Lack of a Clear Disease Modifying Activity of Celecoxib Treatment of End-Stage Knee Osteoarthritis: A Randomized Observer Blinded Clinical Trial

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Abstract

Objective: To evaluate the in vivo disease modifying activity of the selective COX-2 inhibitor celecoxib, compared to no-treatment and naproxen, treating end-stage knee osteoarthritis, using detailed ex vivo tissue analyses.

Methods: Patients (n=172) with end-stage knee OA were randomized to 4 groups and treated for 4 weeks prior to knee replacement surgery: celecoxib 2dd200mg, naproxen 3dd250mg, celecoxib 2dd200mg stopped 3 days prior to surgery, or no-treatment. Cartilage and synovial tissue collected during surgery were analyzed ex vivo, with cartilage proteoglycan release as primary outcome. Additionally, several markers of synovial inflammation and clinical effects were determined.

Results: Data of 138 patients could be analyzed, 34 patients were lost for several reasons. The expression of COX-2 in both cartilage and synovial tissue was statistically significant decreased in patients treated with celecoxib (p=0.017 and p=0.001) respectively, indicating the drug has reached the knee joint within the treatment period. Nonetheless, no significant effect on proteoglycan release, retention or content was found. Synovial inflammation markers did not show any statistically significant decrease although nitric oxide levels in celecoxib treated patients suggest a beneficial effect of celecoxib compared to no treatment. WOMAC scores did not statistically significant improve after treatment; though celecoxib treated patients reported a slightly higher WOMAC pain score compared to non-treated patients.

Conclusion: No direct effect on cartilage upon short term in vivo treatment of knee OA patients with celecoxib could be detected, although decreased expression of COX-2 confirmed its intra-articular availability. Effects on synovial inflammatory mediators and clinical outcome seemed only limited. As such the previous reported disease modifying effects of celecoxib in in vitro and pilot clinical studies could not unambiguously be confirmed.

ABBREVIATIONS

OA: Osteoarthritis; NSAID: Non-Steroidal Anti-Inflammatory Drug; COX-2: Cyclooxygenase-2; mPGES-1: Microsomal prostaglandin E synthase-1; DMOAD: Disease Modifying Osteoarthritic Drug; CVA: Cerebrovascular Accident; TIA: Transient Ischemic Attack; GAG: Glycosaminoglycan; IL: Interleukin; TNF-α: Tumor Necrosis Factor-A; NO: Nitric oxide; WOMAC: Western Ontario and McMaster University

INTRODUCTION

Osteoarthritis (OA) is the most common joint disorder, frequently causing pain, loss of function, and disability in adults [1]. The unknown etiology results in deterioration of structure and function of the whole joint. Treatment of OA is initially conservative, predominantly symptomatic, and is not stopping progression of the disease. Ultimately, in end-stage disease, surgical intervention is indicated. Currently there is no indisputable curative treatment. In fact, the number of joint replacements is increasing exponentially [2,3].

After acetaminophen, non-steroidal anti-inflammatory drugs (NSAIDs) are recommended as 2nd step in the pharmacological treatment of OA [4-6]. Consequently, NSAIDs are among the most commonly used pharmacological agents to alleviate OA symptoms [7]. Although they help relieve symptoms such as pain and inflammation, the direct effects of NSAIDs on cartilage may be of importance in the treatment of joint diseases, specifically when inflammation is relatively mild, as in OA, and treatment is chronic. Direct effects of NSAIDs on cartilage cannot be studied easily in...
clinical trials and are generally ignored in clinical practice. Effects of NSAIDs on inflammation shade their direct effects on cartilage [8]. In addition, (intrinsic) cartilage changes, both catabolic and anabolic, are generally very slow processes in OA. Evaluation of cartilage degeneration by imaging and biomarker evaluation is hampered by the still limited sensitivity of these methods [9]. Although limited in number, different results have been published on direct effects of NSAIDs on cartilage. In vitro and animal in vivo studies reported adverse effects of commonly used NSAIDs, like indomethacin, diclofenac, and naproxen [7,10,11]. Also neutral and beneficial effects have been published, e.g. for piroxicam and aceclofenac [7,11,12]. The number of human in vivo studies is very limited. Indomethacin and diclofenac were reported to accelerate hip and knee osteoarthritis progression [9,13]. As such, the conventional NSAIDs are potentially harmful regarding their direct effects on cartilage.

