroxides and superoxide anions as byproducts[10,11]. Antioxidants are molecules having capacity of preventing or slowing the oxidation of molecules. Oxidation reaction transfers electrons from a substance to an oxidizing agent and produces free radicals thereby causing damage to cells[12]. However, most of the cells contain a complex network of antioxidant metabolites and enzymes that work together to prevent oxidative damage to cellular components such as DNA, proteins and lipids[13].

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Antioxidants either remove or prevent these reactive species before they can damage vital cellular components of the cell or tissues[14]. This study is aimed to evaluate the effect of *T. purpurea* root extract as an antioxidant, anti-inflammatory and potent inhibitor of xanthine oxidase which is mainly involved in formation of uric acid leading to free radical induced damage and gout. Recently, the most important reported biological properties of secondary metabolites are due to their antioxidant abilities, electron transfer capacity, free radical scavenging, chelating abilities, and oxidase inhibitors[15].

Materials and Methods

Plant material and preparation of extract

Roots of *Tephrosia purpurea* were collected during the period of August to September from local forest of Nanded, India. It was taxonomically identified and deposited in department (Voucher No SRT/BT/P/SN/ 101). The roots of plant were shade–dried for a week, grounded by using mortar and pestle. The powder was extracted with methanol (90%) by using Soxhlet apparatus for 4 h. The resultant extract was then filtered, concentrated under reduced pressure at 60 ± 10°C in a vacuum rotator evaporator to dryness and used for activity measurements as per the need[16].

Chemicals

All chemicals, viz. DPPH (1, 1-Diphenyl-2-picrylhydrazyl), ABTS (2, 2-azinobis-3-ethyl benzothiazoline-6-sulfonic acid diammonium salt), AAPH (2, 2'-azobis (2-amino-propane) dihydro-chloride), TPTZ (2, 4, 6-tripyridyl-s-triazine), fluorescein dye, sodium acetate trihydrate, quercetin, EDTA (Ethylene diamine tetra acetic acid), folin-ciocalteu reagent, ferric chloride, acetylsalicylic acid, methanol, chloroform, ethyl acetate, allopurinol, xanthine were purchased from Hi-Media Laboratories Ltd. Mumbai, India and Sigma-Aldrich Chemicals, Co, U.S.A. All chemicals were of AR grade and used without further purification unless stated otherwise. The hen's eggs and cow milk were purchased from local market.

Extraction of phenolic compounds

The extraction of plant root was carried out using various polar and non-polar solvents[17]. According to the method, dried plant material was ground in a Waring blender. It was mixed with a 10-20 volume of 85% methanol. The slurry was placed on a shaker for 24 h. The extract was filtered through a Buchner funnel and the methanol was removed on a rotary evaporator to give crude extract. The aqueous solution was extracted with hexane several times to eliminate lipids. The water fraction was partitioned against chloroform. The remaining aqueous phase was extracted exhaustively with ethyl acetate until the final extract was colorless; the remaining is aqueous extract of roots of plant. All the solvents were removed by evaporation under reduced pressure and the extracts were stored at −20°C until use.

Determination of total polyphenol contents

Total polyphenols were measured using Prussian blue assay method described by Price and Butler[18] modified by Graham[19]. Phenolics were expressed as gallic acid equivalents. Briefly 0.1 mL of *T. purpurea*, root extract samples were dissolved in methanol, 3 ml distilled water was added and mixed, then 1 mL of $K_3Fe(CN)_6$ (0.016 M) was added to each sample followed by the addition of 1 ml of $FeCl_3$ (0.02 M dissolved in 0.1 M HCl). It was immediately mixed using a vortex, and 5 ml stabilizer (30 ml gum Arabic, 1%; 30 ml H^3PO_4, 85% and 90 ml of distilled water) was added to the sample and mixed. The absorbance was measured at 700 nm using a UV/VIS-8500 Techom spectrophotometer. The amount of total polyphenols in different extracts was determined from a standard curve of gallic acid ranging from 0.00 to 200 µg/mL.

