Effect of glucoseamine and its source scales fish on the osteoporosis in rats

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Abstract

Osteoporosis related to various factors including menopause and aging- is the most common chronic metabolic bone disease, which is characterized by increased bone fragility. Although it is seen in all age groups, gender, and races, it is more common in Caucasians (white race), older people, and women. With an aging population and longer life span, osteoporosis is increasingly becoming a global epidemic. This study aims to determine the effects of oral glucosamine and its source as scales fish on biochemical parameters of osteoporosis in rats. Thirty six albino rats, their weight was 150±5g , were divided into six groups , one of them was fed basal diet as negative control while the other thirty rats were injected with 0. 8mg/kg bw Dexamethasone for 3 days to induce osteoporosis, one of them was still fed basal diet withous treated as positive control group . The osteoporotic rats were received 1.5 , 3 g/d of glucoseamine (G3 &G4) and 10 , 20% from scale fish extract(G5&G6) for a period of 28 d.. Results presented that administration of glucoseamin sources showed an improvement in body weight , feed intake and feed efficiency ratio . In the other hand, oral administration of glucosamine showed significant effect on all biochemical measured parameters in osteoporotic rats in both glucosamine sources as compared to positive control group . Therefore, increasing awareness among dietetic source of glucosamine especially scale fish, which, in turn, facilitates increase awareness of the normal populace, will be effective in preventing this epidemic.

Keywards:Glucosamine;scale fish; rats; osteoporosis ; extract

Introduction

Osteoporosis is a progressive systemic skeletal disease characterized by reduced bone mass/density and micro architectural deterioration of bone tissue. Bone formation initially exceeds bone resorption, but by the third decade this has reversed resulting in a net loss of bone mass. This leads to an increased bone fragility and susceptibility to fracture. Many patients presenting with a fracture caused by a fall from standing height or less are not on any kind of therapy and many patients who sustain these fragility fractures are not started on therapy. Osteoporotic (fragility) fractures are fractures that result from mechanical forces that

would not ordinarily result in fracture. Osteoporotic fractures are defined as fractures associated with low bone mineral density (BMD) and include spine, forearm, hip and shoulder fractures. The trouble is osteoporosis is a "silent disease" because there are no symptoms prior to a fracture. However, once a person has broken a bone, their risk of breaking another fragility fracture increases significantly. After the first break, one in eight will break another bone within a year and a quarter within five year **(Hippisley *et al.*, 2009 and Cosman *et al.*, 2014).**

Glucosamine ($C_6H_{13}NO_5$) is an amino sugar and a prominent precursor in the biochemical synthesis of glycosylated proteins and lipids. Glucosamine is part of the structure of the polysaccharides chitosan and chitin, which compose the exoskeletons of crustaceans and other arthropods, cell walls in fungi and many higher organisms. Glucosamine is one of the most abundant monosaccharides. It is produced commercially by the hydrolysis of crustacean *exoskeletons* or, less commonly by fermentation of a grain such as corn or wheat. In the US it is one of the most common non-vitamin, non-mineral, dietary supplements used by adults **(McAlindon *et al.*, 2004 and Imagawa *et al.*, 2011).**

Glucosamine is an aminosaccharide, acting as a preferred substrate for the biosynthesis of glycosaminoglycan chains and, subsequently, for the production of aggrecan and other proteoglycans of cartilage. Because of the essential role aggrecans play in giving the cartilage its hydrophilicity, compounds enhancing synthesis of aggrecans may be beneficial in cases of OA, a disorder characterized by an increase in matrix structural protein turnover, with catabolism being predominant over synthesis . In vitro, glucosamine sulfate **(GS)** has been demonstrated to reduce prostaglandin E2 **(PGE2)** production and interfere with nuclear factor kappa B **(NF_B) DNA** binding in chondrocytes and synovial cells . Glucosamine inhibits gene expression of OA cartilage in vitro . Long-term oral administration of glucosamine sulfate reduces the destruction of cartilage and upregulation of **MMP-3 mRNA** in a model of spontaneous osteoarthritis in Harley guinea pigs. Glucosamine can prevent cytokine-induced demethylation of a specific **CpG** site in the **IL1b** promoter and this is associated with decreased expression of **IL1b** . It was suggested that since glucosamine inhibits both anabolic and catabolic genes, the therapeutic effects of glucosamine might be due to anticatabolic activities, rather than due to anabolic activities. GS is a stronger inhibitor of gene expression than glucosamine hydrochloride **(Reginster *et al.*, 2005 and Taniguchi *et al.*, 2011).**

