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Antimutagenic effects of the ginger extract of reducing chromosomal mutation induced by cyclophosphamide in male rats

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A B S T R A C T

The drug cyclophosphamide (CP) has harmful genetic effects when used with a dose of 5 mg/kg body mass. For this reason, this study was carried out to determine these harmful effects and try to reduce these effects by using the metabolic extract of ginger. Chemotherapeutic treatments are linked to a variety of undesirable outcomes, the most dangerous of which are mutagenesis acts. To measure the possible anti-mutagenic and ginger extract's ability to prevent chromosomal abnormality in bone marrow cells. Four groups, each with seven animals, were used in the experiment. The first group, the control group, consisted of negative control rats fed a regular meal and purified water. In contrast, the second group was given oral cyclophosphamide at a daily amount of 5 mg/kg body mass for four weeks. The third group (the test group), for the duration of four weeks, received 200 mg of ginger extract per kilogram of body weight every day. The fourth group, the protective group, received two phases of treatment: first, 200 mg/kg body mass of ginger extract was given, and then, two hours later, cp (5 mg/kg body mass daily) was received verbally for four weeks. In the CP group, notable changes existed in the chromosome genes of the somatic and mitotic index and an increased frequency of chromosomal changes in terms of structure and number. These results describe the chromosomal effects of CP free radicals. Chromosome aberration is reduced in comparison to the test group when ginger extract is used; chromosomes in bone marrow cells are negatively impacted by CP. Chromosomal studies in bone marrow cells of rats indicate that the CP at 5mg/Kg causes an increase in the level of chromosomal aberrations. These aberrations include ring chromosomes, breaks in the chromosomes and chromatids and in the centromere region, and other aberrations. However, when the ginger extract was given before the CP, the level of chromosomal aberration defects was reduced in comparison with the positive control. The current study concludes that the ginger extract had an anti-mutagenic effect when given at the dose of 200g /Kg before the CP. It can be used as a protective and therapeutic treatment agent since it has a preventive effect against the mutagenic effects of CP.

1 Introduction

Nowadays, oncogenes and tumour suppressor genes are altered by chromosomal Alterations and related events of somatic cells contribute to cancer development in both people and laboratory animals (Paithankar *et al.,* 2014).Aneugenicity,also known as aneuploidy induction, describes the actions of substances that cause a shift (loss or gain) in the number of chromosomes in cells, leaving those cells without an precise multiple of the haploid amount,(Vinardell.2014).After exposure to the test substance in cell cultures, the test substance is introduced to the cultures in addition to and in place of metabolic activation at predefined intervals. After being treated with a chemical that arrests metaphase such as(Colcemid or colchicine), the cells are extracted, stained, and examined under a microscope to check for chromosome abnormalities according to the internationally recognized guidelines in practical genotoxicity for safety assessment.Thein vitro Mammalian Chromosome Aberration Test (IVMCAT) and Cell Gene Mutation Assay are typically employed to determine the clastogenicity and mutagenicity of test agents (EFSA Scientific Committee 2011).A broad range of alterations in chromosomal integrity can be found using *in vitro and in vivo* methods that quantify chromosomal abnormalities in metaphase cells, (Paithankar *et al.,* 2014),finding substances that result in structural chromosomal aberrations in cultivated mammalian cells is the aim of the in vitro chromosome aberration test. There are two types of structural aberrations: chromatid and chromosomal. Based on the numerous hereditary disorders in humans that are caused by chromosomal mutations and related events, "mutagenicity" describes the induction of long-lasting, transmissible alterations in the quantity or composition of a cell's or organism's genetic material, these alterations may affect one gene, a block of genes, a gene segment, or a set of chromosomes, according to the (EFSA Scientific Committee 2011).The agents responsible for structural chromosomal aberrations are referred to as clastogens, Chromosomal breaks, resulting in Chromosome segment loss or restructuring, can be result from a clastogen, assays designed to identify micronuclei or chromosomal abnormalities are suitable for clastogen detection, (Guideline, 2011).

Among the alkylating medications that interfere through nucleic acid transcription and translation is cyclophosphamide, because it damages DNA over time, it also has an impact on cell division, (Davidson *et al.*, 2003).CP is applied to the treatment of cancerous illnesses like leukaemia, lymphoma, myloma, because of its ability to cause various cancers, cyclophosphamide ranked as one of the most harmful substances to human health.This medication is marketed under several trade names, including Cytoxan, Indoxan, and Procytox, (Jing *et al.,* 2005).

