Middle-East Journal of Scientific Research 21 (10): 1893-1897, 2014 ISSN 1990-9233 © IDOSI Publications, 2014 DOI: 10.5829/idosi.mejsr.2014.21.10.84298

Effects of *Psidium guajava* Aqueous Extract on Testosterone and Serum Lipid Profile of Albino Rats

Sushmita Choudhury and M.P. Sinha

University Department of Zoology, Ranchi University, Ranchi-834008, Jharkhand, India

Abstract: The effect of oral administration of aqueous extract of *Psidium guajava* leaves on testosterone and some serum lipid parameters in rats at a dose of 250 mg/kg and 500 mg/kg body weight has been investigated. The extract significantly reduced (p<0.05) total cholesterol concentration, triglycerides and HDL-cholesterol concentration in the serum while it showed no significant effect on serum LDL-cholesterol concentration at all doses administered when compared with control. The recorded data in the present study showed that the testosterone value reduced significantly (p<0.05) from control value (170.62 ± 1.3 ng/dL, 130.94 ± 0.98 ng/dL and 43.80 ± 1.16 ng/dL for control and treated groups respectively) suggesting that the extract may have beneficial effect on serum cholesterol concentration and triglycerides reduction. The reduction in serum testosterone levels indicate significant contraceptive efficacy of *P. guajava* aqueous extract as a sizable reduction in weight of organs like testis, epididymis, prostate and seminal vesicle.

Key words: Psidium guajava · Testosterone · Cholesterol · LDL · HDL · Triglyceride

INTRODUCTION

Use of herbal medicine and related researches are essential for sustainable development particularly in developing countries and human capacity building for healthy population as plants play an essential role in the health care needs for the treatment of diseases and to improve the immunological responses [1-2]. Plant extracts are potentially curative. Some of these extracts have been reported to boost the humoral [3] and cell mediated immunity [4] against viruses [5], bacteria [6], fungi [7], protozoa [8] and cancer [9].

Guava (*Psidium guajava* Linn.), belonging to the Family Myrtaceae, originated in the tropical South America [10] grows wild in Bangladesh, India, Thailand, Brazil, Florida, West Indies, California and also in several other countries [11]. The main constituents of guava are vitamins, tannins, phenolic compounds, flavonoids, essential oils, sesquiterpene alcohols and triterpenoid acids. These and other compounds are related to many health effects of guava [12]. Some authors have found high concentrations of carotenoids (Beta-carotene, lycopene and beta-cryptoxanthin), vitamin C and polyphenols in guava pulp [13, 14]. Lycopene has been correlated with the prevention of cardiovascular damage because of its positive effects on dyslipidemia [15, 16].

Cardiovascular diseases causes some of the main health problems, major ones are coronary heart diseases, stroke and hypertension [17] and elevated plasma lipids are risk factors in cardiovascular problems [18]. Hyperlipidaemia and other abnormal blood lipid profile are largely of genetic origin or due to unwholesome nutritional habits. Lipids like substances accumulate on arterial wall, forming plague, which occlude the vascular lumen and obstruct the blood flow to vital organs such as the heart, brain, liver, or kidney. Obstruction of blood supplies to the heart, brain, liver or kidney cause coronary heart diseases, stroke or kidney failure, as the case may be Gabriel *et al.* [19].

It has also been reported that hypercholesterolemia is a risk factor for cardiovascular diseases such as atherosclerosis and myocardial infarction which are common causes of morbidity and mortality [20]. Increased generation of oxidized LDL is a major factor in the vascular damage associated with high cholesterol levels. Hence, the reduction of lipid profile is considered to be an important therapeutic approach and efforts have been made to identity the lipid lowering effect of various medicinal plants [21]. The prevention of oxidation of low density lipoprotein cholesterol by the antioxidant compounds like poly phenolics and flavonoids is also important in the prevention of cardiovascular diseases and these phytochemicals are present in plants and plant products which are helpful in treating various other diseases [22-26].

