EFFECT OF CURCUMINOID VERSUS DICLOFENAC SODIUM ON MONOCYTES SECRETION OF TUMOR NECROSIS FACTOR–α IN KNEE OSTEOARTHRITIS

Vivin Hudiyanti[1], Nyoman Kertia[2], Sumardi[3]

1. Division of Internal Medicine, Faculty of medicine Gadjah Mada University/Dr Sardjito Hospital
2. Sub Division of Rheumatology, Internal Medicine Dr Sardjito Hospital/ Faculty of medicine Gadjah Mada University Yogyakarta
3. Sub Division of Pulmonology, Intenal Medicine Dr Sardjito Hospital / Faculty of medicine Gadjah Mada University Yogyakarta

ABSTRACT

Background. *Tumor Necrosis Factor-α (TNF-α), a pro-inflammatory cytokine produced by monocyte is increased in osteoarthritis synovial fluid. Curcuminoid from Curcuma domestica Val. suppresses the secretion of TNF-α.*

Objective. *The purpose of this study was to investigate the suppression effect of curcuminoid from Curcuma domestica Val on synovial fluid monocyte's TNF-α secretion compared to diclofenac sodium in knee osteoarthritis*

Methods. *A prospective randomized open end blinded evaluation (PROBE) method was applied. Subjects were patients with knee osteoarthritis visiting Rheumatology Clinic Dr. Sardjito Hospital and Wirosaban Hospital Yogyakarta. Curcuminoid 30 mg three times daily or diclofenac sodium 25 mg three times daily were administered for 4 weeks. The level of TNF-α secreted by synovial fluid's monocytes were measured by ELISA before and after treatment.*

Results. *A total of 80 subjects were enrolled, 39 subjects on curcuminoid treatment groups and 41 subjects on diclofenac sodium group. Seven subjects were dropped out, 5 from the curcuminoid group and 2 from the diclofenac sodium group. There was a significant decrement of TNF-α level during 4 weeks treatment in both groups ($p < 0.001$ respectively). There was no significant difference on TNF-α levels between groups ($p = 0,237$), neither in 50% decrement of TNF-α levels ($p = 518$).*

Conclusion. *The effect of curcuminoid in decreasing TNF- α level on patients with osteoarthritis is similar with sodium diclofenac.*

Keywords*: osteoarthritis-monocyte-TNF-α-curcuminoid-diclofenac sodium*

INTRODUCTION

Osteoarthritis (OA) is a degenerative joint disease that has been related to damage of the joint's cartilage. The manifestations of osteoarthritis such as changes in morphology, biochemistry, molecular and biomedical aspects of cells and matrixes result in fibrillation, ulceration, thinning of joint's cartilage, sclerosis and formation of osteophyte and bone cyst[1].

The prevalence of knee osteoarthritis from radiologic examination in Indonesia is high, approximately 15.5% in man and 12.7% in woman. Due to the high level of prevalence and its chronic progressive characteristic, osteoarthritis has been contributing a big impact, socially and economically, in developed countries as well as in developing countries. In Indonesia, the estimated numbers of elderly who suffers from disability due to OA, is around 1 until 2 million people[2].

The role of cytokines in OA contributes major effects to the incident and disease progressiveness. Tumor necrosis factor-α (TNF-α) and interleukin-1 (IL-1) has been suspected to have responsible as mediators for chondrocyte response in this joint disease[3]. In osteoarthritis, IL-1 and TNF-α activate the degradating enzymes such as metalloproteinase, collagenase, gelatinase and aggrecanase that can generate the inflammatory responses in the joint[4]. Both cytokines, IL-1β and TNF-α, has been shown to play roles in cartilage destruction and inflammatory process'. An

increment of IL-1 β and TNF-α has been found in synovial fluid and cartilage of patients with OA[5].

Osteoarthritis management aimed to control symptoms especially in joint pain of the bone, reduce disability and joint damage progression, enabling patient to continue their living without resistance in function and minimum social impacts. Non-steroidal anti inflammatory drugs (NSAIDs) have an analgesic, anti-pyretic and anti-inflammatory effects. Its mechanism targets the cyclooxygenase (COX) enzyme. One of the NSAID which has often been used in daily practices is diclofenac sodium[6,7].