Determination of flavonoid contents

Flavonoids were quantified using aluminium chloride reagent[20] and measured as quercetin equivalents. One ml of *T. purpurea*, root extract samples were dissolved in methanol, 1 ml of $AlCl_3$ (2%) in methanol was added, and after incubation for 10 min, the absorbance was measured at 430 nm.

Antioxidant activity

DPPH radical scavenging assay

Methanol extract of *T. purpurea* root was used for the preparation of stock solution one mg/l by using ethanol. The stock solution was further diluted and different concentrations (100, 150 and 200 µg/mL) were added to 1.5 ml of 0.1 mM ethanol solution of DPPH (1, 1-Diphenyl-2-picrylhydrazyl). The mixture was shaken vigorously and the absorbance was monitored at 515 nm against a blank after 45 min of incubation (UV–Vis Shimadzu), when the reaction reached a steady state. Quercetin was used as reference compound. The inhibition percentage (%) of

radical scavenging activity was calculated by using the formula, % inhibition = [A_c (o) – A_A (t) / A_c (o)] × 100, where A_c (o) is the absorbance of control at t = 0 min and A_A (t) is the absorbance of antioxidant at t = 1 h. All measurement was done in triplicate[21].

ABTS radical scavenging assay

The 2,2-azinobis-3-ethylbenzothiazoline-6-sulfonic acid diammonium salt (ABTS) 19 mg (7 mM) was oxidized with potassium persulfate 3.3 mg (2.45 mM) overnight in the dark bottle for about 16 h or overnight in dark. The working solution was then diluted with ethanol to an absorbance of 0.75±0.05 at 734 nm. A standard calibration curve was constructed for Quercetin at 0, 0.1, 0.2, 0.3, 0.4, 0.5, 1.0, and 2.0 mM. An aliquot (10 µL) of methanolic extract of *T. purpurea* root (100, 150 and 200 µg/ml conc.) was mixed with 1.0 ml of ABTS radical cation working solution in cuvette and absorbance was read at 734 nm after 30 min. The activity was expressed as the effective concentration of drug necessary to give a 50 % reduction in the original absorbance [22].

ORAC (Oxygen radical absorption capacity) assay

ROO^- was generated by thermo decomposition of 2, 2'-azobis (2-amino-propane) dihydrochloride (AAPH). For this reaction, an automated plate reader with 96-well plates was used. Reaction mixtures in the sample wells contained, in a final volume of 200 µl, the following reagents were dissolved in 75 mM potassium phosphate buffer, pH 7.4, at the indicated final concentrations, Fluorescein was used as substrate (61 nM), methanolic extract of *T. purpurea* root at various concentrations (100, 150 and 200 µg/mL) and AAPH (19 mM). The scavenging effects are expressed as the relative Quercetin equivalent ORAC value, which is calculated according to the following formula, Relative ORAC value = $[(AUC_{sample} - AUC_{blank})/(AUC_{quercetin} - AUC_{blank})] \times$ (mass of quercetin/mass of sample), where AUC is the area under curve[23].

FRAP (Ferric reducing antioxidant power) assay

The stock solutions were prepared by using 300 mM acetate buffer (3.1 g sodium acetate trihydrate and 16 ml acetic acid, pH 3.6). The reagent TPTZ (2, 4, 6-tripyridyl-s-triazine) were prepared by using TPTZ in 40 mM HCl and 20 mM $FeCl_3.6H_2O$ solution. The fresh working solution was prepared by mixing 25 ml acetate buffer, 2.5 ml TPTZ solution and 2.5 ml $FeCl_3.6H_2O$ so as to form the FRAP solution. The working solution (FRAP) was warmed at 37°C before use. Methanolic extract of *T. purpurea* root with different concentrations (100, 150 and 200 µg/mL) were allowed to react with 28.50 µL of the FRAP solution for 30 min in dark condition. Absorbance of the colored product (ferrous tripyridyltriazine complex) was recorded at 595 nm. Quercetin was used as standard and the standard curve was linear between 25 and 800 µM of Torolox. The results were expressed as µM TE/g fresh mass. Additional dilution was needed if the FRAP value measured was over the linear range of the standard curve [24].

