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Biological activity and migration of wear particles in the knee joint: an in vivo comparison of six different polyethylene materials

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Abstract Wear of polyethylene causes loosening of joint prostheses because of the particle mediated activity of the host tissue. It was hypothesized that conventional and crosslinked polyethylene particles lead to similar biological effects around the knee joint in vivo as well as to a similar particle distribution in the surrounding tissues. To verify these hypotheses, particle suspensions of six different polyethylene materials were injected into knee joints of Balb/C mice and intravital microscopic, histological and immunohistochemical evaluations were done after 1 week. Whereas the biological effects on the synovial layer and the subchondral bone of femur and tibia were similar for all the polyethylenes, two crosslinked materials showed an elevated cytokine expression in the articular cartilage. Furthermore, the distribution of particles around the joint was dependent on the injected polyethylene material. Those crosslinked particles, which remained mainly in the joint space, showed an increased expression of TNF-alpha in articular cartilage. The data of this study support the use of crosslinked polyethylene in total knee arthroplasty. In contrast, the presence of certain crosslinked wear particles in the joint space can lead to an elevated inflammatory reaction in the remaining cartilage, which challenges the potential use of those crosslinked polyethylenes for unicondylar knee prostheses.

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1 Introduction

It is known, that the generated wear debris of articulating biomaterials is a major factor in the longevity of total joint arthroplasty, because the released particles can elicit complex biological responses resulting in periprosthetic osteolysis and subsequent aseptic loosening [1–3]. Numerous factors can influence the cellular response to wear particles, including composition, dose, volume, size and shape of the particles [2, 4, 5]. These challenges have prompted investigators to introduce novel bearing materials, such as crosslinked polyethylene (XPE) in an effort to improve wear performance and revision rates.

While there is a wealth of information about the interaction of particles with bone and the consecutive periprosthetic osteolysis [1–3], relatively little is known about the effects of particles on the surrounding soft tissues like synovial layer and cartilage [6, 7]. Therefore, it is useful to investigate potential interactions of new bearing materials with these soft tissues, respectively. To date, XPE is only used in total knee arthroplasty (TKA). But in effort to bring XPE into play especially in unicondylar knee replacement (UKA), in which only one compartment of the joint is replaced, the biological effect of these polyethylenes on remaining cartilage should be investigated.

The hypothesis of this study was that conventional ultra-high-molecular-weight-polyethylene (UHMWPE) and crosslinked polyethylene, similar in size and morphology, lead to similar biological effects in the knee joint: The

synovial layer, the subchondral bone of tibia and femur as well as the articular cartilage had been taken into account. The second hypothesis was that the UHMWPE as well as the XPE particles show a similar migration behaviour in- and outside of the joint, respectively in the tissues around the joint.

2 Methods

2.1 Particle generation and characterization

Four crosslinked polyethylene (XPE) and two conventional polyethylene (UHMWPE) inserts, which differed in virgin powder and/or processing history (Table 1), were utilized for this study. The inserts were tested over 5×10^6 cycles according to the ISO standard using a knee-joint-simulator (Stallforth–Ungethum) [8]. All polyethylene materials articulated against matching cobalt-chromium femoral condyles (Table 1). Whereas the material combinations A–C, E and F are actual product devices, D is an experimental product and not commercially available (Table 1). The setup of this experiment has been described in detail earlier [9]. Before testing, the inserts were left at 37 °C for 30 days to achieve thermal equilibrium and dimensional stability [8]. The applied lubricant which was changed every 6 days was 25 % (v/v) newborn calf serum with 0.1 % (m/v) sodium azide solution in sterile water [10]. The inserts were weighed every 0.5 million cycles. The lubricant was changed every 0.5 million cycles as well.

After 5.0 million cycles the samples were poured together, then particles were separated by the acid digestion method (20 nm-nucleopore-filter [11]), analyzed by scanning electron microscopy (SEM; Leica Stereoscan 420, Leica Microsystems, Wetzlar, Germany) and image analyzer (Leica QWin). The particle analysis of the particles used in the current study has been already characterized before in detail [12]. For all tested groups more than 85 % of the particles were submicron [12]. The mean equivalent circle diameter (ECD) of the particles was 0.39 ± 0.37 and 0.42 ± 0.44 µm for the two UHMWPEs and for the XPEs the mean ECD ranged from 0.33 ± 0.23 µm up to 0.46 ± 0.46 µm [12]. The particles were nearly round in shape and showed a similar Aspect ratio in both groups, XPEs as well as UHMWPEs [12].

Table 1 Types of prostheses and tested tibial inserts

Prosthesis design	Manufacturing process of polyethylene
Scorpio® (Stryker®)	
A	*Crosslinked Polyethylene (XPE)* Fixed-bearing, CR, 24 mm, GUR 1020, 3 × 30 kGy Gamma irradiation and annealing *and sequential irradiated*, Gasplasma sterilised (X3™/Stryker®)
Natural Knee II® (Zimmer®)	
B	*Crosslinked Polyethylene (XPE)* Fixed-bearing, ultra-congruent, 19 mm, GUR 1050, 95 kGy E-beam, remelted, EtO sterilised (Durasul™/Zimmer®)
NexGen® (Zimmer®)	
C	*Crosslinked Polyethylene (XPE)* Fixed-bearing, CR, 14 mm, GUR 1050, 65 kGy E-beam, remelted, Gasplasma sterilised (Prolong™/Zimmer®)
LCS® complete (DePuy®)	
D	*Crosslinked Polyethylene (XPE)* Mobile-bearing, 15 mm, GUR 1020, 50 kGy E-beam, remelted, Gamma sterilised (XPE*/DePuy®:experimental combination: not a commercially available product)
LCS® complete (DePuy®)	
E	*Conventional polyethylene (UHMWPE)* Mobile-bearing, 15 mm, GUR 1020, Gamma sterilised (DePuy®)
Natural Knee II® (Zimmer®)	
F	*Conventional polyethylene (UHMWPE)* Fixed-bearing, congruent, 13 mm, GUR 1050, Gamma sterilised (Zimmer®)

2.2 Elimination of endotoxin and preparation of particles

To avoid any side effects by lipopolysaccharides (LPS), the particles were cleaned from endotoxin by using a recently published method based on ultracentrifugation [13]. For detection of the endotoxin a standard chromogenic LAL assay was used (Limulus Amoebocyte Lysate Test, Lonza®). The particles of each type of polyethylene (group A–F, Table 1) were adjusted in sterile phosphate-buffered saline (PBS) solution, creating a 0.1 vol% concentration [14, 15]. Particle induced cell activation can be influenced by the volume of particles [16–18], thus, only equivalent particle volumes were used to compare the different materials.