The 2nd generation NSAIDs, the selective cyclooxygenase-2 (COX-2) inhibitors (Coxibs) were suggested to have beneficial effects on cartilage volume and defects based on MRI, while non-selective COX-2 inhibitors as comparators were deleterious [14]. Several in vitro studies suggested that selective COX-2 inhibitor celecoxib has beneficial effects on cartilage. Celecoxib was reported to have a favorable effect on overall metabolism of proteoglycans and hyaluronan, making a detrimental effect on articular cartilage during long-term use unlikely [15]. Moreover, reduced production of arthritis-associated mediators and increased production of anabolic indicators by celecoxib in pig chondrocytes has been demonstrated [16]. Celecoxib showed positive effects on turnover of proteoglycans in cartilage tissue explants from OA patients [17-19].

Animal in vivo studies showed variable effects of celecoxib on cartilage. A chondroneutral effect was found in the canine Groove model and a murine OA model [20,21]. Celecoxib reduced Matrix metalloproteinase (MMP) expression and delayed the progress of arthritic damage in a posttraumatic OA mouse model [16]. In an OA model in rats celecoxib reduced the OA-like histological changes and suppressed chondrocyte apoptosis [22]. Studies with intra-articular injections with celecoxib, as an attempt to prevent cardiovascular side-effects, show promising results on cartilage protection [23,24].

By treating patients shortly before joint replacement surgery, the benefit of in vivo treatment of celecoxib was combined with the benefit of ex vivo evaluation of the joint tissues. Using this approach, it was demonstrated that a 3-month celecoxib treatment induced decreased COX-2 and mPGES-1 gene expression by the OA cartilage [25]. Moreover, cartilage proteoglycan turnover, specifically proteoglycan loss, was affected beneficially by 4-week celecoxib treatment prior to joint replacement surgery, while indomethacin showed a tendency towards negative effects [26].

Considering the effects of celecoxib on cartilage, synovial tissue and bone in vitro as well as in vivo, celecoxib and maybe other COX-2 inhibitors were postulated to have disease modifying osteoarthritic drug (DMOAD) activity. This potential is underlined by the continuous interest in celecoxib, with more recent developments such as locally applied sustained release of celecoxib [27,28]. Nonetheless, RCT’s examining DMOAD activity are lacking [29].

Therefore, a randomized clinical trial was warranted. The aim was to evaluate in vivo disease modifying activity of short term celecoxib treatment, with the attempt to modulate proteolytic activity (proteoglycan release) as an early indicator for cartilage repair activity, by treating patients with end-stage osteoarthritis with celecoxib shortly before joint replacement surgery followed by a detailed ex vivo assessment of joint tissues with observer blinded, evaluation.

**MATERIALS AND METHODS**

**Patients**

Patients with severe knee OA (n=172), on the waiting list for total knee replacement surgery at the Sint Franciscus Gasthuis Rotterdam, were included between December 2007 and June 2009. Group size was determined based on the difference in cartilage proteoglycan loss over three days ex vivo incubation between no treatment and celecoxib treated patients in a previous pilot study [26], using α=0.05 and β=0.8.

Exclusion criteria were: total knee replacement for other reasons than OA, history of gastrointestinal bleeding or perforation, increased risk for cardiovascular disease (history of cardiovascular disease like myocardium infarct, heart failure, CVA (cerebrovascular accident) and TIA (transient ischemic attack); untreated/insufficiently treated hypertension; angina pectoris and use of oral anticoagulants), serious liver and/or kidney dysfunction and known intolerance for naproxen. Patients already on NSAIDs had to stop their medication at least 7 days prior to start of the study medication.

