

RESEARCH ARTICLE

ORTHOPAEDIC

THE POTENTIAL FOR STRUCTURAL MODIFICATION HENCE BENEFIT IN OSTEOARTHRITIS OF KNEE BY MINERAL SUPPLEMENTATION –PILOT STUDY

DR HIMANSHU BANSAL* AND DR ANUPAMA BANSAL

Foundation, Rudrapur, Uttarakhand



DR HIMANSHU BANSAL
Foundation, Rudrapur, Uttarakhand

ABSTRACT

The potential for structural modification by natural mineral supplement was evaluated in a pilot perspective, study of six months duration in twenty patients with moderate Osteoarthritis of knee. WOMAC values (subsets and total) showed significant improvement from baseline over the course of six months treatment ($p < 0.001$). The pain free distance covered during a Six minute walk (6MWD) also significantly improved by 140 feet ($p < .001$). Requirement of rescue medication decreased to once a week in 45%. The mean (SD) change for the percentage of painful days was 45.0 (38.7). 25% patients demonstrated improved cartilage thickness by at least 0.01 mm as against 15% who lost thickness at least by 0.01 mm. 55% patients showed improved Cell counts (below 500 microlitre) in Synovial fluid. Hematological and Biochemical safety variables showed that mineral supplement is absolutely safe. The study suggests that natural mineral supplementation helped in synovial metabolism and in reducing cartilage pathology hence had structural efficacy.

KEY WORDS

Osteoarthritis (OA) of Knee, Concentrace Trace Mineral Drops-(CTMD®), WOMAC, Clinical Trials on Osteoarthritis, Food Supplements in Osteoarthritis

INTRODUCTION

Osteoarthritis (OA) is a degenerative disease characterized by focal areas of loss of particular cartilage in synovial joints, subchondral bone changes, osteophyte formation at the joint margins, thickening of the joint capsule and mild synovitis. ACR Guidelines for Medical Management of Osteoarthritis of the knee recommend patient education to reduce weight, quadriceps strengthening exercise, Non-steroidal anti-inflammatory drugs (NSAIDs) and chondroprotection.(1)

NSAIDs are the mainstay of management for pain, but since they may cause serious gastrointestinal and cardiovascular adverse events (2), thus, search for structure modifying therapeutic solutions that could lead to an enhancement of tissue regeneration and the reduction of degenerative mechanisms has been initiated

Inconsistent results for chondro-protection by nutraceuticals like glucosamine, chondroitin, intrarticular hyalran emphasize the associated role of cofactors, vitamins and minerals from diet. (3, 4, 5) Minerals and trace elements like Boron, Zinc, Copper, Selenium, Magnesium, Manganese, Vitamins A, E and C, Niacin, Pantothenic Acid, Omega 3 fatty acids, chondroitin sulphate, Glucosamine, and Sulphur containing amino acids play a significant role in the production of cartilage matrix. In vitro studies suggested that these trace minerals may favorably affect the synthesis of proteoglycans. The trace element copper is an essential cofactor in enzymes such as the collagen cross-linker lysyl oxidase (6, 7, 8, 9).

Naturally occurring minerals such as magnesium, copper, manganese, selenium and zinc have shown anti-inflammatory effects in both animal and human studies. In a rat model

of osteoarthritis, a deficiency of dietary magnesium was demonstrated to enhance the amount of cartilage damage. Furthermore increased magnesium in the diet may influence inflammation through reducing the serum level of the pro-inflammatory protein C-reactive protein [10, 11].

Recent evidence has suggested an excess of reactive oxygen species arising from an imbalance in the antioxidant status of the joint (such as reduced levels of SOD) may result in cartilage degradation and joint remodeling [12].

Anti-oxidant enzyme super oxide dismutase requires copper, zinc and manganese as cofactors. It was demonstrated in the Haqqi model of human cartilage explants that mineral supplementation reduced cartilage degradation in response to IL-1 β , as well as nitric oxide production secondary to the induction of inducible nitric oxide synthase. (13, 14)

Several clinical studies have also reported the effectiveness of minerals in the management of osteoarthritis. (15,16,17,18) However, the results of the available studies were mixed and did not provide confirmed evidence of structural benefit.

This study was intended to explore whether clinical benefit observed with adequate mineral supplementation in osteoarthritis was related to chondroprotective structural modification.

