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Efficacy and Safety Assessment of *Curcuma Longa (L.)* And *Trachyspermum Ammi (L.)* In Experimentally Induced Osteoarthritis Rabbit Model

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ABSTRACT
The objective of current study is to assess the efficacy and safety of Curcuma longa L. and Trachyspermum ammi L. in experimentally induced osteoarthritis (OA) rabbit model. Total 28 male healthy albino rabbits, aged 6-8 months, were assigned into four groups randomly after disease induction. During Phase I, OA was induced using 4% papain solution at first, fourth and seventh days. During Phase II, subjects were treated with liquid extracts of C. longa L. and T. ammi L. in combination and alone. X-ray radiographs were performed at every four weeks intervals and biochemical tests were performed before and at the end of treatment respectively. Results indicate that pain disappeared after day 4 in C. longa L. group. X-ray radiographs showed that OA was reversed and returned to KL score "0" in T. ammi L. group. T. ammi L. considerably decreased the SGOT, SGPT and Alkaline phosphatase levels while BUN, blood urea, serum creatinine and sUA levels were decreased with T. ammi + C. longa group and sUA level was reduced by T. ammi L.. This study revealed that T. ammi L. is most safe and effective agent in inhibition and reversing of OA in experimentally induced rabbit model.
Keywords: *C. longa L.*, Osteoarthritis, Pain, Rabbit, *T. ammi L.*

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INTRODUCTION

Every human being is the author of his own illness or disease, Buddha said. One of these diseases is osteoarthritis. Osteoarthritis also known as degenerative arthritis, degenerative joint disease, or osteoarthrosis, is the degenerative disease of cartilage. The disease manifests first as a molecular derangement (abnormal joint tissue metabolism) followed by anatomic, and/or physiologic derangements (characterized by cartilage degradation, bone remodeling, osteophyte formation, joint inflammation and loss of normal joint function), that can culminate in illness. There are approximately 250 million people have knee osteoarthritis that make up 3.6% of the population [1]. Osteoarthritis (OA) occurs mostly in the knees, hips, hands and spine. Warning signs of osteoarthritis are pain, stiffness in a joint after getting out of bed or sitting for a long time, swelling or tenderness in one or more joints, and a crunching feeling or the sound of bone rubbing on bone. The primary causes of osteoarthritis are injury, obesity, aging and genetic basis while the secondary causes of osteoarthritis include Alkaptonuria, congenital disorders of joints, Hemochromatosis, Wilson's disease, injury to joints or ligaments, obesity, joint infection, inflammatory disease or diabetes [2]. Multiple pathophysiologic mechanisms and etiologic risk factors all contribute to the disease progression and serve as markers of pharmacological and pharmacologic interventions. These effects on the tissues of all three joint partitions marked as osteophyte formation, articular cartilage breakdown, bone marrow lesions, subchondral sclerosis and alterations of the synovium on both biochemical and morphological stages often causing occasional synovitis. Thus, the cytokine-based and molecular events that enterprise joint injury in inflammatory arthritides have gradually appeared as pathogenic paradigms in OA, and will be highly pertinent to the advancement of future OA therapeutics.

Diagnosis of osteoarthritis is based upon clinical examination, history and X-ray analysis. A number of classification systems are used for gradation of osteoarthritis. These include Western Ontario and McMaster Universities Arthritis Index (WOMAC) score and Kellgren-Lawrence grading system. Lifestyle modification including weight loss, exercise, patient education are really good factors in osteoarthritis management. Non-steroidal anti-inflammatory drugs (NSAIDs) are more effective for mild to moderate

symptoms. Oral opioids such as tramadol are also often prescribed. These are only recommended when first line therapy is failed or contraindicated. However, Oral steroids are not usually recommended in the treatment of OA. If problems persist and more conservative management is not beneficial, joint-replacement surgery or resurfacing may be recommended. There is little or insufficient evidence that supports the benefits associated with some supplements, including the Ayurvedic herbal preparations, collagen, ginger, glucosamine, hyaluronic acid etc. However, acupuncture leads to improvements in pain relief on small scale. Therefore, there is no conclusive evidence that may cure the disease.

