Department of Dental Medicine

Prediction of the pain relieving effect of intra-articular glucocorticoid treatment by tumor necrosis factor and serotonin in the synovial fluid.

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Abstract

The aim of this study was to determine which inflammatory mediator, serotonin (5-HT) or tumor necrosis factor is most significant for prediction of local treatment pain with intra-articular glucocorticoid in patients with inflammatory disease of the temporomandibular joint (TMJ).

Twenty patients with TMJ involvement treated with intra-articular glucocorticoid were included in the study. Synovial fluid levels of 5-HT and TNF, TMJ resting pain, movement pain as well as tenderness and pain reflex to digital palpation were assessed. The effects of the treatment were assessed in an interval between 2 weeks and 3 months. The mean duration of TMJ symptoms was 7, 8 years.

This study shows that 5-HT has the highest significance and predictive ability for intra-articular glucocorticoid treatment of inflammatory-related pain in the TMJ in patients with inflammatory disorders of the TMJ.

It can be concluded from this study that pretreatment synovial fluid level of 5-HT but not TNF predicts change (improvement) in TMJ resting and movement pain achieved by intra-articular administration of glucocorticoid in patients with inflammatory disorders of the TMJ.

Authors’ contributions

Author Lisa Callmar has contributed to the project by doing research and a litteratur study for the introduction, selecting patients from a database that are significant for the study, making table 1 and 2 and analyzing the results.

Author Farzaneh Azimi has contributed with evaluating patient materials from database, literature study for background, analyzing results and writing a conclusion.
**Introduction**

The pain relieving effect of intra-articular glucocorticoid treatment is depending on the degree of inflammation in the joint and the presence of inflammatory mediators like TNF and 5-HT (Fredriksson et al 2005, 2006). These two mediators may be independent of each other.

**Pain**

The definition of pain used by the International Association for the study of Pain is as follows; “An unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage.” Certain tissue events cause pain because they activate afferents specialized in the detection of somatic problems or problems in the body. These afferents are known as nociceptive nerves. These nociceptive nerves are activated by extreme stimuli such as strong pressure or stretch, but also tissue damage and inflammation. The word nociception is derived from the Latin word *nocere*, to damage. It is a sensation as a result of the activation of specialized receptors on nociceptive nerves, the so called *nociceptors*. Nociceptive pain occurs as a result of the activation of nociceptors. It could be called somatic *pain*, which is pain in the classic sense of the word, in absence of any psychological influence and without damage of the nervous system.

The afferent innervations of the jaw are dominated by A-delta – and C-fibers.

**A-delta** fibers have a thin diameter of 1-3 micron and are coated with a layer of fatty nerve tissue, myelin, and have a nerve impulse velocity of 5-25m/s.

**C-fibers** have a thinner diameter of 0.2-2 micron, are not myelin coated and have a nerve impulse velocity of 0.1-2m/s.

Because of the velocity difference, the initial activity conduct through A-delta fibers is the first to reach CNS. This pain is distinct, clear and well localized, while pain conducted through C-fibers is diffuse and more difficult to locate.

Some of the afferents are excited already by normal movement of the joint while some are activated first by potential damaging movements. The reason pain may occur just by normal movement is due to lowered pain threshold in peripheral nerves including nociceptors which makes pain occur even by very small pain stimuli.
**Inflammatory pain in the TMJ**

The subjective symptoms of inflammatory joint disorders of the TMJ are dominated by pain spontaneous pain or resting pain or movement pain?

Common clinical sign are tenderness to digital palpation of the joint and restricted mandibular mobility due to fibrous adhesions crepitus of the joint.

When an inflammatory process occurs, production starts of substances/molecules that have a direct activating effect on nociceptors but also increases their sensitivity by lowering the threshold for activation which latter is known as peripheral sensitization.

Nociceptive nerves produce chemicals called neuropeptides in their cell bodies. These neuropeptides are transported to the nerve’s peripheral terminal, where they are stored. When a nerve receives a stimulus that is strong enough for it to depolarize as when an injury is sustained, it releases the neuropeptides into the tissues it innervates. The most well known neuropeptides are substance P (SP) and Calcitonin Gene Related Peptide (CGRP). The effects of SP and CGRP are potent enough to cause and maintain an inflammatory response in absence of any tissue damage. When this happens, it is referred to as a neurogenic inflammation which occurs in e.g. arthritis.

