Synovial fluid sampling from the temporomandibular joint: sample quality criteria and levels of interleukin-1β and serotonin

Per Alstergren, Sigvard Kopp and Elvar Theodorsson

Clinical Oral Physiology, Karolinska institutet, Huddinge, Sweden and Clinical Chemistry, University of Linköping, Linköping, Sweden

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The aims of this study were to compare two sets of quality criteria (SQC A and B) with respect to synovial fluid (SF) sampling and to present temporomandibular joint (TMJ) SF levels of IL-1β and 5-HT. The study comprised 310 TMJ SF samples from 12 healthy individuals (HI) and 59 patients with TMJ inflammatory disorders. Ten HI and 37 patients were selected for investigation of TMJ SF levels and samples were obtained by a push-and-pull method with quantification by vitamin B_{12}. The SQC comprised aspirate weight (AW), dilution factor (DF), blood contamination and hemolysis. IL-1β and 5-HT levels did not differ between the samples that satisfied SQC A or B. The proportion of samples that satisfied SQC A was higher than for SQC B. Patients with polyarthritides had significantly higher TMJ SF concentrations of 5-HT and IL-1β than HI. In conclusion, there is a recovery of TMJ SF of 0.1–0.2 g with the method used and the criteria set with the highest success rate do not differ from the other one with respect to SF levels of IL-1β and 5-HT. This set of sample quality criteria comprised no hemolysis, no or only minor blood contamination, AW >0.5 g and DF <0.98. The higher SF levels in the diseased TMJ (polyarthritides) compared to the healthy joint with respect to 5-HT and IL-1β is of clinical diagnostic relevance and the presence of 5-HT or IL-1β in TMJ SF seems to indicate a pathological joint condition probably of an inflammatory nature. □ *Arthritis; interleukin-1β; range; normal; serotonin; temporomandibular joint*

Per Alstergren, Clinical Oral Physiology, Karolinska institutet, Box 4064, SE-141 04 Huddinge, Sweden. Tel. +468 728 81 70, fax. +468 608 08 81, e-mail. per.alstergren@ofa.ki.se

The research on biochemical events associated with diseases in the musculoskeletal system has increased during the last decades. Knowledge about the underlying pathogenesis on a molecular level of these disorders is fundamental if the ability to diagnose and treat temporomandibular joint (TMJ) disorders is to improve (1). In that sense, synovial fluid (SF) analysis may help the development of more specific diagnostic and prognostic tools as well as more efficient treatment alternatives than those available today by making it possible to identify and quantify markers and mediators of disease. However, biochemical analysis of TMJ SF requires a precise technique for collecting samples of the generally small amount of SF present in this joint (2). So far, the biochemical changes of the TMJ SF have been investigated in only a few studies, probably because of the lack of such a technique.

In a large joint such as the knee joint, SF can readily be obtained by direct aspiration (3, 4). Such samples are preferable, since the true SF concentration of any substance can be calculated directly after analysis. However, in a small joint like the TMJ it is difficult to obtain SF samples consistently by direct aspiration, and, when succeeding, the sample volumes are usually small (2). Several other sampling methods have been used, most of them including washing of the joint with a washing solution, usually saline (1, 5, 6). Washing has been performed as single washing, as "pumping" or with a push-and-pull technique. Single washing means injection of the washing solution and subsequent aspiration; pumping means that the same washing solution is injected and aspirated several times; while push-and-pull means that the washing solution is consecutively injected and aspirated. The major obstacle with the joint washing method has been the unknown relation between SF and washing solution in the aspirate. This unknown dilution by SF has made it impossible to determine the true concentration of its contents, which have made these samples truly valid only for comparison of the relative concentrations of substances in each aspirate.

In two previous studies, hydroxocobalamin (HCA; vitamin B_{12}) has been investigated as external marker for measurement of the dilution of aspirates after saline washing of the TMJ (7, 8). This method has proved to be reproducible and accurate for determination of TMJ SF levels with a low detection limit and it requires a very small sample volume. Since this method was introduced only a few years ago, there are no reference values available for SF levels in the healthy or diseased TMJ regarding mediators or other substances previously studied. Examples of such mediators are neuropeptide Y (5, 9), serotonin (5-HT) (10), interleukin-1β (IL-1β) (6, 11, 12) and prostaglandin E_2 (13).