The population explosion clearly explains that there is an urgent need for development of alternative contraceptive methods. Among men herbal contraceptive help in interfering with the natural production of sperms [27]. Male reproduction is a complex process that involves the testes, epididymis, accessory sex glands and associated hormones. Testes perform two highly organized and intricate functions, called spermatogenesis and steroidogenesis, which are crucial for the perpetuation of life. Spermatogenesis, a highly dynamic and synchronized process, takes place within the seminiferous tubules of the testis with the support of somatic sertoli cells, leading to the formation of mature spermatozoa from undifferentiated stem cells [28]. The interstitial compartment, which comprises Leydig cells, is the site of steroidogenesis in the testis [29]. With herbal contraception getting its acceptance, this field needs to be explored more. Various plants and plant products reported to have antifertility properties [30]. Despite of several studies on the different pharmacological activities of P. guajava, not much has been investigated on its contraceptive and anti-hypercholesterolemic efficacy. Therefore the present study is aimed to investigate the impact of leaf extract of P. guajava on lipid profile variables and serum testosterone in mammalian animal model and along with its impact on serum cholesterol.

MATERIALS AND METHODS

Collection of plant material: The fresh and tender leaves were collected, dried in shade under $28\pm2^{\circ}C$ (for six to seven days and then crushed into coarse powdery substance by using electric grinder. The coarse powdery substance was dried again and was then sieved to get fine powder using the fine plastic sieve, which was then stored in an air tight bottle in the laboratory until required.

Extract preparation: 50 g of the sieved powder was weighed accurately and subjected to extraction in a Soxhlet apparatus at room temperature using ~350 mL

distilled water. The extract obtained was filtered, concentrated in rotary flash evaporator and maintained at 45°C the percentage yield of each extract were calculated and the dried extracts were stored in air tight containers at room temperature for further studies.

Animals: Male Albino rats (175-200 g) were used in the study. They were maintained under standard laboratory conditions at ambient temperature of $25\pm2^{\circ}$ C and $50\pm15^{\circ}$ relative humidity with a 12-h light/12-h dark cycle. Animals were fed with a commercial pellet diet and water *ad libitum*. The experiments were performed after prior approval of the study protocol by the institutional animal ethics committee of Ranchi University, Ranchi (Proceeding no. 46, page no.137).

Experimental design: The animals were randomly assigned into three groups of six rats each as follows:

- Group 1: Received 1mL of distilled water orally
- Group 2: Received 250 mg/kg body weight of *P. guajava* orally.
- Group 3: Received 500 mg/kg body weight of *P. guajava* orally.

Sample Collection: By the end of each experimental period, the rats were reweighed, starved for 24 hours and sacrificed under chloroform anesthesia. 5 mL of blood was collected from each animal by cardiac puncture using sterile needle and syringe. Part of the blood sample was put into test tubes and allowed to clot for 30 minutes before centrifuging at 800g (Wisperfuge, 1384, Samson, Holland) for 5 minutes. The supernatant was used for the lipid analysis.

Analytical procedure: Estimation of total cholesterol, HDL cholesterol, LDL cholesterol and triglycerides was done by cholesterol oxidase-phenol aminoantipyrine method [31]. Estimation of Testosterone was analyzed by method prescribed by WHO [32].

Statistical analysis: All results were expressed as mean \pm standard error of mean (S.E.M.). Data was analyzed using student's t test.

RESULTS AND DISCUSSION

The effect of the oral administration of aqueous extract of *P. gvajava* leaves on body and organ weights is presented in Table 1 which shows that the plant

Table 1: Effect of *P. guajava* leaves extract on body and organ weights of albino rats

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Weights	Group1	Group 2	Group 3
Body weight (g)	185 ±3.2 ^{a, b}	190.5±3.2ª	198.5±4.5 ^b
Testis weight (mg)	1250.5±21ª	1008.2±18ª	980±22ª
Epididymides (mg)	428.3±6ª	380.2±5ª	370±5ª
Seminal vesicle (mg)	760.1±3 ª	690±5ª	678±7ª
Ventral prostate (mg)	360.3±8 ª	348.2±3ª	329±5ª

values are expressed as mean \pm SEM from the experiments, n=6, ^p<0.05, ^p<0.01 relative to control

Table 2: Effect of aqueous leaf extract of *P. guajava* on lipid profile and testosterone of albino wistar rats