MATERIALS AND METHODS

The patients who were above 50 years of age, and presented with symptomatic primary Osteoarthritis of the knee (1), defined by daily pain for the previous three months, analgesics usage at least once a week, less than thirty minutes of morning stiffness and a WOMAC

score of ≤ 75 in the target knee, were included in the study. The food supplement selected was Concentrate Trace Mineral Drops (CTMD). It was selected because of easy availability in market, being a derivative from natural sources and already consumed by masses.

The supplement contained over 72 natural minerals in ionic form, concentrated from the Great Salt Lake in Utah. Half tsf (approx 40 drops) serving contained magnesium 250 mg, chloride 690mg, sodium 5mg potassium 3mg, sulfate 37 mg lithium 295mcg, boron 370 mcg. In addition it also contained naturally occurring varying trace amounts of bromide, carbonate, calcium, silicon, nitrogen, selenium, phosphorus, iodide, chromium, manganese, titanium, rubidium, cobalt, copper, antimony, molybdenum, strontium, zinc, Nickel, tungsten and other elements. Refer Table 1 for typical Mineral Composition of supplement used.

The radiographical eligibility criteria included

Kellgren Lawrence classification for knee Osteoarthritis of grade 0, 1, 2 or 3 (Table 2a), Brandt Radiographic Grading Scale of Osteoarthritis grade 1 and 2 (Table 2b), Ahlback Radiographic Grading Scale of Osteoarthritis of the Tibiofemoral Joint 0 & 1 (Table 2c). If both knees were symptomatic, only the most painful one was taken into account.

The patients with evidence of secondary knee Osteoarthritis, severe Osteoarthritis (JSW < 2 mm), prior intra articular injections and corticosteroids, oral Diacerin within the previous three months, prior to inclusion and patients with clinically significant systemic disease were excluded.

A total of twenty patients were enrolled & advised to take mineral supplement twice daily for six months (24 weeks), on an empty stomach. The dose was gradually increased to 40 drops ($\frac{1}{2}$ tsf) from 5-10 drops a day in a week's time.

Table 1
Mineral composition of supplement

Supplement Facts		
Serving Size $\frac{1}{2}$ teaspoon (about 40 drops)		
Servings Per Container 25		
Amount Per Serving		%DV
Magnesium	250 mg	63%
Chloride	690 mg	20%
Sodium	5 mg	<1%
Potassium	3 mg	<1%
Sulfate	37 mg	†
Lithium	395 mcg	†
Boron	370mcg	†

† Daily Value not Established.

Ingredients: Ionic Sea minerals. Contains no other added ingredients. In addition to the elements listed above, this product contains the following in naturally occurring, varying trace amounts: Bromide, Carbonate, Calcium, Silicon, Nitrogen, Selenium, Phosphorus, Iodide, Chromium, Manganese, Titanium, Rubidium, Cobalt, Copper, Antimony, Molybdenum, Strontium, Zinc, Nickel, Tungsten, Germanium, Scandium, Vanadium, Tellurium, Tin, Lanthanum, Yttrium, Silver, Gallium, Bismuth, Zirconium, Cerium, Cesium, Gold, Beryllium, Hafnium, Samarium, Terbium, Europium, Gadolinium, Dysprosium, Thorium, Holmium, Lutetium, Erbium, Ytterbium, Neodymium, Praseodymium, Niobium, Tantalum, Thallium, Rhenium, Indium and Palladium, plus the other elements found in seawater.

**Table 2. Radiological Criteria for inclusion:
 [(a)Kellgren – Lawrence Grading Scale (b). Brandt Radiographic Grading Scale (c) Ahlback Radiographic Grading Scale of Osteoarthritis of the Tibiofemoral Joint.]**

Grade	Criteria
0	Normal
I	Doubtful narrowing of joint space, possible osteophyte development
II	Definite osteophytes, absent or questionable narrowing of joint space
III	Moderate osteophytes, definite narrowing, some sclerosis, possible joint deformity
IV	Large osteophytes, marked narrowing, severe sclerosis, definite joint deformity

Grade of Osteoarthritis	Description
0	No radiographic findings of osteoarthritis
1	Joint space narrowing < 3 mm
2	Joint space obliterated or almost obliterated
3	Minor bone attrition (< 5 mm)
4	Moderate bone attrition (5–15 mm)
5	Severe bone attrition (> 15 mm)

Grade of Osteoarthritis	Description
0	No radiographic findings of osteoarthritis
1	< 25% joint space narrowing with secondary features
2	50–75% joint space narrowing without secondary features
3	50–75% joint space narrowing with secondary features
4	> 75% joint space narrowing with secondary features