Inflammatory joint disorders, such as RA are complex processes involving peripheral inflammatory mediators and their receptors, sympathetic neurons and the central nerve system. A number of mediators have been shown to have initiating and modulating roles in the inflammatory process, mainly by effects on mediator-specific receptors. The mediators we will put primary focus on in this project are TNF, serotonin, IL-1 and IL-6.

**Tumor Necrosis Factor**

Tumor necrosis factor (TNF) is a potent cytokine that activates macrophages, neutrophiles and eosinophiles. TNF from inflammatory processes stimulate the acute phase response and thereby production of C-reactive protein (CRP) in the liver. TNF is also involved in regulation of immune cells and induce cell death.

TNF is produced mainly by T-cells and macrophages at the site of inflammation and modulates the local inflammatory response. TNF is important as a disease promoting mediator in arthritis joints (Isomäki and Pumonen 1997). TNF levels are increased in the synovial fluid and membrane of patients with RA (Buchan et al. 1998) and in synovial cells. TNF up regulates the production of other pro-inflammatory cytokines such as IL-1 (Brennan et al 1989) and IL-8.
TNF also stimulates PGE$_2$ and collagenase production, which induces cartilage and bone destruction (Dayer et al. 1985). The synthesis of PGE$_2$ is inhibited by glucocorticoids (Kopp 2001).

**TNF and pain**

A major presenting symptom of arthritis is pain. This is caused by the activation and sensitization of nociceptive nerve fibers ("pain fibers") that supply the joint and the ensuing activation of the central nociceptive system. Pain reduction in the course of anti-inflammatory therapy might simply result from an attenuation of inflammatory. Alternatively, anti-inflammatory compounds may neutralize endogenous mediators that cause ongoing pain and/or hyperalgesia by directly activating or sensitizing nociceptive neurons. Both TNFRI and TNFRII have found to be expressed in a large proportion of DRG neurons, indicating that primary afferent neurons may be a target of anti-TNF treatment.

Immunohistochemical examination has shown the presence of TNF alpha in the synovium, e.g. in the lining layer, some endothelial cells and most importantly, in the cells in the cartilage pannus junction. TNF receptors have a similar distribution; (Maini et al 1993). TNF-alpha may have an indirect effect in receptor sensitization by initiating increased production of IL-1a, which has been shown to cause pain and hyperalgesia (Watkins et al 1995, Safieh-Garabedian 1995).

TNF-alpha also has a direct modulatory effect on pain by TNFR on the nociceptive primary afferent fibers to induce ectopic activity. Studies of TNF-alpha in relation to nociception have shown that experimental hyperalgesia can be caused by local or systemic administration of the cytokine. High levels of TNF have been detected in the synovium and synovial fluid of patients with inflammatory diseases. Blocking of the production of TNF has been introduced as a new therapy for patients with inflammatory diseases. In preliminary clinical trials that included patients with RA, anti-TNF antibodies appear to have a significant effect on disease activity, including reduced C-reactive protein and serum amyloid-A production. Therefore, TNF alpha seems to be a possible therapeutic target in patients with RA (Kopp 2001).

**Serotonin**

Serotonin (5-HT) is a neurotransmitter in the central and peripheral nervous system, which mediates nociceptive pain from peripheral tissues.
5-HT is an important endogenous peripheral mediator of pain and inflammation, which is produced in the enterochromaffin cells of the gastrointestinal mucosa and then is absorbed to platelets. In addition, peripheral afferent nerves have been shown to synthesize 5-HT in the cat (Herbert et al. 1992) but not yet in humans. It is also released from activated mast cells.

The known 5-HT receptors comprise seven groups (5-HT 1-7) with a total of 14 subtypes which all belong to the G-protein coupled receptors. Only one, the 5HT3 receptor is ligand-gated to fast ion (Na+/K+) channels (Gyermek 1996, Wolf 2000).

According to Taiwo and Levine (1992), 5-HT produces hyperalgesia by a direct action on primary afferent neurons via the 5-HT1A subset of serotonin receptors, but it also sensitizes sensory neurons by the 5-HT2 receptor (Rueff, Dray 1992). Richardson and Engel (1986) showed that 5-HT participates in the mediation of spontaneous pain from inflamed peripheral tissues by sensitizing or exciting fine afferent units via the 5HT3 receptor.

The 5HT3 receptor is peripherally only located on afferent neurons (Martin & Humphrey 1994).

