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Biochemical alteration of methanolic extract of *Amaranthus spinosus* on liver of Sprague Dawley rats

*Gul T, Singh K, Srivastava A, Ahirwar V

Department of Zoology, Institute of Basic Sciences, Bundelkhand University, Jhansi (U.P.) 284128, India.

*Corresponding Author: tariqgul443@gmail.com

Abstract

Amaranthus spinosus (spiny amaranth) is a tropical plant belonging to the family Amaranthaceae and is used widely as a vegetable and medicinal plant. The present study is carried out to investigate the biochemical role of methanolic extract of *Amaranthus spinosus* on liver of Sprague Dawley rats. Female albino rats were divided into two groups: one group served as control which was fed on normal diet, the second group served as experimental which receives a dose level of 250 mg/kg of methanolic extract of *Amaranthus spinosus* plant. Protein concentration was estimated in liver homogenate using the Lowry method (1951) with bovine serum albumin (BSA) as standard. Also, glycogen concentration was estimated in tested organ by using Seifter method (1950). Results showed significant increase for both acute as well as chronic studies in protein and glycogen contents at 250 mg/kg dose level.

Keywords: *Amaranthus spinosus*; Sprague Dawley rat; liver; protein; glycogen.

Introduction

Amaranthus spinosus Linn. belonging to family Amaranthaceae is commonly known as 'pig weed' or 'spiny amaranth' in English and 'Chaulai' in Hindi. It is an annual or perennial monoecious herb, native to tropical America and found throughout India as a weed in cultivated as well as fallow lands (Anonymous, 1988). Whole plant is used as a traditional medicine to treat diabetes (Vaidyaratnam, 1996). The plant is astringent, diaphoretic, diuretic, emollient, febrifuge and galactagogue (Bown, 1995; Manaadhar, 2002; Usher, 1974). It is also used in the treatment of snake bites (Vaidyaratnam, 1996). Externally, it is used to treat ulcerated mouths, vaginal discharge, nose bleeds and wounds (Bown, 1995). Whole plant is used as laxative (Vaidyaratnam, 1996; William, 1976). The *Amaranthus spinosus* is reported for its anti-inflammatory properties and immunomodulatory activity (Murgan et al., 1993; Olumayokun et al., 2004). The anti-oxidant capacity of *Amaranthus spinosus* was studied in roadside plants which were postulated to be continuously exposed to the high levels of nitrogen oxides and sulphur oxides from automobile emissions (Singh and Dahiya, 2002). Biochemical effect of the *Amaranthus spinosus* plant was studied in the liver of Sprague Dawley rats with the quantitative analysis of protein and glycogen contents.

Materials and Methods

Collection of plant and preparation of extract

The whole plant except root was collected from Bundelkhand University Campus, Parichha Colony and adjacent areas of Jhansi. The plant is air dried and grounded to powdered form. This was subsequently referred to as the plant material. The powdered plant material is extracted with methanol in a Soxhlet extractor and evaporated to dryness yielding a semisolid mass.

Animals

The study was conducted in sexually mature, female Sprague Dawley rats having body weight of (150 ± 10 mg), purchased from CDRI (Central Drug Research Institute), Lucknow. Prior to study, the clearance was obtained from the Animal Ethical Committee (ICMR, Government of India). The animals were acclimatized to the experimental room at temperature of 25-30°C, controlled humidity conditions (50-55%) and 12 hours light and 12 hours dark cycle. It should be necessary that, to keep at least two rats in small sized cage and maximum four rats in large sized cage. They were fed with standard pelleted diet (Amrut Brand Sangli) and water *ad libitum*.

For experimentation, animals were randomly distributed into two groups. One group serves as experimental and other as control. Both groups were having equal number of rats. Experimental group will receive dose at a concentration of 250 mg/kg and control will receive vehicle only. Autopsy of both groups will also be performed on the same day after 5, 7 and 14 days, which is done by chloroform if rat

is to be sacrificed and by diethyl ether if to keep alive.

Biochemical estimation

Protein content was estimated by the method of Lowry et al. (1951) and glycogen content was estimated by the method of Siefert et al. (1950).

Statistical analysis

The results were expressed as Mean ± S.E. significance of differences compared to the control was determined using student's t-test.

Results

Effect on protein content

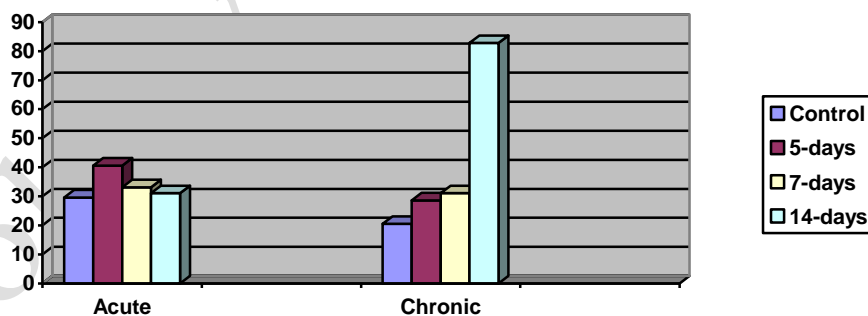
In the present study, effect of methanolic extract of *Amaranthus spinosus* at a dose level of 250 mg/kg was studied on different biochemical parameters like protein and glycogen after 5, 7 and 14 days of duration in liver, with respect to control group. During single administration of the dose, the protein content significantly increases at shorter duration but at longer duration it recouped to the normal level as compared to the control group. During daily administration, the protein content significantly increases both for shorter as well as for longer durations (Table 1, Figure 1).