Table 3
Baseline characteristics (n= 20)of patients–with medial knee OA

Baseline characteristics		(n=20)
Demographic characteristics		
Age (years) mean		54 (7.4)
Sex (n=male)		12
Weight (kg)		70 (12.9)
Height (cm)		162(7.6)
Right knee, No		13
Pain score on 0–100 VAS		
		58.4 (12)
Womac score	total	52-62 (2.4) Mean-51.4
6MWD		1226-1488 Mean 1290
Cartilage thickness (mm)		4.48 -4.52(1.1) Mean 4.50
Joint space width (mm)		4.56-4.64 Mean 4.6
Osteophyte score (4 grades No (%))	0	1
	1	10
	2	8
	3	1
Kellgren and Lawrence score (5 grades), No (%)	0	0 (0)
	1	1
	2	1
	3	17
	4	1
Cell counts no/ microlitre		240-1800 (480)

At the baseline visit, besides vital signs & laboratory tests the subjects were assessed for WOMAC score, 6 MWD, radiological assessment of joint space width, ultrasonological measurement of articular cartilage thickness and cell counts of synovial fluid.

Patients were evaluated weekly for any adverse events and need for any rescue medication (NSAIDs) in the first month followed by monthly evaluation up to six months. WOMAC score and 6 MWD assessments were carried out every month. X-Ray, Ultrasonography, Synovial fluid assessment and Laboratory tests were repeated only at the end of 6 months.

The Western Ontario and McMaster Universities Osteoarthritis Index WOMAC (19) is a widely

used measure of patient’s subjective assessment of pain joint mobility and physical disability. It evaluates 3 dimensions, namely pain, stiffness and physical function with 5, 2, and 17 questions respectively. Total maximum score was 96 and minimum was 0. Each subscale was summated to a maximum score of 20, 8 and 68, respectively.

The six minute walking distance was carried out by marking off a 50 meter distance in an interior hallway and asking subjects to walk as far as they can and as quickly as they can over 6 minutes. The total distance was measured and recorded. (20)

Anteroposterior radiographs of the knee joints were obtained with patients in a weight bearing position, joint fully extended, standing at 1

meter from the X- Ray source, using previously published guidelines. Width was measured of the narrowest point of the JSW (minimal JSW). This progression was defined by a JSW loss of more than 0.50 mm during the study, as previously reported (21, 22). Ultrasonographically, articular cartilage on weight bearing condyle appears as hypo echoic band with sharp anterior and posterior margins. It is thickest over intercondylar area (8 – 10 mm) and thinnest over femoral condyles (average 4 – 5 mm) (23, 24).

Cellularity of synovial fluid was compared by aspirating 2 ml of fluid under aseptic conditions. Normal synovial fluid has cell count of less than 2000/microlitre with a predominance of mononuclear cell. Increased counts up to 5000/microlitre with predominance of polymorphonuclear leucocytes are seen in osteoarthritis. (6)

Tolerability and safety assessments included any symptoms and signs referred by patient and also by laboratory based hematological and

biochemical assays. Adverse effects were categorized as isolated, intermittent or continuous depending on interference with the subject’s daily activities as mild, moderate or severe. Possible causal relationship with the mineral supplement in terms of Definite / Possible / Probable / Non Assessable / None was also arrived at. The study was approved by ethics committee and all the patients were well informed and gave written consent to participate in the study.

RESULTS

A total of 32 patients were screened, of which 20 were considered eligible and enrolled in this pilot study. The most common reason for exclusion was either a lack of radiographic evidence of knee OA or severe disease with a JSW <2 mm at the narrowest point. No patients discontinued the treatment till the end of the study. Table 3 summarizes the main baseline characteristics of the 20 patients.

