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Review Article

BIOACTIVE COMPOUNDS OF ACHYRANTHES

(AMARANTHACEAE) AND THEIR PHARMACOLOGICAL AND PHYTOCHEMICAL USES

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ABSTRACT

The species of *Achyranthes* (fam. Amaranthaceae) is an important herbaceous weed plant has wide range of pharmacological and phytochemical properties. Several bioactive compounds have been isolated from different parts of plants contain ssaponins A, B, sugars like D-glucose, L-rhamnose, oleanolic acid and ecdysterone. These different parts of plant used as expectorant, stomach tonic, laxative, anti-helmintic, diuretic, linthontriptic, sudorific, demulcent, anti-inflammatory and haematinic in indigenous medicine. The plant has antiviral, antibacterial activity, anti-diabetic, spasmolytic to smooth muscle, diuretic and purgative. The plant has medicinal and ethno-botanical properties that make it more valuable in tribal and rural community.

Keywords: Pharmacological, Phytochemical, Amaranthus spinosus, Amaranthaceae, RAPD.

INTRODUCTION

The species of Achyranthes (fam.Amaranthaceae) play vital role as medicinal or ethno-botanical plant in human life. The common species in India Achyranthes aspera L., recognized viz. Latjira (Hindi), Apamargah (Sanskrit), Apang (Bengali), Katalati (Malyalam), Nayurivi (Tamil), Uttaren (Telugu) and Prickly-chaff flower plant (English). The plant grows in a range of climatic conditions but it is frost intolerant. It is commonly grown in all soil types but grows best in well drained and moist situations or in neutral soils. The plant found on disturbed ground, along roadsides, railway lines, neglected land and poorly maintained grazing land or also grown as a weed in a variety of crops and horticultural enterprises. A weed (plants intentionally grown by people in gardens or other cultivated-plant settings due to their economic value or ornamentation) is used in a variety of senses, generally centering on a plant that is not desired within a certain context or unwanted plants in human-controlled settings, especially farm fields,gardens, lawns andparks or any plants that grow, reproduce aggressively,invasively,grow in diverse environmentsquickly.Botanically,the plant is small herb found all over Indiapossessing valuable medicinal properties. It is the plant is widespread inthe world as a weed, in Baluchistan, Ceylon, Tropical Asia, Africa, Australia and America.

THE PLANT

A perennial stiff erect herb, 0.2-2.0 m high, is growing up to 1000 m height. Stems aresquare, leaves elliptic ovate or broadly rhombate, 5-22 cm long, 2-5 cm broad, andadepressed pubescent. The inflorescences are 8-30 cm long, with many single, white or red flower of 3-7 mm wide.The Flowering time of this plant is in summer.

CONSTITUENTS

Compounds in the seeds of *A. aspera* are the saponins A and B (glycosides of oleanolic acid). The carbohydrate components are the sugars D-

glucose, L-rhamnose, D-glucuronic acid (= Saponin A). Saponin B is the ß-Dgalactopyranosyl ester of Saponin A (Hariharan and Rangaswami, 1970). The content of free oleanolic acid in A. aspera roots is 0.54 % (Batta and Rangaswami, 1973, LiX and HuS, 1995), ecdysterone and oleanolic acid has been isolated from the roots. In the unripe seeds saponines, oleanolic acid, amino acids and hentriacontane (a long chained carbohydrate) havebeen found.In the shoots aliphatic an 36. dihydroxyketone 37dihydroxyhenpentacontan-4-on andtriacontanol could be found (Batta and Rangaswami, 1973). Two other long chain compounds, isolated from shoots. characterized the are as 27cyclohexylheptacosan-7-ol and 16-hydroxy, 26methylheptacosan-2-on by chemical and spectral investigations (Misraet al, 1993). The petro extract of the shoots produced a

yellow semi-solid mass. From this a pink coloured essential oil with a pleasant odour and an aliphatic alcohol (penta triacontanol) were found (Gariballa *et al*, 1983). Ecdysterone, (phytoecdysone) was yielded and characterized by its colour and special chemical reactions. Its Contents (g/kg) were 0.25 (seeds), 0.09 (roots), 0.04 (stem, leaves) (Banerji and Chadha, 1970, Banerji *et al*, 1971). Presence of ecdysterone the pronounced insect moulting hormonal activity of this extract of roots has been found (Banerji and Chadha, 1970).