TMJ SF samples will differ in quality due to several factors, including blood contamination with or without hemolysis, small aspirate volumes, large dilution or small recovery of SF. It is therefore important to classify the sample quality in order to exclude inadequate samples.

The first aim of this study was to compare two sets of quality criteria (SQC) (A and B) with respect to SF levels of

Table 1. Age (years), duration (years) of general and temporomandibular joint (TMJ) disease, number of subjects (n) and distribution of male (M) and female (F) subjects

	Mean	SD	*n*	M/F
SQC investigation				
Healthy individuals	40	9	12	7/5
Patients	53	15	59	12/47
Synovial fluid levels				
Age				
Healthy individuals	39	11	10	8/2
Patients				
Polyarthritides	47	13	27	5/22
Local synovitis/capsulitis	65	9	10	0/10
Duration, general disease				
Patients				
Polyarthritides	16	13	27	
Local synovitis/capsulitis	20	14	10	
Duration, TMJ disease				
Patients				
Polyarthritides	9	6	27	
Local synovitis/capsulitis	9	7	10	

IL-1β and 5-HT and proportion of SF samples that satisfy these criteria. The second aim was to present TMJ SF levels for healthy individuals and for patients with TMJ inflammatory disorders concerning IL-1β and 5-HT.

Materials and methods

Subjects

All 310 TMJ SF samples obtained in our clinic with the push-and-pull method in combination with HCA as external marker were used in the investigation of the SQC. These samples were obtained from 12 healthy individuals and 59 patients (Table 1) with TMJ inflammatory disorders (synovitis/capsulitis or polyarthritides) according to the TMJ Diagnostic Classification by the American Academy of Orofacial Pain (14).

In order to establish SF levels of IL-1β and 5-HT, the 10 healthy individuals as well as the 37 patients for whom any of these substances had been analysed were selected from the total group of individuals. The patients belonged to a group with polyarthritides (27 patients) and a group of patients with TMJ synovitis/capsulitis (10 patients) according to the TMJ Diagnostic Classification (14), including pain localized to the TMJ region for a period of at least 6 weeks or tenderness to palpation of the joint laterally or posteriorly. The group of patients with polyarthritides comprised 11 patients with seropositive rheumatoid arthritis (RA), 4 with seronegative RA, 5 with psoriatic arthritis, 4 with ankylosing spondylitis, 2 with systemic lupus erythematosus and 1 patient with Sjögren's syndrome. Patients whose symptoms could be related to disease in other components of the temporomandibular system (e.g. toothache, myalgia and neuralgia) or patients who had been subjected to recent (< 3 month) treatment for TMJ symptoms were excluded. Local infection of the skin over the TMJ was considered a contraindication for arthrocentesis.

Synovial fluid sampling

The SF samplings were performed in accordance with a method described in a previous study (8) by two skilled operators (SK and PA) who had performed and trained together with the sampling procedure before this study was undertaken. Anesthesia of the TMJ was achieved by blocking of the auriculotemporal nerve with 2.0 mL Xylocain® (lidocaine 2%, Astra, Södertälje, Sweden). The TMJ was punctured with a standard disposable needle (diameter = 0.65 mm) inserted into the posterior part of the upper joint compartment. TMJ SF samples were obtained by washing the joint cavity with saline using a push-and-pull technique performed with two syringes, one used for the washing solution to be injected and the other for aspiration, connected to the arthrocentesis needle by a three-way stopcock. The injection solution, which consisted of 75% saline (NaCl 9 mg/mL, Kabi Pharmacia, Uppsala, Sweden) and 25% Behepan® (1 mg/mL, hydroxocobalamin, vitamin B_{12}, Kabi Pharmacia, Uppsala, Sweden), was injected slowly into the joint cavity, 1 mL at a time, and then aspirated. The correct intra-articular position of the arthrocentesis needle could be confirmed by easy flow of the washing solution when injected and aspirated. The total washing solution volume used was 3.5–4.0 mL. The HCA was included in order to measure the amount of washing solution in the aspirate, i.e. for an indirect measurement of the SF content in the aspirate. The samples were compared in a spectrophotometer (Shimadzo UV-160A, Shimadzo Corp., Tokyo, Japan) with a capillary tube system consisting of a capillary tube of quartz (0.7 mm in diameter, 3-µL/sample) and a capillary tube holder (Shimadzo Corp., Tokyo, Japan). The detection limit regarding dilution of the washing solution in the aspirate by this method is 0.9%. The SF concentration of any particular substance (SF) can then be calculated using the formula:

$$C_S = \frac{C_A}{\left(1 - \frac{Abs_{Asp}}{Abs_{Wash}}\right)}$$

where C_S = SF concentration, C_A = aspirate concentration, Abs_{Asp} = aspirate absorbance and Abs_{Wash} = washing solution absorbance.