2.3 Animals

Seventy female Balb/c mice, weighing 18–25 g, were received (Charles River Wiga, Sulzbach, Germany). Seven groups were matched. All mice were assigned to their group randomly: control ($n = 10$), A–F (each with $n = 10$). The letters A–F represent the type of polyethylene as shown in Table 1. Fourteen mice had to be excluded because of complications, such as bleeding or damage to the synovial tissue during surgical preparation, so that each group finally consisted of eight mice.

The mice were killed with 10 mg pentobarbital i.v.-injections after all procedures were completed. During the experiments all national legal forms of animal protection were hold.

2.4 Particle injection, surgical preparation and intravital microscopy

In order to avoid agglomeration of the particles, the suspensions were sonicated for 1 h. Afterwards, 50 µl of each particle suspension (A–F) were injected into the murine left knee by a microcannula (FST Heidelberg, Germany) under sterile conditions. The suspension for the control group solely contained PBS.

Each knee was evaluated 1 week after injection [19–21]. The leukocyte–endothelial cell interactions and the synovial microcirculation were evaluated by intravital fluorescence microscopy to identify the level of inflammatory reaction by injecting intravenously the fluorescent marker rhodamine 6G (Sigma, Deisenhofen, Germany) in a single bolus of 0.15 mg/kg immediately before the measurement [19–21]. The inflammatory reaction was defined by the rolling fraction, the adherent cells and the functional capillary density (FCD; [19–21]). FCD was defined as the length of the rbc perfused capillaries in the observation area (expressed as cm/cm^2), using fluorescent plasma marker FITC–dextran (Sigma, Deisenhofen, Germany, 15 mg/kg body weight) [19–21].

A computer-assisted microcirculation analysis system (Cap-Image) was used for data analysis [22]. Velocity of the rbc in centerline in post-capillary venules, using the unit mm/s, was also measured [22]. Vessel diameter and rbc velocity were evaluated from the rhodamine images, the FCD from the FITC images.

2.5 Histology and Immunohistochemistry

After euthanization, the knee joints were removed, fixed with 4 % paraformaldehyde (24 h, pH 7.2) and incubated (with 4 % EDTA, pH 7.1) for 1 week at room temperature for decalcification. Washing with PBS dehydration with an automatic dehydrator model and coating with paraffin followed. The paraffin embedded joints were cut into 6 µm thin slices and stained with hematoxylin and eosin (HE). The thickness of the synovial membrane in six selected areas of the knee was evaluated (Cap-Image; [23]). Besides a histological score, established by Brackertz et al. [24], was used to classify the synovial tissue [19–21].

For the immunohistochemical staining two slices per knee were stained with the primary antibodys IL-1ß and TNF-α (R&D Systems, Minneapolis, USA). A negative control was generated for each knee as well, using PBS instead of a primary antibody. Semiquantitativley evaluation of each sample with a light microscope (Carl Zeiss Micro Imaging GmbH, Germany) followed. The classification included five groups: Group 1: 0 % positive cells, group 2: <25 % positive cells, group 3: >25–50 % positive cells, group 4: >50–75 % positive cells and group 5 with more than 75 % positive cells in synovial layer, subchondral bone from femur and tibia as well as in the articular cartilage [25].

2.6 Distribution of particles

The localization of the particles was performed using the above described light microscope with polarization filter [26, 27]. Four HE-sections per mouse were evaluated and the amount of the particles in the joint space, the synovial layer and the subchondral bone was determined. The amount of detected particles in each case was categorized into none (no particles present), few, moderate and many. For graphical representation, these values were then converted to the following point system: no particles = 0 points, few particles = 0.5 points, moderate particles = 1 point, many particles = 2 points.

2.7 Statistics

As part of the governmental animal experiment proposal a sample-size calculation was performed analogous to previous studies using this animal model [19, 25]. All measured data showed a non-parametric distribution. For the statistical analysis we applied the Kruskal–Wallis analysis for the differences within the tested groups, followed by a pairwise multiple comparison procedure, for the immunhistochemical data with a Bonferroni correction. The level of significance was set at $P < 0.05$. Within all tests the level of significance of the Bonferroni correction (immunhistochemical data) was at $P = 0.0024$.

3 Results

3.1 Microcirculatory and intravital microscopy measurements

The FCD showed a difference between the control group and the test groups ($P < 0.05$), a difference between the particular test groups could not be determined ($P > 0.05$) as well as any differences in the vessel diameter ($P > 0.05$). Furthermore, there were no differences at all concerning the rbc centerline velocity in all groups (Table 2).

In addition, the fraction of rolling leukocytes as well as the number of adherent cells was increased in all groups compared with the control group ($P < 0.05$; Fig. 1a) without any statistically significant difference between the test groups ($P > 0.05$).

3.2 Histology

The synovial tissues showed increased inflammatory activity in all groups and a similar thickening of the synovial layer

Table 2 Microcirculatory parameters of the intravital microscopic measurements 7 days after intraarticular particle injection in the murine knee joint

	Control group $n = 8$	A $n = 8$	B $n = 8$	C $n = 8$	D $n = 8$	E $n = 8$	F $n = 8$
Vessel diameter (μm)	19 ± 1.2	22 ± 1.2	20 ± 0.7	24 ± 1.0	21 ± 1.0	22 ± 0.7	24 ± 0.7
rbc velocity (mm/s)	0.28 ± 0.02	0.33 ± 0.03	0.34 ± 0.02	0.30 ± 0.03	0.31 ± 0.03	0.31 ± 0.03	0.35 ± 0.02
FCD (cm/cm^2)	368 ± 10	412 ± 10*	423 ± 8*	409 ± 6*	414 ± 7*	420 ± 9*	394 ± 11*