USES

A. aspera is an important medicinal herb found as a weedthroughout India. Almost all of its parts are used in traditional systems of medicines. From the last few decades or so, extensive research work has been done to prove its biological activities and pharmacology of its extracts. Saponins, oleonolic acid, dihydroxy ketones, alkaloids, long chain compounds and many other chemical constituents have been isolated (Srivastav*et al*, 2011).

Sahaiet al (2007) reported that different parts of plant have beenused as expectorant, stomach laxative, anthelmintic, diuretic. tonic. linthontriptic, sudorific, demulcent, antiinflammatory and haematinic in indigenous medicine. Many studies reported (Auniappan and Savarimuthu, 2008) the diverse actions of Achyranthes aspera e.g. antiviral (Khurana and Bhargava, 1970) antibacterial activity (Ikram and Hag, 1980, Farouk et al, 1983) antifertility (Pakrashiet al,1975) anti-diabetic, positive inotropic effect, spasmolytic to smooth muscle (Kapoor and Singh, 1967) diuretic and purgative (Neogi et al, 1970). An aqueous extract of aerial parts showed significant reduction in cholera toxin-induced intestinal hypersecretion in mice (Claeson and Samuelsson, 1989). It is also used indiabetes as hyperglycemia, hyperlipidemia and negative nitrogen balance. Tripathi (2003) reported a wide spread pathological change is thickening of capillary basement membrane, increased vesselwall thickening and cell proliferation resulting in vascular complications like early atherosclerosis; neuropathy and peripheral vascular insufficiency. Vascular complications are the major cause of developing diabetic ulcers. Diabetic ulcers/wounds areslow; non-healing that can last for weeks despite adequate and appropriate care because diabetic wound healing is an enigmatic and debilitating complication and poses a serious challenge inclinical practice.

A. aspera has been used as hypoglycemic, hypolipidemic, haematinic and ingeneral debility in folk medicine. Earlier studies reported its hypoglycemic, hypolipidemic activity on hematological parameters (i.e. RBC, WBC count, Hb%, clotting time, O₂ carrying capacity).These actions may complement hypoglycemic, hypolipidemic actions in treating diabetic complications (Zambare *et al.* 2011).

It is used in the treatment of fever, especially malaria, dysentery, asthma, hypertension. The roots of the A. aspera are reported to have antiimplantation, abortifacient activity and spermicidal activity. The deficiency or excess of trace elements lead to various complications and metabolic disorders in human being. It is useful incough, bronchitis, rheumatism, renal and cardiac dropsy, anddiabetes mellitus. It can stimulate the immunity, enhance the antigen clearance, potentiate antibody production, elevate thyroid hormone levels, decrease hepatic lipid peroxidation and also possesses chemopreventive, spermicidal, antiinflammatory, anti-arthritic. In Chinese traditional medicine, the hot water extract of the plant has been used as an antiarthritic to alleviate arthritic pain. The dried leaf powder mixed with honey is useful in the early stages of asthma. Oleanolic acid is one of the constituents of A. aspera, A. bidentata extract canpromote neuronal growth, protect hippocampal neurons against toxicity, and also has anti-stress and anti-apoptosis activities. Both the plants are found to be a source of many secondary metabolites (Gnanaraj et al. 2012).

A. aspera is rich in Iron, Zinc and protein content can serve as a good dietary supplement especially for diabetes and obese people.Zinc and Iron was found to behigh in this plant, 1.52 ppm and 63.26 ppm along with 0.87 mg/kg ofprotein respectively. It can be a good dietary supplementbecause of the significance of Iron towards providing remedy fordiabetes and its complications including micro angiopathy and atherosclerosis (Swaminathan, 2007) and adequate protein intake is essential for protein loss often thecompensation of encountered in hypoglycaemic conditions (Anonymous, 2004). Zinc, component of many enzymes that involved in the maintenance of several tissue functions (Zargar et al., 1998). The plant act as a slimming supplement for obese patients, due to its increased production of glycogen in liver and its appetitesuppressing property which increases body heat production byactivating thermogenesis and encourages weight loss (Mangal et al. 2009).