Glucosamine is a natural compound found in scales of the fish — the tough tissue that cushions joints. In supplement form, glucosamine is harvested from scales of fish or made in a lab. There are several forms of

glucosamine, including glucosamine sulfate, glucosamine hydrochloride and N-acetyl glucosamine. The most common source of glucosamine used in dietary supplements scales of fish. The hard exoskeletons of crustaceans such as crabs and shrimp contain a substance called chitin. Chitin is a rich source of glucosamine and acts as the starting material for many dietary supplements (**Zhu *et al*., 2018).**

For that, the study aimed to evaluate the effect of different glucosamine levels from fish scales and artificial sources on the healthy status of osteoporosis rats.

Materials and methods

Materials

Glucosamine, Casein, salt mixture, vitamin mixture and biochemical kits were purchased from Gomhoria Co. , Giza, Egypt. Dexamethasone were obtained from Al-Amria Pharmaceutical Industries Company. Scale fish were obtained from Bolti fish which purchased from Menofia Governorate .Male albino rats (150±5g b.w) of Sprague Dawley strain, the animals were obtained from Research Institute of Ophthalmology, Giza, Egypt.

Methods

Preparation fish scales

The extraction of glucosamine was carried as described previously by **Granito *et al.* (2018).** Wash the scale fish , then weigh 200g of and cut into fine dices. Prepare 25ml of 4M ammonium sulphate and spray it on the fine dices. Dry the dices in oven at the temperature of 91^{o}C for 48 hours. Grind the dried dices into fine powder and extract glucosamine with distilled water at room temperature. Centrifuge the sample at 10,000rpm for 10 minutes and collect the supernatant. The supernatant was placed in hot air at 100C for the evaporation. The dried Sample was analyzed for amount of glucosamine extracted by **HPLC**.Each 100 g dried scale fish contained 20% glucoseamine.

Biological investigation

Osteoporosis induction

The rats were injected with 0. 8mg/kg bw Dexamethasone for 3 days until the level of V.D decreased from 31.7 to 6.9 and serum calcium decreased from 12.6 to 4.1 to induce osteoporosis **(Barnes, 2006).**

Animals

Thirty-six adult male albino rats, Sprague-dawley strain weighting (150±5) g each, were used in this study, thirty of rats were osteoporosis. All rats (normal and osteoporosis) were fed standard diet for four consecutive days, then were divided into main two groups each with similar total body weight and were housed individually in the wire cage. All the group of rats were fed the experimental diet for 4 weeks according to the following groups.

Diets

Basal diet was prepared from fine ingredients per 100g. The diet had the following composition :Corn starch 67%, Casein 13%, corn oil 10%, Fiber 5% according to **AIN(1993)**. Salts formula 4% **and** vitamin formula 1% were prepared according the methods of **Hegsted *et al.(* 1941)** and **Campbell (1963)** respectively.

Experimental design

The experimental study was done in Faculty of Home Economics , Menoufia University. Rats were housed in wire cages in room maintained at 25 ± 2 °C and kept under normal healthy conditions.

All rats fed on normal basal diet for one week before starting the experiment for acclimatization. After one – week period, the rats were divided into six groups each with similar total body weight and were housed individually in wire cages (6 rats each), the period of the experiment was **28 days**

Rats groups

– **Group (1)** : Rats were fed on basal diet as a negative control group.
- Group (2): Osteoporotic rats were fed on basal diet as a positive control group .
-Group (3): Osteoporotic rats fed on basal diet containing with glucosamine 1.5 g/day.
- Group (4): Osteoporotic rats fed on basal diet containing with glucosamine 3 g/day.
- Group (5): Osteoporotic rats fed on basal diet containing with 10% of scale fish extract.
- Group (6): Diabetic rats fed on basal diet containing with 20% of scale fish extract.

Each of the above groups was kept in a single wire cage, the diets were introduced to rats in special non-scattering feeding cups to avoid loss of food and contamination. Tap water was provided to rats by means of glass tubes projecting through wire cages from inverted bottles supported to one side of the cage. Rats were weighted at the Beginning and at the end of the experiment Retro - orbital blood samples were collected organs (liver , bones and kidney) of each rats were removed rapidly and were weighted separately.