Cyclophosphamide is frequently used in immunosuppressive diseases and cancer treatment in both the young and elderly, (Al-Niwehee, 2019); (Floyd *et al.,* 2005); (Avendano, & Menendez,2008). By altering DNA purine base cross-linking, CP prevents the production of DNA, RNA, and proteins and the demise of cells that divide quickly, (Wetzels, 2004).Numerous diseases are known to be influenced by free radicals and the lipid peroxides they produce (Ayhanci *et al.*, 2019).Free radicals can lead to chromosomal abnormalities and DNA fragments, (Mott *et al.,* 2021). In many cultivated cells, cyclophosphamide causes sister chromatid exchanges in the metabolic activity and chromosome abnormalities, micronuclei, and gene mutations. Additionally, in rats, Chinese hamsters, and mice. It could lead to in chromosomal harm and micronuclei (Suchitra et al. 2011); (Paithankar *et al.,* 2014). CP controls the immune system by stimulating Th17 cell development,(Viaud *et al.,* 2013).Numerous investigations revealed that anticancer

medications produced chromosomal abnormalities in bone marrow cells (de Oliveira et al. 2020); (Kour et al. 2017).During the past few years, the community of scientists has come to progressively endorse the notion that such meal is capable of being medicine; Diets heavy in plant-based foods and light in prepared meals can help stop or lessen disease,(Chabr *et al.,* 2014);(Visalberghi *et al.,*2021);(Del Carmen *et al.,* 2015).Thus, further research must be done in order to find a possible natural material that can both lessen the toxicity associated with chemotherapy and provide protection against CYP-induced oxidative.(Kaabo& El-Sagheer, 2022).

Zingiber officinale, known as ginger, belongs to the Zingiberaceae family and is herb that has a strong, fragrant flavour. The roots of ginger serve as flavouring in food and drinks and in traditional medicine to cure rheumatism, bronchitis, waist discomfort, and fever. According to (Stoilova, *et al.,*2003), Chinese medicine makes use of ginger rhizomes to treat stomachaches and as a stimulant or tonic when macerated in alcohol. According to reports, plants in this family contain antihypoglycemic, anti-inflammatory, antiulcerating, antioxidant, and antimicrobial properties. The ginger rhizome (Zingiber officinale R., Zingiberaceae family) is a widely used spice. Ginger has both androgenic and antioxidant properties in animal studies (Phan et al. 2005). Vitamins such as B3, B6, volatile oils, acids, resins, choline, folic acid, inositol,gingerol,, pantothenic acid, and sesquiterpene are among the many substances it contains, as well as elements similar k, Mg, Ca, P, K, (Morakinyo *et al.,* 2010).Ginger oil has demonstrated a dominant defensive defending consequence to prevent harm to DNA caused thru H2O2, and it may be utilized as an oxygen radical scavenger and antioxidant (Phan *et al.,* 2005); (Zahedi *et al.,* 2010).and various antioxidants, including flavonoids, flavone glycosides, rutin, alkaloids, terpenoids, and beta-carotene,(Ghasemzadeh *et al.,* 2010).It was shown that phenolic compounds may both stop lipid peroxidation brought on by active oxygen species and scavenge superoxide anion like hydroxyl radicals or superoxide anion. Strong antioxidants like ginger can either prevent or reduce the production of unrestricted radicals.

The goal of the research

1.To assess the mutagenic effects of cyclophosphamide on the abnormality of chromosomes in bone marrow cells

2.To estimate the potential anti-mutagenic and the protection action of ginger extract on chromosomal abnormality in bone marrow cells.

2 Materials and Methods

The current research was completed at the zoology department, faculty of science, Omar Al Mukhtar University**.**

Chemicals: The medication cyclophosphamide was supplied as a 50 mg tablet by Baxter Corp; the medication was given orally to the animal after being dissolved in ultrapure distilled water.

