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Research Article

Chrysin Attenuates Osteoporosis-induced Hyperlipidemia and Oxidative Damage in Ovariectomized Rats

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ABSTRACT

Cardiovascular disease and postmenopausal osteoporosis are age-related diseases with high morbidity and mortality across the globe, especially in the elderly women. This study investigates the potential effects of chrysin (CHR) in osteoporosis-induced hyperlipidemia and oxidative liver damage in ovariectomized rats. Twenty-five female Wistar rats were used; 20 rats were ovariectomized (OVX) while 5 rats were sham-operated. The experimental rats were treated daily for a period of six weeks. CHR treatment alleviated body weight gain ($p<0.01$) in OVX rats. In addition, CHR significantly ($p<0.05$) reduced total cholesterol, triacylglycerol and low-density lipoprotein with a simultaneous increase in high-density lipoprotein levels in OVX rats in a dose-dependent manner in comparison to untreated OVX rats. Moreover, treatment of OVX rats with CHR significantly ($p<0.05$) reduced malondialdehyde level and improved reduced glutathione level, superoxide-dismutase and catalase activities. Furthermore, treatment of OVX rats with CHR significantly ($p<0.01$) suppressed alanine-aminotransferase and aspartate-aminotransferase activities in liver tissue compared to the untreated OVX rats. Conversely, treatment of OVX rats with CHR significantly ($p<0.05$) attenuated reduction in femur bone calcium, phosphorus, magnesium and zinc contents altered by ovariectomy compared with untreated OVX rats. This study demonstrated that CHR reduced symptoms of osteoporosis-induced hyperlipidemia and oxidative damage in OVX rats. Our data suggest that CHR, a natural antioxidant, may potentially protect against postmenopausal osteoporosis linked to cardiovascular disease.

Keywords: *Ovariectomy; Osteoporosis; Hyperlipidemia; Oxidative damage; Chrysin*

INTRODUCTION

Postmenopausal osteoporosis (PO) and cardiovascular diseases (CVDs) are both age-related diseases with high significant morbidity and mortality across the globe, especially in older women. Epidemiological evidence suggests that osteoporosis and CVDs share common pathophysiological and genetic risk factors such as female gender, age, family history and menopause (Anagnostis *et al.,* 2009; Divers *et al.,* 2011). Menopause women are at higher risk of developing CVDs due to estrogen deficiency, dysregulation of lipid metabolism and body fat redistribution through interference with leptin activity (Gao and Horvath, 2008; Sobenin *et al.,* 2016). PO has been implicated as the crucial risk factor in the development of CVDs through elevation of total cholesterol (TC), triacylglycerol (TAG) and low-density lipoprotein cholesterol (LDL-c) levels (Torrens *et al.,* 2009; Basuny *et al.,* 2012). Ovariectomized (OVX) rats are the most common and unsurpassed model used in studying bone disorders among other models because it mimics characteristic features of bone loss in postmenopausal women (Abuohashish *et al.,* 2015). For instance, reduction of estrogen during menopause or induced by ovariectomy may down regulate the expression of genes

required for efficient energy expenditure and genes involved in fatty acid catabolism, which may subsequently induce obesity and other metabolic disorders in postmenopausal women (Al-Safi and Polotsky, 2015). A previous study demonstrated that OVX mice model exhibited decrease expression of peroxisome proliferator-activated receptor (PPAR) gamma which resulted in decrease expression of enzymes involved in the β-oxidation of fatty acids and transcription factors require for lipolysis (Kamei *et al.,* 2005). Postmenopausal osteoporosis is also associated with chronic liver disease which is termed as hepatic osteodystrophy and is mostly linked with alterations of hormonal and inflammatory cytokines regulations as well as calcium and vitamin D metabolism (Handzlik-Orlik *et al.,* 2016; Ibrahim *et al.,* 2021). Consequently, numerous research evidences reported imbalance in antioxidant defense system with bone loss and CVDs (Baek *et al.,* 2010; Savini *et al.,* 2013; Zhang *et al.,* 2013). Previous study reported that estrogen reduces oxidative liver damage through alterations in antioxidative enzymes activities, such as SOD and GPx (Borras *et al.,* 2003). Thus, oxidative stress remains a potential target for managing osteoporosis-induced hyperlipidemia and oxidative liver damage. Bioactive compounds from plant sources have been the hallmark for drugs development. Many natural antioxidant compounds are believed to reduce the risk of several age-linked diseases such as diabetes, hypertension, cancer and osteoporosis (Qi *et al.,* 2017; Zhao *et al.,* 2017). Chrysin (CHR), 5,7-dihydroxyflavone is a dietary phytochemical found in honey, vegetables and bee propolis (bee glue). Many plant extracts like blue passion flower (*Passiflora caerulea*) and some species of mushrooms such as *Pleurotus ostreatus* was reported to contain 40 mg/100g CHR (Jayakumar *et al.,* 2009; Singh and Chaudhary, 2011). A previous study indicated the LD_{50} of CHR was 4350 mg/kg and chronic oral administration of CHR at lower doses of 250-500 mg/kg did not caused any sign of toxicity to rats and is usually classify as GRAS (Generally Recognized as Safe) (Yao *et al.,* 2019). CHR has been linked with several pharmacological properties including anti-oxidant, anti-hypertensive, anti-cancer, anti-inflammatory, anti-diabetic and hepatoprotective effects (Ha *et al.,* 2010; Pushpavalli *et al.,* 2010; Wang *et al.,* 2011; Chen *et al.,* 2012; Liu *et al.,* 2014; Samarghandian *et al.,* 2016). For instance, in a previous study, oral administration of CHR to hypercholesterolemic rats increased enzymatic and non-enzymatic antioxidant parameters (Anandhi *et al.,* 2013). Also CHR modulates impairment of insulin signaling molecules and glucose tolerance (Satyanarayana *et al.,* 2015), decreases hepatic fibrosis (Balta *et al.,* 2015), and inhibits the development of neurodegenerative histopathologies (Durak *et al.,* 2016). Hence, the present study investigated the potential attenuating effect of CHR against osteoporosis-induced hyperlipidemia and oxidative liver damage in OVX rats.