REVIEW OF LITERATURE

Diabetes mellitus (metabolic disorders) characterized by hyperglycemia, hyperglycemia, dyslipidemia, vascular complications and defective metabolism of glucose and lipids characterized by relative orabsolute deficiency of insulin (Porter, and Barrett, 2005, Turneret al 2005, Singh and Chandra, 2009, Udupa et al, 1995) and management of chronic ulcers is another major problem due to the high cost of therapy and the presence of unwanted side effects (Porraset al, 1993, Suh et al. 1998). The American Diabetic Association- 2000 reported that there was marked reduction in RBC count and Hb content of diabetics which may be dueto imbalance in erythropoiesis and anemia associated with diabetes; evidenced in present study by significant (P < 0.05) reduction in the RBC's and Hb count in alloxan induceddiabetic animals. At the same time prolonged clotting time due to disturbed blood clotting cascade because of the delayed expression of clotting factors in diabetics was also observed. Furthermore the natural defense mechanism that operates through WBCs by phagocytosis isobserved to be hampered in diabetics due to reduction in WBC count (Veronelli, 2004). The basic requirement of optimal healing is to minimize tissue damage and provide adequate tissue perfusion and oxygenation, proper nutrition and moist wound healing environment to restore the anatomical continuity and function of the affected (Pierce and Mustoe, 1995). The findings of present study clearly demonstrated that EEAA improves the hematological parameters like RBC, WBC count, Hb%, clotting time, O_2 carrying capacity and color index which may play an important role in offering favorable environment forwound healing. Phytochemical screening of EEAA has revealed presence of many phytochemicals i.e.alkaloids, triterpenoids, saponins, flavonoids, amino sugars and tannins etc. Literature search

have revealed that hypoglycemic and hypolipidemic activity of EEAA is due to presence of flavonoids and saponins as major phytochemicals (Erdman *et al.* 2007, Luo *et al.* 2005). *Achyranthes aspera* whole plant extract shows haematinic along with antidiabetic antihyperlipidemic activity. Our results fortify importance of these plants as an alternative haematinic, antidiabetic-anti-hyperlipidemic remedy (Pierce and Mustoe, 1995).

Health improvement mediated bv "Nutraceuticals" has triggered an increased global interest and the current global market size of herbal Nutraceuticals is estimated between 30 and 60 billion (Annonymous, 2004). In order to contribute to the human health and National economy, attempts were made to evaluate Nutraceutical potentials of certain common plants available from Tamil Nadu, India. Common plants such as A.aspera L, Boerhavia diffusa L, Mukiamadra spatana (L) Roem, Scoparia dulcis, and Phyllanthus madaraspatensis L were collected, identified, authenticated and evaluated for their nutraceutical potential. The shade dried, coarsely powdered and extracted with water and alcohol were subjected to nutraceutical analysis and their nutraceutical valueswere determined. Among the selected plants Mukiamadr aspatana (L) Roem possessed high nutraceutical values.

The ethanolic leaf extract of *A. aspera* by *in vitro*, Hen's Egg Chorioallantoic Membrane method (HET-CAM) Showed increased density of newblood capillaries as compared with control group treated with 0.9% NaCI. The results of leaf extract revealed a significant scope to developa novel broad spectrum of herbal formulations forwound healing and different herbal formulations (Kumar *et al.* 2012).

Great interest in herbal medicine as a potential source of phytopharmaceuticals has created the need to review common factors responsible for major diseases and body disorders. Traditional medicinal herbal remedies in the southern African region have long been used to treat various pain-orinflammation-related symptoms. There is increasing evidence to indicate that both peripheral and central nervous system cells play a prominent role in the chronic inflammatory responses in the body system and anti-inflammatory herbal medicine and its constituents are being proved to be a potent protector against various pro-inflammatory mediators indiseases and disorders. The structural diversity of these medicinal herbs makes them a valuable source of novel lead compounds against the therapeutic molecular targets, cytokines and mediators that have been

newly discovered by the platforms of genomics, proteomics, metabolomics and high through technologies. The basic mechanisms of inflammation and the potential of 123 southern African plant species were found effective as chronic inflammatory disease preventive agents (Iwalewa *et al.* 2007).

