REPORT

Effects of *Camellia sinensis* L. (green tea) extract on the body and testicular weight changes in adult Wistar rate

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Abstract: This research was aimed to study the effects of oral administration of *Camellia sinensis* L. on the testicular and body weights of adult Wistar rats for short and long time periods. The adult Wistar rats were divided into 3 groups (A, B and C). Every group had ten rats. Green tea extract 0.692% (w/v) was given to groups A and B on daily basis. The extracts were prepared fresh and given for a period of ten and thirty days, respectively, while distilled water was given to the group C rats only. The adult Wistar rats were sacrificed on eleventh and thirty-first day of experiment for the particular groups. The testes were dissected out cautiously, free from the supporter tissues and weighed to the adjacent 1 mg. There is no significant difference in the body weight in all 3 groups. Moreover, it was observed that Wistar rat's testicular weight was considerably increased in group B but no major changes were seen in group A. Our results indicated that green tea when given for short period of time may be effective to the testes but has no consequence on Wistar rat's body weight. However, it is indistinct if these alterations are reversible.

Keywords: Green tea, drinking effects, Wistar rats, testicular weight.

INTRODUCTION

Several biological properties have been reported in green tea (*Camellia sinensis* L.) via its active constituents, including antioxidant activity (Morel *et al.*, 1933; Guo *et al*., 1996), anti-inflammatory and antiviral activities, tumor cell growth inhibition and chemo prevention of cancer. (Yang *et al.*, 2000), thyroid peroxidase (Divi & Doergo, 1996), certain enzymes inhibition for instance aromatase (Satho *et al.*, 2002; Godin & Rosegren, 2003), and angiotensin converting enzymes (Actis-Goretta *et al.*, 2006). Dry leaves of *Camellia sinensis* contain predominantly flavanols, polyphenols (30 to 36%), more generally recognized as catechins (Ahmed & Mukhtar, 1999). The principle catechins are epicatechin (EC), apicatechin-3-gallate (ECG), epigallocatechin (ECG) and epigallocatechins-3-gallate (EGCG).

The catechin's effects have been described on the male reproductive system. Laboratory and epidemiological research disclosed a relationship between androgens that the risk of prostate cancer can altered by diet. (Ripple, *et al*., 1997; Clinton & Giovannucci, 1998; Parkin, 2001).

In athymic mice, breast tumor growth and human prostate's tumor can suppress by parenteral injection of EGCG has been described (Liao *et al*., 1995) and the circulating levels of testosterone and leutinizing hormone (LH) reduction in the intact rat (Kao *et al*., 2000). While the catechin's anti-gonadotropic effect is described as a minor issue of EGCG on nutritional regime (Kao *et al*., 2000) or on activity of aromatase (Satho *et al*., 2002; Godin & Rosegren, 2003), at the gonadal level a modulator function might be present.

Herbal medicine treatment is gaining importance globally. Prescription drugs have less approval than traditional herbs in various cultures with rising epidemics of chubbiness. This was typically featured to being protected than drugs, from the patient's point of view. Moreover patients believe that for using herbal medication there is no need for a doctor and it perhaps an appropriate effort to compensate for remedy failure in managing fatness. Some of these alternatives were chitosan (binding resin) and a green tea, which can lowered fat absorptive, leading to precipitate lipids (Herber, 2003). Different studies

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showed that black and green tea contains flavonoids such as myricetin, quercetin and kaempferol, which are all powerful anti-carcinogenics (Herlog *et al*., 1993). The risk of skin, lung, stomach and colon tumors also reduced by flavonoids (Challa *et al.*, 1997).

The extract of turmeric used in a group of herbs as a diabetic cure stimulated fatty liver and confirmed a considerable phospholipid-content, triglyceride and total cholesterol reduction in diabetic rats (Saravanan & Pari, 2005). Leaves of green tea are used locally for preparation and believe as refreshment and for the treatment of nausea, the *Mentha royleana* (Benth) dried leaves are mixed with green tea are taken. Therefore, the present study was undertaken to observe the green tea extract's effects on the adult wistar rat's body and testicular weights for long and short time-period.

MATERIALS AND METHODS

Location and duration of study

The study was conducted at the Histology Laboratory of the Baqai Medical University, Karachi, Pakistan. The preliminary studies, animal acclimatization, drug procurement, actual animal experiment and evaluation of results lasted for a period of 30 to 45 days.