MATERIALS AND METHODS

Chemicals and reagents

Chrysin (5,7-dihydroxyflavone, CAS Number 480-40-0) with molecular weight (254.24 g/mol) and 97% purity, Trichloroacetic acid (TCA), Thiobarbituric acid (TBA) and Ellman's reagent were obtained from Sigma Aldrich Chemical Company (Milwaukee WI, USA), through Bristol Scientific Company Limited, Lagos, Nigeria. Total Cholesterol (TC), Triacylglycerol (TAG), High-density lipoprotein (HDL-c), Low-density lipoprotein (LDL-c), Alanine aminotransferase (ALT), Aspartate aminotransferase (AST) and Alkaline phosphatase (ALP) assay kits were purchased from Randox Chemical Company, United Kingdom. Alendronate, Dimethyl sulfoxide (DMSO), Hydrogen peroxide (H_2O_2), Ketamine, Xylazine, Diclofenac sodium, Povidone-iodine Formaldehyde and Alendronate (ALE); a standard drug against osteopenia were purchased from MUB pharmacy (Local supplier, Zaria, Nigeria). All other chemicals and reagents used in this study were of analytical grade.

Experimental animals

Female Wistar (3-month-old) rats weighing between 160-180 g were procured from the Department of Pharmacology and Therapeutics, Ahmadu Bello University, Zaria-Nigeria and then taken to Animal House, Department of Biochemistry, Ahmadu Bello University Zaria. The animals were kept in well-ventilated laboratory cages at room temperature and 12 hr dark/light cycles for a period of two weeks to acclimatize. The rats were fed with standard rat chow (Growers Pelletized Vital Feeds, Bukuru, Plateau State) and water *ad libitum.* Before the commencement of this study ethical approval was obtained from the Committee on the use of Animal and Care, Ahmadu Bello University Zaria-Nigeria (ABUCAUC/2020/66).

Induction of menopause in rats through ovariectomy

A total of 25 Wistar rats were used in this study. Ovariectomy was carried out on female Wister rats (n=20) by a single midline dorsolateral approach under the aseptic condition described previously (Mada *et al.,* 2017). Each rat was anesthetized through injection of 50 mg/kg body weight ketamine hydrochloride and 12.5mg/kg body weight xylazine via the intramuscular route Then a single midline dorsal skin incision of 3 cm long, approximately halfway between the middle of the back and the base of the tail was made with a scalpel blade on the cleaned area, and the two ovaries were detached individually. The uterine horns were

returned to the peritoneal cavity, and the wound was then closed with an absorbable suture (Vicryl). The remaining rats (n=5) underwent sham operation; this surgical procedure is the same as described for ovariectomy but ovaries were only identified not excise. At the end of ovarietomy and sham operation, the rats were kept in separate cages for 14 days to recover before the commencement of the study.

Animal groupings and treatment

Twenty-five (n=25) female Wistar rats were ovariectomized (n=20) and sham-operated (n=5) to investigate the effect of chrysin against osteoporosis-induced hyperlipidemia, oxidative stress, liver damage and femur-bone mineral contents. The ovariectomized (n=20) and sham-operated (n=5) rats were divided into five groups, consisting of 5 rats each and treated as follows; Group 1: Sham operated rats daily received a vehicle (10% DMSO) orally and served as normal control. Group 2: Untreated OVX rats daily received a vehicle (10% DMSO) orally and served as positive control. Group 3: OVX rats were daily treated orally with 50 mg/kg CHR. Group 4: OVX rats were daily treated orally with 100 mg/kg CHR. Group 5: OVX rats were daily treated orally with 5 mg/kg ALE (Standard drug for treatment of bone loss) via intramuscular. The treatments commenced two weeks after recovery from ovariectomy and sham surgery and continued for a period of six (6) weeks. The choice of dosages for CHR (≤ 100mg/kg bw) and alendronate (5 mg/kg bw) was based on previous literature (Oršolić *et al.,* 2014). The initial body weight of each rat was measured before treatment commenced using electronic weighing balance. Thereafter, weekly body weight was measured and percent body weight gain was determined at the end of study period.