Studies show that 5-HT is a mediator of pain in the TMJ with inflammation and that TMJ resting pain is modulated by the 5-HT3 receptor on primary afferents in the synovial tissue (Voog et al. 2003).

Granisetron acts by selective antagonism of 5-HT3 receptor both peripherally and centrally (Yarker et al 1994).

Ernberg et al. (2000) found that injections of granisetron into the healthy human masseter muscle reduce pain induced by local administration of 5-HT. Granisetron has been shown to have an immediate, short lasting and specific anti-nociceptive effect on TMJ resting pain after intra-articular administration in patients with systemic inflammatory joint disorders (Voog et al. 2003).

No such specific effect on pain provoked by movement or by external pressure was found. Since granisetron is a 5-HT3 receptor antagonist, the superior response might be due to specific 5-HT3 receptor blocking.

These results state/indicate that the 5-HT3 receptor elicits spontaneous pain by chemical activation rather than allodynia/hyperalgesia by mechanical stimulation (Giordano et al 1989).

**Interleukin1**

Interleukin 1 (IL-1) is a cytokine produced by macrophages, monocytes and dendritic cells. IL-1 has a significant role in the mediation of acute and chronic inflammation which leads to inflammatory response and destruction of connective tissue.
Interleukin 6 (IL-6) is a cytokine that acts mostly pro-inflammatory but sometimes anti-inflammatory and is an important mediator of the acute phase response as well as to tissue damage. T-cells and macrophages secrete IL-6 to stimulate immune response.

Glucocorticoid treatment of inflammation

Glucocorticoids (GC) administered locally or systemically suppress inflammation and pain in e.g. rheumatoid arthritis. Glucocorticoids pass through the cellular membrane and bind to glucocorticoid receptors (GCR) in the cytoplasm. Glucocorticoids have an inhibitory effect on inflammatory mediator release from many cell types involved in inflammation such as macrophages, T-lymphocytes, mast cells, dendritic cells and neutrophilic leukocytes. Activated receptors inhibit the expression of genes for pro-inflammatory cytokines, while increasing the expression of genes coding for anti-inflammatory proteins like the IL-10 and IL-1 receptor antagonist. (Fredriksson et al 2006) Treatment with intra-articular glucocorticoid of the temporomandibular joint (TMJ) in patients with RA has proved efficient in alleviating pain and tenderness (to digital palpation of the lateral aspect of the joint) as well as increasing mobility for at least 4-6 weeks after administration. Glucocorticoids might exert therapeutic effects on different pain entities such as resting pain, pain on joint movement, tenderness to digital palpation, or pressure pain threshold, perhaps due to a different molecular mechanism behind each pain entity. The findings in a study made by Fredriksson et al 2005, indicate a local as well as systemic influence of 5-HT on the changes in TMJ pain induced by local glucocorticoid. However, the influence of local and systemic 5-HT has opposite directions, that is, detectable synovial fluid 5-HT predicts pain relief, while high plasma levels of 5-HT predicts less reduction of pain (Kopp et al, 2002). 5-HT detectable in synovial fluid is a positive predictor of pain relief, while high plasma level of 5-HT is a negative predictor of pain relief.

Another study Fredriksson et al (2006) indicates that local TNF-related mechanisms influence the treatment effect of glucocorticoids on TMJ movement pain and that presence of TNF in synovial fluid predicts a positive treatment response in patients with chronic inflammatory TMJ disorders. TMJ movement pain is the clinical variable with the strongest relation to an inflammatory condition of the TMJ as determined by presence of inflammatory mediators in the synovial fluid (Nordahl et al 2000, Alstergren et al 1997) and therefore the most valid clinical sign of TMJ arthritis.
Aims
The aim with this project is to clarify what mediator, 5-HT or TNF, has the highest significance and predictive ability for pain reduction by intra-articular glucocorticoid treatment of inflammatory-related pain in the TMJ.

Specific questions
1. Is synovial fluid pretreatment level of 5-HT or TNF correlated to change in TMJ resting (spontaneous) pain?
2. Is synovial fluid posttreatment level of 5-HT or TNF correlated to change in TMJ resting (spontaneous) pain?
3. Is synovial fluid pretreatment level of 5-HT or TNF correlated to change in movement pain?
4. Is pretreatment SF-5-HT or SF-TNF correlated to change in tenderness of the TMJ?