Colchicine drug :Colchicine is a prescription drug used to treat familial Mediterranean fever and gout. Colchicine has been prohibited from being injected intravenously due to its hazardous effects, yet oral administration of the drug is still common(Wang *et al.,* 2019).The chromosomes shrink due to prolonged exposure to colcemid or high-concentration usage, which raises the proportion of chromosomes at late metaphase. A brief exposure to a high colcemid concentration decreases the total yield of metaphases. A balance between these variables is achieved via optimal exposure, (Lee *et al.,* 1990), was used in the present study because of its ability to arrest cells at the metaphase stage of the cell's replication cycle (mitosis) (Tijio &Whang, 1962), one tablet dissolves in normal saline

Giemsa stain-Methanol -Ethanol-Hypotonic solution-Glacial acetic acid -Sodium chloride -potassium chloride -Puffer phosphate

Hypotonic solution: For 1000ml distilled water 5.6gm potassium chloride
Fixation solution:(glacial acetic acid:Methanol = 1:3),(Paithankar *et al.,* 2014)
Plant material: Ginger rhizome

Experimental Animals

The animal house yielded twenty-eight adult male albino rats for usage in Tobruk City. After that, the rats were moved to the home for animals, at zoology department, University of Omar Al Mukhtar, their weights, ranging from 200- 300 gm, we used to carry out the current work, wood shavings were used for bedding, which will be altered two times minimum a week for the duration of the trial. The rats were placed on a particular diet and given an adaption time of two weeks. Every day, fresh supplies of food and water were provided to the rats. Following the animals' acclimation, they were split up into four primary groups and kept in four cages, each holding seven rats. The rats were chosen at random and have their tails painted to allow for individual identification. After the rats are weighed, the dosage is determined based on their body weight and they are then separated into four groups of seven animals, separately at random, as coming:

Group 1: For four weeks, the only food given to the negative control rats was the regular diet and distilled water.

Group 2: rat got cyclophosphamide at an amount of 5 mg/kg body mass daily by oral for 4 weeks, (Wtwt *et al.,* 2015)

Group 3: rat given ginger extract at a 200-mg dosage /kg body mass,(Badawy *et al.,* 2019);(El-Borm *et al.,* 2023), daily orally for 4 weeks

Group 4:protective group, this group will divide into two steps; given that ginger extract at an amount of 200 mg/kg body weight primarily followed by ginger extract then given cyclophosphamide 2 hours later at an amount of 5 mg/kg body mass every day by oral for 4 weeks.

Table 1: Experimental design

Groups	Types of treatment	Dose	Period of treatment
G	Ginger	200mg/Kg b.w	4 weeks
Control	distilled water and standard diet	2ml	4 weeks
Treatment	Cyclophosphamide	5 mg/Kg b.w	4 weeks
Protective	G then Cyclophosphamide	200 mg/Kg b.w then 5 mg/Kg b.w	4 weeks

Preparation of Ginger extract

The gathered plants was properly cleaned with tap water, then cleaned with distilled water, dried, and ground, adding 200 grams of ginger powder were added to 400 millilitres of ethanol, at a concentration of 95% for maceration. In an opaque bottle, it is hermetically sealed and rotated for 9 hours with continuous shaking every 6 hours. Solvent will be removed from samples by rotary evaporator then filtered and done, kept in the refrigerator for use (Abdul-hanif *et al.,* 2005); (Ajith *et al.,* 2007).

Methods

Chromosomal preparation of bone marrow

The preparation of chromosomes of cells from the bone marrow followed a method (Shubber & Juma, 1999); (Evans et al. 1964) with some modification, three hours before killing the animal, it was injected with 1 ml of colchicine at a concentration of (0.1) mg / ml) into the chlorine cavity Intraperitoneally. And three hours after the injection, the animal was killed. Fixing the animal

on its back above the dissection plate, the lower limbs were washed with an amount of ethyl alcohol 70%.The skin was cut at the thigh area, the thigh muscles were cut, and then the femur was held with forceps from the middle area, and its attachment to the joints was cut. The bone outside the animal was cleaned of muscle residues, and the bone cavity was made. A potassium chloride solution of 8 ml (Hypotonic solution) was placed in a tube and located in a financial bath at 37 °C for a quarter of an hour, then the centrifugation process was conducted for 10 minutes (1000) revolutions per minute in the centrifuge, we used the same procedures as we did with 8ml, but with 4ml, then repeat the same step repeated 2tims(Paithankar et al. 2014). And then the clear was separated from the precipitate, and the precipitate was treated with (3:1 methanol: glacial acetic acid in an amount of twice the quantity, 6 ml of the fixation solution), was placed, and the centrifugation process was conducted again for 10 minutes (1000) cycle's minutes. Then separating the clear liquid and putting 2 ml of the fixation adapter again, followed by another round of centrifugation for 10 minutes at 1000rpm. the precipitate was treated again ,and the cells were dropped onto clean slides from a distance of approximately 60 cm until the cells ruptured and the chromosomes were released . The slides were left for 24 hours (Evans et al., 1964). Then, the slides were stained with Giemsa , and microscopic examinations was conducted using a 1600X magnification lens (Tijio & Whang, 1962).

$$MitoticIndex(MI) = \frac{\text{Mitotic division cells}}{\text{Total No of cells scored}} \text{x}100$$,(Tijio& Whang, 1962).