Collection of femur and liver samples

At the end of six weeks of the study period, rats from all the groups were anesthetized by injection of 50 mg/kg body weight ketamine hydrochloride and 12.5 mg/kg body weight xylazine via intramuscular route. Liver tissue were immediately collected and washed with normal saline, dried by blotting with filter paper and weighed. Subsequently, 1000 mg of liver from each rat was cut into pieces and homogenized with 10 ml phosphate-buffered saline (pH 7.4). The homogenate was centrifuged at 10,000 x g for 15 mins at 4°C. The supernatant was used for biochemical analyses. Also, left femur bones were excised from each rat and cleaned of adherent tissues, then dried in an oven at 100ºC for 12 h. Thereafter, bone mineral contents were measured.

Measurement of lipid parameters, liver enzymes and oxidative stress markers

The concentration of TC, TAG, HDL-c and LDL-c and the activities of ALT, AST and ALP enzymes in liver homogenate of experimental rats were assayed using their respective ELISA assay kits (Randox Chemical Company, United Kingdom) according to the manufacturer's instruction. In addition, malondialdehyde (MDA) concentration in the liver homogenates was determined as thiobarbituric acid reactive substances (TBARS) as described previously (Ohkawa *et al.,* 1979). Reduced glutathione (GSH) level in liver homogenates was measured according to the previously described protocol (Ellman, 1959). Catalase (CAT) activity was determined as described previously by Aeibi and Bergmeyer (1974), while superoxide dismutase (SOD) activity in liver homogenates was determined as described by Fridovich (1975).

Measurement femur bone mineral contents

The dried femora were measured using an electronic weighing balance (Mitutoyo, India) and then ashed in a muffle furnace at 550^0C for 12 h. The ash was weighed and dissolved in 3 ml triple acids (Sulphuric acid: Hydrochloric acid: Nitric acid) in a ratio of 2:1:1 to solubilize the ash, and the content was evaporated using a hot plate. The samples were then transferred to 100 ml volumetric flasks and volume was adjusted to 100 ml with distilled water. Thereafter, 1 ml was taken for measurement of calcium, phosphorus, magnesium and zinc level using an Atomic Absorption Spectrophotometer (Shimadzu AA-680) at their respective wavelength.

Data analysis

Data are expressed as Mean ± SD, (n=5), and analyzed using GraphPad Prism 5.01 version (GraphPad Statistical Software Inc., San Diego, CA, USA). One-way analysis of variance (ANOVA) and Dunnett's post-hoc test was employed to compare the significance of differences between the means of the tests and controls. $P<0.05$ was considered statistically significant

RESULTS

Effect of CHR on body weight in ovariectomized rats

The effect of CHR on body weight in OVX rats was investigated, the data obtained revealed that OVX significantly ($p<0.05$) increased body weight gain compared to the sham control group (Table 1). Meanwhile, when OVX

Table 1: Effect of CHR on body weight gain in ovariectomized rats

ANIMAL GROUPINGS	SHAM + VEHICLE	OVX + VEHICLE	OVX + 50mg/kg CHR	OVX + 100mg/kg CHR	OVX + 5mg/kg ALE
Initial BW (kg)	172.3 3± 2.32	174.67 ± 4.01	173.60 ± 3.74	176.33 ± 5.37	174.50 ± 4.12
Final BW (kg)	201.25 ± 4.61	238.00 ± 8.92$^{###}$	225.40 ± 7.55*	219.00 ± 6.82**	210.00 ± 5.94***
Percentage BW gain	16.78 ± 1.43	36.25 ± 2.17$^{###}$	29.83 ± 1.96*	24.19 ± 1.782**	20.34 ± 1.55***

Data are expressed as Mean ± SD (n=5). $^{###}$P< 0.001, $^{##}$P< 0.01, and $^{#}$P< 0.05 for SHAM group versus untreated OVX group; *P<0.05, **P<0.01, and ***P< 0.001 for untreated OVX group versus treated OVX group. Values bearing different superscript along the row indicated significant difference among the treated groups. SHAM = Control rats, OVX = Ovariectomized rats, CHR = Chrysin, ALE = Alendronate, BW = bodyweight.

rats were treated with different concentrations of CHR, a significant (p<0.05) reduction in body weight gain was observed compared to untreated OVX rats.

Effect of CHR on lipid profile in liver tissue of ovariectomized rats

OVX rats had significant (p<0.05) elevation of TC, TAG and LDL-c levels and significant (p<0.05) reduction of HDL-c level in comparison to rats in sham group (Figure 1A-D). However, administration of CHR to OVX rats significantly (p<0.05) restored all these lipid profile parameters altered by OVX in a dose-dependent relation when compared to untreated OVX rats (Figure 1A-D). The effect of CHR on the lipid profile parameters was almost comparable with 5 mg/kg ALE which is a standard drug used to manage osteoporosis. Thus, CHR could be beneficial against OVX-induced hyperlipidemia in rats.