Parameters	Group 1	Group 2	Group 3
Total Cholesterol (mg %)	66±0.05	57.1±0.5ª	55±0.98 ª
HDL cholesterol (mg %)	32±0.78 ª	30±0.49 ª	29±0.34 ª
LDL cholesterol (mg %)	20.6±.09	20±0.09 ^{ns}	20±0.95 ^{ns}
Triglycerides (mg %)	117±2.7	113±2.3 ns	69±1.67ª
Testosterone (ng/dL)	170.62±1.3 ª	130.94±0.98 ^b	43.80±1.16ª

values are expressed as mean \pm SEM from the experiments, n=6, $^{a}p<0.05$, $^{b}p<0.1$, $^{as} =$ non-significant relative to control

extract suggested possible role of the plant extract as a potential agent in the field of male fertility regulation. Significant weight reduction (p<0.05, p<0.01) of the reproductive organs of treated male rats clearly indicates that the drug caused structural and functional alteration in testes, epididymides, seminal vesicle and ventral prostate.

The effect of the oral administration of aqueous extract of *P. gvajava* leaves on testosterone and serum lipid variables is presented in Table 2. The extract showed no significant changes in serum total cholesterol concentration as well as serum HDL-cholesterol concentration at all doses administered when compared with control. However, the extract significantly decreased (p<0.05) serum triacylglycerol concentration at the dose 500 mg/kg body weight when compared with control and no significant changes were recorded at low dose *i.e* 250mg/ kg body weight. Similarly, the present study shows that the testosterone value reduced significantly (p<0.05, p<0.01) in comparison to the control value (170.62 \pm 1.3 ng/dL, 130.94 \pm 0.98 ng/dL and 43.80 \pm 1.16 ng/dL for control and treated groups respectively).

High blood cholesterol concentration is one of the important risk factors for cardiovascular disease [33]. Thus the reduction in serum total cholesterol concentration effected by the extract is beneficial and may reduce the risk of cardiovascular disease because agents that have the ability to lower cholesterol concentration in the blood have been reported to reduce vascular

resistance by improving endothelial function [33]. Similar alterations in lipid profiles were reported in various other plant extracts such as *Bulbine natalensis* [17], *Bougainvillea spectabilies* leaves [34] and *Fadogia agrestis* stem [35]. The results suggested that *P. guajava* extracts studied showed effect in regulating the cholesterol and triglyceride levels.

Among the plant based contraceptives, inhibition of male fertility after administration of natural substances has been related to decreased spermatozoa density [36]. Also, for male contraception, it is not necessary to stop spermatogenesis, rather to eliminate the fertilizing ability of the spermatozoa by causing changes in the morphology or in the function of the sperm [36].

Saccharum officinarum, Momerdica diocia and Ocimum sanctum are commonly known plants which possess antifertility activities as reported previously [37]. Raji et al. [38] reported that the effects of the ethanol extract of Azadirachta indica stem bark on body and organ weights, sperm morphology, counts and viability, serum levels of testosterone, luteinizing hormone (LH) and follicle stimulating hormone (FSH) were studied in albino rats. Azadirachta indica produced dose-dependent reduction in serum testosterone and LH but no change in FSH levels. In the present study the testosterone values are reducing significantly therefore the aqueous extract may have contraceptive properties. However, such deduction needs to be substantiated by other testicular parameters and histopathological studies.

ACKNOWLEDMENT

The first author acknowledges the financial assistance received by SERB, New Delhi (SB/FT/LS-406/2012) under the FAST TRACK SCHEME FOR YOUNG SCIENTIST.

REFERENCES

- Borchers, A.T., S. Sakai, G.L. Henderson, M.R. Harkey, C.L. Keen, J.S. Stern, K. Terasawa and M.E. Gershwin, 2000. Shosaiko-to and other Kampo (Japanese herbal) medicines: a review of their immunomodulatory activities. J. Ethnopharmacol., 73: 1-13.
- Kumar, A., S. Dandapat, M. Kumar and M.P. Sinha, 2013. Antipathogenic efficacy and aemolytic activity of *Calotropis procera* leaves. World Journal of Zoology, 8(4): 366-370.