During and immediately after the arthrocentesis, blood cell contamination of the aspirate was estimated visually in accordance with the following scale: 0 = no visible blood contamination, 1 = hardly visible blood contamination, 2 = clearly visible blood contamination, and 3 = blood-like appearance of the aspirate. Blood cell contamination, especially by erythrocytes, can be observed in this solution, almost as easily as in pure saline. After aspiration, the weight of the sample (AW) was immediately measured

Table 2. Definition of the two sets (A and B) of sample quality criteria (SQC), which were based on sample hemolysis, degree of blood contamination, aspirate weight (g) and dilution factor

	SQC Set	
Parameter	A	B
Hemolysis	0	0
Blood contamination	0–1	0–1
Aspirate weight	>0.5	>1.0
Dilution factor	<0.98	<0.95

Hemolysis 0 = absent; blood contamination 0 = no, 1 = minor.

using a balance (JW-120, Adam Equipment Co., Milton Keynes, U.K.) the sample was then centrifuged (1500 *g* for 10 min at 4°C) and hemolysis was recorded as absent or present by visual inspection. Twelve microliters of the supernatant, i.e. four capillary tubes, was used for the absorbance measurement. The aspirate and washing solution absorbances were compared and a dilution factor ($DF = Absorbance_{Aspirate}/Absorbance_{Washing\ solution}$) was calculated for each sample. The remaining part of the supernatant was transferred to other tubes, specific for each substance to be analyzed, and stored in a freezer (−85°C) until analysis.

Sample quality criteria

Two sets of SQC based on sample hemolysis, blood contamination, aspirate weight (AW) and DF were compared (Table 2). The recovery of SF (expressed in g) of each sample was calculated using the formula $SF = AW(1 - DF)$.

Analysis of substances

The SF-5-HT was analyzed using a commercially available EIA-kit (No. 0642, Immunotech International, Germany) with a detection limit of 0.5 nmol/L. According to the manufacturer, the intra-assay coefficient of variation is less than 9.4%; the inter-assay coefficient of variation is less than 9.9%.

SF-IL-1β was determined with an ELISA-kit (Cayman Chemical Company, Ann Arbor, MI, USA) with a detection limit of 1.5 pg/mL. Performance characteristics for the analysis are: intra- and interassay coefficient of variation <10%, specificity of IL-1β 100%, of interleukin-1α <0.01%, of interleukin-2 <0.01% and sensitivity <1.0 pg/mL.

Statistics

The Komogorov-Smirnov test was used to test the variables for normal distribution. None of the variables AW, DF, SF recovery, SF-Il-1β or SF-5-HT was normally distributed and non-parametric statistical analysis was therefore applied. For descriptive statistics of the variables, median, intraquartile range (IQR: 75th percentile to 25th percentile) and number of observations were used. The significance of the differences in SF levels between the samples that satisfied SQC B and the samples that satisfied SQC A but not SQC B was tested with the Mann-Whitney U-test. The significance of the differences between the three groups of individuals regarding SF levels was tested according to the Kruskal-Wallis test with Dunn's post-hoc test for multiple comparison where applicable. The significance of the difference in TMJ SF concentrations between the genders was also tested with the Mann-Whitney U-test. The significance of the correlation between age and TMJ SF concentrations was tested with the Spearman ranked correlation test. A significance level of less than 0.05 was considered as significant.