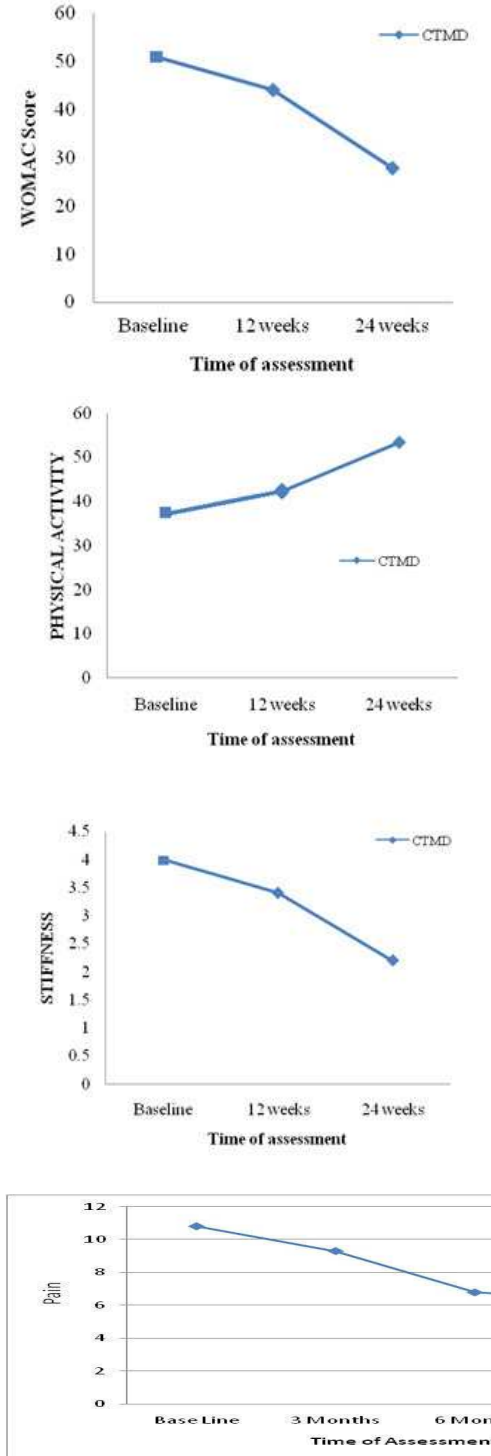
Table 4
Change in WOMAC scores, subscore of pain physical activity, and stiffness from baseline over 6 months of treatment

CTMD (n=20)	Baseline	3months	6m
Total	50- 64 (51.2)	40- 59 (46.2)	29- 54 (41.5)
Pain	10 - 12 (10.8)	8-11 (9.5)	6-10 (8.0)
Stiffness	4 -4 (Av 4)	3-4 (3.5)	2-4 (3.0)
Physica	36- 48 (37.4)	30-40 (35.0)	22-36 (29.0)

Symptomatic outcome measures showed significant improvement from baseline in the WOMAC scores (subsets and total) over the course of six months of treatment ($p < 0.001$). It was noted that after 12 weeks of treatment though the WOMAC scores had marked improvement over baseline but the changes

were not significant ($p > 0.05$). The magnitude of Pain reductions appeared at week 4, but was not significant. Table 4 summarized the change in WOMAC scores, sub score of pain physical activity, &stiffness, 6MWD from baseline over 24 weeks of treatment. (figure 1)

Figure 1
Changes from baseline over 124 weeks of treatment in WOMAC scores (A), subscore of physical activity (B), pain (C) &stiffness (D)



Percentage of responders i.e. the percentage of patients having an improvement of at least 30% in pain on VAS or reduction in pain by WOMAC sub score from baseline to the final visit was 55%.

The pain free distance covered during a 6 minute walk had significantly improved by 140 feet over 6 months (($p < .001$) on treatment with the mineral supplement. (Figure 2)

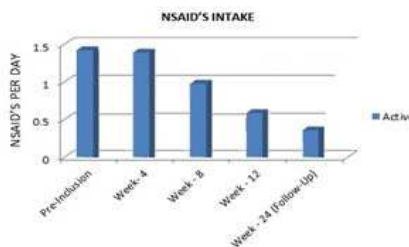
Figure 2
Improvement in six minutes pain free walking distance



An improvement of 100 feet over 6 months was noticed in 5 (25%) patients. All the patients needed Rescue medication at least twice a week (Nsaid –Lornoxicam 8mg SOS) till initial 2 weeks, and the requirement of NSAIDs decreased to once a week in 4 patients (20%), 6 (30%) 9 (45%) by 8, 12, 24 weeks,

respectively. From baseline to the final visit, 7 (35%) patients were NSAIDs free for last 30 days, and the mean (SD) change for the percentage of painful days was 45.0 (38.7). Only 4 patients had no change in the rescue medication requirement. (Figure 3)

Figure 3
Reduction in need of rescue medication



Radiology did not reveal any increase in joint space width (JSW) in any patient. The study group had insignificant deterioration in the JSW (mean SD 0.05, $p > 0.82$) with only 3 patients (15%) with a reduction in JSW > 0.05 mm ($p > 0.90$) hence labeled progress or. Ultrasonographically, at six months the cartilage thickness improved by at least 0.01 mm in 5 (25%) patients as against 3 patients (15%) who lost thickness at least by 0.01 mm. Synovial fluid examination suggested that it helped in restoring synovial fluid rheological properties and synovial metabolism and in

reducing cartilage pathology. 11 (55%) patients had cell counts below 500/ microlitre by 24 weeks compared to $n=4$ (20%) before the treatment.