- Rehman, J., J.M. Dillow, S.M. Carter, J. Chou, B.B. Le and A.S. Maisel, 1999. Increased production of antigen-specific immunoglobulins G and M following in vivo treatment with the medicinal plants *Echinacea angustifolia* and *Hydrastis canadensis*. Immunol. Lett., 68: 391-396.
- Upadhyay, S., S. Dhawan, S. Garg and G.P. Talwar, 1992. Immunomodulatory effects of neem (*Azadirachta indica*) oil. Int. J. Immunopharmacol., 14: 1187-1193.
- Calixto, J.B., A.R. Santos, V. Cechinel Filho and R.A. Yunes, 1998. A review of the plants of the genus Phyllanthus: their chemistry, pharmacology and therapeutic potential Med. Res. Rev., 18: 225-258.
- Boyanova, L. and G. Neshev, 1999. Inhibitory effect of rose oil products on *Helicobacter pylori* growth in vitro: preliminary report. J. Med. Microbiol., 48: 705-706.
- Ali, M.I., N.M. Shalaby, M.H. Elgamal and A.S. Mousa, 1999. Antifungal effects of different plant extracts and their major components of selected aloe species. Phytother. Res., 13: 401-407.
- Sharma, P. and J.D. Sharma, 1998. Plants showing antiplasmodial activity from crude extracts to isolated compounds. Indian J. Malariol., 35: 57-110.
- Wong, C.K., K.N. Leung, K.P. Fung and Y.M. Choy, 1994. Immunomodulatory and anti-tumour polysaccharides from medicinal plants. J. Int. Med. Res., 22: 299-312.
- Pathak, R.K. and C.M. Ojha, 1993. Genetic resources of guava, Vol. I, Fruit Crops, Part 1, In; Advance in Horticulture [C]. Chadha KL, Pareek OP, Editors, Malhotra Publishing House, New Delhi, pp: 143-147.
- Bailey, L.H., 1960. The standard encyclopedia of horticulture [C]. Vol. II. Macmillan Co, New York, pp: 1415.
- Haida, K.S., A. Baron and K.S. Haida, 2011. Phenolic compounds and antioxidant activity of two varieties of guava and rue. Rev. Bras. Ciênc. Saúde., 28: 11-19.
- Oliveira Dda, S., A.L. Lobato, S.M Ribeiro, A.M. Santana and J.B. Chaves, 2010. Carotenoids and Vitamin C during Handling and Distribution of Guava (*Psidium guajava* L.), Mango (*Mangifera indica* L.) and Papaya (*Carica papaya* L.) at Commercial Restaurants. J. Agric. Food Chem., 58: 6166-6172.
- Ordóñez-Santos, L.E. and A. Vázquez-Riascos, 2010. Effect of processing and storage time on the vitamin C and lycopene contents of nectar of pink guava (*Psidium guajava* L). Arch. Latinoam Nutr., 60: 280-284.

- Lorenz, M., M. Fechner, J. Kalkowski, K. Fröhlich and A. Trautmann, 2012. Effects of Lycopene on the Initial State of Atherosclerosis in New Zealand White (NZW) Rabbits. PLoS One, 7: e30808
- Sesso, H.D., L. Wang, P.M. Ridker and J.E. Buring, 2012. Tomato-based food products are related to clinically modest improvements in selected coronary biomarkers in women. J. Nutr., 142: 326-333.
- James, D.B., O.A. Kadejo, C. Nwochiri and C.D. Luca, 2013. Determination of Phytochemical Constituents of the Aqueous Extracts of the Leaves, Stem Bark and Root Bark of *Vitex doniana* and its Effects on Lipid Profile of Albino Rats. British Journal of Pharmacology and Toxicology, 4(6): 210-214.
- Yakubu, M.T. and A.J. Afolayan, 2009. Effect of aqueous extract of Bulbine natalensis Baker on haematological and serum lipid profile of male wistar rats. Ind. J. Expt. Sci., 47: 283-288.
- Gabriel, O., N. Harrision, O. Okey and A. Ukoha, 2008. Changes in Lipid and Haematological profile Of Aqueous Ethanolic Extract of *Alstonia boonei* In Rats. The Internet J. Hematol., 4(1): 1-8. DOI: 10.5580/2877.
- William, C., M.D. Cromwell, D. James and O. Otvos, 2004. Low density lipoprotein particle number and risk for cardiovascular disease. Curr. Aetheroscle. Rep., 6: 381-387.
- Onat, A., G.S. Arci, M.M. Barlan, H. Uyarel, Uzunlan and V. Sansoy, 2004. Measures of abdominal obesity assessed for visceral adiposity and relation to coronary risk. J. Obesity Related Metab. Disord., 28: 1018-1025.
- Choudhury, S., L. Sharan and M.P. Sinha, 2012. Phytochemical and Antimicrobial Screening of *Psidium guajava* L. leaf Extracts against Clinically Important Gastrointestinal Pathogens. J. Natural Prod. Plant Resour., 2(4): 524-529
- Choudhury, S., L. Sharan and M.P. Sinha, 2012. Antibacterial efficacy and phytochemistry of methanolic leaf extracts of *Mangifera indica* Linn. The Ecoscan; special issue, 1: 419-423.
- 24. Choudhury, S., L. Sharan and M.P. Sinha, 2013. Pharmacological Efficacy of Some Medicinal Plants used for Treatment of Gastrointestinal diseases. Proceedings of National Seminar on Ecology, Environment and Development, 25-27 January, The Ecoscan. Special issue, 3: 111-116.
- Choudhury, S., L. Sharan and M.P. Sinha, 2013. Phytochemical and antimicrobial standardization of the methanolic leaf extracts on Murraya koenigii Linn. Archives des Sciences, 66(3): 67-80.