The anti-inflammatory activity of an alcohol extract of *A.aspera* was tested on carrageenin induced hind paw oedema and cotton pellet granuloma models in albino male rats. The animals were fed with an alcohol extract at various dose levels (125, 250, 375 and 500 mg/kg). Diclofenac sodium was used as a standard drug. The alcohol extract (375 and 500 mg/kg) showed the maximum inhibition of oedema of 65.38% and 72.37% at the end of 3 h with carrageenin-induced rat paw oedema, respectively.Using a chronic test, the extract exhibited a 40.03% and 45.32% reduction in granuloma weight (Vertichelvan *et al.* 2003).

Efficiency of *A. aspera* was evaluated in Swiss albino mice after treated with mineral oil. The anti-cancerous activity of plantleaves was tested against mineral oilinduced cancer in mice which indicated that the ether extract at the concentration of 3 mg/ ml was very effective in reducing the cancer symptoms (Geetha*et al.*2010).

Plants and plant-based medicaments are the of many of the modern basis pharmaceuticals. The radical scavenging activity of the different extracts of root, stem, leaf and inflorescences wasevaluated by DPPH assay and the antibacterial activity against Staphylococcus aureus a grampositive and Escherichia coli a gram negative bacterium was studied by Agar well cut diffusion method. All of the extracts exhibited different antioxidant and antibacterial activities and theactivities varied from solvent to solvent and the activities were concentration and time dependant. The antioxidant and antibacterial activities were compared with the positive control using ascorbicacid and qualitative phytochemical gentamycin. A analysis was carried out and found to possessbioactive compounds like alkaloids, glycosides, terpenoids, steroids, flavonoids and tannins (Blaulah et al. 2011).

The antifungal activity and minimum inhibitory concentration (MIC) of various plant extracts indifferent solvents such as hydro-alcohol (50 % v/v) and hexane of plants traditionally used as medicines Valeriana as jatamansi (Sugandhbala), Coleus barbatus (Patharchoor), Berberis aristata (Kingore), Asparagus racemosus (Satrawal), Andrographis paniculata (Kalmegha), A. aspera (Latjiri), Tinospora cordifolia (Giloei), Plantago depressa (Isabgol) were evaluated against Aspergillus niger and Candida albicans. Hydro-alcoholic extracts of all the plants were found to have maximum antifungal activity in comparison to hexane extracts. Hydro alcoholic extracts of Andrographis paniculata and A. aspera showed maximum potency against Aspergillus niger and Candida albicans at highest MIC value of 0.5 and 0.3mg/ml respectively. Hexane extracts of Andrographis paniculata showed highest MIC value of 0.7 mg/ml against A. niger (Mathur et al.2011).

An ideal Anti-microbial is perpetual and never ending in any system of medicine and becomes mandatory as the microbial evolve newer methods of resisting the antimicrobial activity of the existing drugs in the market. To improve theQuality of therapy and hence the quality of life of the patient, It is require to find out newer molecules, which are patient friendly, reasonably affordable, least toxic and available inplenty. Still fight for identifying such an ideal drug to fight common organisms that inhabitate human systems. The Siddha system of Medicine is regarded as the most unique system because of its exclusive Materia Medica. Among the various sources of drugs, salt based preparations are note worthy. The herbal salts mentioned in Siddha literatures include Madar (Calotropis gigantean), Indian marsh mallow (Sida mauritiana), Rubbish plant (Acalypha indica), Plantain tree (Musa paradisiaca) and Drumstick tree (Hyperanthera moringa). The superiority of these herbal based salts are that they are easily available, economical, time tested and have no side effects. It is used in the treatment of lipid disorders in the Indian system of medicine. The saponin extract of Achyranthes asperahas both hypolipidemic and weight reducing effects on high fat diet fed rats (Lathaet al. 2011).

BIOTECHNOLOGICAL APPROACH

The latest studies reported that extract of A.aspera was found to enhance the induction of ovalbumin specific humoral antibody response in mice, on intra-peritoneal injection of extract with ovalbumin specific humoral along antibody. The antibody response was evaluated by passive cutaneous anaphylaxis and ELISA for IgE and other classes or subclasses of antibodies, respectively. The adjuvant property of the extract was further examined in different strains of mice and a significant elevation of the OVAspecific IgG antibody response in all strains tested was found. When the extracts of different parts of the herb were tested, the seed and root extracts appeared to exhibit relatively higher activity. It has been established and validated for

determination. Detection and quantification were of oleanolic acid in methanolic extract of *A. aspera* roots and leaves by HPTLC method was performed by densitometry at $\lambda = 529$ nm. In roots and leaves methanolic extract oleanolic acid content was found 0.37% and 0.13 %w/w respectively.