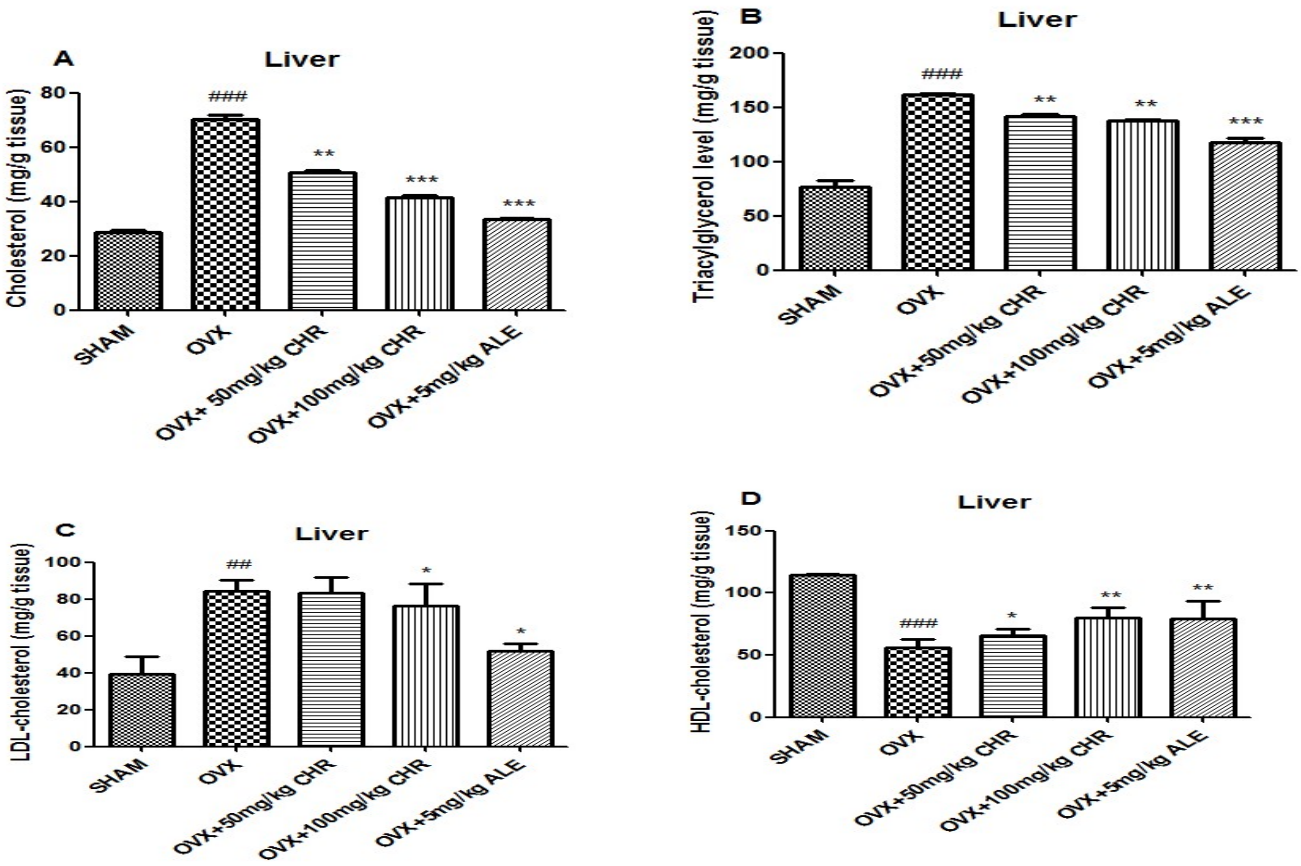

Figure 1: Effect of chrysin on lipid profile parameters in liver tissue of ovariectomized rats.

A) Total Cholesterol, B) Triacylglycerol, C) Low density lipoprotein (LDL-c), D) High density lipoprotein (HDL-c)

Data are expressed as Mean ± SD (n=5). $^{###}$P< 0.001, $^{##}$P< 0.01, and $^{#}$P < 0.05 for SHAM group versus untreated OVX group; *P< 0.05, **P<0.01, and ***P< 0.001 for untreated OVX group versus treated OVX group. Bars bearing different superscript indicated significant difference among the treatment groups. SHAM = Control rats, OVX = Ovariectomized rats, CHR = Chrysin, ALE = Alendronate.

Effect of CHR on oxidative stress markers in ovariectomized rats

The present data demonstrated a significant ($p<0.05$) increase in MDA level and decreased in GSH level of OVX rats compared to the sham control group (Figure 2A and B). Conversely, treatment of OVX rats with different doses of CHR, significantly ($P<0.05$) reversed the alteration of these parameters in a dose-dependent manner in comparison to untreated OVX rats (Figure 2A and B). This result again demonstrates the antioxidative property of CHR and could play potential role in in managing postmenopausal osteoporosis.