The aims and objectives of our study are to determine the efficacy of *Curcuma longa (L.)* and *Trachyspermum ammi (L.)* alone and in combination in osteoarthritis, and to study the safety of *Curcuma longa (L.)* and *Trachyspermum ammi (L.)* in experimentally induced osteoarthritis rabbit model.

MATERIAL AND METHODS

Chemicals and Reagents

Papain 4% solution, *T. ammi* extract, *C. longa* extract, Normal Saline.

Subjects

Total twenty-eight male healthy albino rabbits (*Oryctolagus cuniculus)*, aged 6-8 months, were selected and divided into four groups. The subjects were housed under normal temperature (21 $\pm$ 1^{o}C) and humidity (65-70%) conditions with 12 hours light-dark cycle. Deer grass (*Muhlenbergia rigens*), carrots (*Daucus carota*) and water were given as food *ad libitum*.

Tests and Experiments

Pain score was calculated using Rabbit Grimace Scale (RbtGS). An RbtGS score was calculated by averaging intensity ratings for each action unit (AU). These AUs include orbital tightening, nose shape, cheek flattening, whisker change, and ear position. No pain was scored 0, moderate pain 1 and severe pain was scored 2. An RbtGS difference score (relative to baseline/"no pain") can then be calculated for each subject, and averaged across a group. Routine radiographic examination of the knee joint consisted of standard projections were performed. Anterio-posterior radiographs of the knee were obtained. Global joint assessment was done according to Kellgren and Lawrence Grading Scale (KL) as explained in the Atlas of standard radiographs of arthritis [3]. Knee classified as grade 2 or higher defined OA. To ensure safety of treatment, Liver function tests, Renal function tests and Complete blood count was performed.

The present study was divided into two phases. During Phase I, OA was induced experimentally by methods as specified by Bentley (1971). During Phase II, subjects were treated with liquid extracts of *Curcuma longa (L.)* and *Trachyspermum ammi (L.)* according to dosage schedule as specified in Table 2. Plants were identified and authenticated from the Department of Botany, University of the Punjab, Lahore - Pakistan with voucher no. 2435-16. X-ray radiographs were made at every four weeks intervals and biochemical tests were performed before and at the end of treatment respectively. Pain score was calculated using RbtGS.

Table.2. Dosage Schedule

Group ID	Group Names	Group Type	No. of Subjects	Plant used	Dose per Kg PO (body weight)
I	*T. ammi*	Experimental	n=7	*T. ammi*	5mg BD
II	*C. longa*	Experimental	n=7	*C. longa*	10mg BD
III	*T.ammi+C.longa*	Experimental	n=7	*T.ammi+C.longa*	2.5+5mg BD
IV	Control	Controlled	n=7	Water	N.A

Statistical Analysis

Data were evaluated using the IBM statistical package for social sciences software (SPSS) version 22.0. One way ANOVA was performed to determine significance and interaction among groups utilizing post hoc tests with Tukey range test. The value $p<0.05$ was considered significant.

Ethics

Research protocol was approved by the Institutional Animal Ethical Committee of Hajvery University, Lahore-Pakistan.

RESULTS

Total twenty-eight male, healthy albino rabbits (*Oryctolagus cuniculus)*, aged 6-8 months, were selected and divided into four groups. Subjects were treated with *Curcuma longa (L.)* and *Trachyspermum ammi (L.)* alone and in combination. Pain score was observed with the start of treatment. Pain score was calculated using Rabbit Grimace Scale (RbtGS).

Pain Score

It was observed that signs/AUs of pain were found from day 0 to day 4. This theme of pain score is depicted in Pareto Chart of Pain score in Figure 1 among all four groups.