Data are given as mean ± SEM

Rbc red platelet count, *FCD* functional capillar density

* $P < 0.05$

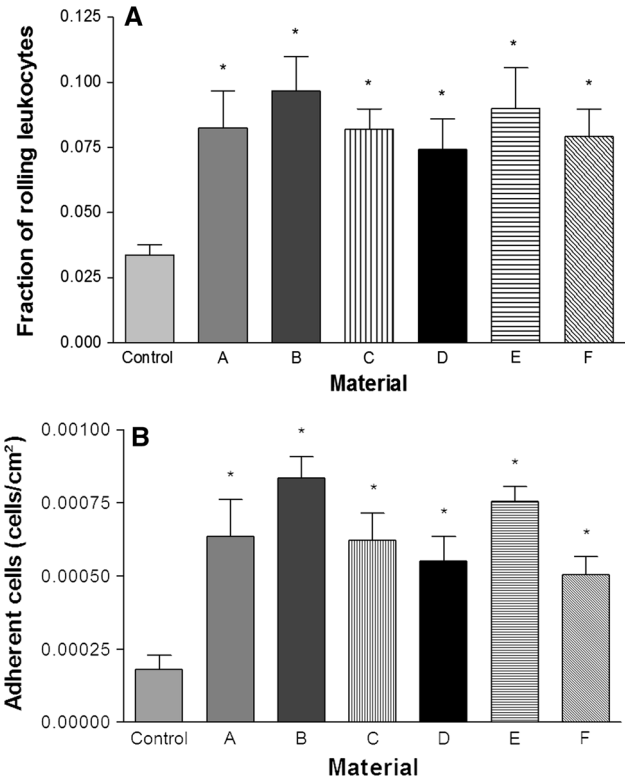

Fig. 1 Results of the leukocyte-endothelial cell interaction by intravital microscopy after intraarticular injection of different polyethylene wear particles suspensions. **a** Fraction of rolling leukocytes. **b** Adherent cells The values are illustrated by mean ± SEM. *$P < 0.05$. There was no difference between the testing groups *A–F* (*A–D* crosslinked polyethylene; *E* and *F* UHMWPE; $P > 0.05$)

compared with the control ($P < 0.05$) without any difference between the different polyethylenes ($P > 0.05$; Table 3). All particle-stimulated groups showed an enhanced cellular infiltration in the synovial layer (Fig. 2).

3.3 Immunohistochemistry

In the synovial tissue of the murine knee joint there was an increased expression of IL-1β in all test groups in relation to the control ($P < 0.001$; Fig. 3a). Furthermore, the cells

Table 3 Histopathological Score of the synovial tissue according to Brackertz et al. [24] and measurement of the synovial membrane

	Histopathological score (median)	Thickness of synovial membrane (μm; mean ± SEM)
Control	0	46 ± 6
A ($n = 8$)	1.3*	100 ± 5*
B ($n = 8$)	1.5*	120 ± 6*
C ($n = 8$)	1.8*	118 ± 4*
D ($n = 8$)	1.8*	121 ± 6*
E ($n = 8$)	1.5*	115 ± 6*
F ($n = 8$)	1.5*	91 ± 3*

There was no significant difference within the test groups, but all groups differed significantly from the control group (* $P < 0.05$)

of the murine synovial layer of all groups except of C and F expressed significantly more TNF-α than these of the control mice ($P < 0.0001$; Fig. 3b). In addition to this, a disparity between the different polyethylenes concerning the release of TNF-α in the synovial layer was not detected ($P > 0.005$; Figs. 3b, 4).

In the subchondral bone all polyethylene groups secreted more IL-1β than the control group ($P < 0.002$; Fig. 5a). The TNF-α release was also significantly increased in all test groups in comparison to the control group except group F ($P < 0.001$; Fig 5b). Figure 6 illustrates visually as an example the elevation of the secretion of IL-1β in the murine bone.

In the articular cartilage IL-1β was not increased in the experimental groups A–F ($P > 0.0024$), whereas TNF-α was significantly increased in the experimental groups A ($P \leq 0.001$) and B ($P \leq 0.001$; Figs. 7, 8).

3.4 Distribution of particles around the joint

The XPE particles of the groups A and B were predominantly localized in the joint space. For the other XPEs (group C and D), however, absolutely no particles were seen in the joint space, but mainly in the subchondral bone and also in the synovial tissue. In the UHMWPE groups