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REFERENCES

- 1. Anonymous. Nutrition Principles and Recommendations in Diabetes, diabetes care, 2004, 27, supplement 1, January 2004.
- 2. Auniappan A and Savarimuthu I. Medicinal Uses and Pharmacological Actions of Five commonly used Indian medicinal plants: A Mini-Review. Iranian Journal of Pharmacology & Therapeutics. 2008;7(1):107-114.
- 3. Banerji A and Chadha MS. Insect moulting hormone from Achyranthes aspera. Phytochemistry. 1970;9:1671.
- 4. Banerji A, Chintalwar GJ and Joshi NK. Isolation of ecdysterone from Indian plants. Phytochemistry. 1971;10:2225-6.
- 5. Batta AK and Rangaswami S. Crystalline chemical components of some vegetable drugs. Phytochemistry. 1973;12:214-6.
- 6. Blaulah AG, Sadiq MA and Santhi JR. Antioxidant and antibacterial activity of Achyranthes aspera an in vitro study. Scholars Res Lib. 2011;3(5):255-262.
- Claeson P and Samuelsson G. Screening of some Somalian medicinal plants for anti diarrhoeal effects in mice. Phytotherapy Res. 1989;3:180-183.
- Erdman Jr JW, Balentine D, Arab L, Beecher G, Dwyer JT, Folts J, Harnly J, Hollman P, Keen CL, Mazza G, Messina M, Scalbert A, Vita J, Williamson G and Burrowes J. Flavonoids and Heart Health: Proceedings of the ILSI North America Flavonoids workshop May 31– June 1, 2005, Washington, DC1–4. Journal of Nutrition. 2007;137(3):718 -737.

- 9. Farouk A, Bashir AK and Salih AKM. Antimicrobial activity on certain Sudanese plants used in folkloric medicine, Screening for antibacterial activity. Fitoterapia, 1983;54: 3-7.
- 10. Gariballa Y, Iskander GM, Daw EL and Beit A. Investigation of the alkaloid components in the Sudan Flora III. Fitoterapia. 1983;54:269-72.
- 11. Geetha P, Narayanan KR and Murugesan AG. Screening the anticancerous efficacy of Achyranthes aspera L. using animal model Swiss albino mice. J Biomed Sci and Res. 2010;2(4): 231-235.
- 12. Gnanaraj WE, Antonisamy JM, Mohanamathi RB and Subramanian KM. In vitro clonal propagation of Achyranthes aspera L. and Abutilon indicum using nodal explants. Asian Journal of Tropical Biomedicine. 2012;1-5).
- 13. Hariharan V and Rangaswami S. Structure of saponines A and B from the seeds of Achyranthes aspera. Phytochemistry. 1970;9:409-414.
- 14. Ikram M and Haq I. Screening of medicinal plants for antimicrobial activity. Firoterapia. 1980; 51:281-284.
- 15. Iwalewa EO, McGaw LJ, Naidoo V and Eloff JN. Inflammation: the foundation of diseases and disorders. A review of phytomedicines of South America Origen used to treat and inflammatory conditions, AJB. 2007;6(25):2868-2885.
- 16. Kapoor VK and Singh H. Investigation of Achyranthes aspera Linn. Ind J Pharma. 1967;29:285-88.
- 17. Khurana SMP and Bhargava KS. Effect of plant extracts on the activity of three papaya viruses. J Gen Appl Microbiol. 1970;16:225-230.
- Kumar HK, Deshmukh GR, Reddy PV, Sureshrao BS and Valluru L. Preliminary investigation of Angiogenic property of ethanolic leaf extract of Achyranthes aspera using chorioallantoic membrane model. IJDDR. 2012;4(1).
- 19. Latha PB, Vijaya T, Reddy IRM, Ismail M and Rao SD. Therapeutic efficacy of Achyranthes aspera saponin extract in high fat diet induced hyperlipidaemia in male wistar rats. A. J of Biotechech. 2011;10(74):17038-17042.
- 20. Li X and Hu S. Determination of oleanolic acid in the root of Achyranthes bidentata from different places of production by TLC-scanning.