Table 1: Showing effect of single and daily administration of methanolic extract of *Amaranthus spinosus* on liver of female albino rats indicating level of protein (mg/100mg).

S. No.	Duration	Acute	Chronic
1	Control	29.6±3.90	20.50±1.50
2	5-days	40.6±2.0*	28.60±3.50
3	7-days	33.1±2.5	31.10±5.0
4	14-days	31.1±5.0	82.80±5.80*

Values are given as Mean ± SE of 6 animals in each group. Values are statistically significant at p>0.05.

Figure 1: Illustrating Table 1.



Effect on glycogen content

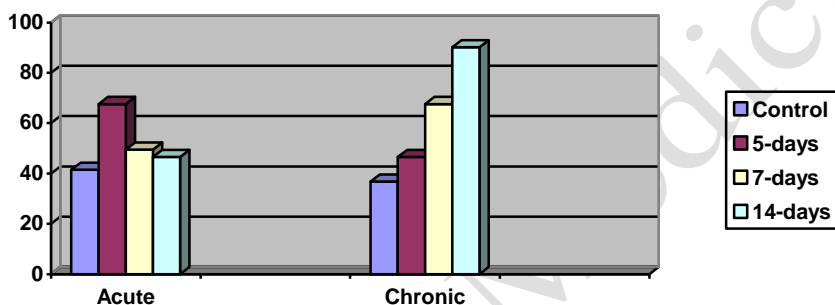
During single administration of the dose, the glycogen content significantly increases at an early duration but at longer duration it recouped to the normal as compared to the control group

in the tested organs. While during daily administration, glycogen content significantly increases both at shorter as well as longer durations (Table 2, Figure 2).

Table 2: Showing effect of single and daily administration of methanolic extract of *Amaranthus spinosus* on liver of female albino rats indicating level of glycogen (mg/100mg).

S. No.	Duration	Acute	Chronic
1	Control	41.5±3.4*	36.8±5.1*
2	5-days	67.6±47.0	46.6±1.6
3	7-days	49.5±4.4	67.6±47.0
4	14-days	46.6±4.3	90.1±7.7

Values are given as Mean ± SE of 6 animals in each group. Values are statistically insignificant at $p < 0.05$.

Figure 2: Illustrating Table 2.

Discussion

As the seeds of the *Amaranthus spinosus* plant contains high protein content which may be responsible for the increase in protein content (Teri, 2009). Proteins are considered as the building blocks of the tissues. The plant acts as a good antioxidant due to the presence of some chemical constituents like flavonoids, phenolic compounds, tannins and amino acids, since glycosylation of proteins is an oxidation reaction, due to the antioxidant activity of the plant it should be able to prevent this reaction that means the plant is having capacity to increase the protein content of the albino rats (Asgary et al., 1999). The *Amaranthus spinosus* plant also shows hepatoprotective activity, due to which it elevates serum enzymatic levels of Serum Glutamate Oxaloacetate Transaminase (AST), Serum Glutamate Pyruvate Transaminase (ALT), Serum Alkaline Phosphatase (SALP) which is involved in the transport of metabolites across the cell membrane, protein synthesis, enzyme synthesis and glycogen metabolism and total bilirubin, which may be responsible for the increase in protein as well as glycogen contents in the tested organ. This study suggested that the possible activity may be due to the presence

of flavonoids, phenolic compounds, betalains, tannins etc. in the ethanolic extract of *Amaranthus spinosus* (Zeashan et al., 2008). Similarly, Orafidiya et al. (2004) studied the acute and sub chronic toxicity of the essential oil of *Ocimum gratissimum* L. leaf, through oral and I.P. route and it was suggested that oral administration leads to less hepatic injury there by not affecting the biochemical parameters to a significant level. A number of reports are also available about the increase in the protein contents due to administration of herbal drugs and extracts (Gupta et al., 1980; Shukla et al., 1987).

However, Nair et al. (1988) and Dogar et al. (1988) have reported that the ethanolic extract of *Strobilanthes heyneams* (leaves) and morphine hydrochloride caused significant decrease in protein contents in liver and kidney. Even Mehta (1991) reported in *Datura metel* and Kushwah (1992) reported that neem oil also causes significant depletion in protein contents in vital and female reproductive organs of albino rats. This depletion may be probably due to toxic nature of the medicinal plants. The rise in protein content due to the administration of *Amaranthus spinosus* may alter certain key

enzymes which are needed for protein synthesis.

As the plant is reported to be rich in carbohydrate content this may be probably responsible for the increase in glycogen content (Sawangjaroen and Sawangjaroen, 2005). The plant also acts as a good antioxidant due to the presence of some chemical constituents like flavonoids, phenolic compounds, betalians, tannins, saponins and glycosides. Due to these constituents the plant extract may alter certain enzymes which may increase the glycogen content (Asgary et al., 1999). Similarly, Vijayanarayana et al. (2007) reported that extract of rhizome of *Curculigo orchioides* causes significant increase in uterine glycogen content. Similarly, a number of reports are available about the increase in the glycogen content due to the administration of herbal drugs (Shukla et al., 1987; Gupta et al., 1980), which will support the present study.

Though, Sujin et al. (2009) reported that *Gymnema sylvestre* causes significant decrease in glucose content in kidney. *Amaranthus spinosus* plant has been reported to contain chemical constituents, which possess antioxidant property so it might have increased the glycogen content in liver due to single and daily administrations.

Conclusion

Overall, the results of the present study indicate that *Amaranthus spinosus* plant demonstrated a significant change in protein and glycogen contents. Thus, at normal therapeutic doses *Amaranthus spinosus* is considered to be safe for the treatment in liver problems.

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