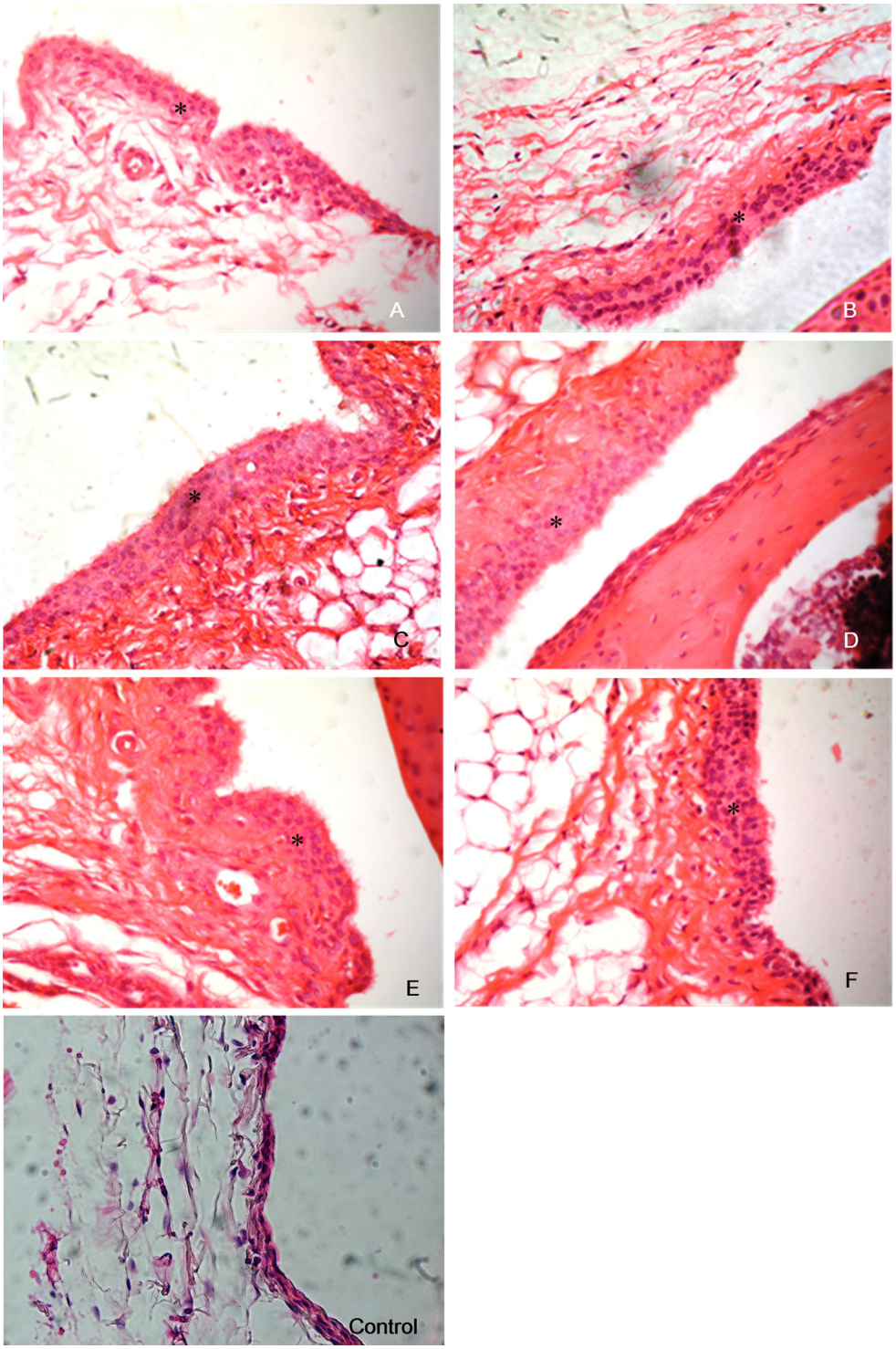

Fig. 2 Histological appearance of the synovial tissue of murine knee joints 1 week after injection of different polyethylene wear particles. A marked thickened synovial cell layer with cellular infiltration (marked by *asterisk*) can be seen in all the testing groups (**a**–**f**), compared to the control group. Magnification ×400

particles of group E were almost evenly distributed in the joint space and partially in the subchondral bone, while almost no particles were seen in the synovial layer. In group F, the particles were predominantly seen in the subchondral bone, partly in the synovial layer and no particles in the joint space (Fig. 9).

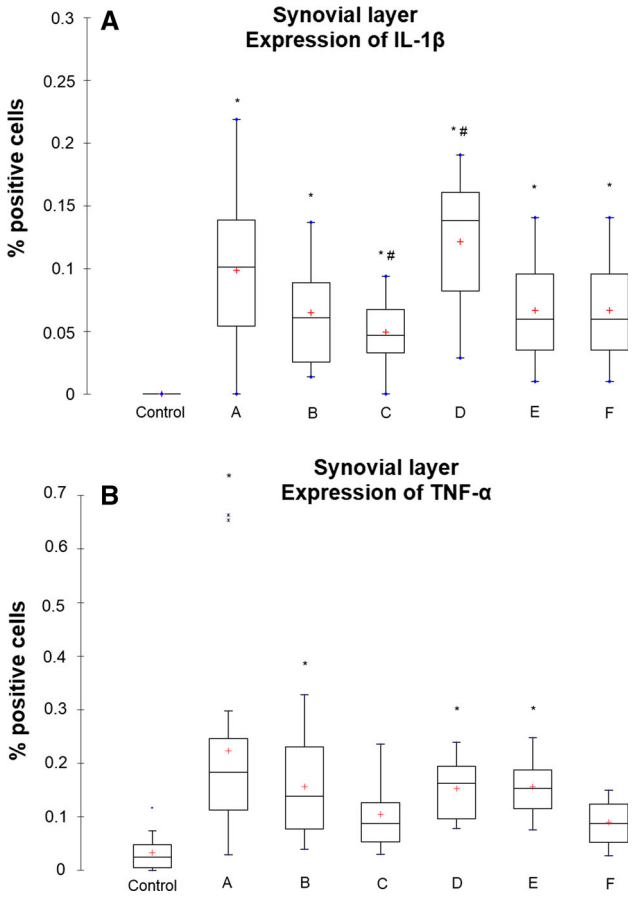

Fig. 3 Elevation in the release of IL-1β (**a**) and TNF-α (**b**) in all polyethylene groups compared to the control in the murine synovial layer, shown as a *boxplot*. The positive cells are given in percent. *Significant difference compared to the control group (PBS) ($P < 0.0024$). #Significant difference between the testing groups ($P < 0.0024$)

4 Discussion

The aim of this study was to test the biological activity of conventional (UHMWPE) and crosslinked polyethylene (XPE) particles on the synovial tissue, the articular cartilage as well as the subchondral bone of the femur and tibia. The hypothesis of this study was that UHMWPE and XPE, similar in size and morphology, lead to similar biological effects in the knee joint. The second hypothesis was that the UHMWPE as well as the XPE particles migrate similarly in- and outside of the joint as well as in the tissues around the joint.

The first hypothesis has been partially confirmed. Wear particles of XPE and UHWMPE showed similar biological activity in the synovium and adjacent bone, however, differences were observed with articular cartilage. The second hypothesis has to be rejected. Although XPE and UHMPWE wear particles of similar size and shape were used, different particle distributions around the joint were detected, clearly dependent on the material that was used.

The results of the present study show that there is a similar inflammatory reaction in the synovial tissue as well as in the subchondral bone by contact with UHMWPE and XPE particles without any effect of the different manufacturing process of these polyethylenes. The intravital microscopy findings could be confirmed by the histological findings.

The cytokines IL-1 and TNF-α were chosen as important parameters of a macrophage activation and because of that as parameters for the osteolytic potential [2, 28–30]. The current immunohistochemical analysis showed a close match to the intravital microscopic and histological evaluations. IL-1 was increased in both the synovial layer and bone marrow in all experimental groups compared with the control ($P < 0.0024$). TNF-α was detected in the subchondral bone for all experimental groups except group F ($P \leq 0.001$). An increase was also reflected in the synovial layer, which was, however, in groups C and F not significant ($P > 0.0024$). Thus, it will be understood that reference to the up-regulation of TNF-α and IL-1 all applied polyethylenes lead to increased macrophage activation [2, 28, 29]. Furthermore it can be assumed, that XPE leads to similar macrophage activation and thereby initiation of the osteolytic cascade as well as conventional UHMWPE. Overall, the similar inflammatory results in the present study for XPE as well as UHMWPE in context with the lower wear rates of the XPE inserts published in a recent study [9] allow the conclusion that XPE can be recommended for the use in bicompartimental knee arthroplasty.

In the present study, across materials, particles had similar size and shape parameters. Thus, an effect of particle size or shape cannot account for the differences in migration behaviour. Thereby, it can be concluded that crosslinking as itself seems to have no effect on the inflammatory response.

The biological activity of wear particles has been studied most in in vitro macrophage models [31–37]. However, since the inflammatory response to wear particles is an extremely complex process with many cells are involved [2], it is essential to measure the biological activity of wear particles even in in vivo models.

With the model used the beginning of the inflammatory response is certainly adequately measurable. The initiation of the inflammatory response to wear particles by leukocyte recruitment had already been confirmed in other studies [38, 39]. In addition, this model can perform intraarticular intravital microscopic measurements in the synovial tissue and then be compared with histological and immunohistochemical methods. Intraarticular particle application represents an important point, since this is the only way to simulate the intra-articular release of wear particles including the distribution of the particles by the synovial fluid in the joint space [40, 41].