Materials and methods
All patients with synovial fluid and blood samples analyzed for inflammatory mediators (TNF, 5-HT, IL-1, IL-6) before and up to 12 weeks after intra-articular glucocorticoid treatment were selected from the data base at the department of Clinical Oral Physiology.

The patient background data are shown in Table 1.

Patients
This study comprised 20 patients (Table 1), one male and nineteen females, with a mean age of 47.1 years. The selected patients had different forms of chronic inflammatory TMJ disease, such as seropositive/seronegative RA (n = 4/5), ankylosing spondylitis (n = 3), osteoarthritis (n = 1), psoriatic arthropathy (n = 2), chronic unspecific polyarthritis (n = 1), Sjögren’s syndrome (n = 2) or Marfan’s syndrome (n = 2). TMJ inflammatory disorder was present bilaterally or unilaterally according to the diagnostic classification by the American Academy of Orofacial Pain (Okeson et al. 1996). The mean duration of TMJ symptoms was 7.8 years. The study was approved by the local ethical committee at Karolinska University Hospital in Huddinge, Sweden (142/02 and 176/91).
### TABLE 1. Demographic and background data of patients.

<table>
<thead>
<tr>
<th></th>
<th>Min</th>
<th>Median</th>
<th>Mean</th>
<th>25th</th>
<th>75th</th>
<th>Max</th>
<th>SD</th>
<th>n</th>
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<tbody>
<tr>
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<td>23</td>
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<td>39</td>
<td>55</td>
<td>67</td>
<td>10,7</td>
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<tr>
<td>Males/females</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1/19</td>
</tr>
<tr>
<td>Duration of general disease</td>
<td>years</td>
<td>0</td>
<td>5</td>
<td>7,8</td>
<td>1,8</td>
<td>13,8</td>
<td>25</td>
<td>7,8</td>
</tr>
<tr>
<td>S-5-HT</td>
<td>&gt; 20 IU</td>
<td>17</td>
<td>1147</td>
<td>1233</td>
<td>680</td>
<td>1330</td>
<td>2877</td>
<td>845</td>
</tr>
<tr>
<td>P-TNF</td>
<td>(pg/mL)</td>
<td>2,7</td>
<td>14,9</td>
<td>21,6</td>
<td>13,5</td>
<td>32,3</td>
<td>64</td>
<td>16,4</td>
</tr>
<tr>
<td>S-CRP</td>
<td>(pg/mL)</td>
<td>0</td>
<td>0</td>
<td>6,2</td>
<td>0</td>
<td>8,3</td>
<td>63</td>
<td>15,1</td>
</tr>
</tbody>
</table>

n = number of observations

### Treatment

Examinations were made before and after treatment. At the first examination, glucocorticoid was injected into the TMJ after the synovial fluid sampling. A volume of 0.7 mL of the glucocorticoid methylprednisolone (40 mg/mL) with Lidocaine (10mg/mL) added (Depo-Medrol cum lidocaine; Pfizer AB, Täby, Sweden) was injected into the upper joint compartment of the TMJ. The effect of treatment was analyzed within an interval of 2 to 12 weeks.

### Pain assessments before and after treatment

#### Resting (spontaneous) pain

The patients were questioned about the intensity of resting pain in the TMJ. They were presented a visual analogue scale (VAS; 1-100) where the end-point to the left of the 100 mm line was marked “No pain” and the right with “Worst pain ever experienced” and the patients marked out on the line where it corresponded to the degree of their complaints.

Movement pain in the TMJ during maximum mouth opening was assessed by the patient according to a numeric rating scale (NRS; 0-10). Movement pain was also expressed as number of painful movements, i.e. pain during maximum mouth opening.
opening, laterotrusion to right and left, protrusion as well as retraction (0-5).

Presence of tenderness to digital palpation (allodynia/hyperalgesia) in the TMJ region as well as pain reflex upon palpation was assessed (0-2) over the lateral and the posterior aspect (via the external meatus) of the joint on each side. Two units were scored if the tenderness or pain reflex was bilateral.

**Mandibular mobility**

Mandibular mobility was measured as maximum voluntary mouth opening (MVM) and was measured in mm between the incisal edges with vertical overbite added.

**TMJ crepitus**

Presence of TMJ crepitus was assessed bilaterally (0-2) by digital palpation.