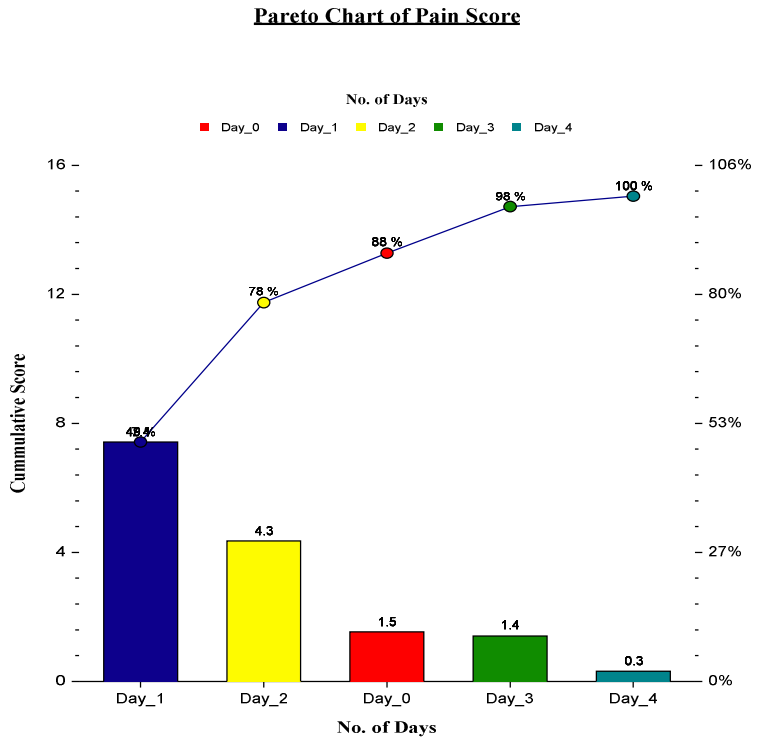


Figure.1. This Pareto Chart depicts Pain Score from Day 0 to Day 4. X-axis shows number of days and y-axis shows cumulative score against days. Figure shows that highest cumulative score was observed on day 1 and least was observed on day 4.

Pareto chart shows that pain was found severe in Day 1 and after continuation of therapy it was disappeared in next few days. A little pain was found at day 4. Consequently, it was observed that Group II (*C. longa*) treatment was found more effective in reducing pain as compared to other treatment groups as illustrated in Figure 2.

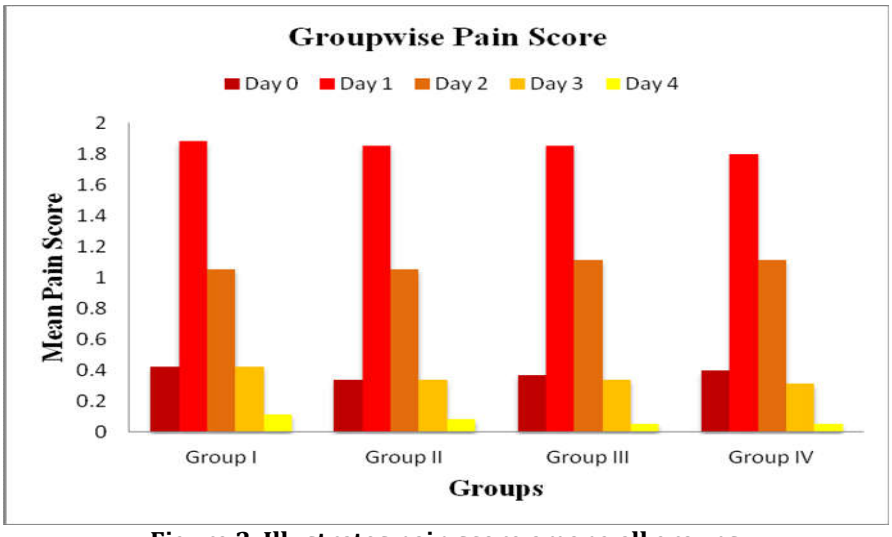


Figure.2. Illustrates pain score among all groups

Radiographic Imaging

To assess efficacy of treatment protocols, the radiographs were examined that had been acquired using a commercially available digital detector. All digital images were evaluated on a medical-grade flat-screen monitor. An experienced radiologist scored each radiograph for the presence and severity of osteoarthritis using a KL scale. These X-Ray anterio-posterior radiographs showed that bilateral osteoarthritis was induced in rabbits' knees. After induction of disease, anterio-posterior radiographs revealed that bilateral osteoarthritis was developed in knees of rabbits as shown in Figure 3. According to

KL grading system, grade 3 osteoarthritis was developed in right knee and grade 4 was developed in left knee. However, it varied within groups and between both knees.