The study was conducted according to the declaration of Helsinki and received ethics approval of the hospital. A written informed consent was given by each patient before participating in the study.

**Study design**

Patients were randomized to one of four treatment groups 4 weeks prior to total knee replacement surgery. Patients were randomized to celecoxib 2dd200mg (until surgery), celecoxib 2dd200mg until 3 days before surgery (to control for the obligated stop of naproxen treatment 3 days before surgery), and naproxen 3dd250mg until 3 days before surgery, or no treatment (controls). Because of its platelet-inhibiting effect, the use of naproxen had to be stopped 3 days prior to surgery. All 4 groups included 43 patients. Because of the increased risk for gastrointestinal adverse effects with the use of naproxen, all patients also received omeprazol 20mg o.d.

The no-treatment group used no NSAIDs during the study period. To check for treatment compliance patients were interviewed whether medication was taken.

Four weeks of treatment was considered sufficient to alter cellular (proteolytic) activity in cartilage based on previous in vitro experiments [17], where changes in proteoglycan release, the primary outcome of this study, are enabled within one week.
Clearly a 4 week-treatment was anticipated to be too short to alter cartilage tissue structure significantly.

Tissue collection

After 4 weeks of treatment, at total knee replacement surgery, femoral cartilage (both medial and lateral) with underlying bone and synovial tissue was obtained. Tissue was kept in phosphate buffered saline for maximum 4 hours during transport. At the University Medical Center Utrecht (UMCU) tissue was processed under laminar flow conditions. Investigators performing experiments and analysis were fully blinded to patients’ clinical data and medication use. Blinding was guaranteed until all data of all patients were gathered.

From weight bearing areas of the femoral condyles all cartilage was cut aseptically at full thickness from the underlying bone. These slices were cut into full thickness squares.

From each donor 24 samples were taken randomly. Twenty samples were weighted (range 5-15mg, accuracy 0.1mg) and incubated ex vivo for 3 days in culture medium for biochemical analyses (37°C, 5% CO₂). Four samples were fixed for histochemistry. Additionally, four synovial tissue samples (range 50-150mg, accuracy 0.1mg) from each donor were taken randomly. Two were incubated ex vivo for 3 days in 4 ml culture medium for biochemical analyses (37°C, 5% CO₂). The supernatants of these cultures were harvested and rendered cell-free by centrifugation (1300g). Two samples were fixed for histochemistry.

Remaining cartilage and synovial tissue of the control group and the full celecoxib group was used for COX-2 expression analyses. The limited amount of tissue available limited the analyses of COX-2 for cartilage to 18 treated patients and 25 non-treated patients and for synovial tissue to 31 treated patients and 25 non-treated patients.

Macroscopy and histochemistry

Macroscopic cartilage damage and synovial tissue inflammation were evaluated on digital high-resolution photographs of femur surface parts and synovial tissue, by two observers blinded for source of photographs (scores adapted from [20]). Severity of cartilage damage was graded from 0-3: 0=fibrillation or focal degeneration, 1=degeneration at multiple locations, 2=degeneration at multiple locations with focal lesions, and 3=degeneration throughout the tissue with severe focal lesions and focally full cartilage abrasion. Synovial tissue inflammation was graded from 0 to 2 for color, angiogenesis and fibrillation: 0=none, 1=slightly, 2=strong.

For histochemistry, cartilage and synovial tissue samples were fixed in 4% phosphate buffered formalin with 2% sucrose. Cartilage was stained with Safranin-O fast green-iron haematoxylin. Synovial tissue samples were stained with Haematoxylin-Eosin. Both tissues were sliced and from each sample 3 slices were used for histological scoring. Cartilage damage was scored using modified Mankin score [30,31]. Synovial tissue inflammation was graded using modified Goldenberg and Cohen score [32,33].