Statistical Analysis

With Minitab software (version 22.0), statistical analyses were performed, and a difference was deemed significant if P <0.05, the three replicates' mean ± standard deviation is presented as the result. One-way ANOVA with a significance level of P <0.05 and Tukey's test were used to assess statistically significant differences in the experiments conducted in the different tests.

3 Results

The average mitotic index (MI) of male rats' bone marrow cells after CP (5 mg/kg) treatment

Table (2) displays the average Mitotic index (M1) of bone marrow cells of the rats subjected to cyclophosphamide at an amount of 5 mg/kg body mass daily by oral for 4 weeks. It shows a significant difference in the value of (MI) when comparing group 3 (group treated with ginger at an amount of 200 mg/kg body mass daily by oral for 4 weeks) or group 4 (group given ginger with a 200 mg/kg dosage then cyclophosphamide treatment at an amount of 5 mg/kg) with the group treated by cyclophosphamide at an amount of 5 mg/kg. The value of the difference among the levels reached 12.0000 at a significant level (p= 0.001), while the differences among the control and cyclophosphamide group were no significant.

Table 2: Mean of MI of bone marrow cells of male rats in all groups

Groups	N	Mean ± SD
G (200 mg/kg)	35	39.11±11.46[a]
CP (5 mg/kg)	30	27.14±11.62[b]
N.C	15	29.23±15.18[ab]
G 200 + CP 5(mg/kg)	25	34.33±13.03[ab]

(N.C): Negative Control group. (G) : Ginger group. (CP) :cyclophosphamide group.

N: Number slides. SD: Standard Deviation.

The significance level is usually denoted as P. P-value < 0.05 means the difference between groups is statistically significant. In table, superscript letters like a, b, and ab indicate significance levels between groups. Group G (a) is significantly different from Group CP (b). Group G 200 + CP 5 (ab) is not significantly different from either G or CP because it shares letters with both.

ANOVA of MI of bone marrow cells from the experiments male rats

The significant differences of mitotic index of bone marrow cells of rats between groups were measured by detecting their mean differences using the ANOVA tests and P value < 0.05 was regarded to be significant. For each group, the variance analysis, significant differences (F = 5.56, p = 0.001) between means of the value of (MI) was very high as presented in (Table 3). The interval plot of (MI) mean of bone marrow cells of the rats of each group were calculated using individual standard deviations (Figure 1).

Table 3: ANOVA of MI of bone marrow cells of the experiments male rats

Source	DF	AdjSS	AdjMS	F-Value	P-Value
Groups	3	2590	863.3	5.56	0.001
Error	101	15677	155.2		
Total	104	18267			

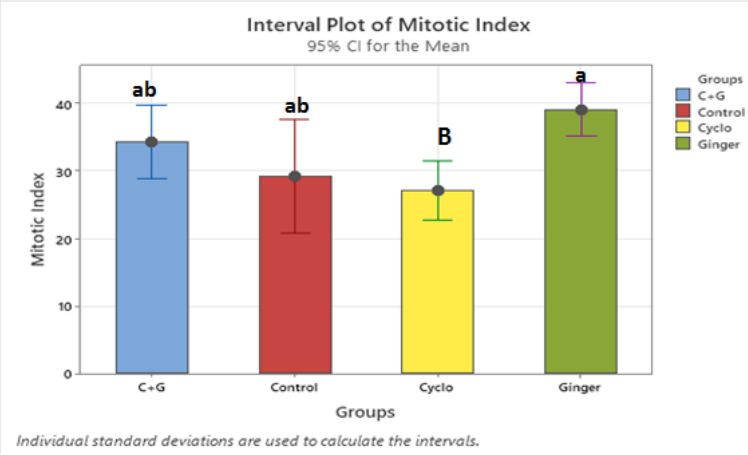

Figure1: MI in the male rats' bone marrow cells in all groups

-The error bars represent the 95% Confidence Intervals for the mean mitotic index in each group.