- Choudhury, S., L. Sharan and M.P. Sinha, 2013. Antidiarrhoeal potentiality of leaf extracts of *Moringa oleifera*. British J. Appl. Sci. Tech., 3(4): 1086-1096.
- Gediya, S., C. Ribadiya, J. Soni, N. Shah and H. Jain, 2011. Int. J. Curr. Pharm. Rev. Res., 2(1): 47-53.
- Hess, R.A. and L. Renato de Franca, 2008. Spermatogenesis and cycle of the seminiferous epithelium.Adv. Exp. Med. Biol., 636: 1-15.
- Osinowo, O.A., 2006. Introduction to Animal Reproduction. Sophie Academic Services Limited, Abeokuta, Nigeria 1st edition, pp: 91.
- Odusoga, A.O., O. Ifabunmi and O. Ayokunle, 2014. Evaluation of oral administration of aqueous leaf extract of *Momordica charantia* on fertility hormones of adult male Wistar rats. Global journal of pharmacology. 8(2): 150-153.
- Rifa, N. and G.R. Warnick, 2006. Lipids, lipoproteins, apolipoproteins and other cardiovascular risk factors. In: Burtis CA, Ashwood ER, Bruns DE, Editors. Tietz text book of clinical chemistry and molecular diagnostics. 4th edition, New Delhi: Elsevier's, pp: 942-960.
- 32. WHO, 1987. Method manual. Program for the provision of matched assay reagents for the radioimmunoassay of hormones in reproductive physiology fifth editor. Geneva: World Health Organization.

- Adebayo, J.O., A.A. Adesokan, L.A. Olatunji, D.O. Buoro and A.O. Soladoye, 2005. Effect of Ethanolic extract of *Bougainvillea spectabilis* leaves on haematological and serum lipid variables in rats. Biochem., 17: 45-50.
- Yakubu, M.T., M.A. Akanji and A.T. Oladiji, 2007. Haematological evaluation in male albino rats following chronic administration of aqueous extract of *Fadogia agrestis* stem. Pharmacog. Mag., 3: 34-39.
- 35. Sharma, N. and D. Jacob, 2001. Antifertility investigation and toxicological screening of the petroleum ether extract of the leaves of *Mentha arvensis* L. in male albino mice. J Ethnopharmacol., 75: 5-12.
- 36. Nikkanen, V., K.O. Soderstrom, S. Tuusa and U.M. Jaakkola, 2000. Effect of local epididymal levonorgestrel on the levorgestrel on the fertilizing ability of male rat, a model for post-testicular contraception. Contraception, 61: 4001-6.
- 37. Singh, A. and A. Kala, 2011. Duration dependent effect of plants extracts on Hematology Histopathology Hormonal profile and Sperm parameters of rats: An approach for male contraceptive development. Double Helix in Pharmaceutical Research, 1(1): 1-9.
- Raji, Y., U.S. Udoh, O.O. Mewoyeka, F.C. Ononye and A.F. Bolarinwa, 2003. Implication of reproductive endocrine malfunction in male antifertility efficacy of *Azadirachta indica* extract in rats. African Journal of Medicine and Medical Sciences, 32(2): 159-165.