Animals

For this experiment, thirty mature individual wistar rats weighing between 200-250gms were obtained and maintained in the animal dwelling of the Baqai Medical University, Karachi. They were split up into 3 groups A, B and C. Every group had ten rats. Group A and B were the treatment groups, while group C considered as control group. Each group was kept in separate cage and fed with routine diet. At the beginning of the study, rats were allowed to settle down for 7 days. Ethical acceptance from Baqai Medical University was taken as per policy of research and ethical committee of the university.

Drug preparation and administration

Camellia sinensis L. extract was prepared according to Wang *et al.* (1992) with minor modification. Dry *Camellia sinensis* L. leaves (5g) were immersed for five minutes in 100 ml of hot saline (90°C). It was cooled to room temperature and then sieved. The resultant clear solution is like to tea brews used by human beings. The residue (solid matter) of this mixture was determined by 10 ml of drying samples at 100°C in an oven for overnight and the dehydrated solid matter obtained was then weighed. The dry mass of green tea leaves was detected to be 0.0692gm for 0.005 kg, the concentration production of this mixture is 0.692% (w/v). On a daily basis, the extracts were prepared freshly. This preparation consist of around EGCG represents $\geq$50%, 0.4% theobromine, 8.0% caffeine, and 27% catechins, of total catechins (19). The green tea (*Camellia sinensis)* was given to group A as drinking liquid about 2% aqueous extract, at the same time with the animal dwelling drinking water (Challa *et al.*, 1997) for 10days and the same extract was given to group B for the period of 1 month, while the Group C was considered as the control.

Body and testicular weights determination

After the final dose (24hour), the rats were weighed and dissected out by cervical displacement on the 11th day for group A and 31st day for group B after the particular treatment. The testes were dissected out cautiously, free from the supporter tissues, and on Sartorius electro balance, weighed up to the adjacent 0.001g.

STATISTICAL ANALYSIS

The testicular and body weight's data were stated as the mean $\pm$SD. The Student's T-test was used for relationship among the treated groups and control. Significant value was considered statistically for differences with values are less than 0.05 (Mahajan, 1997).

RESULTS

General findings

The rats of all groups were not shown any gross dissimilarity after completion of experimental procedure. There were not recorded any anticipated killings. The animal's behavior was found similar in all three groups i.e., A, B and C.

Effect of green tea on body weight

Mean body weight of the control and green-tea-treated rat's are given in table 1. The same increase in body weight had evident after mean body mass comparison of the control and treated rats before and after treatment in both the short and long term treated groups. A 25% increase in rat's mean body weight was observed in comparison to preliminary weight.

Effect of green tea on testicular weight

The data obtained from the controlled and green tea treated rat's mean testicular weight are shown in table 1. The mean testicular weight of rats was significantly increased in groups C and B but there was no significant difference in groups C and A by using T-test analysis technique.

DISCUSSION

The present investigation showed that there were no variations among the control and treated rats on gross/morphological changes. There was a gain in the body weight of controlled rats but no effect on somatic growth was observed in both treated groups after oral administration of green tea for shorter and longer periods. Same findings were observed by Rosário *et al*., (2008) in

Table 1: Effect of Green Tea on the body and testicular weight of rats following oral administration for shorter and longer periods of time

Group	Body weight (g)			Testicular weight (g)
	Initial	Final	% Increase	
A	200.00 ± SD	250.00 ± SD	25	0.606±0.365
B	201.00 ± SD	249.00 ± SD	23.88	0.780±0.101
C	202.00 ± SD	251.00 ± SD	24.25	0.522±0.117

Values are expressed as mean ± SD; obviously different statistically from the control at $p< 0.05$, T-test. Data is based on 10 replicates.

which wistar rat's body weight showed no significant increase in body weight after consumption of green tea.