Blood sampling

From all the previously mentioned groups, blood samples were collected after 12 hour fasting at the end of the experiment .Using the retro - orbital method by means of a micro capillary glass heparinized tubes, blood was collected into a dry clean centrifuge tube and left to clot in a water bath (37°C)at room temperature for half an hour. The blood

was centrifuged for 10 minutes at 3000 r.p.m. to separate the serum. Serum was carefully aspirated and transferred into clean quit fit plastic tubes and kept frozen at (20ºC) until the time of analysis (**Yenson,1994)**.

Biological Evaluation

All rats were weighted once weekly and feed intake was also calculate daily. At the end of the experiment, biological evaluation of the different diets was carried out by determination of body weight gain (BWG), feed efficiency ratio **(FER)** according to **Chapman *et al.*, (1959).** Using the following formulas.

BWG = Final weight – Initial weight

$$\textbf{FER} = \frac{\text{Feed Intake (g)}}{\text{Gain in body weight (g)}}$$

Biochemical analysis

The following determination were carried out for all serum samples:

Enzymatic determination of plasma glucose was carried out calorimetrically according to the method of **Yound (1975)**. Serum aspartate transaminase (AST), alanine transaminase (ALT) and alkaline phosphatase (ALP) activities were estimated with the Randox reagent kit using 2, 4-dinitrophenylhydrazine as substrate according to the method described by **Reitman and Frankel (1957).** Kidney function parameters including urea, uric acid and creatinine were determined according to **Patton& Crouch(1977); Fossati *et al*.(1980) and Bonsens& Taussky(1984)** respectively. Blood glucose was carried out according to the method of **Tietz (1976)** .Calcium and phosphorus levels were quantified using determined by Atomic absorption spectrophotometer according to **Ohishi *et al.* (1993)** . Serum levels of calcitonin, parathyroid were measured using methods of Chantraine et al., 1979and osteocalcin was measured by **Garcia-Carrasco *et al.*, (1988)** . **BMD** was measured according to Archimedes principle of **Craciun *et al.* (2000).**

Statistical analysis

All results obtained in the present study were evaluated by One-way ANOVA test and the values were expressed as means ± SD and values of $P < 0.05$ were considered statistically significant **(Snedecor and Cochron, 1982).**

Results and disscution

Effect of feeding different levels of glucosamine and scale fish on feed intake, feed efficiency ratio and body weight gain.

The effect of feeding different levels of glucosamine and scale fish on feed intake, feed efficiency ratio and body weight gain are shown in

table (1). Concerning the feed intake, it were 14.9±0.38 g / day for control (-) and , 8.38±0.38 g/ day for control (+). However, the feed intake for groups 3, 4, 5 and 6 decreased compared to control (-) and increased as compared to control (+).

The feed efficiency ratio were 0.097 ±0.001 for control (-), while the values decrease in groups control (+),3, 4 and 6, respectively. They were 0.085 ±0.007, 0.095 ± 0.006, 0.095 ±0.02and 0.095 ±0.02 respectively. However, such decrease is statistically significant while group 5 was nonsignificant with control group.

Results of body weight gain were 42.27 $\pm$ 2.51 g. for negative control group and decreased in the other groups.This decrease is statistically significant when compared with negative control and positive control.

These results are in agreement with those reported by **Pavelka *et al.* (2002)** who reported that, glucosamine was able to improve feed intake. it can cause weight gain

The feed efficiency ratio decreased in positive control compared to the others. This decrease is statistically significant. These results are in agreement with those reported by **Usha *et al.*, (2004)** who reported that; glucosamine can able reduced the cardiovascular disease risk, diabetes and obesity.

Table (1): Effect of feeding different levels of glucosamine and scale fish extract on feed intake, feed efficiency ratio and body weight gain.

Parameters	Animal groups					
	Negative control G1	Positive control G2	1.5 g glucosamine G3	3g glucosamine G4	10% scale fishG5	20% scale fishG6
Feed intake g/d	14.9^{a}± 0.38	8.38^{d} ±0.38	9.78^{c} ±0.35	12.2^{b}± 0.33	8.54^{d} ±0.35	11.83^{c} ±0.03
BWG g/28d	40.27^{a} ±2.51	20.12^{d} ±3.46	23.17^{c} ±4.82	32.41^{b} ±7.56	23.28^{c} ±4.69	31.58^{b} ±5.71
FER	0.097^{a} ± 0.001	0.085^{c} ± 0.007	0.095^{b} ± 0.006	0.095^{b} ± 0.02	0.097^{a} ±0.001	0.095^{b} ±0.02

Each value represents the mean of 6 rats ± SD. Values significantly different compared to diabetic group: $P \leq 0.05$. Same letter means non-significant.