Right Knee	Left Knee

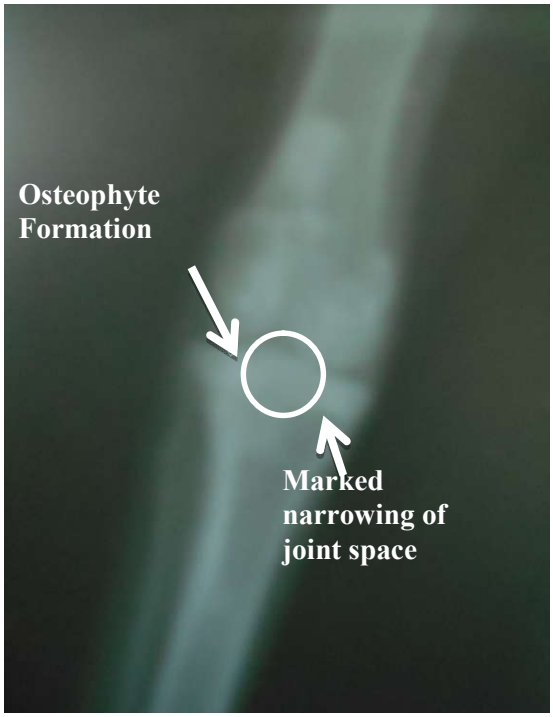
Figure.3. Shows the comparison of right and left osteoarthritis knee joint; Narrowing of joint space; osteophyte formation is clearly visible. Right knee shows grade 3 and Left knee shows grade 4 KL score.

After four weeks of treatment, radiographic imaging revealed that osteoarthritis disease progression was reduced to KL score "2" in *T. ammi (L.)* and *C. longa (L.)* groups. However, combination therapy (*T. ammi + C. longa*) was found less beneficial with KL score "3" and control group scored "4" as revealed in Figure 4 and 5.

a. Group I: *T. ammi*

b.* Group II: *C. longa

c. **Group III: *T. ammi + C. longa***



d. Group IV: *Control*

Figure.4 (a - d). Reveals comparison of Right Knees of all groups after 4 weeks of Treatments

a. Group I: *T. ammi*

b.* Group II: *C. longa

c. **Group III: *T. ammi + C. longa***

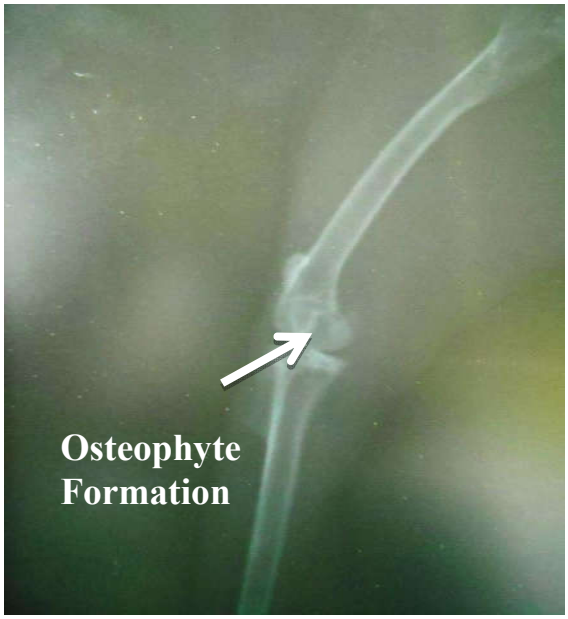
d. Group IV: *Control*

Figure.5 (a - d). Reveals comparison of Left Knees of all groups after 4 weeks of Treatments

After eight weeks of treatment, radiographs depicted that osteoarthritis was reversed and returned to KL score "0" in *T. ammi (L.)* while in *C. longa (L.)* group, disease progression was stopped to KL score "1". However, in combination therapy (*T. ammi + C. longa*), treatment effects observed were not much significant and had KL score "2". On the other hand, control group addressed at KL grade "4" as illustrated in Figure 6 and 7.