Xanthine oxidase isolation and purification

Xanthine oxidase was isolated by slight modification of previously described methods. All XO purification steps were carried out at 4°C. Ethylene diamine tetra acetic acid (EDTA), sodium salicylate and 2-mercaptoethanol (all at 0.1 mmol/lit final concentrations) were added to milk and to all buffers. Cow milk (1 lt) stored at –20°C in deep freezer (Elico, Mumbai), was thawed and iso-butanol (170 ml/lt of milk), previously cooled at –20°C, and solid $(NH_4)_2SO_4$ (to 35% saturation) were added and stirred it for 3 h, the mixture was then centrifuged at 18,000 rpm for 30 min. The aqueous bottom phase was collected and $(NH_4)_2SO_4$ was added to 60 % saturation. The floating upper precipitate was re-suspended in the same buffer and dialyzed overnight in a dialysis bag against, 0.2 mol/lt sodium phosphate buffer (pH 6.0). The precipitate remaining after dialysis was removed by centrifugation at 30,000 rpm for 30 min and supernatant again precipitated by addition of ammonium sulphate (70%) with continuous stirring. The precipitate was collected by recentrifugation at 9000 rpm for 15 min. The precipitate was dissolved in 6-7 volume of 0.05 M potassium phosphate buffer (pH 7.9) containing 0.1mM (EDTA). The purification was further carried out by 38% v/v and then 47% v/v cold acetone. The precipitate was separated by centrifugation and dissolved in 0.1mM EDTA containing deionised water. The final step of purification was accomplished by dialysis against deionised water for 38 h at 4°C. The extract dialyze itself was used immediately as crude enzyme for the measurement of enzyme activity and protein estimation[25,26].

Protein measurement

The protein content of the enzyme was determined from the standard curve of BSA by Folin Lowry

method[27]. One unit of enzyme activity was defined as the amount of enzyme responsible for production of 1 µmol of uric acid in 1ml of assay volume per min at 25°C at pH 7.5 the enzyme activity was measured by a standard method[28]. In this method the reaction mixture contained 2.0 ml of potassium phosphate buffer (50 mM, pH 7.5), 1 ml of xanthine (0.15 mM) as substrate, 0.1 ml of native XO (0.25U/ml) and 0.5 ml of NAD^+ (0.5 mM) as reducing agent. All the contents were mixed by swirling and incubated at 37°C for 10 min. The increase in absorbance at 295 nm after the enzyme addition confirms the production of uric acid from substrate xanthine. The formed uric acid concentration was estimated by standard curve of uric acid spectrophotometirically. The blank was prepared without enzyme solution.