Effect of feeding different levels of glucosamine and scale fish extract on some organs weight

Table (2) represents the effect of feeding different levels of glucosamine and its source on some organs weight. The liver weight of G1 as a control group recorded 4.03 ±0.04 g, there were significant difference between negative control and the other groups. The positive control was the highest weight value and G4 was the lowest one .There were significant differences among groups 3,5 and positive control and the same statical effect between G4 and G6 which fed on3g glucosamine and 15% scale fish. The kidney weight for negative control group was 0.55 ± 0.02 g, there is no significant difference between positive control and groups 3 and 5 .while group 4 recorded nonsignificant changes as compared with negative control group. The total bone weight **(**femur, tibia and vertebrae) of the negative control group was the highest weight in opposite of positive control group which was the lowest value .Adding glucose amine and its source as fish scale led to improve the bone weight , from the same table , it could be noticed that the significant reaction between positive control and the others while there is no significant between G3 and G6 . The obtained results were in the same way of **Vajranetra (2016)** who found that, glucosamine maintained body weight gain and liver organ. Also, **Bailey and Turner (2004)** reported the glucose amine improve the activity of organs weight of diabetic rats especially pancreas and kidney.

Table (2): Effect of feeding different levels of glucosamine and scale fish extract on some organ weight (g.).

Parameters	Animal groups					
	Negative control G1	Positive control G2	1.5 g glucosamine G3	3g glucosamine G4	10% scale fish G5	20% scale fish G6
Liver weight g	4.03^{c} ±0.04	5.36^{a} ±0.57	5.16^{a} ±0.35	4.48^{b} ±0.28	5.23^{a} ±0.14	5.02^{b} ±0.04
Kidney g	0.55^{c} ±0.02	0.69^{a} ±0.23	0.65^{a} ±0.29	0.53^{c} ±0.21	0.66^{a} ±0.01	0.61^{b}±0.09
Bones (femur, tibia and vertebrae) **g**	22.34^{a} ±0.02	10.41^{e} ±0.61	13.53^{c} ±0.61	19.46^{b} ±2.03	12.31^{d} ±0.62	15.48^{c} ±0.57

Each value represents the mean of 6 rats ± SD. Values significantly different compared to diabetic group: $P \leq 0.05$. Same letter means non-significant.

Effect of feeding different levels of glucosamine and scale fish extract on blood glucose (mg/dl).

Blood glucose of osteoporosis rats as affected by different levels of glucosamine and its source is illustrated in **table (3).** Blood glucose level was 89.8 ±0.87 mg/dl for negative control while positive control was the highest which recorded 150.1 ±1.2 mg/dl . All groups were

higher than negative control and lower than positive control. There were significant differences between all groups. Group 4 which fed on diet with 3% showed the lowest value. Traumatic injuries, such as breaking a bone, can lead to stress hyperglycemia. This effect can be present with or without diabetes. Insulin signaling in bone favors whole-body glucose homeostasis by activating osteocalcin. Insulin signals osteoblasts, bone cells responsible for bone formation, which tell osteoclasts, bone cells responsible for resorption, to destroy old bone. Next , the acidic (low pH) conditions created by the osteoclasts activates osteocalcin inside the bone. Finally , the active osteocalcin released from bone travels to the pancreas and stimulates the release of more insulin**(Ferron *et al.*, 2008 and Otani *et al.*, 2015). Huang *et al.*, (2017)** discovered that a hormone released by bone – known as osteocalcin – can regulate glucose levels. Osteocalcin turns on the production of insulin in the pancreas and improves the ability of other cells to take in glucose. These results are in agreement with those reported by **Zhu *et al.*(2018)** who reported that, glucosamine offered marked protection, it brought down the level of blood sugar. **Vajranetra (2016)** suggested that, glucose lowering effects are most often associated with the high level dose of glucosamine which effect on glucose and insulin levels. Another important report indicated that glucosamine can reduce glucose in diabetic rats to near the normal level (**Reginster *et al.*, 2001**).