Average cell count reduced to 240/ microlitre from a value of 480/ microlitre at the start of study.

No significant change in Blood pressure (systolic and diastolic), respiration rate and pulse rate were noted from screening of the study, at the 5 intermediate evaluation points till the study's conclusion. Only 2 subjects' i(10%) experienced adverse effects with Mineral

supplementation at these doses. The adverse effects were related to upper GI discomfort , mild to moderate in intensity and completely resolved by 8 -12 weeks . A summary of

Hematological and Biochemical safety variables is depicted in Table 5 shows that this mineral supplementation was absolutely safe

Table 5
safety Assessments-- haematological and biochemical parameters observed during the study

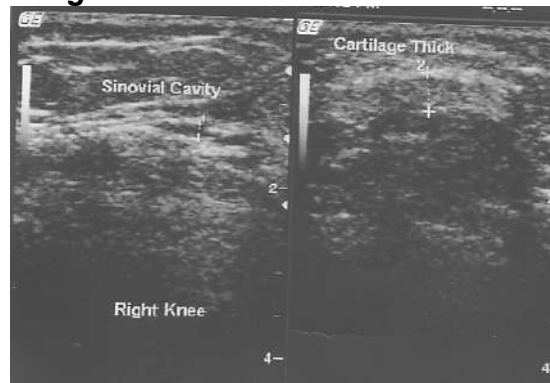
	Screening	Follow-up at 24 week
Laboratory Tests:		
Hematology:		
CBC: Hemoglobin	11.4	11.6
PCV	38	40
Platelet Count/mm ³	200- 400	210-400
TLC/mm ³	4.2-9.4	4-9
Absolute neutrophil count/mm ³	5-7	5-7
Absolute lymphocyte count/mm ³	2-2.	2-2.4
Absolute RBC count millions/mm ³	4.0-4.4	4-4.2
SGPT	16- 18	16-18
SGOT	18-20	16-20
Serum Creatinine	0.8- 0.9	0.8-0.9
Glucose (F)	80-	80-100

Figure -4
Ultrasound images of patients showing increase in cartilage width

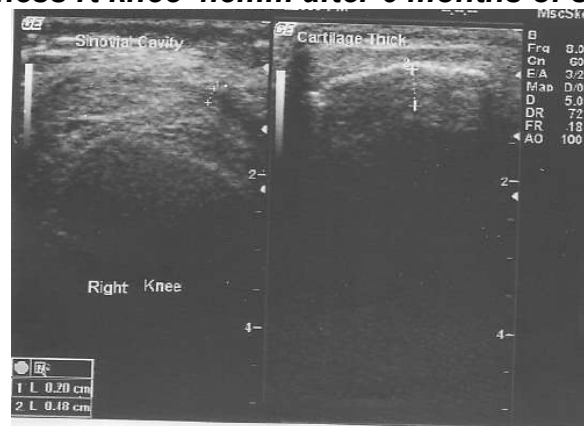
A: Patient 1:
cartilage thickness rt knee-4.7mm after 6 months of supplementation



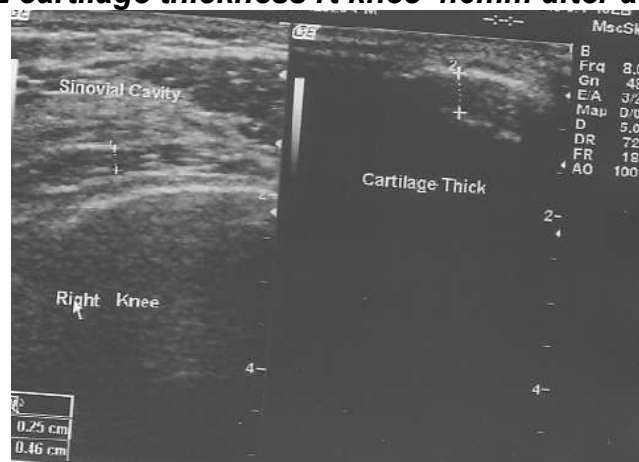
Patient 1 cartilage thickness rt knee-4.6mm after at baseline



***Patient 2
cartilage thickness rt knee-4.8mm after 6 months of supplementation***



Patient 2 cartilage thickness rt knee-4.6mm after at baseline



DISCUSSION

Recent documentation of an increased risk for cardiovascular disease and stroke with COX-2 inhibitors and significant gastrointestinal, renal

complications and premature deaths associated with non-selective COX inhibitors (2), along with the NSAIDs which provided only symptomatic

relief but did not abrogate the disease process, and has raised a need for search of better and safer alternatives.