Xanthine oxidase inhibition assay

Assays were performed by using the better physiological condition for optimization of enzyme activity at variable pH ranging from 2 to 9 and temperature from 10°C to 80°C. The pH effect was studied by using phosphate buffer of varying *p*H. Effect of variable substrate (xanthine) concentration (1 mg/ml to 5 mg/ml) was studied for determining the stability of enzyme substrate complex in terms of kinetic parameters K_m and V_{max} values. The optimum pH, temperature and substrate concentration was examined by the same procedure used for enzyme activity study[29].The inhibitory activity of *T. purpurea* root extract was determined using the standard inhibitor allopurinol (5 mM), which was then diluted with phosphate buffer (0.1 mM) to obtain 5, 10, 15, 20 and 25 µM solutions. *T. purpurea* root extract was dissolved in 1 mM phosphate buffer to make concentrations 25, 50, 75 and 100 µg/ml, which was then diluted with 0.1 mM of phosphate buffer to obtain different concentrations for kinetic study. The inhibitory activity of *T. purpurea* root extract was determined using a slight modification of reference methods. Briefly: 7.0 µl of xanthine oxidase solution (0.4U/ ml) were added to 130 µl phosphate buffer (0.1 M, pH 7.8). The reaction mixture was then incubated at 37°C for 10 min. The 70.0 µl of 40 µM xanthine solution was added to the mixture and the absorbance was recorded at 295 nm spectrophotometrically. The blank solution was prepared in an analogous way, but instead of the enzyme, it contained 7 µl of phosphate buffer solution. The test was performed in triplicate. The sample tests were performed in following way in which 7.0 µl of xanthine oxidase solution (0.4U/ml) was added to 80.0 µl of phosphate buffer (pH 7.8, 0.1M) and different concentrations of *T. purpurea* root extract (25, 50, 75 and 100 µl) were treated in the same way as the control. Four µl of phosphate buffer solution was used instead of xanthine oxidase solution (0.4 U) as blank. The test was performed in triplicate. Enzyme inhibitory activity was determined by quantifying the amount of uric acid formation from xanthine in the reaction mixture. The assay mixture contains both *T. purpurea* root extract and xanthine. Both the inhibitor and substrate concentrations were maintained identical for the reaction. Xanthine oxidase activity was expressed as % inhibition of xanthine oxidase, calculated as (1-B/A) × 100, where A is the change in absorbance of the assay without the *T. purpurea* root samples. Δ Abs with enzyme, Δ abs without enzyme and B is the change in absorbance of the assay with the *T. purpurea* root sample (Δ abs with enzyme - Δ abs without enzyme). The enzyme kinetics was similar to xanthine oxidase assay methods[30,31].

Statistical analysis

All determinations were conducted in triplicate and all the results were calculated as mean ± standard deviation (SD). Statistical analysis was performed using Student's *t*-test for significance and analysis of variance (ANOVA) followed by Dunnett's test were done for the multiple comparison of the effect of different extracts. Values of $P < 0.05$ were considered statistically significant.

Results

The values of total polyphenols and flavonoids in *T. purpurea* are shown in Table 1. There was a wide range of polyphenolphenol concentrations in *T. purpurea*. The value varied from 10.12 ± 0.11 to 56.12 ± 2.32mg equivalent gallic acid/g lyophilizate and from 02.21 ± 0.42 to 28.17 ± 3.2 mg equivalent quercetin/g lyophilizate for flavonoids, respectively. The highest level of polyphenols and flavonoids were recorded in the methanol extract followed by chloroform extract and ethyl acetate extract, while the total polyphenol and flavonoid levels were particularly low in the aqueous extract. It is well known that polyphenols are widely distributed in plants; they are sometimes present in surprisingly high concentrations especially in medicinal[32]. The methanol extract of *T. purpurea* revealed significant free radical scavenging activities (Table 2). The overall range of observed antioxidant scavenging activity of *T. purpurea* extract at 2 µg/ml were, ABTS

Table 1—Total polyphenol and flavonoid contents of *Tephrosia purpurea* root extracts

Extracts	% yield (w/w)	Total polyphenols (mg equivalent gallic acid/ g lyophilizate)	Total flavonoids (mg equivalent quercetin/ g lyophilizate)
Methanolic	10.25 ± 0.80	56.12 ± 2.32	28.17 ± 3.2
Chloroform	02.12± 0.65	98.64 ± 0.45	18.02 ± 0.67
Ethyl acetate	04.20 ± 0.24	48.97 ± 1.23	12.45 ± 0.65
Aqueous	8.81 ± 0.08	10.12 ± 0.11	02.21 ± 0.42

Values are represented as Mean ± S.D. *n*=3.

Table 2—Antioxidant activity of *Tephrosia purpurea* root extracts

Concentration of root extracts (µg/ml)	% Antioxidant activity (µm TE or QE/g FM) ABTS	DPPH	FRAP	ORAC
100	38.90± 5.3	26.82± 1.6	34.90± 2.6	24.80± 1.8
150	42.84± 4.2	28.17± 1.4	35.80± 5.1	25.40± 1.5
200	44.86± 7.2	31.21± 2.8	38.80± 4.9	27.60± 7.2
Quercetin (as standard)	40.80± 3.8	25.60± 6.5	32.25± 2.4	26.20± 6.1

Values are represented as Mean ± S.D. *n*=3.

