INTRODUCTION

The androgenic anabolic steroids that occur naturally are synthesized in the testis, ovary and adrenal gland from cholesterol via pregnenolone and bind to receptors that are specifically occurring in reproductive tissues, muscles and fat bodies (Mooradian et al., 1987). Synthetic anabolic steroids are mainly related to the principal male hormone testosterone (Haynes and Murad, 1985). Below is a brief description of the androgenic anabolic steroids studied for their potential genotoxic effects and the natural plant products (antioxidants) used to ameliorate their genotoxic effects on chromosomes.

Androgenic Anabolic Steroids

(i) Stanozolol

Stanozolol is a synthetic steroid similar to the naturally occurring androgen called testosterone. It is used in the treatment of many disorders such as anemia and hereditary angioedema (Gannon, 1994). Athletes and bodybuilders commonly use this anabolic steroid for performance enhancement (Elashoff et al., 1991).

![Stanozolol structure](image)

Its large oral bioavailability is due to a C17 alpha-alkylation principle which allows the hormone to survive the first pass through liver metabolism. At high dosage, Stanozolol could exert a proliferative effect on liver cells (Boada et al., 1999). Precocious prostate cancer has been reported after a
long term steroid abuse (Roberts and Essenhigh, 1986). Hepatic cancer has also been linked to anabolic steroidal abuse (Overly et al., 1984).

Due to reports linking prostate and liver cancers to high steroid dosage and long term steroid abuse, stanozolol was studied for its genotoxic effect on human lymphocyte chromosomes using chromosomal aberrations and sister chromatid exchanges, both in the presence and absence of metabolic activation, and with and without NADP.

(ii) Trenbolone

This is also a synthetic steroid used frequently by veterinarians on livestock as a promoter of growth in animal husbandry (Richold, 1988). Trenbolone compounds have not yet been approved by the Food and Drug Administration, USA for use by humans due to their considerable negative side effects, although bodybuilders use the drug illegally to increase body mass and strength. Cases of prostate and hepatic cancers have been associated with long term anabolic steroid abuse (Roberts and Essenhigh, 1986; Overly et al., 1984). Trenbolone compounds increase nitrogen uptake by muscles after metabolization, leading to increased rate of protein synthesis (http://en.wikipedia.org/wiki/trenbolone, 2007).

![Trenbolone molecule](Image)

Prostate and hepatic cancers associated with anabolic steroid abuse has led to the study of Trenbolone at different doses for genotoxicity in cultured human lymphocytes using chromosomal aberrations and sister
chromatid exchanges as parameters, in the presence as well as absence of metabolic activation system, with and without NADP.

(iii) Norethandrolone

This is an oral anabolic androgenic steroid (AAS) synthesized and approved under the brand name of Nilevar and used in bodybuilding. It is used in the treatment of hormonal diseases for promoting muscle regrowth in patients. It has moderate progestagenic activity and shows liver toxicity as a 17-alkylated steroid (IPCS, 1993).

![Norethandrolone structure](image)

The International Agency on Cancer (IAC), on the basis of epidemiological studies, classifies androgenic anabolic steroids mainly as probably carcinogenic (Group 2A) (Martelli et al., 2003). Hepatic and prostate cancers have been reported after long term steroidal abuse (Roberts and Essenhigh, 1986; Overly et al., 1984). Keeping in view the above reports, Norethandrolone was studied for its genotoxicity using chromosomal aberrations and sister chromatid exchanges as parameters in human lymphocytes in vitro.

(iv) Oxandrolone

It is also an oral anabolic androgenic steroid, synthesized, approved and sold as Anavar and used in bodybuilding (IPCS, 1993). It has moderate progestogenic activity and shares liver toxicity issues common to 17-alkylated steroids. It is used in the treatment of anemia and hereditary angioedema.
(Gannon, 1994) and also prescribed for promoting muscle regrowth in the patients suffering from involuntary weight loss in treating osteoporosis.

![Testosterone propionate structure](attachment:structure.png)

Cases of hepatic and precocious prostate cancers have been linked to long term AS abuse (Roberts and Essenhigh, 1986; Overly et al., 1984). Such reports linking cancers to AAS abuse has led to our study of oxandrolone for its genotoxic effect on cultured human lymphocytes, applying the chromosomal aberrations and sister chromatid exchanges as parameters.