In osteoarthritis, the exact mechanism remains controversial. It is suggested that there is a mismatch between regenerative and degenerative forces that govern cartilage formation, partly because of a malfunction in proteoglycan synthesis (6). The rationale for a general therapeutic application of mineral supplementation stems from this hypothesis.

Mineral supplementation can be of most benefit if minerals are in balance with other elements they interact with. Ionic form balance of these mineral elements has to be maintained for better results. Trace elements, bound in chelated form, are more readily available to the body and less likely to interact and interfere with each other during absorption. (7,9)

The dose of the mineral supplement was determined based on available literature about daily dietary allowance of various minerals (6). Patients with severe Osteoarthritis were excluded as they have severely damaged cartilage. Obviously the number of healthy synoviocytes in these patients is poor, which eventually means that there are not enough healthy cells to act upon and would not benefit from conservative treatment including dietary and viscosupplementation.

The onset of benefits is evident from week 3 or 4 onwards, there was an improvement of 9.6% over baseline walking distances at 24 weeks. Although, these distances appear to be small, our subjects indicated that the ability to walk even a little bit further was important to them. Need of rescue medication was reduced by 45%. This early onset of benefits as early as one week in some patients is not inconsistent with the in-vitro studies demonstrating the protection of human cartilage degradation induced by IL-1 β . However, the present study does not directly assess whether protection of against cartilage degradation was associated with the therapies, nor is it likely that a substantial change in joint architecture would occur in this timeframe (10, 11, and 12).

Our extensive literature search did not yield any study that present objective clinical data showing beneficial effect of mineral supplementation in joint health. Thus, we compared our results with subjective data of other double blind placebo controlled studies (11, 12, 13, 14) and found them comparable.

Some of the comparable studies (15,16,17,18) included Joy L Frestedt et al (n = 50, improvement in WOMAC P < 0.001, 6 MWD of 7% over 3.5%), Mark JS Miller, (n = 91 improvement in WOMAC Total 38 – 43% versus 27% and VAS scores after 8 weeks (p < 0.001), 28 - 23% lower use of rescue medication, Jacquet A (significant less use of analgesics (P < 0.001) with a group mean difference of -10.0 (95% CI: - 4.9 to - 15.1). Mean WOMAC scores for pain, stiffness and function in the active arm were significantly different (P < 0.001) and showed benefits in Osteoarthritis as noted in a separate the potential to act as disease modifying agents in osteoarthritis.

Synovial fluid examination suggested that mineral supplement helped in restoring synovial fluid rheological properties and synovial metabolism and in reducing cartilage pathology. Joint fluid is not considered an optimal source of biomarkers because of the difficulty in obtaining repeated samples. Average cell count significantly decreased to 240/microlitre.

MRI revealed the entire spectrum of Osteoarthritis related abnormalities in knee as it allowed assessment of soft tissue structures, cartilage and bone lesions and it could be used to monitor cartilage damage in-vivo but reproducibility was expensive. Ultrasonography on the other hand is a simple relatively inexpensive method to depict early changes of synovium and articular cartilage in patients with joint disease. Studies comparing MRI and Ultrasound modalities showed that there was a significant correlation between MRI and Ultrasound techniques for evaluating cartilage changes in patients with Osteoarthritis. (23,24). However some studies suggested that total cartilage volume and mean thickness were relatively insensitive to diseases progression

though there are some conflicting results. There was evidence to suggest that Osteoarthritis caused regional changes in cartilage structure with some regions exhibiting thinning or loss of cartilage while swelling occurred elsewhere on articular cartilage. For this reason localized measures of cartilage thickness were likely to provide fuller picture changes in cartilage during disease process (25,26)

Conventional radiograms were commonly used to assess the severity of articular involvement. However alterations appeared late. In early disease, structural changes in OA joint were difficult to study because of relative insensitivity of radiographs. The groups had insignificant deterioration in the JSW (mean (SD) (0.05) ($p>0.82$) as compared to some studies using placebo arm which normally reported significant deterioration in the JSW to a tune upto 0.55mm (mean (SD) -0.09 , $p<0.01$) (3)

By the time patients presented with symptoms and initiated treatment, all of the other tissues in the joint, including the subchondral bone, synovium, joint capsule, periarticular muscles, sensory nerve endings, and ligaments are affected. Treatment development continues to focus on cartilage and just treating the cartilage alone by throwing new cartilage into the mix or protecting remaining cartilage from being further damaged, which is not going to solve the problem (27).