Effect of CHR on antioxidant enzymes in ovariectomized rats

There was a significant ($p<0.05$) decrease in SOD and CAT activities in OVX rats in contrast to the sham-operated group ((Figure 2C and D). However, when OVX rats were treated with different concentrations of CHR, a significant ($p<0.05$) increase in both SOD and CAT activities were observed compared to the untreated OVX group. These data were comparable with the data obtained for ALE (Figure 2C and D).

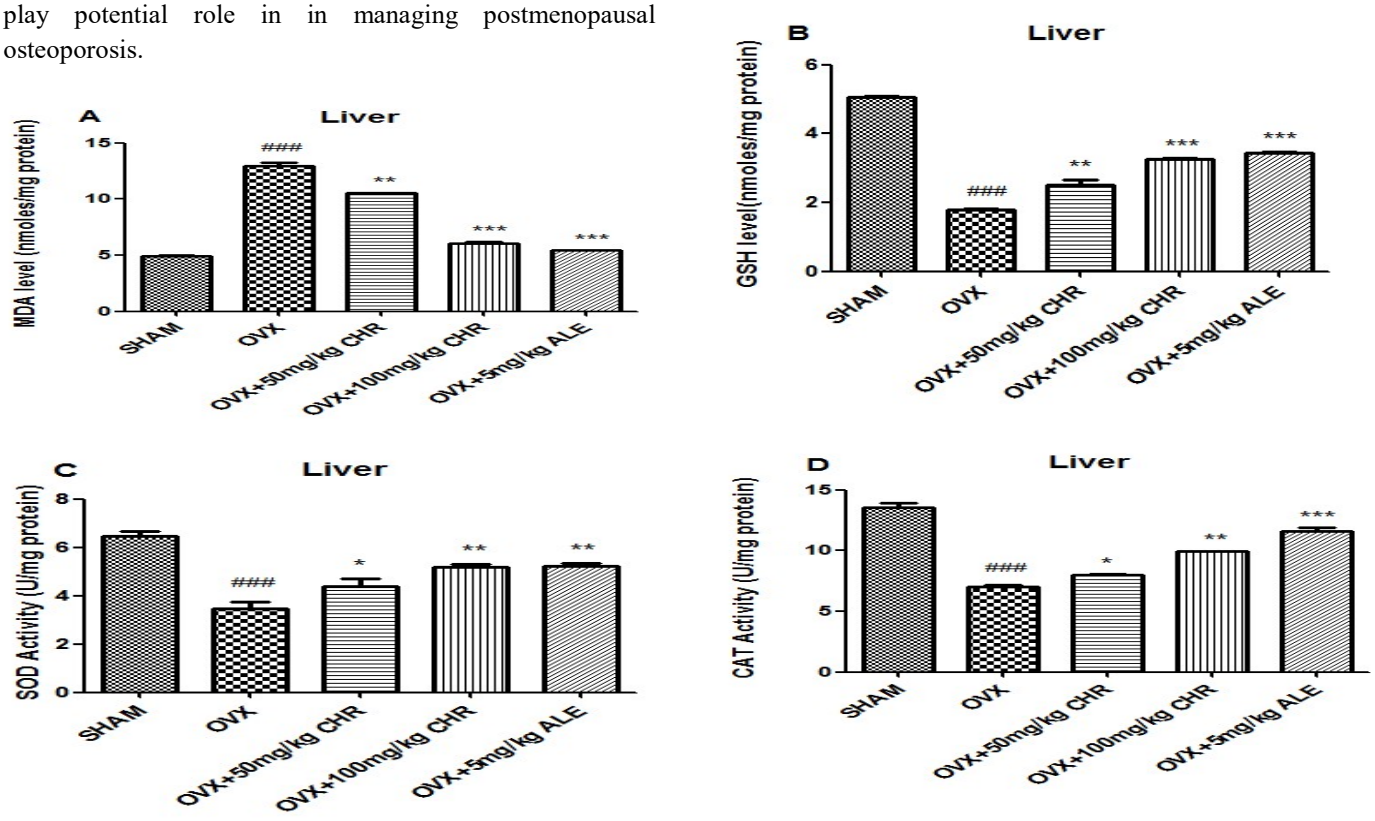

Figure 2: Effect of chrysin on oxidative and antioxidative stress markers in ovariectomized rats. A) Malondialdehyde (MDA) level, B) Glutathione (GSH) level, C) Superoxide-dismutase (SOD) activity, D) Catalase (CAT) activity

Data are expressed as Mean ± SD (n=5). $^{###}P < 0.001$, $^{##}P< 0.01$, and $^{#}P< 0.05$ for SHAM group versus untreated OVX group; *P<0.05, **P<0.01, and ***P<0.001 for untreated OVX group versus treated OVX group. Bars bearing different superscript indicated significant difference among the treatment groups. SHAM = Control rats, OVX = Ovariectomized rats, CHR = Chrysin, ALE = Alendronate.