Results

Sample quality criteria

The distributions of AW and DF of the total 310 investigated samples are shown in Figs 1 and 2. The median AW for all samples was 1.20 g (IQR = 1.53) and the median DF was 0.940 (IQR = 0.103). For the 32 samples from the healthy individuals the median AW was 0.97 g (IQR = 0.95), for the 204 samples from the patients with polyarthritides it was 1.24 g (IQR = 1.66) and for the 74 samples from patients with synovitis/capsulitis it was 1.49 g (IQR = 1.76). The 32 samples from the healthy individuals had a median DF of 0.897 (IQR = 0.223), for the 204 samples from the patients with polyarthritides the median DF was 0.940 (IQR = 0.095) and for the 74 samples from patients with synovitis/capsulitis it was 0.944 (IQR = 0.111). The differences in AW and DF between the two patient groups and the healthy individuals were not significant. Five samples showed a DF of 0.33–0.50, all of which had a blood contamination of grade 3. In 41

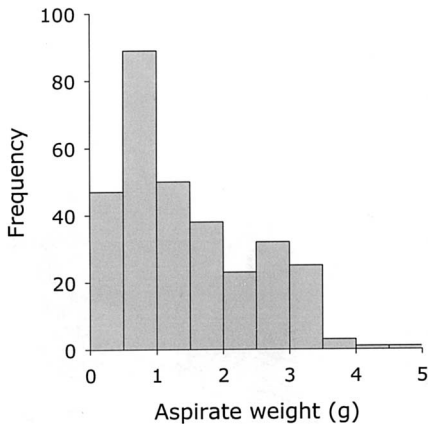

Fig. 1. Distribution of aspirate weights (g) in 310 temporomandibular joint synovial fluid samples from 71 individuals obtained by the push-and-pull technique with hydroxocobalamin as external marker.

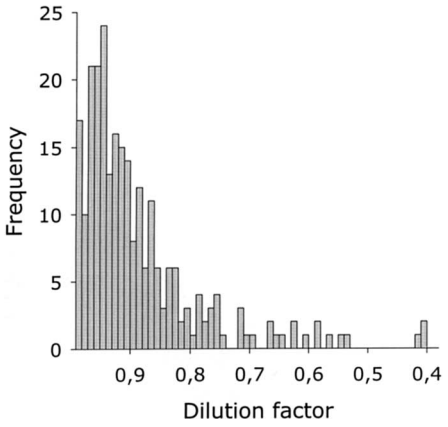

Fig. 2. Distribution of dilution factors in 310 healthy and diseased temporomandibular joint synovial fluid samples from 71 individuals obtained by the push-and-pull technique with hydroxocobalamin as external marker.

samples (13%), the DF was 1.000, i.e. no dilution of the washing solution was detected. In three samples the AW was above 4.0 g and these samples were not contaminated with blood. Two-hundred-and-fifty-four samples (82%) had a blood contamination grade of 0 or 1, and 81 samples (13%) had a blood contamination grade of 2. Four samples (1.3%) showed signs of hemolysis.

The SF recovery and the proportion of the samples that satisfied the two SQC sets are shown in Fig. 3. The differences between the SF levels in the samples that satisfied SQC B and the samples that satisfied SQC A but

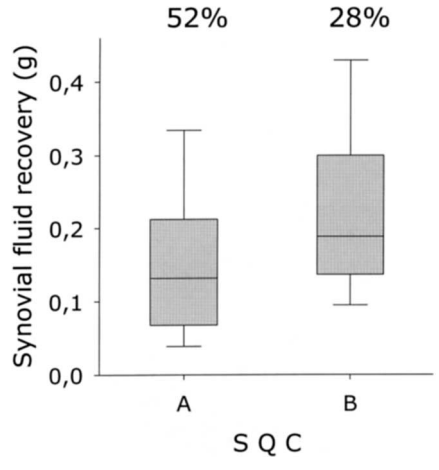

Fig. 3. Distribution of synovial fluid recovery in 310 temporomandibular joint synovial fluid samples obtained by the push-and-pull technique with hydroxocobalamin as external marker that satisfied the respective set of sample quality criteria (SQC). The figures above the graph denote the proportion of the samples that satisfied the respective criteria.

Table 3. Proportion (%) of samples that did not satisfy the respective set (A and B) of sample quality criteria (SQC) according to causes

	SQC Set	
Cause	A	B
Blood contamination	33	14
Dilution factor	33	24
Aspirate weight	13	21
Dilution factor + aspirate weight	15	29

not SQC B were not significant for 5-HT ($P = 0.768$) or IL-1β ($P = 0.611$).