**Synovial fluid sampling from the temporomandibular joint**

TMJ synovial fluid samples were obtained by washing by washing the joint cavity with saline using a push and pull technique (Alstergren et al. 1999). The washing solution, consisting of 78% saline (NaCl 9 mg/ml, Pharmacia Upjohn, Uppsala, Sweden) and 22% hydroxocobalamin (Behepan 1mg/ml; Pharmacia Upjohn, Uppsala, Sweden), was slowly injected into the posterior part of the upper joint cavity approximately 1 ml at a time and then aspirated. The total volume of the washing solution was 4 ml. The hydroxocobalamin was included in order to determine the amount of synovial fluid in the aspirate by comparing the spectrophotometric absorbance of the aspirate and that of the washing solution. The synovial fluid level was then calculated. Only samples that fulfilled previously established sample quality criteria were included in the statistical analysis (Alstergren et al., 1999).

**Mediator analysis**

The concentrations of investigated mediators were determined by commercially available enzyme-linked immunosassays with highly specific antibodies (5-HT: Kit nr 0642 Immunotech International; TNF: TNF-alpha EASIA, Medgenix, B 6220 Fleurus) (Alstergren et al, 2008).
Statistics

Correlations were tested with the Pearson product moment correlation coefficient when variables had normal distribution and Spearman rank correlation coefficient ($r_s$) when not. The probability used for statistical significance was $p< 0.05$. Multiple stepwise regression analysis was used to estimate the relative ability of SF-5-HT and SF-TNF to predict treatment effects.

Results

The change of studied pain variables after administration of glucocorticoid into the TMJ is shown in Table 2.

SF-5-HT and SF-TNF were independent according to correlation analysis ($r_s = -0.09, n=20, p=0.692$. Multiple stepwise regression with both variables showed that SF-5-HT was the strongest and only significant predictor for change after treatment in resting pain of the TMJ ($r^2=0.22, n=18, p=0.028$).

Also post-treatment level of SF-5-HT was positively and significantly correlated to change in TMJ resting pain ($r=0.52, n=18, p=0.026$), but not SF-TNF.

Change in TMJ pain upon mouth opening after treatment was positively correlated to and predicted by pretreatment level of SF-5-HT ($r=0.62, n=20, p=0.003$), but not TNF ($r=-0.02, n=20, p=0.950$). Multiple stepwise regression with both variables showed that SF-5-HT was the strongest and only significant predictor for change in this pain ($r^2=0.36, n=20, p=0.003$).

Change after treatment in lateral or posterior tenderness to digital palpation or pain reflex to digital palpation of the TMJ was not significantly correlated to or predicted by pretreatment levels of neither SF-5-HT nor SF-TNF.
TABLE 2. Clinical variables: Inflammatory mediators in TMJ synovial fluid for patients prior to start of local treatment with glucocorticoids as well as after 12 weeks of treatment.

<table>
<thead>
<tr>
<th>Pretreatment</th>
<th>After treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Median</td>
</tr>
<tr>
<td>SF-5-HT</td>
<td>22,9</td>
</tr>
<tr>
<td>SF-TNF</td>
<td>0</td>
</tr>
<tr>
<td>MVM</td>
<td>36,3</td>
</tr>
<tr>
<td>Spontaneous pain</td>
<td>50</td>
</tr>
<tr>
<td>Pain on maximum mouth opening</td>
<td>3</td>
</tr>
<tr>
<td>Tenderness to digital palpation and pain reflex to digital palpation</td>
<td>3</td>
</tr>
</tbody>
</table>

n = number of observations
Discussion

This study shows that pretreatment level of 5-HT in the synovial fluid was the strongest and only significant predictor for changes in TMJ resting and movement pain after intra-articular treatment with glucocorticoid in comparison with TNF, i.e. a high pretreatment level of 5-HT predicts reduction of pain. According to correlation analysis, 5-HT and TNF in the synovial fluid were independent of each other. In addition, post treatment levels of 5-HT in the synovial fluid were positively correlated to changes in TMJ resting pain, while there was no such relation to clinical treatment response for synovial fluid levels of TNF.