-The letters above the bars (a, b, ab, B) represent statistical groupings:

-Groups sharing at least one letter are not significantly different from each other.

-Groups with no letters in common are significantly different.

Ginger (a) has the highest mitotic index and is significantly higher than Cyclo (B), which has the lowest index.

C+G (ab) and Control (ab) are not significantly different from each other or from Ginger or Cyclo , indicating their mitotic index falls in an intermediate range.

Cyclo (B) is statistically distinct from Ginger (a), suggesting a suppressive effect on mitosis.

Male rat bone marrow cells with aberrant chromosomes following CP therapy (5 mg/kg)

The results which gained from this study to evaluate the types of chromosome mutations are given in (Figure 2) which show several kinds of

chromosome abnormalities comparing to negative control male rats.

Control group:

Generally, the chromosome of the rat is simple rod-type in germ cells and somatic cells, both cells contain some smaller chromosomes carrying a constriction in each at its middle portion or near its extremity. The chromosome numbers of the albino rats (Rattus norvegicus) is 42 in diploid and 21 in haploid in the germ cells, figure (2) present the typical chromosomes form of the rats in this study comparing to CP group.

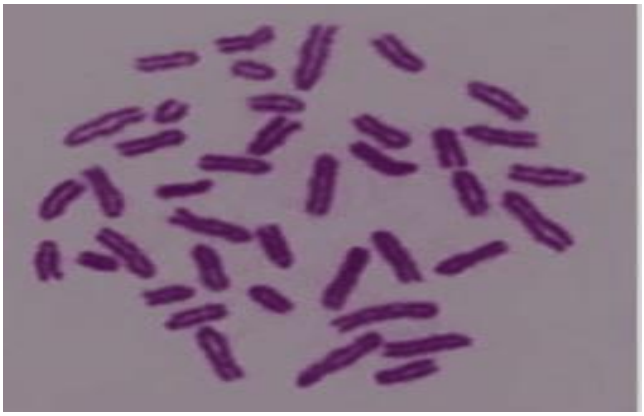

Figure 2: Rat bone marrow cells with normal chromosomes (Giemsa stain1600X)

Cyclophosphamide group:

Male rats were orally given CP (5 mg/kg) once a day for four weeks. Investigation of the bone marrow cells treated with CP revealed that CP resulted in mutations in the chromosome contrast with the animals under control. Data in Figures (3,4,5,6,7,8)showed that the quantity and kind of chromosomal abnormalities in the bone marrow cells of rats treated with CP included various chromosomal abnormalities, such as centromere region fractures, chromosomal breaks, chromosomal adhesions, ring chromosomes, low chromosome number, excessive chromosome number, degenerate chromosomes, irregular shapes, chromatid fractures, and chromosomal fractures.

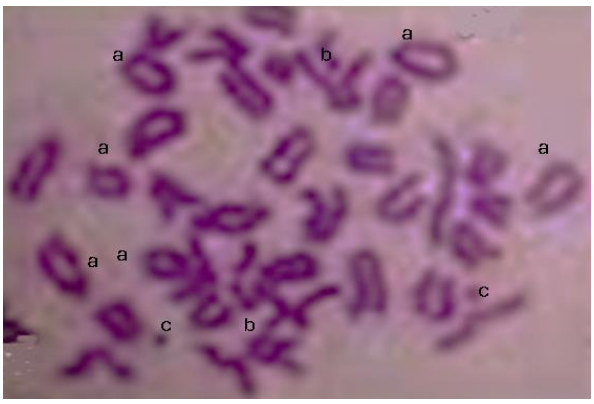

Figure 3: Chromosomal aberrations in the bone marrow of rats from the CP treatment group (Giemsa stain1600X)

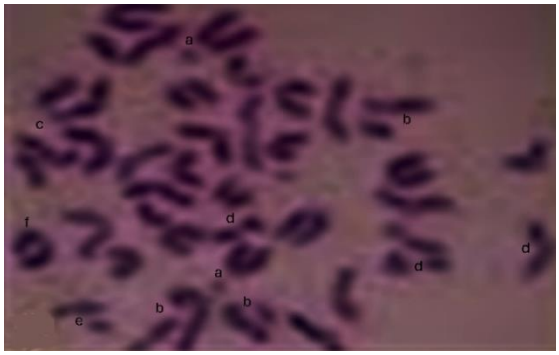

Figure 4: Chromosomal aberrations in the bone marrow of rats from the CP treatment group (Giemsa stain1600X).