Two observers blinded to the source of the samples, scored all samples and the averages of observers and samples were taken as representative score of each donor. When observers scored >1-point difference, consensus was sought.

Intra-articular COX-2 expression

As a control whether the drug had actually reached the joint, COX-2 expression in cartilage and synovial tissue was evaluated by Western Blot analysis. Total protein was extracted by crushing cartilage and synovial tissue samples using sonification (ART-MICCRA D-1, Müllheim, Germany) and RIPA lysis buffer (Thermo Fisher Scientific Inc., Waltham, MA, USA). Supernatant was collected and the concentration of each sample was determined using a BCA protein assay kit (Thermo Fisher Scientific). Proteins were denatured, separated by 7.5% SDS-PAGE and transferred to nitrocellulose membrane 0.45um (Bio-Rad Laboratories Inc., Hercules, CA, USA). Membranes were incubated overnight with primary antibodies against COX-2 (polyclonal rabbit anti-human, 1:1500, Santa Cruz Biotechnology, Inc., Dallas, TX, USA) and marker protein GAPDH (monoclonal mouse anti-human, 1:500, Santa Cruz Biotechnology, Inc.).

Membranes were washed and incubated with secondary antibodies. For GAPDH a rabbit anti-mouse IgG HRP-labelled antibody (1:1000, DAKO Agilent, Agilent technologies. Glostrup, Denmark) was used and for COX-2 a goat anti-rabbit IgG HRP-labelled antibody (1:10.000, Santa Cruz Biotechnology, Inc.). Protein bands of GAPDH (37kDa) and COX-2 (69-72kDa) were visualized using the Proxima C16 Phi+ (Isogen Life Science, de Meern, The Netherlands). Protein expression was analyzed by Total Lab 1D software (Totallab, Newcastle, UK), comparing COX-2 band volumes to GAPDH (marker protein) band volumes.

Cartilage biochemistry

Twenty randomly taken cartilage samples of each donor were used for biochemical analysis.

Proteoglycan release, release of newly formed proteoglycans (as a measure of retention of proteoglycans) and content were determined as described before [26,31,34]. Values for total proteoglycan release were normalized to proteoglycan content of explants and expressed as percentage GAG release (%). Values for cartilage proteoglycan content were normalized to wet weight of the cartilage sample and expressed as milligrams of GAG per gram wet weight of cartilage tissue (mg/g).

Values for release of newly formed proteoglycan were normalized to proteoglycan synthesis rate, and expressed as percentage of newly formed proteoglycans.

Synovial tissue inflammation

Interleukin-1β (IL-1β), tumor necrosis factor-α (TNFα) and nitric oxide (NO) levels were determined as described before [26]. IL-1β and TNFα were determined by ELISA and expressed as pg/ml per mg synovial tissue and NO levels were determined using the standard Griess reaction and expressed as uM per mg (wet weight) synovial tissue.

Clinical outcome

Patients were asked to fill out a Western Ontario and McMaster University (WOMAC) questionnaire to evaluate pain, stiffness, and function, before and after medical treatment [35].
Calculations and statistical analysis

Patients were not blinded for medication. As such clinical outcome, based on patient report was not blinded. For all other parameters, the trial was fully blinded until all assays were performed and all data were collected.

In all cases, multiple data from each patient (e.g. the results of 20 cartilage samples per patient obtained at random and handled individually) were averaged and taken as a representative value for that patient. This is done to compensate for focal differences in composition and bioactivity of the tissue (especially in case of severe OA).

Because of a drop-out of 34 patients, intention to treat and per protocol analyses were performed. Data from per protocol analyses have been given in figures and table and did not differ from those of intention to treat analyses.