**(v) Testosterone propionate**

It is a very strong and androgenic compound having a high anabolic effect. Its users gain strength rapidly without increase in the body weight. Precocious prostate cancer and liver cancer have been associated with long term anabolic steroid abuse (Roberts and Essenhigh, 1986; Overly et al., 1984). It is used for treating hormonal diseases, for boosting muscle regrowth in the patients, as also in bodybuilding (IPCS, 1993).
The drug TP has been shown to induce cell transformation in Syrian Hamster Embryo (SHE) cells (Tsutsui et al., 1995). The TP treatment for a long duration leads to biomyosarcomas in the vas deferens or uterus of Golden Syrian Hamsters (Hudson et al., 1998). This drug has an enhancing effect on phalloidin-induced liver toxicity in mice (Muraoka et al., 1988). TP also has a direct toxic effect in treating cell cultures neonatally in rat (Welder et al., 1995). Such reports of TP toxicity has led to the study of its genotoxic potential using cultured human lymphocytes in the presence as well as absence of metabolic activation system, with or without NADP, applying CAs and SCEs as parameters.

Other similar estrogens and synthetic progestins among steroids have been reported to be genotoxic in human lymphocytes in vitro and in mouse bone marrow cells (Dhillon et al., 1994; Hundal et al., 1997; Joosten et al., 2004; Siddique and Afzal, 2004a,b,c,d; Siddique and Afzal, 2005a; Siddique et al., 2005a,b; Siddique et al. 2006a).

The genotoxic effect of steroids can be ameliorated by the use of antioxidants and natural plant products (Ahmad et al., 2004; Siddique and Afzal, 2005b; Siddique et al., 2005b; Siddique et al., 2006b; Siddique et al., 2007 a,b,c; Beg et al., 2007; Siddique et al., 2008a,b).

Natural Plant Products

(i) Gingerol

It is a key constituent of the plant ginger (Zingiber officinale) and is chiefly responsible for the pungency of ginger (Hasenohrl et al., 1998). It shows antioxidant activities through scavenging of superoxide and hydroxyl radicals and by inhibiting lipid peroxidation (Kikuzaki and Nakatani, 1993). The compound 6-gingerol is the major pharmacologically active component inducing apoptosis in cancer cells (Lee and Surh, 1998; Lee et al., 1998).
Gingerol was studied for its antioxidant activity in different cases of genotoxicity induced by methylmethanesulphonate (MMS), norethandrolone and oxandrolone, in cultured human lymphocytes in the presence as well as absence of metabolic activation system (S9 mix) on frequencies of CAs and SCEs.

(ii) Genistein

It is an anticarcinogenic polyphenol found in soybean seeds as glycosides. It has antioxidant properties and is involved in modulation of cell proliferation and transformation (Muster et al., 1997). It is effective in quenching free radicals produced by toxic agents and protects cells against oxidative damage especially with respect to DNA (Foti et al., 2005; Lee et al., 2000).

Epidemiological and animal model studies have shown a link between a soy-rich diet and reduced incidence of breast and prostate cancers (Dixon and Ferreira, 2002). Several studies have suggested that genistein has a
protective effect against lipid peroxidation of low and high density lipoproteins (Patel et al., 2001; Ferretti et al., 2004). In the present case, the antigenotoxic effect of geinstein was studied on the frequency of CAs and SCEs induced by MMS (in the absence of metabolic activation) and cyclophosphamide (in the presence of metabolic activation) in human lymphocytes in vitro.

(iii) Epicatechin gallate (ECG)

It is a catechin and polyphenolic antioxidant plant metabolite found in abundance in green, black, and oolong tea, derived from the tea plant *Camellia sinensis* (Balentine et al., 1998). Catechins have been studied for their health benefits in humans as well as the animals.

![ECG structure](image)

Reduction in atherosclerotic plaques (Chyu et al., 2004) and in carcinogenesis was seen in vitro (Mittal et al., 2004) and also in vivo. ECG was studied for its antioxidant effect on the CAs and SCEs induced by TP, both in the presence and absence of metabolic activation system in human lymphocytes in vitro.

(iv) Epigallocatechin-3-gallate (EGCG)

Epigallocatechin-3-gallate (EGCG), a compound closely related to ECG, is also a catechin and polyphenolic antioxidant plant metabolite found in
abundance in various types of tea, derived from the tea plant *Camellia sinensis* (Balentine et al., 1998).

![Chemical structure of EGCG](image)

It helps protect the skin from ultraviolet radiation-induced genotoxic damage and tumor formation (Katiyar et al., 2007). EGCG was studied for its antimutagenic effect on the CAs and SCEs induced first by MMS and CP and then, in the next experiment, by Stanozolol and Trenbolone, both in the presence and absence of metabolic activation system in human lymphocytes in vitro.