Other limitations of this study were its short duration (24 weeks), lack of assessment for remnant effects after treatment stoppage and limited sample size. Additional study of longer treatments in a greater numbers of subjects

would be helpful to verify the treatment effect for mineral supplementation and to explore the lack of significant treatment effect and its efficacy may have been under demonstrated within this 24 week study period.

However in this pilot study mineral supplementation slowed cartilage damage progression thus confirming chondroprotection activity of the supplement. It was clear that this mineral supplement was indeed safe, as there were no changes in various clinical and laboratory measures of safety in this 6 month study. Supplementation was efficacious, particularly compared to baseline conditions, but there were also clear difficulties in determining a sustained disassociation from placebo which warrants further study.

CONCLUSION

Therapeutic approaches to cartilage lesions are a new challenge. A number of viable options have been made available over the years to address problems concerning cartilage damage. An alternative approaches for the management of Osteoarthritis are desirable, natural mineral supplement alone or in combination with other nutraceuticals improves joint health and hence provide a significant relief of osteoarthritis symptoms. However, carefully conducted randomized, prospective studies for each of these nutraceuticals should be conducted to validate the safety and efficacy of cartilage regeneration.

REFERENCES

- 1 -ACR. Recommendations for the medical management of osteoarthritis of the hip and knee: 2000 update. American College of Rheumatology Subcommittee on Osteoarthritis Guidelines. *Arthritis Rheum* 2000;43:1905–15.
2. Graham DJ, Campen D, Hui R, Spence M, Cheetham C, Levy G, Shoor S, Ray WA. Risk of acute myocardial infarction and sudden cardiac death in patients treated with cyclo-oxygenase 2 selective and non-selective non-steroidal anti-inflammatory drugs: nested case-control study. *Lancet*. 2005;365:475–81. [PubMed]
3. Listrat V, Ayral X, Patarnello F, Bonvarlet J P, Simonnet J, Amor B, *et al*. Arthroscopic