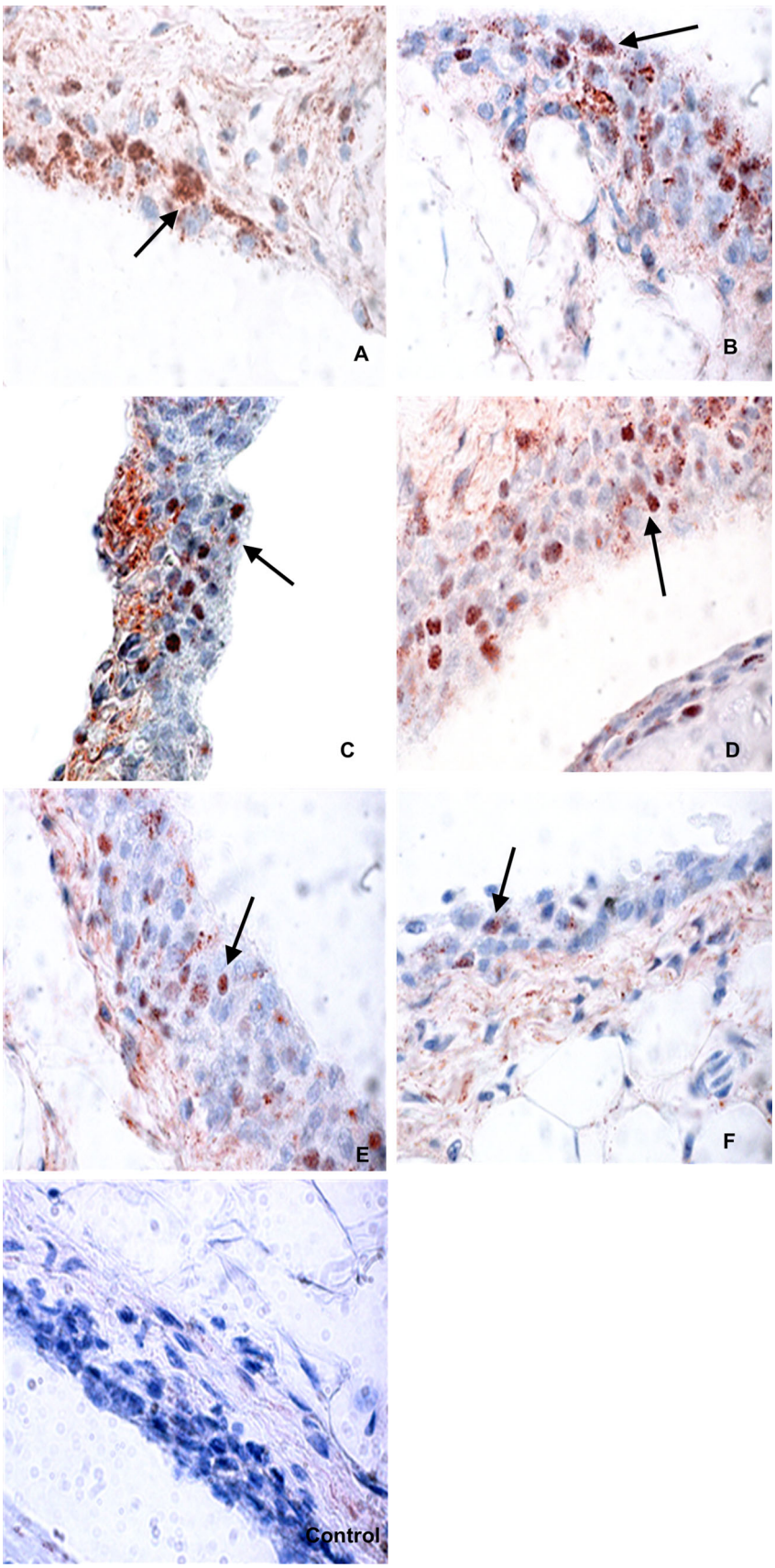

Fig. 4 The synovial tissue of the testing groups (**a**–**f**) and the control stained with TNF-α. In contrast to the control, present all test groups positive stained cells (marked by *arrow*) in the synovial layer. Magnification ×1000

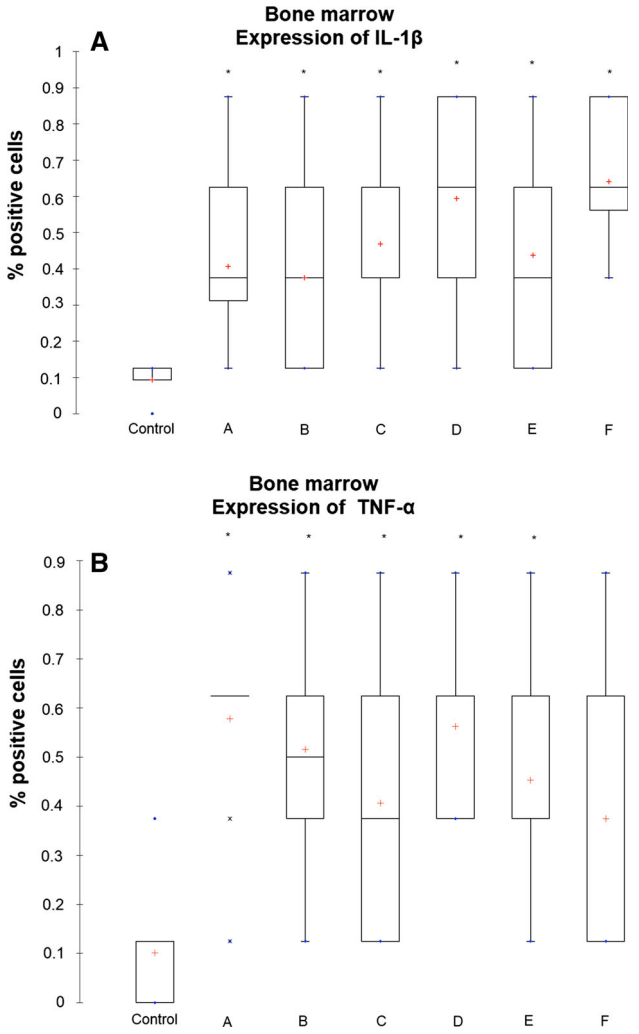

Fig. 5 Increase in the expression of IL-1β (**a**) and TNF-α (**b**) in all polyethylene groups compared to the control (PBS) in the adjacent bone marrow, shown as a *boxplot*. The positive cells are given in percent. *Significant difference compared to the control group (PBS) ($P < 0.0024$)

However, there is no conclusion on the differences in the biological effects of wear particles from XPE compared to UHMWPE [42]. Illgen et al. [43] were able to demonstrate increased osteolysis after 6 days using XPE particles of the same size. However, they used no simulator particles, but irradiated commercial Ceridust® powder with 100 and 400 kGy, so that in this case effects of the different material as well as the higher radiation dose can influence the results. Moreover, the study does not indicate in which way and to what extent a subsequent heat treatment of polyethylene was carried out after irradiation, so it is not clear whether the increased rates of osteolysis are due to charge any of these radicals during irradiation. In addition, in the present study, the initial inflammatory reaction to XPE was measured in contrast to Illgen et al., so that the results are not directly comparable. Furthermore, their measurements were extraarticular, away from the primary effector of the particles took place.