a. Group I: *T. ammi*

b. **Group II: *C. longa***



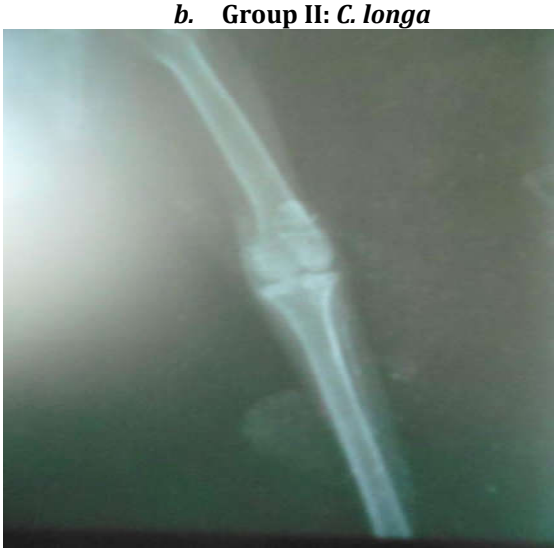
c. Group III: *T. ammi + C. longa* **d. Group IV: *Control***

Figure.6 (a - d). Illustrates comparison of Right Knees of all groups after 8 weeks of Treatments

a. Group I: *T. ammi*

b.* Group II: *C. longa

c.* Group III: *T. ammi + C. longa



d. Group IV: *Control*

Figure.7 (a - d). Illustrates comparison of Left Knees of all groups after 8 weeks of Treatments

Biochemical Analysis

To assess the safety of treatment protocols, various biochemical tests were performed. These tests include liver function tests, renal function tests and complete blood count.

Liver function tests: Liver function tests include serum bilirubin (s.bilirubin), serum glutamic-pyruvic transaminase (SGPT), serum glutamic-oxaloacetic transaminase (SGOT) and alkaline phosphatase. It was found that *C. longa L.* (8.00 ± 0.279) significantly reduced the s.bilirubin level in comparison with *T. ammi L.* (9.00 ± 0.227) with a *p = 0.045* as shown in Figure 4.15. However, SGOT enzyme was least affected by *C. longa (L.)*. *T. ammi L.* (78.00 ± 0.723) considerably decreased the SGOT levels as compared to *T. ammi + C. longa* (78.57 ± 1.849) and *C. longa L.* (90.00 ± 1.951) groups with a relatively high level of significance (*p = 0.000*). Similar is the case with SGPT, *T. ammi L.* (84.85 ± 1.242) consistently reduced the SGPT level than *C. longa L.* (102.00 ± 0.690), *T. ammi + C. longa* (88.85 ± 0.737) and Control (212.28 ± 15.092) groups. Alkaline phosphatase level was ample reduced by *T. ammi L.* (82.85 ± 0.508) as compared to *C. longa L.* (85.71 ± 0.184) group. However, other treatment groups showed not much improvement. In this case, *T. ammi L.* was also found statistically significant as compared to *C. longa L.* (*p = 0.000*). In short, it was witnessed that liver function enzymes were significantly reduced by *T. ammi (L.)* group among all treatment. Therefore, this study showed that *T. ammi (L.)* is found most hepatoprotective in all defined treatments.

Renal function tests: Renal function tests include blood urea nitrogen (BUN), blood urea, serum creatinine (s,creatinine) and serum uric acid (sUA). It was observed that BUN and blood urea levels were decreased with *T. ammi + C. longa* group (28.57 ± 0.368, 60.00 ± 0.577) while *T. ammi L.* (29.42 ± 1.231, 66 ± 0.816) and *C. longa L.* (33.85 ± 0.594, 69.28 ± 2.146) were least affected by BUN and blood urea levels respectively. However, *T. ammi (L.)* was statistically significant than *C. longa (L.)* in case of BUN (*p = 0.006*) while it showed relatively high level of significance in association to *T. ammi + C. longa* group (*p = 0.010*) in blood urea. Similarly, s.creatinine levels were decreased by *T. ammi + C. longa* group among all treatment groups (*P-value 0.009*). Conversely, sUA level was reduced by *T. ammi L.* (*p = 0.000*) in all therapy regimens. Consequently, this study depicted that *T. ammi + C. longa* group found more nephroprotective among all groups irrespective of sUA which was greatly reduced by *T. ammi (L.)*.