- evaluation of potential structure modifying activity of hyaluronan (Hyalgan) in osteoarthritis of the knee. *Osteoarthritis Cartilage* 1997;5:153–60.
4. Reichenbach S, Sterchi R, Scherer M, Trelle S, Buergi E, Buergi U, et al. Meta-analysis: chondroitin for osteoarthritis of the knee or hip. *Ann Intern Med.* 2007;146:580-90. [PMID: 17438317]
 5. McAlindon T. Why are clinical trials of glucosamine no longer uniformly positive? *Rheum Dis Clin North Am.* 2003;29:789–801. doi: 10.1016/S0889-857X(03)00064-4.
 6. Harrison's principle of internal medicine. 14th edition vol. 1 and 2 ; 489-492/446-447/1931
 7. Gaby AR. Natural treatments for osteoarthritis. *Altern Med Rev.* 1999;4:330–341
 8. Stephen Holt. Alternative and Complementary Therapies. *Bone and Joint Health.* April 1998, 4(2): 101-108.
 9. McAlindon TE, Biggee BA. Nutritional factors and osteoarthritis: recent developments. *Curr Opin Rheumatol.* 2005;17:647–652. doi: 10.1097/01.bor.0000175461.57749.46. [PubMed] [Cross Ref]
 - 10- Connor JR, Manning PT, Settle SL, Moore WM, Jerome GM, Webber RK, Tjoeng FS, Currie MG. Suppression of adjuvant-induced arthritis by selective inhibition of inducible nitric oxide synthase. *Eur J Pharmacol.* 1995;273:15–24. doi: 10.1016/0014-2999(94)00672-T. [PubMed] [Cross Ref]
 - 11 -Henrotin Y, Kurz B, Aigner T. Oxygen and reactive oxygen species in cartilage degradation: friends or foes? *Osteoarthritis and cartilage / OARS, Osteoarthritis Research Society.* 2005;13:643–654. doi: 10.1016/j.joca.2005.04.002. [PubMed] [Cross Ref]
 - 12 King DE, Mainous AG, 3rd, Geesey ME, Woolson RF. Dietary magnesium and C-reactive protein levels. *Journal of the American College of Nutrition.* 2005;24:166–171. [PubMed]
 - 13 Miller MJS, Ahmed S, Bobrowski P, Haqqi TM. Suppression of human cartilage degradation and chondrocyte activation by a unique mineral supplement (sierrasil™) and a cat's claw extract, vincaria® *J Amer Nutr Assoc.* 2004;7:32–39.
 - 14 .Shakibaei M, Kociok K, Forster C, Vormann J, Gunther T, Stahlmann R, Merker HJ. Comparative evaluation of ultrastructural changes in articular cartilage of ofloxacin-treated and magnesium-deficient immature rats. *Toxicologic pathology.* 1996;24:580–587. [PubMed]
 15. Frestedt JL, Kuskowski MA, Zenk JL: A natural seaweed derived mineral supplement (Aquamin F) for knee osteoarthritis: a randomised, placebo controlled pilot study. *Nutr J.* 2009 Feb 2;8:7.
 16. Jacquet A, Girodet PO, Pariente A, Forest K, Mallet L, Moore N. Phytalgic, a food supplement, vs placebo in patients with osteoarthritis of the knee or hip: a randomised double-blind placebo-controlled clinical trial. *Arthritis Res Ther.* 2009;11(6):R192. Epub 2009 Dec 16.
 17. Joy L Frestedt, 1 Melanie Walsh, 2 Michael A Kuskowski, 3 and John L Zenk 1
A natural mineral supplement provides relief from knee osteoarthritis symptoms: a randomized controlled pilot trial *Nutr J.* 2008; 7: 9.
 18. Miller MJ, Mehta K, Kunte S, Raut V, Gala J, Dhumale R, Shukla A, Tupalli H, Parikh H, Bobrowski P, Chaudhary J. Early relief of osteoarthritis symptoms with a natural mineral supplement and a herbomineral combination: a randomized controlled trial [ISRCTN38432711]. *Journal of inflammation (London, England).* 2005;2:11. [PMC free article] [PubMed]
 - 19 -Bellamy N Buchanan WW et al. Validation study of WOMAC: A health status instrument for measuring clinically important patient relevant outcomes to antirheumatic drug therapy in patients with osteoarthritis of the

- hip or knee. *J Rheumatol*.1988; 15: 1833-1840.
20. Lequesne M , Brandt K, Bellamy N, Moskowitz R, Menkes C J, Pelletier J P, *et al*. Guidelines for testing slow acting drugs in osteoarthritis. *J Rheumatol Suppl*1994;41:65–71 ; discussion 72–3; erratum, 2395. 24
- 21 Ravaud P , Auleley G R, Chastang C, Rousselin B, Paolozzi L, Amor B, *et al*. Knee joint space width measurement: an experimental study of the influence of radiographic procedure and joint positioning. *Br J Rheumatol*1996;35:761–6. 25
- 22 Ravaud P , Giraudeau B, Auleley G R, Drape J L, Rousselin B, Paolozzi L, *et al*. Variability in knee radiographing: implication for definition of radiological progression in medial knee osteoarthritis. *Ann Rheum Dis*1998;57:624–9. 26
- 23 S.tarhan,Z.unlu,C.Goktam: Magnetic resonance imaging and ultrasonographic evaluation of patients with knee osteoarthritis : a comparative study. *ClinRheumatology* 2003 22:181-188
- Chaojeannie :ultraonography in Osteoarthritis of knee :recent advances and prospects for the future. *Current opinion in rheumatology* 2008, 20 (5)
- C.J. Taylor,Tomos G Williams,Zai Xiang Gao John C Walterton; corresponding articular cartilage thickness measurement in knee joint by modeling the underlying bone (commercial in confidence:IPMI 2003,LNCS2732,PP126-135,2003
- +*Rudert, MJ; *Alliman, KJ; *Lundberg, HJ; *El-Khoury, GY; *Brown, TD; *Saltzman, CL ARTICULAR CARTILAGE THICKNESS MEASUREMENT WITH MRI AND MULTI-DETECTOR COMPUTED TOMOGRAPHY (MDCT) 49th Annual Meeting of the Orthopaedic Research Society Poster #0571
- .Felson DT, Kim YJ. The futility of current approaches to chondroprotection. *Arthritis Rheum.* 2007;56:1378-83. [PMID: 17469094] 27