Table 3 gives the main reasons why samples did not satisfy the respective criteria sets. The main reasons in SQC A were blood contamination (33%) and high DF (33%) and for SQC B a combination of low AW and a high DF (29%).

Synovial fluid levels

Table 4 gives the TMJ SF levels of IL-1β and 5-HT according to the two SQC sets. The SQC sets were similar concerning the SF levels, both for the healthy individuals and for the patient groups.

Figs 4 and 5 show the TMJ SF levels of IL-1β and 5-HT in the healthy individuals and the patients with polyarthritides as well as synovitis/capsulitis according to SQC A. According to both SQC A and B, IL-1β and 5-HT were undetectable in the samples from the healthy individuals. The patients with polyarthritides had significantly higher TMJ SF concentrations of 5-HT ($P < 0.001$) than the healthy individuals according to SQC A. The patients with polyarthritides also had significantly higher TMJ SF concentrations of IL-1β ($P = 0.015$) than the healthy individuals as well as compared to the patients with synovitis/capsulitis according to SQC A.

There were no significant correlations between age and AW, DF, SF recovery, SF-IL-1β or SF-5-HT in the patients with polyarthritides. Neither was there any significant difference in AW, DF, SF recovery, SF-IL-1β or SF-5-HT between the genders in this group. The other two groups were not analyzed in these respects because of a low number of observations.

Discussion

In this study, quality criteria sets for TMJ SF sampling used in combination with the push-and-pull technique and vitamin B_{12} as a marker were investigated for clinical use. One set of criteria that resulted in a success rate of 52% was considered as the most useful. The study further indicates that 5-HT and IL-1β are undetectable in TMJ SF from healthy individuals. The higher SF levels in the diseased TMJ (polyarthritides) compared to the healthy

Table 4. Temporomandibular joint synovial fluid levels of interleukin-1β (IL-1β) and serotonin (5-HT) in healthy individuals and patients with TMJ disorders according to the two sample quality criteria (SQC) sets (A and B)

	SQC A			SQC B		
	Median	IQR	*n*	Median	IQR	*n*
IL-1B (pg/mL)						
Healthy individuals	0	0	10	0	0	4
Patients						
Polyarthritides	0	27	35	0	28	18
Local synovitis/capsulitis	0	0	10	0	0	7
5-HT (nmol/L)						
Healthy individuals	0	0	6	0	0	3
Patients						
Polyarthritides	35	29	15	32	24	10
Local synovitis/capsulitis	15	11	4	15	11	4

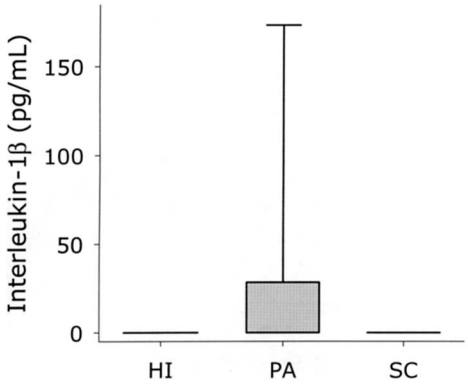

Fig. 4. Box-plot showing the temporomandibular joint (TMJ) synovial fluid concentration of interleukin-1β (10th, 25th, 50th, 75th and 90th percentile) in 10 healthy TMJs (HI), 35 TMJs with polyarthritides (PA) and 10 TMJs with synovitis/capsulitis (SC) according to SQC A. There was a significant difference between the healthy individuals and the patients with polyarthritides ($p < 0.001$) as well as between the two patient groups ($p < 0.001$).

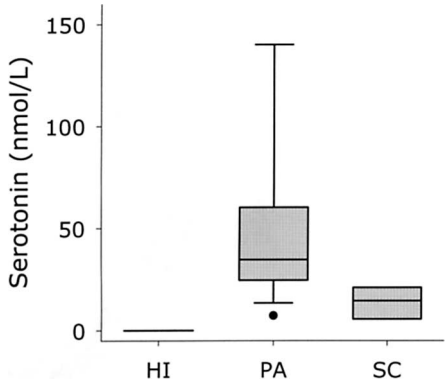

Fig. 5. Box-plot showing the temporomandibular joint (TMJ) synovial fluid concentration of serotonin (10th, 25th, 50th, 75th and 90th percentile) in 6 healthy TMJs (HI), 15 TMJs with polyarthritides (PA) and 4 TMJs with synovitis/capsulitis (SC) according to SQC A. There was a significant difference between the healthy individuals and the patients with polyarthritides ($p < 0.001$).

joint with respect to 5-HT and IL-1β is of clinical diagnostic relevance and the presence of 5-HT or IL-1β in TMJ SF seems to indicate a pathological joint condition of an inflammatory nature.