Complete Blood Count: Biochemical tests performed for complete blood count include Haemoglobin (Hb), White blood cells count (WBCs), Red blood corpuscles count (RBCs), Platelet count (PLT), Neutrophil count (Neut), Lymphocyte count (Lym), Monocytes count (Mon), Eosinophil count (Eosin), Basophil count (Baso), Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH), and Mean corpuscular haemoglobin concentration (MCHC). *C. longa (L.)* group (10.71 ± 0.088) showed relatively high level of haemoglobin followed by *T. ammi (L.)* group (10.40 ± 0.125). However, *C. longa (L.)* was found statistically significant as compared to *T. ammi + C. longa* group (*P-value 0.000*). White blood cells count was significantly lowered by *T. ammi (L.)* and it was least affected by *T. ammi + C. longa* group with a significant level (*p = 0.000*). Red blood cells were not much decreased in number with *T. ammi L.* (5.32 ± 0.041) as compared to *T. ammi + C. longa* (3.58 ± 0.209), *C. longa L.* (5.09 ± 0.037) and Control (3.27 ± 0.129) groups. Results were statistically significant with *T. ammi (L.)* in relation to *T. ammi + C. longa* (*p = 0.000*). *T. ammi L.* (158.57 ± 1.151) was assessed effective in platelet count as it did not much decrease the platelets with respect to *T. ammi + C. longa* (85.42 ± 0.947) group. Similar observation came from *C. longa L.* (153.42 ± 1.087) group but its effectiveness was found less in relation to *T. ammi (L.)*. Results indicated that *T. ammi (L.)* was statistically significant than *T. ammi + C. longa* (*p = 0.000*). Differential leucocytes count was observed high in *T. ammi + C. longa* group irrespective to Neutrophils and Lymphocytes which were found high in number in *T. ammi (L.)* and *C. longa (L.)* groups respectively. Statistical analysis revealed Neutrophils, Lymphocytes and Monocytes counts were found significant in *T. ammi L.* than *C. longa L.* (*p = 0.000, p = 0.001, and p = 0.000*) groups respectively. *T. ammi (L.)* has comparatively high MCV after *T. ammi + C. longa* group (*p = 0.000*). Results were not much significant between *T. ammi + C. longa* and *C. longa* (*p = 0.89*). However, it was found statistically significant between *T. ammi L. and C. longa L.* (*p = 0.044*). On the other hand, MCHC was statistically significant in *C. longa L.* group in comparison with *T. ammi L.* group (*p = 0.002*).

DISCUSSION

Diseases started even before the existence of human beings [4]. So when the civilization began, the biggest threats for human were diseases. Man has made several sincere efforts for the search of new drugs for in order to cure and control the different diseases. One of these diseases that run through life is osteoarthritis [5, 6]. OA is a degenerative joint disease that has classical signs and symptoms of pain, stiffness, inflammation and crepitus. Moreover, our study also included joint space narrowing, osteophyte formation and bone spurs. Rabbits were considered as an animal model for osteoarthritis alternative to

humans [7-9]. The objective of current study was to assess the efficacy and safety of *T. ammi (L.)* and *C. longa (L.)* alone and in combination.