Curcuminoid is secondary metabolite derived from *Curcuma domestica Val*. and temulawak which has been used as flavor and herbal remedies component. Curcuminoid has been known for their activity in inflammatory process and elimination of mild bone pain and low side effects to the gastro intestinal tract[8].

The aim of the study is to investigate the effect(s) of curcuminoid from *Curcuma domestica Val*. compared to diclofenac sodium in its ability to suppress the secretion of TNF-α by synovial fluid monocytes of the knee affected by osteoarthritis.

SUBJECTS AND METHOD

This research was established in a clinical trial entitled as the Prospective Randomized Open And Blinded Evaluation = PROBE. The research was enrolled from May to September 2007 in Rheumatology Clinic, Dr. Sardjito Hospital

The inclusion criteria includes patients who suffer knee OA that was diagnosed based on ACR 1986 criteria, have synovial fluid sample from the knee joint with minimum volume of 2 cc, agreed to participate in this research by signing informed consent. Exclusion criteria includes impairments in liver function (increment of serum transaminase enzymes more than 2 times of the normal of upper limit) and kidney function (serum creatinin > 2 mg/dl), has history of peptic ulceration or duodenum ulceration, in anticoagulant drug(s), had undergone surgery on the affected knee, had received corticosteroid injection or viscous supplement at the knee joint in the past 3 month, or if there is any contraindication for intra-articular injection. Thirty subjects were included in each group.

Methods. The participants received information and explanation about the study purposes and objectives. Patients who had been using analgesic drug were obliged to stop the consumption of the drug for at least 7 days. Randomization was done on 6 block. The subjects were divided into 2 groups. The synovial fluids of the knee joint were measured to investigate the level of TNF-α. The subject in the treatment group were given capsules containing 30 mg curcuminoid from Curcuma domestica Val. three time 1 capsule daily and the subject in the control group were treated with capsules containing 25 mg of diclofenac sodium three time 1 capsule daily. After 4 weeks treatment, measurement of the synovial fluid in the knee joint was repeated to investigate the level of TNF-α rate secreted by monocyte in synovial fluid. The measurement of the ability of synovial fluid monocytes to secrete TNF-α were accomplished by monocytes cell culture in a culture media. The ELISA method was performed to assess the TNF-α level in culture media before starting and after 4 weeks treatment.

Data of the study subjects is presented in means and standard deviations. The mean differences of TNF-α level in the curcuminoid group and the natrium diclofenac group was analyzed with t-test where 95% confidence interval and p value <0.05 was considered as significant. If pre-requisites terms and conditions for the application of t-test were not fulfilled, then the Mann-Whitney test was used. Changes in TNF-α rate before and after therapy was analyzed with paired t-test where 95% confidence interval with p value <0.05 was considered as significant. When criterias for paired t-test was not fulfilled, a Wilcoxon signed rank test was applied.

RESULTS

There were 80 subjects who fulfilled inclusion without exclusion criteria. Five subjects from curcuminoid group were dropout: one subject was hospitalized because of urinary tract infection, one subject was hospitalized because of chronic

obstruction pulmonary disease, one subject had joint swelling and pain, one subject had hematuria because of bladder mass, one subject stopped the therapy without any reason. From diclofenac sodium group there was one subject stopped the therapy because of heart burn after 1 week therapy and one subject has no synovial fluid in the end of the study.

Table 1. Data base of subjects with normal distribution

Variable	Curcuminoid group (n=39)	Diclofenac group (n=41)	95% CI	P
Sex				
• Female	24	29		0.385*
• Male	15	24		
Weight (kg)	62.91±11.38	63.28±11.64	-5.50-4.76	0.886**
Height (cm)	154.36±7.37	154.78±7.67	-3.77-2.93	0.803**

Abbreviation : * X^2-test ; ** t-test ; CI: Confident Interval; SD : Standard Deviation

Tables 1 showed the data of subjects. In the curcuminoid group the proportion of female was 61.54% while in the group of diclofenac sodium the proportion of female was 70.73%. The difference of body weight between both groups was not statistically significant with p=0.385. The body weight of the curcuminoid group was 62.91±11.38 kg while in diclofenac sodium group was 63.28±11.64 kg. The difference of the weight average between both group was not statistically significant (p= 0.886). The average of height in curcuminoid group was 154.36±7.37 cm while in diclofenac group was 154.78±7.67 cm with no significant difference.