(44.86± 7.2), DPPH (31.21± 2.8), FRAP (38.80± 4.9) and ORAC (27.60± 7.2) percent, respectively to that of standard (quercetin) were, DPPH (25.60± 6.5), ABTS (40.80± 3.8), FRAP (32.25± 2.4) and ORAC (26.20± 6.1). The freshly purified milk XO from different species showed an ultraviolet/visible spectrum with three major peaks at 280, 325, 450 nm, with A280/A450 (protein to flavin, PFR) ratio of 5.3, 5.2 and 5.1 for bovine, sheep and human, respectively, indicating a high degree of purity. Run on SDS-PAGE, purified enzymes showed quite similar patterns with one major band of approximately *M*r 150 KDa. Traces of degradation bands appeared on storage. This is analogous to the well-studied bovine, human, and camel enzymes[33-35]. The XO activity of purified sheep milk (121.9 nmol of urate/min/mg protein) was low relative to that of the bovine milk enzyme (1227 nmol of urate/min/mg protein), but higher than that of human XO (73.8 nmol of urate/min/mg protein). This large difference in activity between milk XO is due to the intrinsic nature of the enzyme from different species, especially the existence in a molybdenum-deficient form. Such an inactive form represents more than 97% of HXO, 81% of SXO and 44% of BXO[36]. The methanol roots extract of *T. purpurea* demonstrated xanthine oxidase inhibitory activity at 25, 50, 75 and 100 µg extracts/ml (Table 3). The inhibition is a dose dependence effect with minimal XO inhibitory activity 40% at 25 µg extracts/ml and maximum 99%

Table 3—Xanthine oxidase (XO) inhibitory activity of methanolic extract of *Tephrosia purpurea*

Samples	Xanthine oxidase inhibition (%)			
T. purpurea extracts conc. (µg/mL)	25 µg/mL	50 µg/mL	75 µg/mL	100 µg/mL
	40.00± 2.6	62.42± 1.8	86.22± 2.2	99.00± 1.2
Allopurinol conc. (µg)	25 µg	50 µg	75 µg	100 µg
	25.20± 1.8	40.10± 3.5	70.5± 4.1	95.50± 3.2

Values are represented as Mean ± S.D. *n*=3.

Table 4—Vmax and Km of *Tephrosia purpurea* root extracts for xanthine oxidase inhibition

Samples	Concentrations (µg/mL)	V_{max} (µg/min)	K_m (µg/mL)
T. purpurea root extracts	25	0.046	0.18
	50	0.036	
	75	0.034	
	100	0.030	
Allopurinol	100	0.043	0.21

V_{max} is a maximum velocity; K_m is a concentration at 50 % V_{max}

inhibition at 100 µg/ml. The inhibition percentage by 100 µg/ml of extract is maximum to that of standard drug Allopurinol. The inhibition of XO results in a decreased production of uric acid as measured spectrophotometrically at 295 nm. The kinetic parameters expressed in terms of V_{max} and K_m. The value of V_{max} decreases as the concentration of root extract increased while the K_m value is constant for all samples it indicate that the inhibition is noncompetitive type of inhibition (Table 4).The inhibition revealed by the root extract of *T. purpurea* is greater than that of allopurinol a potent inhibitor of xanthine oxidsase.

Discussion

In the present study, the evaluation of *T. purpurea* root extracts for antioxidant and xanthine oxidase inhibitory activity were evaluated. The results presented for methanol extract of *T. purpurea* indicate that the ABTS, DPPH, FRAP and ORAC assays exhibited comparable antioxidant activity. *T. purpurea* revealed high scavenging activity by ABTS method showing maximum percent inhibition to that of quercetin. The ABTS method showed high reproducibility and highest correlation with both, plant extract and quercetin as reference. The DPPH radical was widely used as the model system to study the scavenging activities of several natural compounds such as flavonoids, tannins, phenolic and anthocyanines. In this assay, the electrons become paired off and solution loses colour stochiometrically