Seneca, Roman philosopher and playwright (4BC-65) said, "*Remember that pain has this most excellent quality. If prolonged it cannot be severe, and if severe it cannot be prolonged*". Similar findings are observed in our study while working with RbtGS. Pain was severe on day 1 from the start of treatment and diminished after day 4 as shown in Figure 1. Pain is significantly reduced more in *C. longa L.* group in comparison with other experimental groups (Figure 2). These findings are also parallel to Matsumiya, Sorge [10]. Their finding concluded that postoperative pain in mice, lasted for 36 to 48 hours, and appeared to show relative exacerbation during the early dark (active) photo-phase. Our findings are also supported by Madhu, Chanda [11] who demonstrated the safety and efficacy of *C. longa L.* as a useful treatment option for patients with primary painful knee OA in human. Consequently, it is found from present study that both *T. ammi L.* and *C. longa L.* are useful in reducing OA pain, however, *C. longa L.* exterminates pain more rapidly than any other conventional herbal or allopathic pharmacological treatment.

Effectiveness of therapy was assessed by performing X-ray radiographs that were taken at the start of treatment, after 4 and 8 weeks of treatments. OA score was calculated using KL grading scale [3]. After four weeks of treatment, radiographic imaging revealed that osteoarthritis disease progression is reduced to KL score "2" in *T. ammi L.* and *C. longa L.* groups. However, combination therapy (*T. ammi + C. longa group*) is found less beneficial with KL score "3" and control group "4" as revealed in Figure 4 and 5. After eight weeks of treatment, radiographs depicted that osteoarthritis is reversed and returned to KL score "0" in *T. ammi L.* while in *C. longa L.* group, progression of disease is reduced to KL score "1". However, in combination therapy (*T. ammi + C. longa group*), treatment results observed are not much significant and have KL score "2". On the other hand, control group addressed at KL grade "4" as illustrated in Figure 6 and 7. This revealed that combination therapy probably has a competitive antagonistic effect. Conversely, *T. ammi L.* group not only restricted the disease progression but also it reversed the disease. *T. ammi L.* was found very effective ingredient in reversing of primary OA while *C. longa L.* is observed beneficial in slowing of disease progression. High molecular weight hyaluronan (HA) effects on cartilage degeneration were studied in OA model of rabbit knee by Kikuchi *et al.* (1996). This study compared HA80, HA190 and saline. Scanned electron micrographs of femoral cartilage revealed that degeneration of cartilage was less severe with HA190 than with saline. This suggested that intra-articular administration of higher molecular weight HA is more efficacious than lower molecular weight HA, in the rabbit model, in the inhibition of cartilage deterioration in early OA [12]. A study was done by Amiel, Toyoguchi [13] to investigate the long-term effect of sodium hyaluronate (Hyalgan) on osteoarthritis progression in a rabbit model. Their study estimated the HA effects after a 26-week follow-up period, and shown that two courses of HA injections provided a benefit compared with one course of injections for slowing OA progression. A randomized, double blind, placebo controlled, crossover study was carried out to assess the safety, efficacy and tolerability of *Boswellia serrata* Extract (BSE) in knee OA. All drug treatment groups reported lowering of knee pain, increased walking distance and knee flexion. Yet, the frequency of knee joint swelling was decreased with radiologically no change. BSE was well tolerated by the patients except for minor gastrointestinal adverse events. BSE is recommended in the knee OA patients with possible therapeutic use in other arthritis [14]. Another study was conducted by Permuy, Guede [15] to assess the effects of glucosamine and risedronate alone or in combination in an experimental rabbit model of osteoarthritis. This animal study concluded that glucosamine sulfate and risedronate treatment alone or in combination may be able to stop cartilage swelling. The risedronate treatment could partially stop the fibrillation and the inflammation of synovial membrane as well as modify the orientation of trabeculae in healthy and in osteoarthritic knees. However, this study did not correlate much to our findings as risedronate was not found better in secondary OA as our study speaks to primary and secondary bilateral OA in rabbit models.