Table 2. Data base of subjects without normal distribution

Variable	Curcuminoid group (n=39) mean±SD (median)	Diclofenac group (n=41) mean±SD (median)	P
Age (years)	64.05±8.83 (66)	64.56±8.86 (64)	0.700*
History of OA (years)	2.94±3.48 (2.00)	3.40±2.38 (3.00)	0.052*
TNF-a (pg/ml)	53.95±61.35 (37.85)	69.35±113.14 (26.01)	0.192*

Abbreviation : **Mann-Whitney test*; SD: Standard Deviation

Table 2 showed the age average of curcuminoid group is 64.05±8.83 years while in diclofenac is 64.56±8.86 years. The age between both groups is not significantly different (p= 0.700). The average history of osteoarthritis for curcuminoid group is 2.94±3.48 years while in diclofenac group is 3.40±2.48 years (p=0.052). The average level of TNF-α before treatment in curcuminoid group is 53.95±61.35 pg/ml while in diclofenac group the level is 69.35±113.14 pg/ml (p= 0.192).

Table 3. Level of TNF-α before and after 4 weeks treatment

	TNF-α (pg/ml) Before treatment Mean±SD (Median)	TNF-α (pg/ml) After treatment Mean±SD (Median)	P
Curcuminoid group (n=34)	54.92±65.43 (36.06)	25.84±37.66 (11.27)	p<0.001*
Natrium diklofenak group (n=39)	55.28±95.94 (24.90)	19.05±18.61 (11.04)	p<0.001*

Abbreviation : **Wilcoxon signed ranks test*; TNF-α : *Tumor Necrosis Factor*-α;
SD : Standard Deviation

Table 3 showed the averages and the median of TNF-α level before and after 4 weeks therapy. The decreasing of TNF-α level in the curcuminoid group is from 52.92±65.43 pg/ml to 25.84±37.66 pg/ml. The decreasing of TNF-α level in diclofenac group is from 55.28±95.94 pg/ml to 19.05±18.61 pg/ml. The data is not in normal distribution, causing Wilcoxon signed ranks test to be used. The difference before and after treatment was significant (p<0,001).

Table 4. Mean of difference between TNF- α level before and after 4 weeks treatment

	Paired t-test	95% CI
Curcuminoid group (n=34)	0.013	6.6 – 51.6
Diclofenac group (n=39)	0.017	6.9 – 65.6

Abbreviation: CI: Confidential Interval

Table 4 showed the mean of difference of decreasing TNF-α level before and after 4 weeks treatment for curcuminoid group and diclofenac group. The mean of difference of decreasing TNF-α level before and after treatment for the curcuminoid group was statistically significant (p=0.013; 95% CI 6.6 – 51.6) while in diclofenac group it is also statistically significant (p=0.017; 95% CI 6.9 – 65.6).

Table 5. Mean of difference of TNF-α level before and after 4 weeks treatment

Variable	Curcuminoid group (n=34) mean±SD (median)	diclofenac group (n=39) mean±SD (median)	P
Mean of difference of TNF-α level before and after 4 weeks treatment (pg/ml)	29.07±64.42 (19.97)	36.22±90.66 (12.94)	0.237*

Abbreviation : * *Mann-Whitney* test; TNF: *Tumor Necrosis Factor*; SD: Standard Deviation

Table 5 showed the mean of difference of TNF-α in curcuminoid group before and after 4 weeks treatment is 29.07±64.42 pg/ml while in the diclofenac group is 36.22±90.66 pg/ml. The Mann-Whitney analysis showed no significant difference between decreasing of TNF-α level between the curcuminoid group and diclofenac group with p=0.237.

Table 6. Fifty percent decreasing of TNF-α level between groups

Variabel	Curcuminoid goup (n=34)	Diclofenac group (n=39)	P
50% decreasing of TNF-α level	20	20	0.518*
Less than 50% decreasing of TNF-α level	14	19	

Abbreviation: * X^2-test

Tables 6 showed no significant difference of 50% decreasing of TNF-α level between both group (p= 0518).