Prediction

High pretreatment level of 5-HT in the synovial fluid predicted reduction of TMJ resting pain and pain upon mouth opening, which is in agreement with another study (Fredriksson et al 2005). Resting pain and pain upon mouth opening in TMJ-patients with detectable levels of SF-5-HT will thus be expected to decrease as an effect of glucocorticoid injection into the joint. Taiwo and Levine, 1992 suggested that pain and hyperalgesia could be two different entities, spontaneous pain being mediated by the 5-HT3 receptor. The most probable location for the 5-HT3 receptors is the peripheral afferent nerve terminals (Voog et al, 2000). Giordano and Rogers (1989) found that 5-HT3 receptor antagonists had a greater analgesic effect on chronic pain than on acute pain in a peripheral animal inflammation model, indicating the involvement of peripheral 5-HT3 sites in chronic inflammatory pain. 5-HT3 receptors have also been suggested to respond only to chemical stimulation (Giordano and Dyche, 1989) with only resting pain. The reason for pain relief might be blocking of 5-HT receptors, especially R5-HT3, in the synovial membrane since an increased level of 5-HT is found in the synovial fluid after treatment.

Pretreatment level of synovial fluid TNF was not found to be predictive for change of TMJ movement pain in this study which is in contrast to the study by Fredriksson et al (2006). The reason for this difference may be the different group of individuals that were observed in each study. The conclusion of the study of Fredriksson et al indicates that presence of SF-TNF in the synovial fluid predicts a treatment effect on TMJ movement pain, which is reduced in synergy with the level of TNF in the synovial fluid. The treatment effects of local glucocorticoid on TMJ and other joint pain are well known (Fredriksson et al,
2005; Fredriksson et al 2006 ), but Kopp et al (1991) showed that also the number of tender muscle regions were reduced and the maximum voluntary mouth opening was increased after glucocorticoid treatment. However, pretreatment synovial fluid levels of 5-HT or TNF did not predict changes after treatment of lateral or posterior tenderness to digital palpation of the TMJ nor pain reflex to digital palpation. This is in agreement with another study (Kopp et al 2005), which showed that treatment response of TMJ pain but not pressure pain threshold of the TMJ was associated with changes in anti-inflammatory cytokines and receptors in synovial fluid and plasma. The mechanisms behind the pressure pain threshold and tenderness to palpation are therefore probably different from those of TMJ pain at rest or on movement (Taiwo and Levine, 1992; Alstergren and Kopp, 1997). These results also indicated that TMJ pressure pain threshold and tenderness to palpation are modulated by systemic rather than local inflammatory mechanisms. Fredriksson et al (2005) have also reported different pain mechanisms behind TMJ resting pain and lateral tenderness. Results from another study (Fredriksson et al 2006) accordingly showed that there were no treatment effects observed on presence of palperbral pain reflex to digital TMJ palpation or TMJ pressure pain threshold after local glucocorticoid treatment.

Consequently, local glucocorticoid seems to have different effects on different pain entities, which in part may be due to different origins of these pain entities, where TMJ pain on maximum mouth opening is probably most related to intra-articular pain mechanisms (Alstergren and Kopp 1997).

Patient sample

The number of patients investigated was modest due to mandatory synovial fluid analysis of 5-HT and TNF before and after treatment with a reasonable time interval in between. Only patients with systemic inflammatory disorders were included, which means that the results are valid for a specific group of patients. However, this patient group is important and often subjected to intra-articular glucocorticoid treatment. The systemic inflammatory disease activity was low according to CRP in this patient sample and the influence of systemic inflammatory factors on the results can be considered low.
Methods

The clinical procedures are standard in this field and the synovial fluid sampling method including quantification of the mediators with vitamin $B_{12}$ as tracer is reliable and well investigated. The assays used to detect and quantify 5-HT and TNF in synovial fluid and blood serum/plasma are also well known.

General discussion about the value of predictors before glucocorticoid treatment

There is a general need to identify predictors of treatment effect of pain in order to be able to choose the most effective treatment for the individual patient. In this study it was shown that reduction of spontaneous/resting and movement pain in the TMJ can be expected after intra-articular administration of glucocorticoid in patients with high synovial fluid levels of 5-HT. Glucocorticoid injection have been used for a long time as anti-inflammatory drug for patients with inflammatory TMJ disorders and is a relative safe method with less risk in comparison with other drugs. Nevertheless, it should only be used when it can be expected to be effective, e.g. when SF-5-HT is high. According to this study 5-HT has a great importance in the inflammation process and for pain in the TMJ, but further studies of TNF are needed in this subject.

Conclusions

It can be concluded from this study that pretreatment synovial fluid level of 5-HT but not TNF predicts change (improvement) in TMJ resting and movement pain achieved by intra-articular administration of glucocorticoid in patients with inflammatory disorders of the TMJ.

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References