When assumptions for parametric testing were not met a log transformation of the variables was performed. Statistical evaluation of effects of treatment was performed using MANOVA tests followed by Dunnett’s post-hoc tests for further analysis of differences and Independent Sample T-tests for analysis of COX-2 expression (SPSS software, version 21.0 for Windows, Chicago, IL, USA). P-values less than or equal to 0.05 were considered statistically significant.

The primary hypothesis was that 4 weeks’ in vivo treatment of knee OA patients with celecoxib can alter proteoglycan release of cartilage (beneficially) via direct or indirect in vivo effects of celecoxib.

RESULTS

Patient characteristics

All 172 patients were included and randomized to four treatment groups. After randomization, a total of 34 patients were lost to follow-up due to several reasons, summarized in Figure 1. Statistically, there was no selective drop-out in either of the treatment groups. In total, 138 patients could be evaluated.

Patient characteristics for each of the treatment groups (per protocol analyses) are provided in Table 1. For none of the 4 groups demographic data differed significantly from all randomized patients (data not shown). None of the characteristics was significantly different between the 4 groups.

Expression of COX-2

In order to evaluate whether treatment led to a decrease in the main target of NSAIDs, cyclooxygenase (COX)-2 activities, ex-vivo COX-2 levels in cartilage and synovial tissue were determined by Western Blot (Figure 2). The control group and celecoxib until surgery group were considered as most extremes and evaluated.

Figure 1 Patients from inclusion until analyses in the different treatment groups.
Table 1: Patient characteristics for the different treatment groups.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Treatment group</th>
<th>No Celecoxib</th>
<th>Celecoxib full</th>
<th>Celecoxib -3 days</th>
<th>Naproxen -3 days</th>
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<td>(n=39)</td>
<td>(n=34)</td>
<td>(n=33)</td>
<td>(n=32)</td>
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<tr>
<td>Age (year)</td>
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<td>22 / 12</td>
<td>20 / 13</td>
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<td>2.8 (0.7)</td>
<td>3.1 (0.6)</td>
<td>3.1 (0.7)</td>
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<td>X-ray joint space narrowing, Altman[37]</td>
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<td>X-ray osteophytes, Altman</td>
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<td>Cartilage</td>
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<td>2.2 (0.1)</td>
<td>2.3 (0.1)</td>
<td>2.2 (0.1)</td>
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<td>4.8 (0.2)</td>
<td>4.5 (0.1)</td>
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<td>38.1 (3.1)</td>
<td>44.1 (3.4)</td>
<td>43.7 (2.8)</td>
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</tbody>
</table>

Patient characteristics presented as means ± SEM (except for gender * which is presented as female/male ratio). There were no statistically significant differences between the treatment groups for any of the baseline characteristics amongst groups and compared to the originally randomized patients in each group. K&L: Kellgren and Lawrence. WOMAC: Western Ontario and McMaster University.

A statistically significantly decrease in COX-2 expression in both cartilage (35%, p=0.017) as well as synovial tissue (41%, p=0.001) was found, indicating that the drug had reached the knee joint in amounts able to decrease COX-2 amount in both tissues.

Proteoglycan parameters of the treatment groups

Despite the fact that joint tissue COX-2 was reduced in the celecoxib treated patients this was not reflected in a changed ex-vivo total proteoglycan release (Figure 3A), the primary outcome parameter of this study. No statistically significant effects on proteoglycan loss could be found (-7.5%, p=0.878 and -5.6%, p=0.990 for treatment until surgery and stopped 3 days in advance, respectively. The naproxen group showed no change in proteoglycan loss (-0.5%, p=0.978) (Figure 3B).
Similar results were seen in the ex-vivo retention of newly formed proteoglycans in the tissue; +9.0% for celecoxib until surgery, +14.7% for celecoxib stopped 3 days in advance and -0.4% for naproxen treated patients (all not statistically significant). As anticipated for only a 4-week treatment period, ex-vivo proteoglycan content was not statistically significant different between the 4 treatment groups (Figure 3C).