DISCUSSION

Diclofenac sodium inhibit the NFκβ in inducing monocyte to produce TNF-α. As unselective COX-2 inhibitor diclofenac is able to inhibit the activation of COX-2. Diclofenac also able to block the peroxisome proliferators activated receptor-γ (PPAR-γ)[9]. Existence of these inhibition activity, diclofenac is able to suppress the production of TNF-α. This research showed the decreasing of TNF-α level in both treatment group. Decreasing the mean of difference of TNF-α level was not significant different between the curcuminoid group and diclofenac group, meaning their activities are comparable.

The reason of dropout in curcuminoid group looked not correlated to the effect of curcuminoid, except for the joint swelling and the pain.

Kertia et al (2005) have run the research regarding the influence of combination of curcuminoid from Curcuma domestica Val and essential oil of temulawak rhizome to the leucocyte count in synovial fluid of patients with osteoarthritis compared to piroxicam at random blind. The result showed that both of drugs suppressed the leucocyte count significantly and comparably[10]. Chan (1995) has run the in vitro research and proved that the curcuminoid can reduce the TNF-α production[11]. This research also showed that 50% decreasing of TNF-α level was not significantly differ between both groups.

CONCLUSION

The effect of curcuminoid in decreasing TNF- α level on patients with osteoarthritis is similar with sodium diclofenac.

REFERENCES

1. Klippel, J.H. Mediator of Inflamation Tissue Destruction and Repair. In J.H. Klippel, L.J. Crofford, J.H. Stone, C.M. Weyand (eds). *Primer on the Rheumatic Diseases*. 12th Edt. Arthritis Foundation, Georgia. 2001. pp:51-88.
2. Soeroso, J., Isbagio, H., Kalim, H., Broto, R., Pramudiyo, R. Osteoarthritis. In A.W. Sudoyo, B. Setiyohadi, I. Alwi, M. Simadibrata, S. Setiati (eds). *Buku Ajar Ilmu Penyakit Dalam* Jilid II Edisi IV. Fakultas Kedokteran Universitas Indonesia.2006. pp:1205-1211.
3. Noormartany. Gambaran Petanda Biologi Cairan Tubuh pada Penyakit Osteoatritis. In R. Pramudiyo, R.G. Wachjudi, L. Hamijoyo (eds). *Osteoarthritis Course*. Bandung. 2005. pp:32-43.
4. Sharma, P., Singh, N., Singh, V., Shrivastava, A. Bio-Immunology of Osteoarthritis. *J Immunol Immunopath*. 2002: 4(1&2):20-28.
5. Faloon, W. Arthritis Update : Drugs That Inhibit COX-2 May Cause Tissue Damage. *About Joint Health*. 2001:1-6

6. Departemen Kesehatan (Depkes). Pedoman Penggunaan Obat Opioid dalam Penatalaksanaan Nyeri. *Departemen Kesehatan RI Direktorat Jendral Pelayanan Kefarmasian dan Alat Kesehatan*. 2002. pp 42.
7. Ikatan Reumatologi Indonesia (IRA). *Panduan diagnosis dan pengelolaan Osteoarthritis*. 2004. pp:5-15.
8. Chainani-Wu, N. Safety and Anti-inflamatory Activity of Curcumin: A Component of Tumeric *(Curcuma longa)*. *J Alternat Compl Med*. 2003: 9(1):161-168.
9. Ari, S.L., Strier, L., Kazanov, K., Elkayam, O., Lechtenberg, D., Caspi, D., Arber, N. Curcumin Synergistically Potentiates the Growth-Inhibitory and Pro-Apoptotic Effects of Celecoxib in Osteoarthritis Synovial Adherent Cells. *Rheumatology*. 2006:5:171-177.
10. Kertia, N., Sudarsono, Imuno, A.D., Mufrot, Catur, E., Raharjo, P., Asdie, A.H. Pengaruh Pemberian Kombinasi Minyak Atsiri Temulawak dan Ekstrak Kunyit Dibandingkan Dengan Piroksikam Terhadap Angka Leukosit Cairan Sendi Penderita dengan Osteoarthritis Lutut. *Majalah Farmasi Indonesia*. 2005:16(3):155-161.
11. Chan, M.M. Inhibition of Tumor Necrosis Factor by Curcumin, a Phytochemical. *Biochem Pharmacol*. 1995:49(11):1551-1556.