Sample quality criteria sets

The criteria sets selected were chosen by logical and practical reasoning. Hemolysis would influence measurement of all mediators contained in erythrocytes and it would probably also interfere with the photometric measurement of HCA. The presence of blood in the aspirate interferes with the estimation of the SF proportion in the aspirate. Blood contamination might also interfere with analysis depending on which substance is to be analyzed. For example, quantification of 5-HT is more sensitive to blood contamination than IL-1β since the level of 5-HT in serum is probably several hundred times higher than in the SF (10). We therefore only accepted no or hardly visible blood contamination in this study, despite the fact that blood contamination is difficult to avoid completely upon arthrocentesis of inflamed joints. An AW of less than 0.5 g might mean that only the part of the injection solution that remains in the dead-space of the needle and three-way stop-cock during the washing procedure is recovered. Since the dead weight of the washing solution in the push-and-pull setup is approximately 0.4 g, the AW should exceed this figure. The vitamin B_{12} method has a detection limit of 0.9% dilution when used with a washing solution consisting of 25% Behepan and measured with the capillary tube system in a spectrophotometer at a wavelength of 350 nm. The content of plasma or sodium-hyaluronate in the SF does not influence the detection limit of the absorbance measurement (8). In theory, this means that a DF equal to or less than 0.99 could be used. However, in the SQC sets a DF of less than 0.98 or less than 0.95 was investigated since a very high DF is likely to increase the measurement error. In addition, blood contamination or low AW would also increase the measurement error and cause the aspirate content to be less representative of the SF content.

Both AW and DF showed a large variation. One explanation for these large variations could be technical difficulties during the arthrocentesis, in turn due to adhesions, pannus tissue or fibrosis. The main variation in DF, however, is probably due to the variation in joint effusion volumes, e.g. a large effusion volume causes a low DF.

SQC A showed the highest success rate of sampling, but SQC B resulted in the highest recovery of SF. However, the SF levels of IL-1β and 5-HT in the samples that satisfied SQC B were not significantly different from the samples that satisfied SQC A but not SQC B. This means that SQC A can be considered to be the most useful SQC set due to its higher success rate. With SQC A, more than 50% of all samples obtained by the push-and-pull technique could be used for analysis. There was a large variation of SF recovery, but in 90% of the samples that satisfied SQC A, the recovery was approximately 18–420 µL (15) with a median of 119 µL. This is more than the mean of 50.4 µL (SD: 56 µL, $n = 9$) reported by Aghabeigi et al. (2) to be present in the symptomatic TMJ. The quantification of SF volume was made with orally administered aspirin as a marker, whereby the concentrations of salicylate in plasma and in saline aspirates of the TMJ were compared and a DF was calculated. Two problems that may be encountered by this method are hypersensibility towards salicylate and an uneven distribution of salicylate between the plasma and SF compartments. Our results are similar to what Shibata et al. (16) on average obtained from patients with TMJ disk derangement or osteoarthritis with their direct aspiration technique. Shibata et al. (16) obtained TMJ SF samples in 70.4% of the aspirations on 101 TMJs and the mean AW was 0.146 g (SD: 0.387 g). An explanation for the difference in success rate between the latter study and ours might be the different patient categories included as well as different criteria for successful sampling.