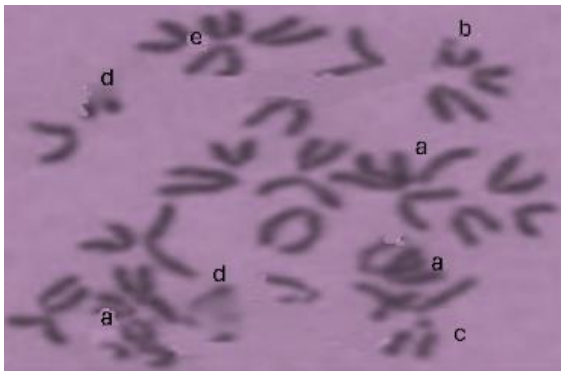

Figure 5: Rats receiving CP treatment have chromosomal abnormalities in their bone marrow cells (Giemsa stain 1600X)

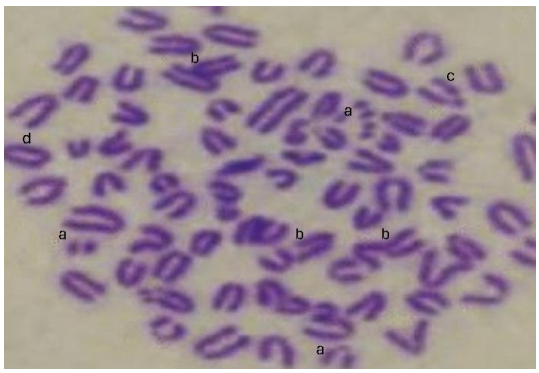

Figure 6: Chromosomal aberrations in the bone marrow of rats from the CP treatment group (Giemsa stain 1600X). A. chromosome fractures

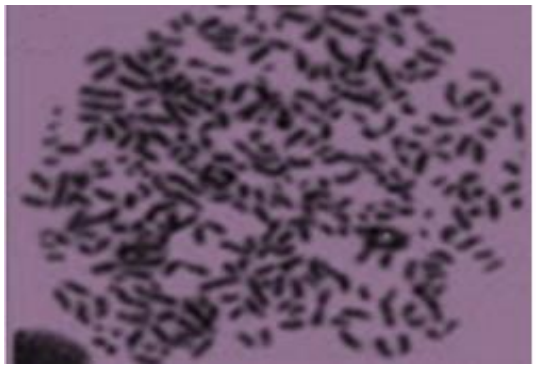

Figure 7: Chromosomal aberrations in the bone marrow of rats from the CP treatment group (Giemsa stain1600X). Excessive chromosome numbers (polyploidy)with many chromosomal anomalies

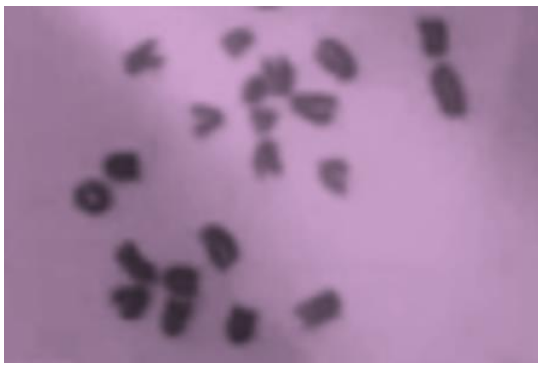

Figure 8: Chromosomal aberrations in the bone marrow of rats from the CP treatment group (Giemsa stain1600X).Low chromosome numbers

Protective group

The results of the bone marrow of male rats given ginger extract and then CP (G 200 + CP 5mg/kg) of body weight revealed that almost all of the chromosomes were normal like to those results showed by the control group (Figure 9). The capability of ginger extract to prevent the negative consequences of CP in the chromosomes of bone marrow cells of rats might act as an antioxidant agent to prevent chromosomal damage and DNA degeneration.

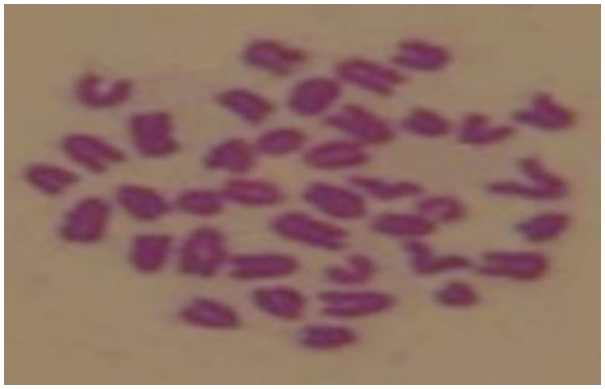

Figure 9: chromosomes of bone marrow cells of the rats near to normal features of protective group (Giemsa stain1600X).