Synovial tissue inflammation

In order to evaluate inflammatory activity of synovial tissue, ex vivo IL-1β, TNFα, and NO production of synovial tissue was measured (Figure 4). NO levels of celecoxib treated patients suggested a decrease but were not statistically significant changed (-18.3%, p=0.164). For IL-1β and TNFα production by synovial tissue no (statistical) differences between groups were found either. As such, no clear anti-inflammatory effects could be demonstrated upon treatment with celecoxib.

WOMAC score

Although this study was not designed to evaluate clinical efficacy, changes in WOMAC scores before and after treatment were evaluated. One of the celecoxib treatment groups reported an improvement in WOMAC pain score, compared to the no treatment group (p=0.056 for celecoxib stopped 3 days in advance). The other celecoxib group showed a comparable trend (p=0.275). One can argue whether these changes were clinically relevant as actual changes are very small and patients were not blinded. Naproxen treatment did not show a change in WOMAC pain score (Figure 5A).

Comparable trends were obtained for the other WOMAC subscales (Figure 5B, 5C) and the total WOMAC score (Figure 5D).

DISCUSSION

The present randomized controlled observer blinded trial could not confirm the previously reported disease modifying osteoarthritic drug effect of a 4-week celecoxib treatment of severe OA prior to joint replacement surgery, despite using good powered group sizes (based on the pilot study [26]). The celecoxib group showed the most profound effect on proteoglycan release, the primary endpoint. Nonetheless, this effect was minor and not statistically significant. Similarly, the decreased NO production upon celecoxib treatment appeared not statistical significant. Very limited clinical benefit was reported by the patients receiving celecoxib. Taken together, the reported disease modifying effects of celecoxib in vitro and effects in small size clinical studies could not unambiguously be confirmed, however these findings are based on end-stage knee OA and might not hold for early OA.

No altered cartilage proteoglycan loss (via a direct effect on chondrocytes or indirectly via changing synovial cell activity)

![Figure 3 Cartilage proteoglycan parameters.](image)

The differences between the four treatment groups in percentage proteoglycan release (A; primary outcome), release of newly formed proteoglycans (as a measure of retention of these newly formed proteoglycans) (B), as well as proteoglycan content (C). Results are presented for each individual patient (dots) and means ± SEM (dash with whiskers). According to the hypothesis, proteoglycan releases (total and newly formed) were anticipated to be decreased in the celecoxib treated groups. Values for percentage decrease compared to untreated patients are given.
Figure 4 Ex vivo inflammatory mediator release of synovial tissue.

The synovial tissue ex vivo release of the inflammatory mediators IL-1β (A), TNFα (B), and NO (C) measured in the 3-day synovial tissue culture media of the four treatment groups. Results are presented for each individual patient (dots) and means ± SEM (dash with whiskers).

Figure 5 The effects of treatment on WOMAC scores for the 4 treatment groups.

The differences between the four treatment groups in change in WOMAC pain (A), WOMAC stiffness (B), WOMAC function (C), and WOMAC total (D) scores. Changes are calculated as baseline scores – follow-up scores after a treatment period of 4 weeks. Results are presented for each individual patient (dots) and means ± SEM (dash with whiskers). P-value is given for WOMAC pain in celecoxib until 3 days before surgery compared to no treatment group.
upon short-term treatment in this RCT could be demonstrated. This despite a decreased COX-2 expression in cartilage and synovial tissue demonstrating drug bioavailability within this short-term RCT, although this evaluation was limited to the patients who received full celecoxib treatment or no treatment for reasons mentioned previously. Extrapolating this to the other treatment groups these data suggest that the absence of effect on proteoglycan turnover cannot simply be explained by a lack of drug bioavailability in the joint during the 4-week treatment.

