School of Life Sciences, Devi Ahilya University, Indore, India

Hypoglycaemic effects of some plant extracts are possibly mediated through inhibition in corticosteroid concentration

S. Gholap, A. Kar

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Dr. Anand Kar, Thyroid Research Unit, School of Life Sciences, Devi Ahilya University, Khandwa Road Complex, Indore – 452017, India

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To unravel the possible mechanism of glucose lowering activity, effects of ten different plant extracts in the regulation of serum cortisol and glucose concentrations were evaluated in male mice. While the extracts of *Inula racemosa*, *Boerhaavia diffusa* and *Ocimum sanctum* decreased the serum concentration of both cortisol and glucose, *Aegle marmelos, Azadirachta indica* and *Gymnema sylvestre* extracts could exhibit hypoglycaemic activity without altering the serum cortisol concentration. It appears that the hypoglycaemic effects of former three plant extracts are mediated through their cortisol inhibiting potency, whereas the mechanism for other plant extracts could be different. Lipid-peroxidation was not enhanced by any of the plant extracts (some were in fact, antiperoxidative in nature). As *I. racemosa*, *B. diffusa* and *O. sanctum* exhibited antiperoxidative, hypoglycaemic and cortisol lowering activities, it is suggested that these three plant extracts may potentially regulate corticosteroid induced diabetes mellitus.

1. Introduction

Type-II diabetes mellitus or non-insulin dependent diabetes mellitus (NIDDM) may result from chronic elevation of serum corticosteroids (Ganong 1995). Although some plant extracts are known to act as antihyperglycaemic and antistressor agents (Andallu and Radhika 2000; Asolkar et al. 1992; Chattopadhya 1993; Gilani et al. 1994; Mungatiwar et al. 1997; Shivarajan and Balchandran 1994; Srivastava et al. 1985; Tripathi and Chaturvedi 1995), their importance in the regulation of serum corticosteroid concentration is not known, despite the fact that chronic stress is a cause for hyperglycaemia. Cortisol, a stress hormone has been reported to increase gluconeogenesis and to decrease the peripheral glucose utilization, very often leading to diabetes mellitus (Ganong 1995). Also, excess endogenous or exogenous cortisol commonly results in an increase in serum glucose concentration and potentially induces diabetes mellitus (Jeffers et al. 1991). Therefore, it can be presumed that a plant extract, inhibiting cortisol secretion may ameliorate hyperglycaemia. Unfortunately, no investigation was made untill today on the regulation of cortisol concentration by any plant extract. Therefore in the present study, an attempt was made to reveal the possible inhibition of cortisol as well as glucose concentrations by ten different plant extracts, if any. Simultaneously, lipidperoxidation (LPO) was studied to rule out the possible toxic effect in the liver, the major target organ of any drug metabolism.

2. Investigations, results and discussion

In this investigation, effects of ten different plant extracts on the alteration in serum cortisol and glucose concentration were evaluated. Simultaneously, hepatic LPO, superoxide dismutase (SOD) and catalase (CAT) activities were also investigated.

Results revealed that the serum concentration of glucose decreased significantly in mice, receiving extracts of *A. marmelos, A. indica, C. pluricaulis, G. sylvestre*, *I. racemosa, B. diffusa* and *O. sanctum*. However, a concomitant decrease in serum cortisol was observed in animals treated with the extracts of the last three plants only.

Neither of the plant extracts enhanced the hepatic LPO, which was rather significantly decreased by the extracts of *A. marmelos, A. sativum, C. pluricaulis. G. sylvestre*, *M. oleifera, O. sanctum* and *W. somnifera* as indicated earlier (Kar and Panda 2003). The activity of SOD and CAT was also enhanced by most of these plant extracts.

From these results it is evident that some plant extracts such as *I. racemosa*, *B. diffusa* and *O. sanctum* are capable of decreasing serum cortisol as well as serum glucose concentrations indicating their possible efficacy in the amelioration of adrenal corticoid induced hyperglycaemia.