A considerable increase in mean testicular weight was observed among the control and the longer period treated rats but no variation was seen in mean testicular weight in the short period treated group of rats when green tea was administered. In fact, a decrease or increase in absolute or relative weight of an organ after administrating a drug or chemical is a sign of the lethal consequence of that chemical (Simon *et al*., 1995; Maina *et al*., 2008). Moreover it generally gives a positive reproductive risk evaluation by the weights of male reproductive organs in experimental studies (Raji *et al*., 2005) and the germinal elements and tubules accounts for about 98 percent of the testicular mass while testicular size for spermatogenesis, is the primary evaluation (Sherines and Howards, 1978). The present study on the basis of data obtained indicated that no changes were seen in testicular weight in control animals and in rats when green tea was administrated for shorter period of time. However, when given for longer period significant increase in mean testicular weight was noticed. Therefore, both the control and short time treated rats were not shown any change in testicular weight in the testis proposed that the green tea administration had no lethal consequence on this organ, but the long time treated rats showed increase in testicular weight indicated that the green tea may have lethal consequence on the testis during the longer period.

There are studies that the herbal tea mixture administration including Foliumnelumbinus, Semen cassia, Radix polygoni multiflori, Gynostemma pentaphyllum and testicular oedema was caused by Green tea which has been associated to aspects for instance testosterone (Maddocks & Sharpe, 1989), vascular abnormality and direct consequence of GnRH agonists, (Bergh *et al*., 1988), Human Chorionic endogenous LH or Gonadotrophin (hCG) (Bergh *et al*., 1990), lymphatic obstruction, inflammation and salt retention (Robbins & Cotran, 2004). The Sertoli cells are produced Seminiferous tubule fluid, and re absorption by efferent ductules is regulated its volume (Turner *et al*., 1984). It has also been revealed that fluid of testicle exist in 2 partitions, the interstitium and the seminiferous tubules (Setchell *et al*., 1994). From the testicular vasculature, chiefly Interstitial fluid initiates and lymphatic drainage, physiological osmotic pressure differences, vascular permeability, changes in blood flow is influenced its volume (Bergh *et al*., 1988). It has been shown that after radiation, fluid production of testicular interstitial is increased (Laporte *et al*., 1985) and the use of the drugs such as procarbazine (Delic *et al*., 1986) dibromochloropropane (DBCP) (Kluwe *et al*., 1983) and busulfan (Udagawa *et al*., 2001) increases volume. Correspondingly it has been determined that the germinal epithelium cells sloughed the efferent duct's obstruction or the ducts themselves can lead to increase in weight of testis because of accumulation of fluid (Hess *et al*., 1991; Nakai *et al*., 1993), result that might equalize the germinal epithelium depletion effects on testis weight.

Usually, green tea induces its antiviral and anti-inflammatory activity leading to peroxidation of lipid (Yang *et al*., 2000) oxygen free radicals, alkylating generation (Morel *et al*., 1993). The accumulation of lipid peroxides is lethal to the leading to alteration in disintegration of cellular organelles, membrane structure, and permeability (Muler & Ohnesorge, 1982). On the basis of these reports, it is assumed that use of the green tea stimulates the increase of testicular weight when green tea was used on long-term basis. This finding is almost inline with the previous study who also claimed that green tea consumption for longer period tends to increase in testicular weight.

Green tea protects against stomach cancer by inactivating the bacterium *Helicobacter pylori*. It also protects against oesophageal cancer by destroying cancer-causing nitrosamines (Murray, 2000). In addition to significant anti-cancer and antibacterial properties, catechins, the chemicals green tea contains, lower cholesterol levels in the blood and improve lipid metabolism (Weil, 1997).

Increased use of green tea is reported to decrease blood levels of total cholesterol and triglycerides and butter HDL: LDL ratio. The persons who took more than ten cups of green tea had lower amounts of hepatological markers in their blood, alanine transferase, ferritin and aspartate aminotransferase. It may be protective against cardiovascular disease (Imai, 1997).

The testes damage can be identified as a change in weight only after longer period dose than those wanted to

generate considerable effect in other gonadal condition evaluation (Berndtson, 1977; Foote *et al*., 1986; Ku *et al*., 1993). The findings drawn from the present study regarding the changes in testicular weight of the animal concluded that administration of green tea had no harmful effects in testicular weight and structure in the control and treated rats probably due to the nature of chemicals exist in this herbal material or when given for a shorter period of time.

Our study recommends that green tea when given for shorter period had no effect on testicular weight but if administered for longer period, it caused a considerable increase in weight of testicle. It is yet doubtful, if these alterations are reversible. Further studies should be undertaken in humans to ensure green tea's metabolic effects.

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