Effect of CHR on hepatic enzymes activities in liver tissue of ovariectomized rats

The effect of CHR on the ALT, AST and ALP enzyme activities in liver tissue of OVX rats was investigated. The result indicated that ALT and AST enzyme activities were significantly ($p<0.05$) lowered in liver tissue of OVX rats when compared to sham-operated rats (Figure 3A-C). However, AST activity was not significantly ($p>0.05$) different in liver tissue of OVX and sham-operated groups (Figure 3A-C). Interestingly, when OVX rats were treated with 50 mg/kg and 100 mg/kg doses of CHR, a significant ($p<0.05$) increase in ALT and AST activity were recorded in comparison with the untreated OVX rats. However, CHR administration did not showed significant ($p>0.05$) effect on ALP activity between OVX rats and sham-operated groups (Figure 3A-C).

Effect of CHR on femur bone mineral contents in OVX rats

The present data indicated that calcium, phosphorus, magnesium and zinc were significantly ($p< 0.05$) reduced in femur of OVX rats in comparison to the sham control group. Although, the treatment of OVX rats with different concentrations of CHR significantly ($p< 0.05$) ameliorated

the reduction of these mineral contents in OVX rats compared to the untreated OVX rat group (Figure 4A-D).

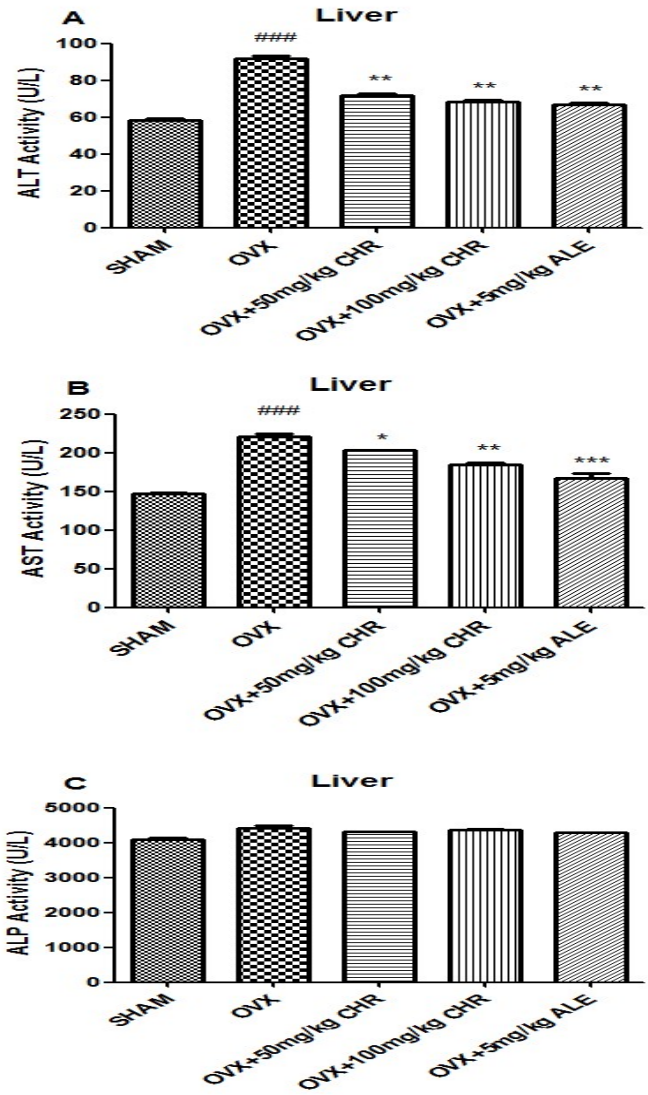

Figure 3. Effect of chrysin on hepatic enzymes activities in liver tissue of ovariectomized rats A) Alanine aminotransferase, B) Aspartate aminotransferase, C) Alkaline phosphatase.

Data are expressed as Mean ± SD (n=5). $^{###}P < 0.001$, $^{##}P < 0.01$, and $^{#}P < 0.05$ for SHAM group versus untreated OVX group; *P<0.05, **P<0.01, and ***P<0.001 for untreated OVX group versus treated OVX group. Bars bearing different superscript indicate significant difference among the treatment groups. SHAM = Control rats, OVX = Ovariectomized rats, CHR = Chrysin, ALE = Alendronate.