While, hypoglycaemic activities of *I. racemosa* and *O. sanctum* had been reported earlier (Chattopadhya 1993; Tripathi and Chaturvedi 1995), nothing is known about their cortisol lowering activity so far. In fact, the present report appears to be the first one on some plant extracts which can decrease both serum cortisol and glucose concentrations. However, the extracts of *A. marmelos*, *A. indica*, *C. pluricaulis* and *G. sylvestre* could not exhibit marked cortisol lowering, but hypoglycaemic activity. Although three of these plants were earlier reported to lower blood glucose concentrations (Asolkar et al. 1992; Srivastava et al. 1985), their mode of action was not ascertained.

No significant alteration in serum cortisol concentration by some plant extracts at least indicates that the hypoglycaemic activity of those plant extracts is not mediated

Table 1: Details of investigated plants indicating their family, parts used, type of extract, yield, dose and route of administration

Plants with family	Part used	Type of extract	Herb: Extract (in parts)	Dose (mg/ kg)	Route of administration	Reference
Aegle marmelos (Rutaceae)	Leaf	Aqueous	6 : 1	166	Oral	(Rao et al. 1995)
Allium sativum (Liliaceae)	Bulb	Alcoholic	5 : 1	500	Oral	(Bordia et al. 1996)
Azadirachta indica (Meliaceae)	Leaf	Aqueous	10 : 1	40	Oral	(Panda and Kar 2000)
Boerhaavia diffusa (Nyctaginaceae)	Root	Aqueous	8 : 1	150	Oral	(Rawat et al. 1997)
Convolvulus pluricaulis (Convolvulaceae)	Root	Alcoholic	10 : 1	16	i.p.	(Panda and Kar 1997)
Gymnema sylvestre (Asclepidaceae)	Leaf	Aqueous	10 : 1	600	Oral	(Shrivastava et al. 1985)
Inula racemosa (Compositae)	Root	Alcoholic	3 : 1	400	Oral	(Tripathi and Chaturvedi 1995)
Moringa oleifera (Moringaceae)	Leaf	Aqueous	8 : 1	175	Oral	(Tahiliani and Kar 1999)
Ocimum sanctum (Labiatae)	Leaf	Aqueous	5 : 1	500	Oral	(Panda and Kar 1998)
Withania somnifera (Solanaceae)	Root	Alcoholic	5 : 1	1400	Oral	(Panda and Kar 1999)

Table 2: Effects of different plant extracts on serum glucose (mg/dl) and cortisol (µg/dl) concentrations and hepatic LPO (nM of MDA formed/h/mg protein), SOD (units/mg protein) and CAT (µM of H_2O_2 decomposed/min/mg protein) activities in male mice

Groups	Glucose	Cortisol	LPO	SOD	CAT
Control	73.28 ±2.44	0.477 ±0.009	0.676 ±0.028	6.08 ±0.22	59.11 ±1.17
Aegle marmelos	55.08*** ±2.13	0.470 ±0.007	0.566** ±0.022	6.15 ±0.13	68.56** ±2.85
Allium sativum	73.23 ±1.17	0.454 ±0.013	0.441*** ±0.022	6.34 ±0.21	65.64*** ±0.89
Azadirachta indica	54.06*** ±2.20	0.460 ±0.003	0.689 ±0.039	6.87* ±0.15	66.33*** ±0.98
Boerhaavia diffusa	63.56* ±3.55	0.400*** ±0.007	0.659 ±0.034	6.87** ±0.13	58.47 ±2.85
Convolvulus pluricaulis	47.65*** ±3.48	0.490 ±0.007	0.554* ±0.039	6.72* ±0.19	66.25* ±2.13
Gymnema sylvestre	49.89*** ±1.15	0.422 ±0.025	0.553* ±0.038	6.74* ±0.09	67.42* ±3.47
Inula racemosa	44.13*** ±3.60	0.360*** ±0.008	0.688 ±0.047	6.90 ±0.39	75.14*** ±1.09
Moringa oleifera	71.11 ±3.46	0.451 ±0.027	0.601* ±0.015	6.65* ±0.11	67.68*** ±0.964
Ocimum sanctum	67.15* ±1.35	0.392*** ±0.008	0.555** ±0.011	6.01 ±0.11	74.93*** ±1.07
Withania somnifera	66.55 ±2.08	0.452 ±0.013	0.556** ±0.017	6.83* ±0.16	78.45*** ±3.01