Beyond that, there is lack of data documenting the clinical performance of XPE in knee replacement [44]. Overall, there are only few studies showing clinical results over the first years [45–47]. Hodrick et al. [46] and Minoda et al. [47] conducted comparative studies of XPE and UHMWPE. Minoda et al. [47] reported no revisions, osteolysis or loosening using XPE in a follow-up of 2 years. In the study by Hodrick et al. [46], the XPE group ($n = 100$) had two tibial radiolucencies and no sign of loosening or polyethylene wear at an average of 6 years, whereas the UHMWPE group ($n = 100$; mean follow-up 7 years) had 20 patients with radiolucencies and four patients with tibial loosening.

In the present study it was shown that the particles may migrate already within a week by the synovial membrane and synovial tissue to the bone marrow. After reviewing the current literature, no publication could be found that could show the pathway of the particle from the point of origin (intra-articular) into the synovial layer and the bone marrow exactly in vivo.

So far, the transport of wear particles to the implant-bone interface, where bone resorption takes place, remains unclear [48]. One hypothesis is that there will be micro fissures between the prosthesis and bone, so that the particles pass through the synovial fluid into these fissures [26, 30, 49]. On the other hand, there was already evidence that particles can penetrate bone or be transported by bone in vitro [48, 50]. Due to the present study the notes may be confirmed that particles were transported from the joint into the synovial membrane and the subchondral bone within 7 days without micro fissures. Based on the present study, the question arises whether the particles are not solely transported from the joint into the synovial membrane but also to the bone and then initiate osteolysis there. Against the theory of particle transport via ingress of synovial fluid into micro gaps speaks the fact that wear particles were also found in lymph nodes and distant organs [27, 51].

Moreover, for the first time a different localization of different polyethylene particles in the surrounding tissue was detected. For particle localization, a polarizer was used [26, 27]. To represent the amount of particles semiquantitatively and make them comparable with each other, the particles were slightly, moderately, and much divided into each of the areas of joint space, synovial layer and bone marrow. This methodology was developed itself, as there is no literature for this purpose.

It was notable that in the XPEs A and B, many particles were localized in the joint space, whereas in the XPEs C and D, however, the particles were mainly transported from the joint space into the adjacent bone and also the synovial

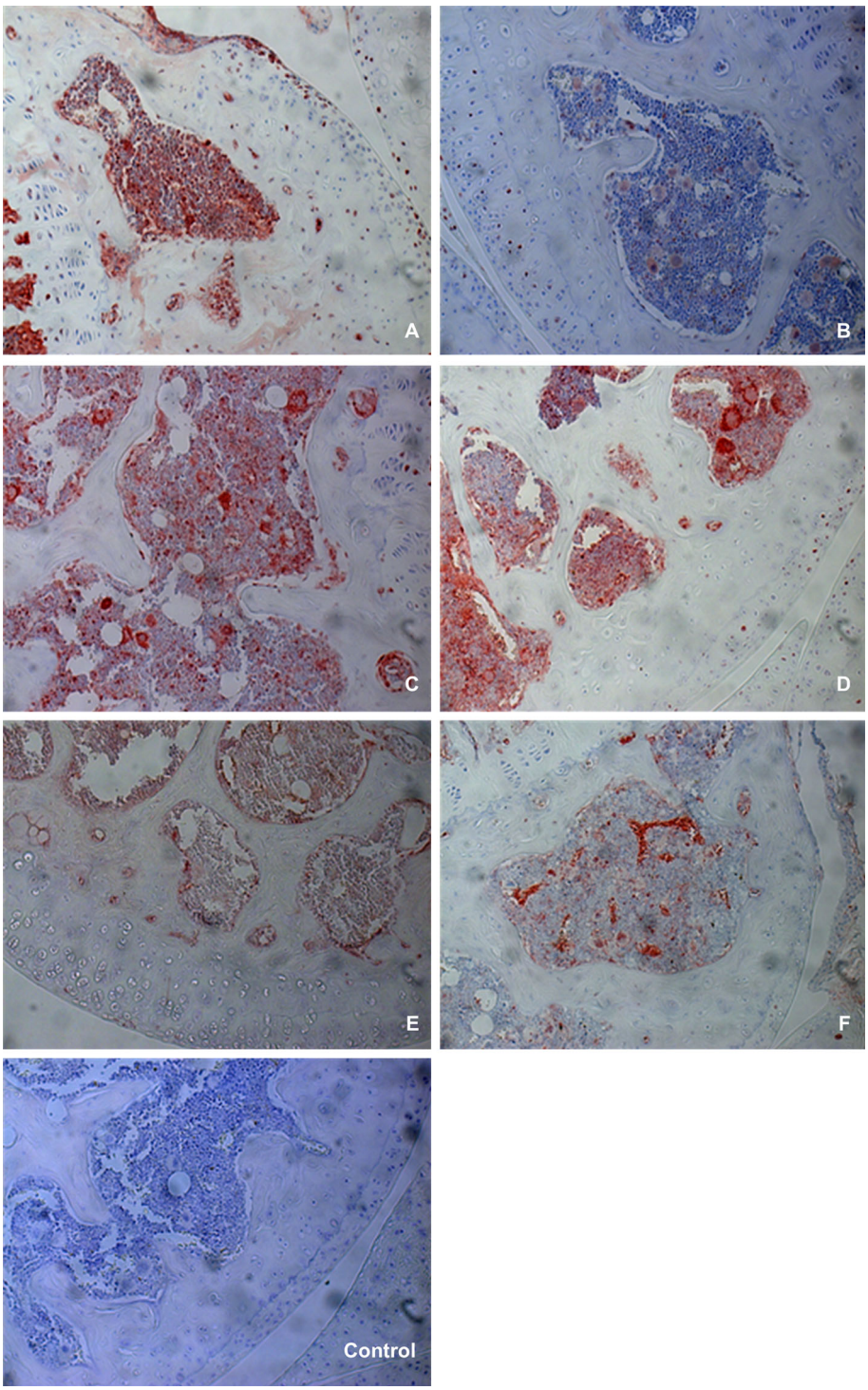

Fig. 6 Adjacent bone marrow of the murine tibia and femur stained with IL-1β. All particle-stimulated groups demonstrate many positive, red stained bone marrow cells whereas the control shows mainly negative *blue* bone marrow cells. Magnification ×200