depending on the number of electrons taken up. In case of ABTS assay the inhibition of the absorbance of the radical cation $ABTS^+$, which has a characteristic long wavelength absorption spectrum[37]. The results of antioxidant scavenging activity by ABTS revealed that the *T. purpurea* root extracts inhibited or scavenged the $ABTS^+$ radicals significantly as compared to DPPH, FRAP and ORAC. The FRAP assay mainly depends on the reducing capacity of Fe^{3+}- Fe^{2+} conversion and serve it as an significant indicator of its potential antioxidant activity. The antioxidant activities have been attributed to various reactions, binding of transition metal ion catalysts, decomposition of peroxides, prevention of continuous proton abstraction and radical scavenging activity[38]. The possible reason for these results may be due to the presence of active constituents of *T. purpurea* which may be polar or non polar compound like coumarins, flavonoids, flavanones, isoflavones, rotenoides, etc. The kinetic analysis using Lineweaver–Burk plot revealed that the root extracts of *T. purpurea* displayed high inhibitory activity. The pattern of inhibition is a type of non competitive type of inhibition in presence of *T. purpurea* were in V_{max} is decreased and K_m appears to be unaltered with respect to Xanthine as substrate (Figure 1). It indicates that the binding of extract may occur with the free enzyme or the enzyme–substrate complex. The significant inhibition of XO by root extracts of *T. purpurea* may suppress the production of active oxygen species or uric acid *in vivo* under the conditions that xanthine oxidase works. The inhibition percentage by root extracts of *T. purpurea* is comparable to that of allopurinol (20 µg/ml), a therapeutic drug used to treat gout. *T. purpurea* root extracts exhibited the significant antioxidant activity in the tested *in vitro* assays. This study indicates that the herb may contain phytochemicals which may have great potential to act as free radical scavenger to reduce the oxidative stress and may prevent the diseases caused due to over production of reactive oxygen species or free radicals in living beings. The bio-guided fractionation of the active phytochemicals from *T. purpurea* root extract is being carried out, by aiming the formulation of a safer and efficient drug to prevent the oxidative stress and related diseases. The root extract of herb revealed potent Xanthine oxidase inhibition activity.

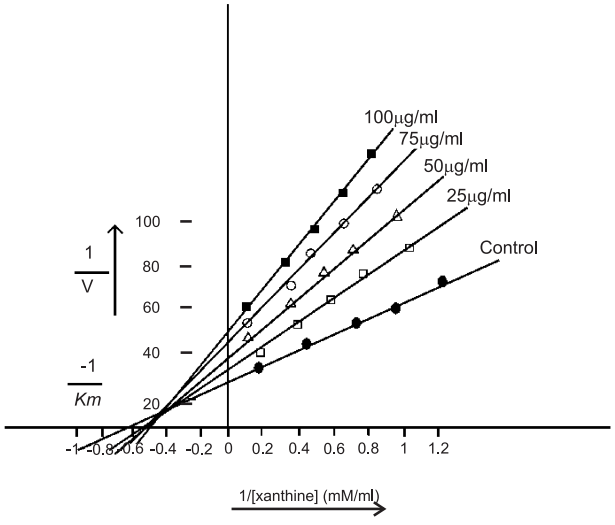

Fig.1—Lineweaver-Burk plots: Inhibition of Xanthine oxidase by *T. purpurea* root extract (□-Δ-o-■) and Allopurinol (•) with Xanthine as the substrate

Conclusion

It is concluded that the phytochemicals present in this plant extract may also be useful for the treatment of gout and hyperuricemia, which are correlated with the ethnobotanical data on the use of these plants in Indian folklore and Ayurveda. The data investigated in this study provides the basis for further investigation on this plant to isolate the active constituents and drug developments against the disease related to oxidative stress, inflammation and gout. Xanthine oxidase converts hypoxanthine to xanthine and finally to uric acid, which can accumulate and lead to hyperurecemia associated with gout. Thus specific inhibitors of xanthine oxidase are expected to be therapeutically useful in treatments of hyperurecemia associated with gout and kidney stones.

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