To assess the safety of all treatment protocols under investigation, various biochemical tests were also performed, i.e., liver function tests (LFTs), renal function tests (RFTs) and complete blood count (CBCs). LFTs are routinely used to estimate liver disease severity, toxicity and hepatic dysfunction [16-18]. It was found that *C. longa L.* significantly reduced the s. bilirubin level in comparison with *T. ammi L.* ($p = 0.045$). However, SGOT enzyme was least affected by *C. longa L*. while *T. ammi L.* considerably lowered the SGOT levels as compared to *T. ammi + C. longa* and *C. longa L.* groups with a relatively high level of significance ($p = 0.000$). Similar is the case with SGPT, *T. ammi L.* consistently reduced the SGPT level than *C. longa L.*, *T. ammi + C. longa* and Control groups. ALP level was ample reduced by *T. ammi L.* as compared to *C. longa L.* group. In this case, *T. ammi L.* was also found statistically significant as compared to *C. longa L.* ($p = 0.000$). In short, it is witnessed that liver function enzymes were significantly reduced by *T. ammi L.* group among all treatments. Normal bilirubin level [19, 20] indicates normal liver function while normal levels

of liver enzymes in our study indicate that all treatment protocols did not damage the hepatocyte or caused hepatitis. This confirms the safety of our treatment protocols. An equivalence study was carried by Tugwell, Wells [21] to compare the topical diclofenac solution with oral diclofenac in symptomatic treatment of knee OA. They found that oral diclofenac group developed abnormal liver function tests, hemoglobin, and creatinine clearance. However, these pathological parameters were found normal by Stammers, Sibbald [22] when they were working to assess cod liver oil efficacy as an adjuvant to non-steroidal anti-inflammatory drug treatment in OA management. Therefore in relation to other studies on OA, current study showed that *T. ammi L.* is most hepatoprotective in all defined treatments.

RFTs involved are blood urea nitrogen (BUN), blood urea, serum creatinine (s. creatinine) and serum uric acid (sUA). It is observed that BUN and blood urea levels are decreased with *T. ammi + C. longa* group while *T. ammi L.* and *C. longa L.* are least affected by BUN and blood urea levels respectively. However, *T. ammi L.* is statistically significant than *C. longa L.* in case of BUN ($p = 0.006$) while it showed relatively high level of significance in association to *T. ammi + C. longa* group ($p = 0.010$) in blood urea. Similarly, s. creatinine level is decreased by *T. ammi + C. longa* group among all treatment groups. Conversely, sUA level was reduced by *T. ammi L.* ($p = 0.000$) in all therapy regimens. Consequently, this study depicted that *T. ammi + C. longa* group found more nephro-protective among all groups irrespective of sUA which was greatly reduced by *T. ammi L*. Hence, this study reveals that *T. ammi L.* is nephro-protective next to combination therapy (*T. ammi + C. longa* group).

Biochemical tests performed for CBCs of all treatment groups were Haemoglobin (Hb), White Blood Cells count (WBCs), Red Blood corpuscles count (RBCs), Platelet count (PLT), Neutrophil count (Neut), Lymphocyte count (Lym), Monocytes count (Mon), Eosinophil count (Eosin), Basophil count (Baso), Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH), and Mean corpuscular haemoglobin concentration (MCHC). Results indicate that *T. ammi L.* found safe in all defined treatments as it improved RBCs, PLT, Neut counts and MCV. Although it showed comparatively less effect on WBCs than *C. longa L.* but its safety is revealed in Hb next to *C. longa L.* Hence, results direct that *T. ammi L.* is most safe in a row to *C. longa L.* and *T. ammi + C. longa* groups. Consequently, *T. ammi L.* is recommended in the patients of knee OA with possible therapeutic use and reversing of disease progression with no gastrointestinal disturbances. These parameters of blood chemistry were also observed by Puhl, Bernau [23] and Doig, Purbrick [24] for the safety of their treatment protocols.

CONCLUSION

This study was conducted with the objective to evaluate the safety and efficacy of *T. ammi L.* and *C. longa L.* alone and in combination. The outcomes of this study has revealed that *T. ammi L.* is most safe and effective agent in inhibition and reversing of OA in experimentally induced rabbit model. It is the first agent found most effective in disease progression as well as harmless for gastrointestinal tract. Because most drugs used for OA disturb the gastrointestinal tract like celecoxib and piroxicam, therefore, we recommend *T. ammi. L.* for the treatment of OA.

FUTURE CONSIDERATIONS

Results in this study offer confirmation that *T. ammi L.* is an effective therapeutic approach in the rabbits with OA. Future studies should address human populations with more sensitive approaches and dose recommendations.

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