Data are mean ± S.E.M. (n = 7).*, $P < 0.05$; **, $P < 0.01$ and ***, $P < 0.001$ as compared to the respective control values

through changes in cortisol concentrations. On the other hand *I. racemosa*, *B. diffusa* and *O. sanctum* exhibited both cortisol and glucose lowering ability, suggesting that their effects on blood glucose concentration might have resulted from their cortisol lowering efficacy, as a high level of cortisol is commonly associated with hyperglycaemia (Jeffers et al. 1991).

The decrease in cortisol concentration by the plant extracts could either be due to an inhibition of adrenocorticotrophin (ACTH) secretion in pituitary glands or could be a direct action at the level of the adrenal cortex. Whatever may be the mode of action of these plant extracts, from the present findings it appears that the hypoglycaemic effects of *I. racemosa*, B. *diffusa* and *O. sanctum* are possibly mediated through their cortisol lowering efficacy and therefore these plant extracts may potentially ameliorate corticoid induced hyperglycaemia. The observations on hepatic LPO, SOD and CAT activities also indicate the safe nature of these plant extracts at the present doses. In fact, most of the plants seem to be anti-peroxidative in nature.

3. Experimental

3.1. Plant extracts

In this study, 10 different plants, namely *Aegle marmelos*, *Allium sativum*, *Azadirachta indica*, *Boerhaavia diffusa*, *Convolvulus pluricaulis*, *Gymnema sylvestre*, *Inula racemosa*, *Moringa oleifera*, *Ocimum sanctum* and *Withania somnifera* were considered as they had been reported earlier to exhibit antistressor/hypoglycaemic activity (Andallu and Radhika 2000; Asolkar et al. 1992; Chattopadhya 1993; Gilani et al. 1994; Mungatiwar et al. 1997; Shivarajan and Balchandran 1994; Srivastava et al. 1985; Tripathi and Chaturvedi 1995). Plant extracts were obtained from Amsar Private Limited, Indore (MP), India. Details of the plant extracts including the doses used and route of administration (taken from earlier studies, done in relation to some other functions) are given in Table 1.

3.2. Animals

Swiss albino healthy male mice, around two month old (28 ± 2 g) were obtained from Institute of Animal Health and Veterinary Biologicals, MHOW (MP), India, and were maintained at constant temperature (27 ± 1 °C) and photo schedule (14 h light: 10 h dark) with the provision of food (Golden Feeds, New Delhi, India) *ad libitum* and free access to drinking water.

3.3. Experimental design

The experiment was conducted after an acclimation period of seven days. Animals were divided into eleven groups. Each of group having seven animals each and initial body weight of each one was recorded. Group I receiving 0.1 ml of vehicle served as control. Each experimental animal received its daily dose in the form of 0.1 ml of a specific plant extract as mentioned in Table 1. Extracts were administered every day between 1000 and 1100 h to avoid circadian interferences and the treatment was continued for 15 days. On the last day, after an overnight fast, the mice were weighed and sacrificed by cervical dislocation. Blood samples were collected between 800 and 900 h of the day and clear serum samples were stored at −20 °C, until assayed for glucose and cortisol concentrations.

3.4. Parameters studied

Serum cortisol was estimated by radioimmunoassay (RIA) using a DSL 2000 ^{125}I kit (Diagnostic systems laboratory, USA). The inter-assay and intra-assay coefficient of variation were 6.5 and 7.9% respectively. The lower limit of sensitivity was 0.11 µg/dl at 95% confidence limit. The estimation of serum glucose was done by the modified glucose oxidase method (Hugget and Nixon 1957) using a kit purchased from Qualigens, Mumbai, India. Estimations of hepatic lipid peroxidation (LPO), superoxide dismutase (SOD) and catalase (CAT) activities were done according to protocols followed earlier in our laboratory (Panda and Kar 1998).

3.5. Statistical analysis

The data were analyzed for significance using analysis of variance (ANOVA) followed by Student's t-test. A P value of 5% and less was considered as significant (Senedector and Cochran 1967).

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