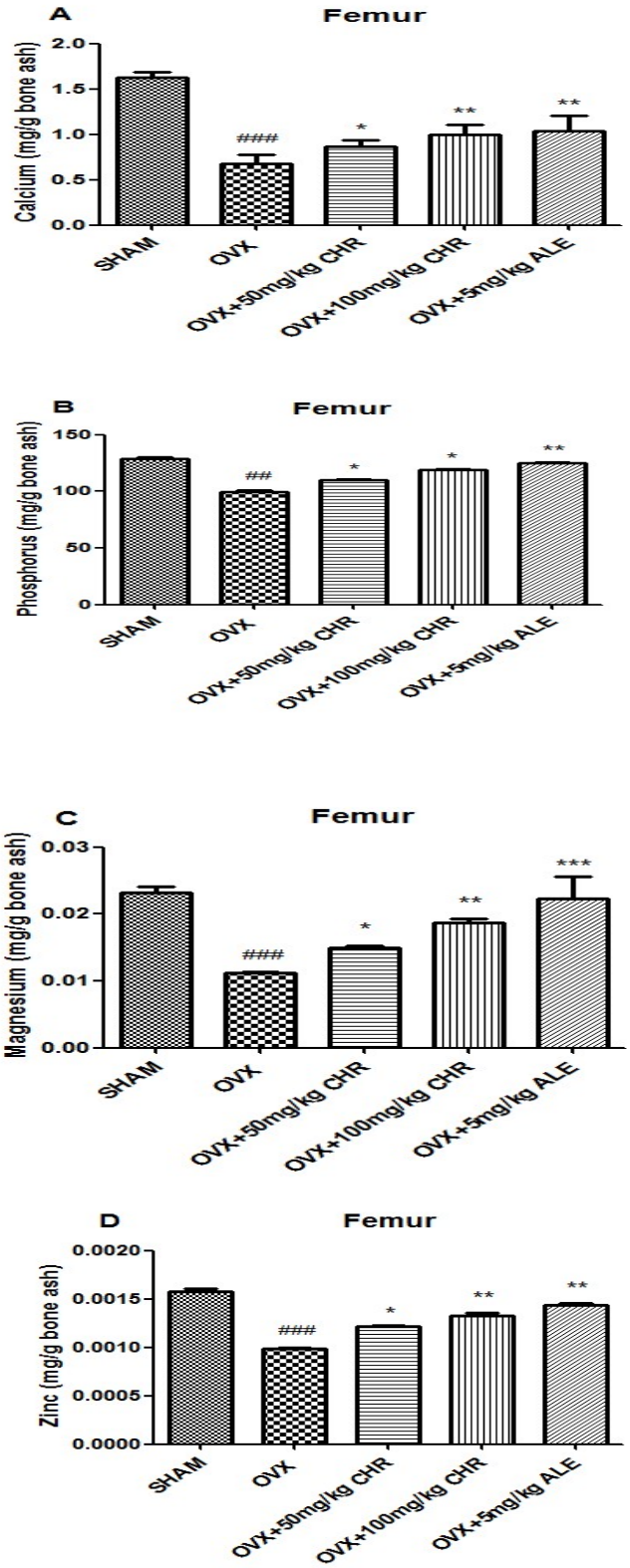

Figure 4: Effect of chrysin on femur bone mineral contents in ovariectomized rats A) Calcium, B) Phosphorus, C) Magnesium, D) Zinc

Data are expressed as Mean ± SD (n=5). $^{###}P<0.001$, $^{##}P<0.01$, and $^{#}P<0.05$ for SHAM group versus untreated OVX group; *P<0.05, **P<0.01, and ***P<0.001 for untreated OVX group versus treated OVX group. Bars bearing different superscript indicated significant difference among the treatment groups. SHAM = Control rats, OVX = Ovariectomized rats, CHR = Chrysin, ALE = Alendronate.

DISCUSSION

Postmenopausal osteoporosis has been implicated in the pathogenesis and development of hyperlipidemia. For instance, several studies have reported positive correlation between postmenopausal osteoporosis and development of CVDs through dyslipidemia, obesity and excessive oxidative damage which may impair liver functions (Lin *et al*., 2014; Tian *et al.,* 2015; Sankar *et al.,* 2019; Ko and Kim, 2020). This study demonstrated significant increase in body weight gain OVX rats, an indication of successful OVX and the onset of menopause condition in rats (Choi, 2009; Nian *et al.,* 2009). Treatment of OVX rats with CHR reduced the excessive body weight gain. The results obtained from this study are consistent with previous studies which reported that OVX was indeed linked to drastic increase in the body weight gain in OVX rats due to estrogen deficiency and excessive fat deposition (Shuid *et al.,* 2011; Hwang *et al.,* 2016). The uterus histopathology analyses demonstrated that treatment of OVX rats with CHR partly suppressed atrophy of endometrial lumen. This finding suggests that CHR may have beneficial effect on uterus growth and development. Thus, CHR could be helpful against osteoporosis-induced uterine atropy. Moreover, previous studies have shown positive correlation between osteoporosis and high levels of TC, LDL-c and high TC-to-HDL-c ratio (Derby *et al.,* 2009; Kim *et al.,* 2011). Similarly, another study also reported that TC-to-HDL-c ratio increased in menopause and it could serve as a better indicator of CVDs than TC alone (Zago *et al.,* 2004). Therefore, menopausal women are at high risk for developing CVDs due to dysregulated lipid metabolism and estrogen deficiency (Anagnostis *et al.,* 2015), since the receptors for estrogen and androgen are expressed in both visceral and subcutaneous adipocytes and thus, can affect lipid profile in premenopausal and postmenopausal women (Rhee *et al.,* 2008).