A great advantage of the set-up of this RCT is the opportunity to perform a full detailed biochemical analysis of the articular cartilage and other joint tissues while the treatment was given in vivo. A drawback is that in addition to the direct effects on cartilage (as evaluated thus far in vitro), there will be an effect on synovial tissue inflammatory activity, with expectedly indirect effects on cartilage. Moreover, the cartilage tissue studied involves severely damaged (end stage disease) tissue.

Patients that received no treatment showed proteoglycan retention, release, and content typical for osteoarthritic cartilage [26,31]. But unanticipated, proteoglycan release was not statistically significant changed by treatment. This suggests that earlier studies using a similar protocol may have been biased by group size [26]. However, the present RCT, blinded for the primary outcome evaluation, has also its limitations: the condition of cartilage before start of treatment is unknown, enabling only unpaired evaluation. In case values were already different before start of medication despite randomization, which could be the case in patients on long-term NSAID treatment before inclusion for example, changes due to treatment might not have become apparent. It is expected that histochemical and radiographic evaluation of cartilage after treatment is representative of cartilage condition before start of treatment, as it is unlikely that histochemical grade and X-ray (Altman) score will change significantly in a relative short period of 4 weeks. It should be kept in mind that cartilage tissue studied involves severely damaged (end-stage diseased) tissue. Only hyaline cartilage that could be cut from the joint surface after replacement surgery was used. This is the limitation of the approach using in vitro treatment with extensive detailed ex vivo evaluation of tissues obtained after replacement surgery. It could be interesting to evaluate similar parameters in healthy or less severely damaged cartilage, as in earlier stages of OA, when treatment with NSAIDs is considered. But in such an approach biopsies need to be taken.

Others reported on in vivo studies in which celecoxib did seem to have a chondro protective effect. An explanation for differences between those studies and ours could be duration of celecoxib treatment: 4 weeks prior to knee replacement surgery (based on our pilot study [26]) versus 3 months up to 3 years in other studies [25,36]. Although patients were told not to use any NSAIDs beside their treatment regime it cannot be ruled out that patients may have used NSAIDs (occasionally) on top of their prescribed dosage. When this happened more often in the no treatment group, because they were without pain medication compared to the treatment groups, this could have caused the lack of significant differences in the clinical outcome.

Another possible explanation of lack of clear statistically significant effects could be the number of patients who dropped-out of the study. Based on a previous pilot study we included 172 patients with a power of 0.86. The loss of 34 patients led to a power of 0.82, a minimal decrease, not expected to be the sole cause of the absence of an effect. This is supported by the fact that there are no differences in outcome when the study is evaluated by intention to treat analyses vs. per protocol analyses.

Both celecoxib treated groups show a tendency towards a lower percentage proteoglycan release compared to the no treatment group. Nonetheless these effect sizes noticed were not in the magnitude of the results shown in previous work, both in vitro as in vivo [17-19,26]. However, it should be kept in mind that a decrease of 5% or more in proteoglycan loss in 4 weeks can be of clinical relevance in case of long term treatment. Another (indirect) measure of treatment efficacy is the inhibition of proinflammatory cytokine release; in addition to analgesic effects, NSAIDs are known to have anti-inflammatory effects. This is best seen in the synovial lining of the affected knee. Both celecoxib and naproxen treatment where unable to lower the release of IL-1β and TNFα by synovial tissue, but NO production could be diminished. A decrease in IL-1β and TNFα might have been expected, however in end-stage disease inflammation might be limited.

Corroborating the very mild effects of celecoxib on biochemical level, analgesic effects of celecoxib treatment evaluated by WOMAC scores were also limited. It might be that 4 weeks of medication is critical, and a minimum (and maybe too short) duration to observe clinical effects, although in clinical practice sufficient.

CONCLUSION

In conclusion, the present randomized controlled observer blinded clinical trial demonstrates no clear decrease in cartilage tissue proteoglycan release as surrogate for cartilage repair activity (via DMOAD activity) by a 4-week celecoxib treatment in end stage disease, despite sufficient intra-articular availability of the drug. No adverse effects were found either.

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