layer. Since the particles had similar size and shape relationships, this cannot be used to explain the different distribution of the particles in the joint space and the surrounding tissue. The different sterilization methods give no pattern that could explain the separation by the method of sterilization. The radiation doses of the XPEs were similar for A and B as well as for C and D, which could explain the localization. However, these irradiation doses were not similar to those of the UHMWPEs, so that the distribution pattern cannot be explained solely on the irradiation dose. A possible explanation for the similar distribution may lie in surface structures, such as similar

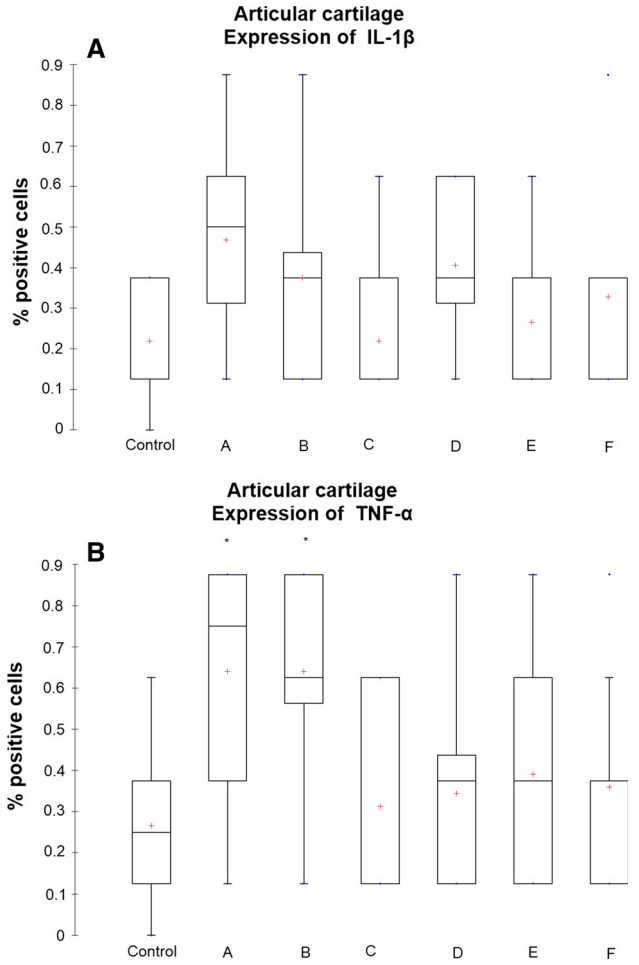

Fig. 7 Expression of IL-1β (**a**) and TNF-α (**b**) in all polyethylene groups compared to the control (PBS) in the articular cartilage, shown as a *boxplot*. The positive cells are given in percent. *Significant difference compared to the control group ($P \leq 0.001$)

surface roughness or surface charge, by which the particles are faster or slower phagocytized and thus resulting in a different distribution in the surrounding tissue.

In contrast to TKA, in UKA only one compartment of the joint is replaced. Thus, there remains cartilage in the opposite compartment of the knee. When XPEs are to be used also in UKA, the biological effect on the chondrocytes of these materials must be investigated. The effect of polyethylene particles on the remaining cartilage is unclear. The effect of particles on chondrocytes has been studied only in a very few studies [6, 7, 52]. Chang et al. found that chondrocytes are able to incorporate polyethylene particles and then increased nitric oxide (NO) and prostaglandin E 2 (PGE 2) was produced. They concluded that polyethylene particles lead via increase of NO and PGE 2 to cartilage degeneration and ultimately to osteoarthritis [6]. Castillo et al. [7] observed that chondrocytes phagocytosed latex particles as evaluated by confocal microscopy and flow cytometry. In a very recent study by Park et al. [52] the authors proved in vivo effects of conventional UHMWPE particles, which were produced by a micro-cutting process, on soft tissues like cartilage and synovium. They conclude that UHMWPE particles per se lead to detrimental effects in these tissues. As these particles show a largely different size distribution compared to wear particles generated for this study, the expressiveness of the study by Park et al. is not directly transferable on the results presented in this investigation, as there definitively smaller particles can be found [12].

In the current study, no significant upregulation of IL-1 in all experimental groups, XPEs as well as UHMWPEs ($P > 0.0024$), could be shown in the cartilage. However, the expression of TNF-α in the articular cartilage was significantly increased ($P \leq 0.001$) in the XPE groups A and B. One possible explanation for that fact could lie in the particle distribution: The most fraction of these particles could be found in the joint space and therefore these particles had direct contact with the chondrocytes. Polyethylene E, as the third material, which was also found to be increased in the joint gap, however showed no significant increase in TNF-α in the articular cartilage compared to the control group. The differences in these three materials in relation to the expression of TNF-α in the chondrocytes may be attributed to the fact that the difference of the materials is located in the irradiation. While the groups A and B are highly irradiated XPEs, E is a conventional UHMWPE. This fact must be pursued in further studies, as the effects on the remaining cartilage using these XPEs may be relevant in unicondylar knee prostheses.

In summary it can be stated that the current model certainly is not a model of osteolysis, which is a limitation of this study, but it has to be discussed which part of the reaction cascade of inflammatory response to wear particles would actually be measured with the respective model [53]. Additionally, for the animal experiments no implants were used. The wear particles solely were injected into the murine knee joint. Thus, it is not possible to investigate the reaction between the bone–implant interface. Using this inflammatory reaction based animal model, it is only indirectly possible to draw conclusions concerning osteolysis. Furthermore, to verify even clearer results, larger sample sizes would have been necessary, which was not possible due to the applicable regulations by law for animal experiments.

In conclusion, according to the data of this study, crosslinked polyethylenes have to be suggested for TKA. Especially the combination of similar inflammatory reactions of XPE and UHMWPE particles and the previously described lower wear rates using XPE [9] promise lower osteolytic reactions and consecutive increased survival rates of those TKAs.