Table (3): Effect of feeding different levels of glucosamine and scale fish extract on blood glucose (mg / dL).

Parameters	Animal groups					
	Negative control G1	Positive control G2	1.5 g glucosamine G3	3g glucosamine G4	10% scale fish G5	20% scale fish G6
Blood glucose	89.8^{f} ±0.87	150.1^{a} ±1.2	135^{c} ±4.6	111.1^{e} ±0.5	140.9^{b} ±4.3	128.6^{d} ±2.7

Each value represents the mean of 6 rats ± SD. Values significantly different compared to diabetic group: $P \leq 0.05$. Same letter means non-significant.

Effect of feeding different levels of glucosamine and scale fish extract on liver enzymes

The effect of feeding different levels of glucosamine and its source on liver enzymes is shown in **table (4).** The negative control presented a level of 25.1 ± 0.27 u/L for aspartames amino transferase (**AST**) , while positive control (+) presented 40.2 ± 0.11 U/L. There is no significant differences between negative control (-) and groups 4 and 6. Also, There is no significant differences between groups 3 and5.

In the same table, data showed that the negative control recorded 29.8 ± 0.31 U/L for alanine amino transferase (**ALT**), while positive was more than the negative control and the other groups which recorded

48.9 ±5.51 U/L. There is no statistical differences between groups 3 and 5 and the same effect was found between 4 and 6 .

In case of alkaline phosphates (**ALP**), the negative control was the lowest value while the positive one recorded the highest level .Groups 3 and 5 showed statistical differences when compared with both controls while, there is no significant differences between each other. Also, Groups 4 and 6 showed no statistical differences between each other.

Osteoporosis poses a major burden on health care systems worldwide, and its importance is bound to increase even further because of the demographic transition . Decreased bone mineral density (**BMD**) predisposes individuals to fragility fractures that often result in a loss of self-sufficiency and permanent disability and are indeed associated with substantial excess mortality . Early identification of subjects at particular risk of developing osteoporosis would allow targeted interventions and could help to prevent manifest osteoporosis and its complications. Evidence accumulated in recent years has shown that physiological serum levels of liver enzymes (within and just above the normal range) are associated with an increased risk of incident cardiometabolic disease and mortality from various causes in the general population . It is also well known that chronic liver disease is associated with profound adverse effects on bone health and homeostasis (**Owens *et al.*, 2004).**

These results are investigated by **Kirkham and Samarasinghe (2009)** who mentioned that, glucosamine affected of the activities of aspartame amino transferase (AST), alanine amino transferase (ALT), alkaline phosphates (ALP), and they were significantly decreased in both plasma and liver. Also, **Huskisson (2008)** showed that glucosamine can improve the functions and kept them near to the normal levels. While, **Smith and Dillon (2009)** stated that there is no effect of glucosamine on the liver activity.

Table (5): Effect of feeding different levels of glucosamine and scale fish extract on liver enzymes (U/L)

Parameters	Animal groups					
	Negative control G1	Positive control G2	1.5 g glucosamine G3	3g glucosamine G4	10% scale fish G5	20% scale fish G6
AST(U/L)	25.1^{c} ±0.27	40.2^{a} ±0.11	32.1^{b} ±2.50	25.1^{c} ±1.56	35.5^{b} ±0.21	27.7^{c} ±3.15
ALT(U/L)	29.8^{d} ±0.31	48.9^{a} 5.51±	44.4^{b} ±2.51	37.7^{c} ±3.52	45.4^{b} ±4.5	38.7^{c} ±2.25
ALP (U/L)	80.1^{d} ±0.97	107.7^{a} ±2.41	98.1^{b} ±4.01	89.7^{c} ±5.16	100.7^{b} ±4.16	93.7^{c} ±1.35

Each value represents the mean of 6 rats ± SD. Values significantly different compared to diabetic group: $P \leq 0.05$. Same letter means non-significant.

Effect of feeding different levels of glucosamine and scale fish extract on kidney functions (mg/dl)

The effect of feeding different levels of glucosamine and scale fish on kidney function is illustrated in **table (6).** Concerning creatinine, group (1) showed statistical difference all groups. There is no significant between group received 1.5% glucosamine and group fed on10% scale fish and also, the groups 4 and 6 had the same statical effect .