In SQC A, the main reasons for not satisfying the criteria were blood contamination and high DF, which together caused 66% of the exclusions, whereas for SQC B the reason was a combination of low AW and high DF. An explanation for this difference is that SQC B excluded more samples than SQC A based on AW and DF. These reasons are important to consider if the method is to be further improved. There are at least three ways to avoid high DF during sampling: to use smaller washing solution volumes, to wait longer before aspiration or to use repeated injections and aspirations with the same solution, i.e. pumping (6). The use of a smaller washing-solution volume or to wait longer before aspiration might, however, make the washing less efficient, i.e. cause a lower absolute recovery of molecules, or cause loss of washing solution by diffusion into the surrounding tissue. The use of repeated washings with the same solution might decrease the DF, although there is a risk of increased loss of washing solution and iatrogenic stimulation of mediator release. Repeated washings should therefore be tested in a controlled way in the future. To avoid or minimize the risk of blood contamination, the arthrocentesis should be as non-traumatic as possible, a factor which largely depends on the operator's skill and experience. If bleeding occurs, the procedure should be terminated and postponed for a period of time (at least 10–15 min) or, even better, be retried on a later occasion.

Synovial fluid levels

According to both SQC A and B, IL-1β and 5-HT seem to be undetectable in the SF of the TMJ in healthy individuals. In addition, patients with polyarthritides have higher SF levels of 5-HT than healthy individuals. These patients also seem to have higher SF levels of interleukin-1β than healthy individuals as well as patients with local synovitis/capsulitis. The fact that none of these mediators was detected in healthy SF is of clinical interest. The presence of either of them thus indicates a pathological condition in the TMJ probably of an inflammatory nature.

Methodological considerations

The saline washing was performed in the upper TMJ compartment in this study. Access to the lower joint compartment by an arthrocentesis needle is more difficult to achieve than access to the upper compartment due to the anatomical situation, i.e. that the disk is tightly attached to the condyle. An attempt to access the lower compartment therefore increases the risk of trauma to the tissue as well as bleeding. The size of the upper compartment is also larger, which is an advantage.

In this study, the correct arthrocentesis needle placement, i.e. in the superior joint compartment, was confirmed by an easy flow of washing solution upon injection and aspiration. Another way of confirming an intra-articular needle placement is to inject washing solution until a resistance is felt, usually after 1–2 mL for the TMJ. However, the reason for not using this technique was to avoid the trauma of distention of the inflamed synovial membrane, which could iatrogenically stimulate the release of inflammatory mediators into the SF or cause bleeding. The presence of fibrous adhesions and pannus tissue in arthritic joints, especially in patients with polyarthritides, is not uncommon. Such changes may increase the risk of trauma and intra-articular bleeding even when resistance is felt after injection of only a small volume.

A total of 3.5–4 mL washing solution was injected. Due to the difficulties with saline washing in some patients with arthritis (fibrous adhesions and pannus tissue), the injected washing solution could only be partially recovered. The non-recovered portion probably diffuses into pannus tissue cavities or into the surrounding inflamed capsular tissue with increased permeability.

It is not possible to make a total aspiration of SF, neither with the direct method nor with saline washing. The vitamin B_{12} method, as used in this study, cannot thus be used for determination of the total SF volume in the TMJ and that was not the intention of this study. The samples analyzed contain recovered volumes of SF that have been

obtained in a similar and standardized way. The problems with the mixing of the washing solution and the SF in the joint and the large dilution of synovia in the aspirates are two factors that can perhaps be solved. We have presented some suggestions for improvements of the method, which we are going to test in the near future.

The vitamin B_{12} method is capable of detecting a dilution of less than 1% when it was tested in vitro; i.e. a DF of 0.99 is valid with respect to the DF. Neither plasma nor hyaluronan influenced the determination of dilution, at least up to 16% dilution (7, 8). However, the relatively large dilution in some samples may increase the measurement error of the analysis of a target substance. In order to reduce this risk, the DF limits 0.98 and 0.95, respectively, was used in the SQC sets. In our opinion, this means that the SF levels presented can be used as a quantitative measurement of the target substances.

Conclusion

This study shows that for both the tested sets of criteria there is a recovery of TMJ SF of 0.1–0.2 g (approximately 100–200 μL). The most useful set of criteria with the highest success rate does not differ from the other one with respect to SF levels of IL-1β and 5-HT. This set of sample quality criteria comprised no hemolysis, no or only minor blood contamination, AW >0.5 g and DF <0.98. The study further indicates that IL-1β and 5-HT are undetectable in TMJ SF from healthy individuals. The higher SF levels in the diseased TMJ (polyarthritides) compared to the healthy joint with respect to IL-1β and 5-HT is of clinical diagnostic relevance and presence of IL-1β or 5-HT in TMJ SF seems to indicate a pathological joint condition probably of an inflammatory nature.

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