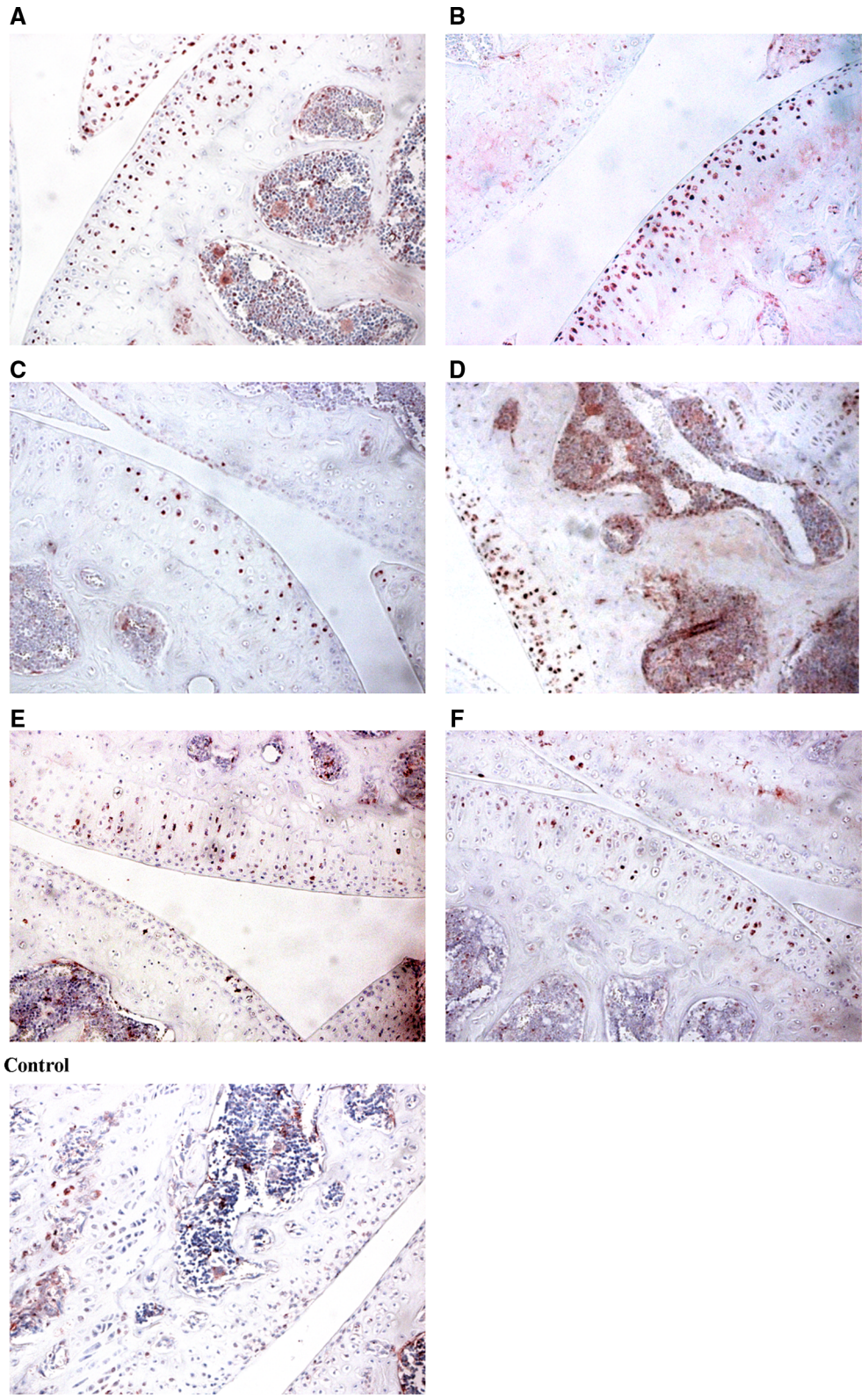

Fig. 8 Articular cartilage stained with TNF-α. The XPE groups A and B present many positive, red stained chondrocytes. Magnification ×200

The presence of certain crosslinked wear particles in the joint space elevates the inflammatory reaction in the remaining cartilage. If XPEs are planned as alternative bearing material in UKA, this reaction on the cartilage must be further investigated and must not be underestimated.

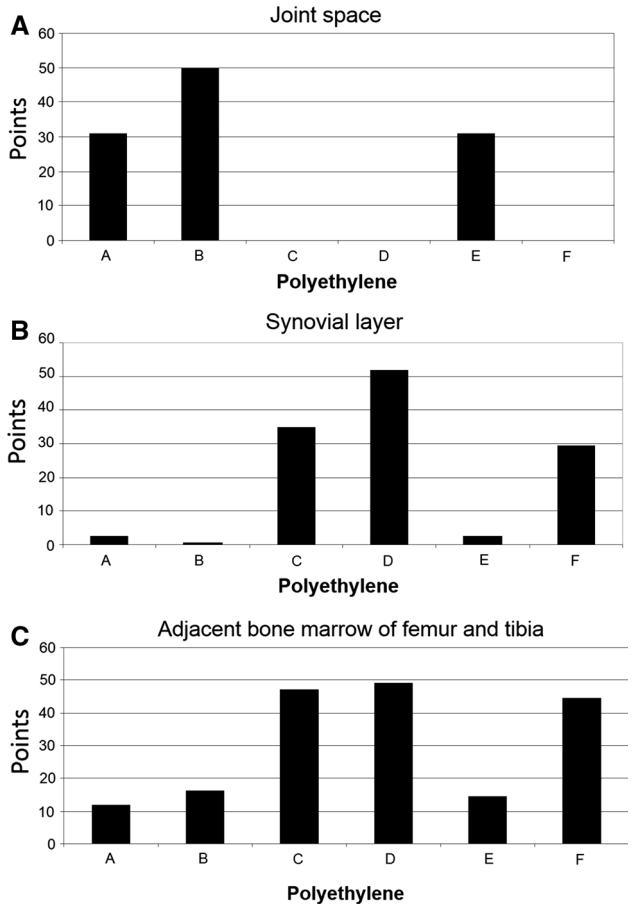

Fig. 9 Distribution of wear particles out of different polyethylenes around the joint. It can be found a clear distribution according to the different materials

Whether the different particles distributions influence the inflammatory osteolytic potential on long term needs to be clarified in further studies. As there is no data on this aspect in the literature, it must be examined whether the retention of particles in the joint space ultimately leads to an increased inflammatory response and to the maintenance of this process or the rapid removal into the bone lead to an increased osteolytic potential.

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