Ginger group:

As we know, ginger rhizomes are used as a spice in food and drink, and they are also used in treated to cure rheumatism, stomach-aches, and waist pain, Therefore, male rats were orally giving a daily an amount of ginger extract (200mg/kg) of bodyweight for four weeks. Results of this group proved that majority of chromosomes were typical and nearly identical to the outcomes that the control group had shown.

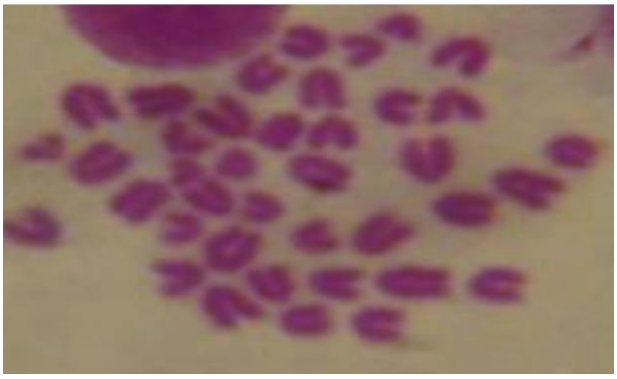

Figure 10: chromosomes of bone marrow cells of the rats near to normal features of Ginger group (Giemsa stain1600X).

4 Discussion

The Food and Drug Administration (FDA) recommends cyclophosphamide (CP), a bi-functional alkylating agent, for the treatment of various rheumatologic illnesses as well as benign and malignant diseases(Matz & Hsieh, 2017). Cyclophosphamide, an antimitotic medication, is known to impact cell proliferation, it may also obstruct primordial germ cells' (PGCs') migration and proliferation from the yolk sac to the vaginal ridges(Ray & Potu, 2007).Through metabolic activation, cytochrome P450 enzymes in hepatic cells convert cyclophosphamide to its reactive intermediates, phosphoramide mustard and acrolein (Foley,1961);(Hales,1982). This medication's active metabolites are alkylating substances that cross-link DNA to obstruct RNA transcription and DNA synthesis,(Ray &Potu, 2007); (Huttunen et al., 2011).Cyclophosphamide targets quickly-dividing cells with its lethal action. Because cyclophosphamide continually exits the germline cell pool and inhibits the formation of new Leydig cells, it is especially harmful to spermatogenic lineage cells (Colvin,1999); (Jahnu kainen *et al.,* 2011). Research conducted on humans revealed that cyclophosphamide treatment caused long-term harm to the male gonadal, resulting in decreased hormone synthesis and infertility from spermatogonia depletion (Ridola *et al*., 2009);(Nurmio, *et al*., 2009). The main causes of cyclophosphamide's toxic effects on the testis were oxidative stress on Sertoli cells and seminiferous tubules, which hampered spermatogenesis and androgenesis and caused germ cell apoptosis (Rezvanfar *et al*., 2008); (Turk *et al*., 2010).Furthermore, cyclophosphamide treatment of the mice led to decreased weight in the testes and epididymis, fewer spermatogonia inside the seminiferous tubules, lower testosterone levels, and finally sterility (Rezvanfar *et al*., 2008). Studies have demonstrated that when an adult male animal takes cyclophosphamide, the weight of their reproductive organs decreases and their ability to reproduce is also affected (Trasler *et al*., 1986); (Ichikawa et al. 2010). In actuality, the quantity of germ cells produced determines testicular weight, and a drop in weight may be a sign of a decline in this cell production (Katoh *et al*., 2008). It was demonstrated in the research of (Tripathi & Jena,2008), that the injection of cyclophosphamide causes an aberration in the morphology of the sperm head. According to research by (Selvakumar *et al.,* 2006);(Ilbey *et al*., 2009), treating male rats with CP results in defective sperm and death. According to (Çeribaşi *et al*., 2010), (Nayak *et al*., 2015).