For albumin levels , the values of tested groups showed non significant differences when compared to both of controls.

In case of uric acid , the negative control (+) group showed the lowest value while the positive was the highest . There is no statistical difference among negative, fourth and the sixth groups for increasing the glucosamine level .

The positive control group showed high level of urea nitrogen . The levels of urea decreased by increasing the receiving of glucosamine and its source There is no significant differences between groups 3 and 4.

Studies showed that disturbance parathyroid hormone levels, abnormal calcium and phosphate metabolism, and vitamin D deficiency can have negative effect on renal functions **(Fransen *et al.*, 2015**). Mentioned results in the presents study showed that, the treatment of glucosamine decreased blood glucose and blood urea levels and had no effect on the serum proteins and albumin levels (**Huskisson, 2008).**

Also, in the present study showed that glucosamine can reduce serum urea and creatinine levels and confer a protective effect on the kidney (**Park *et al.*,2021).**

Table (6): Effect of feeding different levels of glucosamine and scale fish extract on kidney function (mg / dl).

Parameters	Animal groups					
	Negative control G1	Positive control G2	1.5 g glucosamine G3	3g glucosamine G4	10% scale fish G5	20% scale fish G6
Creatinine mg/100ml	0.69^{d} ±0.31	1.26^{a} ±0.21	1.12^{b} ±0.07	1.07^{c} ±0.02	1.14^{b} ±0.21	1.09^{c} ±0.14
Albumin mg/100ml	3.84^{a} ±0.152	3.81^{a} ±1.05	3.79^{a} ±0.05	3.75^{a} ±2.31	3.81^{a} ±1.32	3.82^{a} ±0.01
Uric Acid mg/100ml	2.35^{d} ±0.15	2.95^{a} ±0.22	2.82^{b} ±1.00	2.75^{b} ±0.11	2.85^{b} ±2.5	2.66^{c} ±1.2
Urea Nitrogen mg/100ml	25.2^{c} ±1.15	40.46^{a} ±0.1	35.76^{b} ±1.3	28.21^{c} ±0.7	35.15^{b} ±2.1	29.11^{c} ±1.2

Each value represents the mean of 6 rats ± SD. Values significantly different compared to diabetic group: $P \leq 0.05$. Same letter means non-significant.

Effect of feeding different levels of glucosamine and scale fish on serum calcium, serum phosphorous ,BMD and some hormones.

Data in **table (7)** shows the effect of different levels of glucosamine and its source on serum calcium, serum phosphorous ,BMD and some hormones. Serum calcium level in negative control group was significantly higher when compared with osteoporosis rats (C+) . Meanwhile, the effect of different levels of glucosamine and its source on calcium level for osteoporosis rats were improved compared to control (+) group. There is no significant between groups 3 and 6.

According to the same table, the level of serum phosphorus was decreased in positive control group when compared to negative one, while their level increased by increasing the levels of treatment glucoseamine. The increasing was nonsignificant in G4 as compared to negative control group.

As for the level of serum Parathyroid hormone for positive control showed significant differences as compared with the other groups. Both in case G3&G5 and G4&G6 were nonsignificant between each other. Calcitonin levels showed a gradual increase with the increase of supplement level and G4 and G6 revealed nonsignificant differences.

For serum Osteocalcin hormone level, the effect of different levels of glucosamine and its source were significantly decreased compared to control (+) group. There is no significant between groups 3and 5.

BMD for negative control presented the high level while the positive control group recorded the lowest level . The means with glucoseamine and its source at the levels 3% and 10% respectively are 15%nonsignificant.Also, the same effect between the level 1.5 % and 15%.Osteoporosis is a metabolic bone disease characterized by loss of bone mass thus making the bone more susceptible to fractures. The results showed that the concentration of calcium level which is considered the principal stimulus for the secretion of calcitonin by C-cells. When blood calcium is lowered, the stimulus for calcitonin secretion is diminished. Also the actions of PTH and calcitonin are antagonistic on bone resorption. Further, evidence of osteoporosis resulting from ovariectomy was indicated by the present elevation of PTH level. The elevated level of PTH causes additional loss of BMD. Moreover, hyperparathyroidism which may cause calcium change associated with a compensatory rise in PTH resulting in calcium release from the skeleton thus causing bone loss in osteoporosis rats **(Pavelka *et al.*, 2003).**

Glucosamine appears to help reduce the weakening of bones by supporting healthy bone growth. it is most important for the formation

of hyaluronic acid, chondroitin sulfate as well as keratan sulfate, which are—aside from the collagen fibers—the most important components of the extracellular matrix of the articular cartilage and the synovial fluid (**Reginste *et al.*, 2001 and Towheed *et al.*, 2005).**

Table (7): Effect of feeding different levels of glucosamine and scale fish extract on serum calcium, serum phosphorous ,BMD and some hormones.