ZhongguoZhong Yao ZaZhi. 1995;20(8):459-60.

- 21. Luo L, Yin HJ, Zhang Y, Jiang YR, Liu Y and Shi DZ. Effect of ginseng fruit saponins on insulin sensitivity index in high fat-fed rats.Journal of Chinese Integrative Medicine. 2005;3(6):463-465.
- 22. Mangal A and Sharma MC. Evalution of certain Medicinal plants for antiobesity properties, Indian Journal of Traditional Knowledge. 2009;8(4):602-605.
- 23. Mathur A, Singh, R, Yousef S, Bhardwaj A, Verma SK, Babu P, Gupta V, Prasad GBKS and Dua VK. Antifungal activity of some plants extract against clinical pathogens. Pelagia Res Lib Advances in Applied Sci Res. 2011;2(2):260-264.
- 24. Misra TG, Singh RS, Pandey HS. Two long chain compounds from Achyranthes aspera. Phytochemistry. 1993;33:1:221-3.
- 25. Neogi NC, Garg RD and Rathor RS. Preliminary pharmacological studies on achyranthine. Indian J Pharm. 1970;32:43-46.
- 26. Pakrashi A, Basak B and Mokerji N. Search for anti-fertility agents from indigenous medicinal plants. Indian J Med Res. 1975;63:375-378.
- 27. Pierce GF and Mustoe TA. Pharmacologic enhancement of wound healing.Annual Review of Medicine. 1995; 46:467-481.
- 28. Porras-Reyes BH, Lewis WH, Roman J, Simchowitz L and Mustoe TA. Enhancement of wound healing by the alkaloid taspine defining mechanism of action.Soc Exp Biol Med. 1993;203:18-25.
- 29. Porter JR and Barrett TG. Monogenic syndromes of abnormal glucose homeostasis: clinical review and relevance to the understanding of the pathology of insulin resistanceand cell failure. J Med Gene. 2005;42:893-902.
- 30. Sahai S, Pavithran P and Barpujari I. BIOPIRACY: Imitations not innovations, Gene Campaign, 2007;27.
- 31. Singh MP and Chandra SS. Wound healing activity of Terminalia chebula in

experimentally induced diabetic rats. Int J Pharm Tech Res. 2009;1(4):1267-1270.

- 32. Srivastav S, Singh P, Mishra G, Jha KK and Khosa RL. A. aspera an important medicinal plant: A review J. Nat. Prod. Resour. 2011;1(1):1-14.
- 33. Suh DD, Schwartz IP, Canning DA, Snyder HM, Zderic SA and Kirsch AJ. Comparison of dermal and epithelial approaches to laser tissue soldering for skin flap closure. LasersSurg Med. 1998;22:268–274.
- 34. Swaminathan S. The Role of Iron in Diabetes and Its Complications, DIABETES CARE. 2007;30:7.
- 35. Tripathi KD. Essentials of Medical Pharmacology. 5th ed. JPB. 2003;235.
- 36. Turner RC, Cull CA, Frighi V and Holman RR. Glycaemic control with diet, sulfonylurea, metformin: Progressive requirements for multiple therapies. 2005;12:281-312.
- Udupa AL, Kulkarni DR and Udupa SL. Effect of Tridax procumbens extracts on wound healing. Int J Pharmacognosy. 1995;33:37-40.
- 38. Veronelli A, Vardaro D, Laneri M, Paganelli M, Ranieri R, Folli F, Koprivec D and Pontiroli AE. White blood cells in obesity and diabetes. Diabetes Care. 2004;27:2501-2502.
- 39. Vertichelvan T and Jagadusan M. Effect of alcohol extract of Achyranthes aspera on acute and subacute inflammation. Phytotherapy Research. 2003;17:77-79.
- 40. Zambare M, Bhosali AU, Somani RS, Yegnanarayan R and Talpate AK. Effect of treatment with aethano extract on various hematological and biochemical parameters in Alloxam Induced Diabetic Rats. IJPFR. 2011;1(1):42-52.
- 41. Zargar AH, Shah NA, Masoodi SR, Laway BA, Dar FA, Khan AR, Sofi FA and Wani AI. Copper, zinc, and magnesium levels in non insulin dependent diabetes mellitus. Postgrad Med J. 1998; 74:665-668.