Mean of mitotic index (MI) of cells in the bone marrow of male rats in all groups

Chemotherapy is an effective cancer treatment, yet there have been a number of side effects documented so far, (Matz&Hsieh,2017). Cyclophosphamide, an antimitotic medication is known to impact cell proliferation. (Ray& Potu, 2007).In comparison to other groups, the current study found that Cyclophosphamide considerably ($p < 0.001$) reduced the mitotic index of bone marrow cells (27.14). The MI mean was increased, after the animals received ginger extract treatment (39.11) also it reached 34.33 when ginger extract was administered to the rats initially followed by Cyclophosphamide. In addition, the mean of MI of the negative control was abet increased (29.23) compared to Cyclophosphamide group, through metabolic activation, cytochrome P450 enzymes in hepatic cells convert cyclophosphamide to its reactive intermediates, phosphoramide mustard and acrolein (Foley,1961);(Hales, 1982). This medication's active metabolites are alkylating substances that cross-link DNA to obstruct RNA transcription and DNA synthesis (Ray&Potu,2007) ; (Huttunen *et al.,* 2011).Cyclophosphamide's cytotoxic action targets cells that divide quickly,(Al-Niwehee,2019). was abet increased (29.23) compared to Cyclophosphamide group, this study in agreement with study by(Al-Niwehee,2019),In contrast to the negative control

(39.33).It is shown that administering CP alone to the rats decreased their MI (23.33), the MI was (34.33) when the extract was administered prior to the CP. Whereas the mean MI was (30.66) and (25.66) when the extract was administered both with and following the (CP) respectively. Chromosomal abnormalities in the bone marrow of male rats in all groups. An investigation of chromosomal abnormalities in rat bone marrow cells revealed that CP was responsible for several abnormalities. The current results are generally consistent with a parallel study on the effect of CP on the chromosomes of mammary cells through its induction of numerous chromosomal abnormalities in the meristem cells of mouse embryos (Fernández *et a*l., 2003).As well as sister chromatid exchange in rat liver cells (Grüngreiff.2016), and in the bone marrow of rats (Popov *et al*., 2011); (Sushma & Devi,2015);(Kour et al., 2017). As for the ability of ginger extract to decrease the percentage of these deformities, it can be explained in light of the previously mentioned mechanism regarding its protective effects in reducing DNA degradation.(Phan *et al*.,2005);(Zahedi *et al*. 2010) ; (Ghasemzadeh *et al*., 2010), This result also confirms the findings of, indicated that ascorbic acid, beta-carotene, terpenoids , polyphenols and alkaloids, such as flavones glycosides, flavonoids, and rutin . An effective antioxidant compounds, that is found in ginger and has the ability to prevent chromosomal damage and mutation caused by mutagenic and carcinogenic substances and factors (Morakinyo *et al*., 2010).

The role of ginger plant

Worldwide, medicinal plants are a significant source of income. Ginger has been used in Chinese and Ayurveda medicine to treat nausea, diarrhea, upset stomach, and heart problems, (Shukla &Singh, 2007). It also encourages the gall bladder to secrete bile and masks the taste of medications (Kato *et al*., 1994);(O'Hara *et al.,* 1998), reduce s arthritic joint discomfort; beneficial for treating lung and cardiac conditions, (Opdyke,1974),cure for throat infection, cough, and cold, (Awang,1992).

By inhibiting cyclooxygenase-1 and cyclooxygenase-2, ginger reduces the production of prostaglandins .(Kumar& Sharma.,2014). Results of the current study confirmed that ginger extract was effective against CP harmful and has the ability to protect the bone marrow chromosome as well as sperm of rats from mutations and DNA damage. According to a previous study, ginger oil has a dominant protective effect against H2O2 induced DNA damage. It may be utilized as an antioxidant and scavenger of oxygen radicals (Phan *et al*., 2005); (Zahedi *et al*., 2010). Another study proved that phenolic compounds in ginger were discovered to scavenge superoxide anions (al-Yahya *et al*., 2022).

5 Conclusions

This study concluded that assessing the mutagenicity of CP on the experimental animals and the potential anti-mutagenic action of ginger extract was as follows:
This study confirmed that CP has genotoxic effects in the studied biological systems.
, CP as a medication for cancer clearly caused a wide range of motions of chromosomes of bone marrow cells in male rats. This study proved that ginger extract exhibited anti-mutagenic effects at selected dose make it an important plant to protect and treat the harmful effects of chemical materials such as CP. Using CP as cancer treatment exposes human beings lives to danger because it causes DNA damage

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