Parameters	Animal groups					
	Negative control G1	Positive control G2	1.5 g glucosamine G3	3g glucosamine G4	10% scale fish G5	20% scale fish G6
Serum calcium (mg/dl)	8.99 a ±0.76	6.33 d±1.11	6.89 c±0.99	7.57 b±0.75	6.51 d±0.57	7.04 c±1.03
Serum phosphorous (mg/dl)	3.21 a ±0.005	2.15 c±0.65	2.33 c ±3.5	2.98 a±10.96	2.06 c ±9.6	2.76 b ±2.5
Parathyroid (pg/ml)	15.1 d ±2.1	25.5a ±1.5	22.11 b ±0.76	19.33 c ±2.08	23.53 b ±1.5	20.66 c±6.02
Calcitonin (pg/ml)	4.6 5 a ±0.05	3.12 d ±0.11	3.46 c ±0.16	4.06 b± 0.85	3.33 c ±1.5	4.01 b ±6 0.27
Osteocalcin (pg/ml)	3.99 e±0.16	5.48 a ±0.87	5.12 b±0..55	4.36 d±1.03	5.23 b±0.64	4.78 c±0.36
BMD (g/cm^2)	0.272 a ±0.05	0.149 d±0.08	0.167 c±0.003	0.199 b±0.02	0.159 c±0.01	0.192 b±0.01

Each value represents the mean of 6 rats ± SD. Values significantly different compared to diabetic group: $P \leq 0.05$. Same letter means non-significant.

Conclution

Glucosamine consumption with recommended dose as drug or natural supplement can improve the biological and biochemical parameters of osteoporotic rats especially the responses of the rat serum calcium and other hormones which related to bone structure.

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تأثير الجلوكزامين ومصدرة قشور الاسماك على هشاشة العظام في الفئران

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ملخص البحث

هشاشة العظام المرتبطة بعوامل مختلفة بما في ذلك انقطاع الطمث والشيخوخة هي اكثر امراض العظام الأيضية المزمنة شيوعا ، وعلى الرغم من انه يظهر في جميع الفئات العمرية مع اختلاف الجنس والأعراق ، الا انه أكثر شيوعا عند القوقازين وكبار السن والنساء.

هدفت الدراسة التعرف على مدى تناول الجلوكوزامين ومصدره قشور الأسماك على المؤشرات البيوكيميائية لهشاشة العظام في الفئران. تم تقسيم 36 فأر من اناث الفئران البيضاء (وزن 150± 5 جم) بطريقة عشوائية الي 6 مجموعات : تم تغذية الأولى على الوجبة الأساسية كمجموعة ضابطة سالبة وتم حقن بقية الفئران ب 0.8 / ملجم / كجم من وزن الجسم ديكساميثازون لمدة 3 أيام للأصابة بهشاشة العظام تم استخدام المجموعات الخمس ك مجموعة ضابطة موجبة واستمر تغذيتها على الوجبة الاساسية فقط واعطاء بقية الفئران 1.5 و 3 جم / يوم و 10 و 20 % من مستخلص قشور الأسماك لمدة 28 يوم.

اظهرت النتائج أن اعطاء مصادر الجلوكوزامين اظهر تحسنا في وزن الجسم ، المأخوذ الغذائي ونسبة الاستفادة من الغذاء كذلك حدوث تأثير معنويا في جميع المؤاشرات البيوكيميائية في الفئران المصابة بهشاشة العظام مقارنة بالمجموعة الضابطة لذلك يوصي بتناول مصادر غنية الجلوكوزامين للوقاية من حدوث هشاشة العظام عند كبار السن وخاصة السيدات.

الكلمات المفتاحية : الجلوكوزامين – مستخلص قشور الأسماك – المؤاشرات البيوكيميائية – هشاشة العظام – ديكساميثازون