Referees Report ob Revised manuscript: JAMPS_24382. **Ameliorative Effects of Skimmed Milk and Tuna Fish in the Dietary Management of Osteoporotic Female Rats; A Comparative Study**

General remarks:

The script is much improved; there are many minor grammatical and spelling mistakes. Have tried to rake through and indicate these below. Extra edits still needed.

The whole Methods and Materials must be re-scripted in the journal style for reading, without numerical headings. See notes on lines 76-146 below.

There is an overall lack of discipline in checking and no due diligence to detail. For example (☹1) they start with 66 rats; ten (10) are separated group 1, leaving the other 50 which are accounted for. Total 60; what happened to the other 6 rats? (2) Also see Lines 175, 218 a1nd 242. Where does "sardine powder" come in?? This is a spurious confounder and an inconsistency which casts doubt on the integrity of the whole experiment. Kindly check and correct this variable.

Lines 329-377: DISCUSSION: This section is verbose, over-written and needs to be shortened by at least half. Linking words like meanwhile shd be omitted. **Over analysis leads to paralysis.** High first class protein combined with adequate calcium in the diet favors minimizing developing OP. Each statement selected shd bear in mind that the statement(s) shd support this, as this insight is the authors.

Figures: Obviously much effort went into obtaining these histopathology slides. The slides show clear changes. But these are labeled as "Photo"; they are not photographs, but colored micrographs. They must be correctly referred to as **Figures** in the text, and also labeled as a **Figure** with an explanatory proper legend. See notes below.

This is a good piece of research and shd be improved before proceeding to publishing; it still needs sound detailed revision.

Specific Comments:

The Ms script is in black. Comments in red. Suggestions in blue.

Line 10: Aim: This study was carried out to comparatively investigate the ameliorative effects of skimmed milk and whole dried Tuna fish in osteoporotic female rats. Grammar. Prejudgment shd be avoided. Suggest: This investigation was carried out to compare the moderating effects of skimmed milk and whole dried Tuna fish in osteoporotic female rats.

Line 12: Method: Sixty six female albino rats. What strain? Sprague –Dawly? Wistar?; you must mention the strain here, to ensure genetic homogeneity. Some rats may be bred to be OP-prone. This is mentioned later in the M&M of the Ms..

Line 27: The best results were observed of rats fed diet supplemented with 10% skimmed milk. Suggest: The best results were observed in rats fed diet supplemented with 10% skimmed milk.

Line 34:. Osteoporosis (OA) is a disease… Osteoporosis (OP) is a disease

Line 34-36: Osteoporosis (OA) is a disease predominantly involving deterioration of articular cartilage which is marked clinically by a gradual onset of pain, stiffness and loss of mobility in synovial (weight-bearing) joints. Does this apply to rats? In humans the intramembranous and cartilaginous derived bone, (as cortex and trabeculae) and the juxtaposed bone forming cartilage are also affected. Suggest: Osteoporosis (OP) is a disease predominantly involving deterioration of cartilage and bone; the loss of articular cartilage is marked clinically by a gradual onset of pain, stiffness and loss of mobility in synovial (weight-bearing) joints.

Line 36: The therapeutic approach is mainly at the relief of symptoms. **Suggest:-** The therapeutic approach is targeted mainly at the relief of symptoms.

Line 41: Osteoporosis is a major complication in patients who requires chronic glucocorticoid treatment [4]. Sense not clear. **Suggest:** Osteoporosis is a major complication in patients who are usually treated with chronic glucocorticoid treatment [4].

Line 47: … that about 44 million Americans or 55 % of people 50 years and alder are both osteoporosis and low bone mass [7]. Grammar and spelling. **Suggest:** …that about 44 million Americans or 55 % of people 50 years and older are both osteoporotic with low bone mass [7].

Line 83: (without head) was …(without heads) were…

Line 93: Table 1: This has a heading. It also needs an **explanatory legend at the bottom** which must include all the notes on the minerals and vitamins. **Suggest: Table 1: Composition (g/kg) of the basal diet (BD).** Note: Group 2 had skimmed milk, Group 4 and Group 5 had 15g/kg and 30g/kg respectively. **Vitamins etc etc**

Lines 76-145. Change of style and numerical headings. THIS IS AN ARTICLE for a scientific journal to be read; this seems to have been exerpted from a thesis or lab-report. The headings must be clear and numerals minimized or removed.

Line 175: 5% sardine fish powder….the variable is dried Tuna fish!!! Where does sardine powder come in??

Line 218: Table 5 shows the effect of skimmed milk and dried sardine fish….. the variable is Tuna!!! Where does sardine powder come in??

Line 242: Table 6 shows the effect of skimmed milk and dried sardine fish powder on calcium…….. the variable is Tuna!!!

Line 301: Photo 2: Photomicrograph of femur of experimental rats from positive control group showing thin bone cortex and dilatation of bone marrow cavity (H and E x 200). **Suggest:- Figure 2:** Photomicrograph of femur of experimental rats from positive control group showing thin bone cortex (black arrow) and dilatation of bone marrow cavity (H and E x 200).

Line 306: Photo 3: Photomicrograph of femur of rats from positive control group showing cracks and necrosis in bony cortex (H and E x 200).**Suggest: Figure 3:** Photomicrograph of femur of rats from positive control group showing cracks and necrosis (black arrows) in bony cortex (H and E x 200).

Lines 287: Photo 1: These are micrographs of histology slides. Suggest: Figure 1,2,3,4,5,6,7,8, etc. Each Figure must have its own legend: **Suggested examples: Figure 1:** Photomicrograph of femur of experimental rats from negative control group showing no histopathological changes . White horizontal lines of scale are discernable. (H and E x 200).

Figure 2: Photo 2: Photomicrograph of femur of experimental rats from positive control group showing thin bone cortex and dilatation of bone marrow cavity (H and E x 200). **Suggest: Figure 2:** Photomicrograph of femur of experimental rats from positive control group showing thin bone cortex (arrow) and dilatation of bone marrow cavity (H and E x 200).

Line 348 {31]….[31]

Lines 465;466;467;470;472;476. Refer to Figures 1;2;3;4;5;6; etc

Line 482: These findings show that skimmed milk and Tuna fish are effective in positively moderate metabolic dysfunctions associated with osteoporosis in DOPI rats. 10% skimmed milk dietary supplementation is more therapeutically effective compared with the other three methods. This may have implications for humans, which indicates for more research into this important field of medicine. **Suggest: .** These findings show skimmed milk and Tuna fish are effective at positively moderating metabolic dysfunctions associated with osteoporosis in DOPI rats. 10% skimmed milk dietary supplementation is more therapeutically beneficial when compared with the other three methods. This may have implications for humans, which indicates for more research into this important field of medicine.