The present study demonstrated that TC, TAG, and LDL-c were elevated in OVX rats while HDL-c level was decreased. Upon treatment of OVX rats with CHR substantially attenuated the alterations in lipid profile parameters suggesting that CHR exert hypolipidemic effect. These data were supported by previous studies which reported that N-acetyl-cysteine, an antioxidant compound possesses hypolipidemic effect (Razmjou *et al.,* 2018; Mada *et al.,* 2020). Lowering of cholesterol absorption and reduction of oxidative damage could in part be among the possible mechanism through which CHR may reduce OVX-induced hyperlipidemia in OVX rats. Previous studies on rodents reported that other flavonoids such as naringenin (0.02% supplements in diet) and CHR (10 mg/kg orally) exhibit anti-hyperlipidemic effect (Chanet *et al.,* 2012; Zarzecki *et al.,* 2014). Oxidative stress has been demonstrated as crucial pathogenic factor in the development of osteoporosis amongst menopause women (Kim et al., 2003). Lack of estrogens during menopause is recognized as the main cause of increase in oxidative damage due to elevation of lipid peroxidation, alterations of enzymatic and non-enzymatic antioxidant system (Maroti *et al.,* 2010). The present study indicated that OVX rats had elevated MDA and lower GSH levels while the activities of CAT and SOD were considerably reduced. These alterations in selected oxidative and antioxidant biomarkers were substantially reversed in OVX rats when treated with different concentrations of CHR. This data further supported the antioxidant activity of CHR. The antioxidant property of CHR reported in this study could be attributed to the structure-function property; this is because CHR possesses hydroxyl groups which may be responsible for scavenging free radicals, decrease lipid peroxidation and thus enhances the antioxidant defense system in OVX rats. The liver is an essential organ involved in several complex metabolic functions including synthetic and detoxification process. Postmenopausal osteoporosis has been associated with liver diseases and about 30% of individuals with chronic liver disease are found to be osteoporotic (Handzlik-Orlik *et al.,* 2016).

The most common clinical biomarkers used in diagnosing liver damage are ALT, AST and ALP activities among others (Mani and Natesan, 2018). The present findings showed a substantial increase in the activities of ALT and AST but without significant change in ALP activity in liver tissue of OVX rats. Expectedly, treatment of OVX rats with chrysin, the activities of ALT, AST and ALP enzymes were reversed, which suggested that CHR exhibits hepatoprotective effect and could, ameliorates osteoporosis-induced liver damage and dysfunction. The present data was in conformity with the previous findings which reported that CHR showed hepato-protective activity by reducing tumor necrosis factor-α (TNF-α) expression (Subramaniam *et al.,* 2015). Also bioactive compounds derived from the plant with phytoestrogens-like activity are believed to possess beneficial effects against postmenopausal osteoporosis and by extension may reduce the risks associated with hepatic toxicity in rats (Sathiavelu *et al.,* 2009; Hermenean *et al.,* 2017). Bone is dynamic and metabolically active tissue consisting of inorganic compounds such as Ca, P, Mg, Zn, and Na among others, which are involved in matrix mineralization and increase bone strength (Mada *et al.,* 2018). Reduced bone mineral density (BMD) is regarded as a strong predictor for osteoporosis fracture risk and epidemiological study have linked CVDs with the increase in bone loss (Baldini *et al.,* 2004; Tankó *et al.,* 2005;

Sennerby *et al.,* 2009). In addition, the present investigation showed that femur bone calcium, phosphorus, magnesium and zinc contents were altered in OVX rats. Remarkably, CHR administration prevents demineralization of the femur bone in OVX rats which indicates potential beneficial effect of CHR on bone mineralization possibly through down regulation of abnormal bone remodeling thereby reducing the risk of osteoporotic fractures (Kanis *et al.,* 2009). This data was consistent with the previous study which also reported that bone mineral contents especially calcium was reduced in OVX rats but recovered after administration of medicinal herbs (Elkomy and Elsaid, 2015). Therefore, this study demonstrated the potential beneficial effect of CHR against OVX-induced hyperlipidemia and oxidative stress in postmenopausal osteoporotic rat model.

CONCLUSION

In conclusion, the present study among surgically induced menopause rats suggests that CHR; a natural antioxidative compound may attenuate osteoporosis-induced hyperlipidemia and oxidative liver damage perhaps through alleviating oxidative stress in OVX rats. Thus, CHR could be a potential health promoting agent in oxidative stress associated disorders. However, the molecular mechanism through which CHR mediates the beneficial effect remains to be elucidated.

Future Directions/Limitations

This study investigated the role of CHR in mitigating osteoporosis-induced hyperlipidemia in OVX rats. We have reported that CHR exhibits antioxidative and antihyperlipidemic effects in OVX rats. This study is preliminary and thus further research are required especially on the effects of CHR on femur bone mineral density (BMD), expression of bone-resorbing cytokines genes and detailed molecular mechanisms through which CHR exert the beneficial effect.

AUTHORS' CONTRIBUTIONS

SBM and SOI contributed to the conception and design of the work; MAS, NAG, AS, ABH and AMJ contributed to the acquisition, analysis and interpretation of data; SBM, MAS, SOI, MMA and AG drafted the work and substantively revised the manuscript. All authors gave approval for the final version submitted.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest

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