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# EXPERIMENTAL OSTEOARTHRITIS OF THE KNEE IN RABBITS INDUCED BY ALTERATION OF THE LOAD-BEARING

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Numerous experimental methods for inducing degenerative changes of the articular cartilage in animals have been described. Only in a few cases have attempts been made to obtain conditions which encourage the development of a slowly progressive degeneration analogous with the predisposing factors known from clinical work (Hulth et al. 1970). It is generally agreed that abnormal mechanical conditions predispose to degenerative changes in the misloaded joint.

The aim of the present work was to investigate whether it is possible experimentally, by means of valgus angular osteotomy of the tibia in rabbits, to induce degenerative changes in the cartilage in the knee joint analogous with osteoarthritis in man.

## MATERIAL AND METHODS

Six adult rabbits weighing 3.2 to 3.5 kg were used for the experiments.

*Operative procedure:*

Observing sterile precautions and under Fluothane anaesthesia preceded by 0.5 ml Combélène i.m., osteotomy was performed on the left tibia. The operation was made by means of medially arched incision and the proximal area of the tibia was laid open subperiostally. Transversal osteotomy was then performed by sawing just distal to the tibial tuberosity, and at the same time a small wedge was removed laterally from the bone with its base, resulting in a valgus position of the tibia of approximately 30°. The rudimentary fibula was partially resected. The position was fixed with a five-hole plate with four screws; prior to fixation the plate was bent to make it conform to the valgus position. The wound was flushed with Nebacetine and closed with nylon in fascia and skin. No other immobilization was made except the internal fixation. There were no postoperative complications.

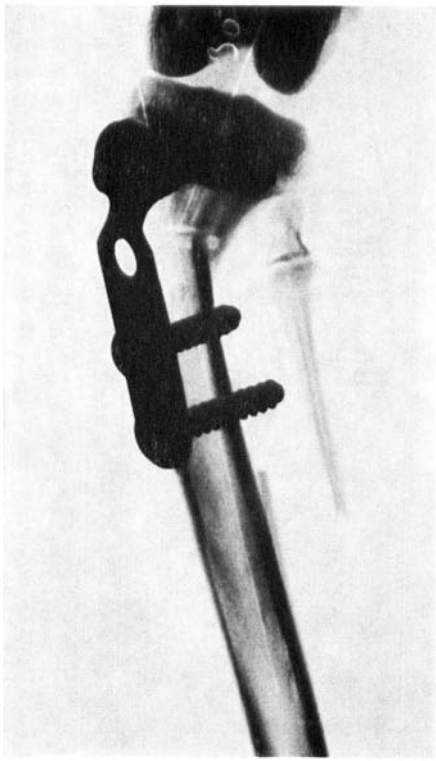

*Figure 1. Radiograph from case 13 immediately after the operation illustrates osteotomy of the upper portion of the tibia with 30° of lateral angulation. Fixation with a plate and 4 screws.*

The position was controlled radiologically post-operatively (Figure 1), and during the investigation period radiographical control took place fortnightly.

The animals were killed 10 to 12 weeks after the operation. In continuation of the killing, microangiography was carried out with Micropaque containing Indian ink through the abdominal aorta on the heparinized animal. The hind parts of the animal were fixed in 90 per cent alcohol, which was replaced after 24 hours by 40 per cent alcohol.

The specimens were evaluated macroscopically with regard to changes of the components of the knee joint, especially the cartilage, and the valgus position was measured.

*Specimens:*

Histological specimens were made from six cartilage-bone blocks from each knee joint, as shown in Figure 2. Specimens from the unoperated side were used for comparison. Pieces A and B were from the medial and lateral tibial condyle, respectively, and pieces C and D from the medial and lateral femoral condyle. Specimen F is the patella, and E (consisting of bone only) was included in order to study whether the osteotomy had healed histologically. The specimens were embedded, without previous decalcification, in methyl-metacrylate containing Plastoid N and afterwards cut by Jung microtome into sections with a thickness of 5 $\mu$. From each

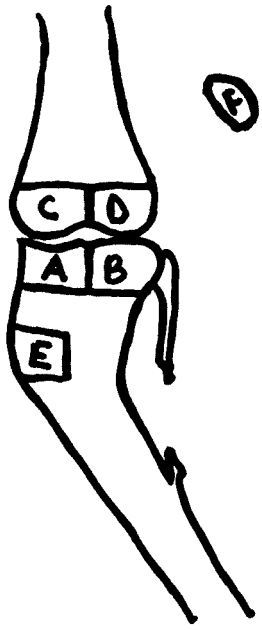


*Figure 2. Diagram illustrating the different bone-cartilage specimens taken for histological examination. A: from medial tibial condyle; B: from the lateral tibial condyle; C: from the medial femoral condyle; D: from the lateral femoral condyle. F indicates patella and E the specimen taken from the site of the osteotomy.*

block, serial sections were made in three places perpendicular to the cartilage. The specimens were stained by haematoxylin-eosin and toluidine blue 0.1 per cent in 30 per cent alcohol.

## RESULTS

Examination prior to the killing showed that all osteotomies had healed clinically. Healing had also been observed radiologically in all cases and no radiological signs of osteoarthritis were seen. In one case (No. 14) a flexion contracture of 10–20° was found in knee and ankle joints.

Macroscopical examination disclosed that the valgus angulation varied from 29 to 36° (Table 1). Visible changes of the articular cartilage had taken place on the lateral tibial condyle in one case only (No. 13), where the cartilage was found to be yellowish and soft in spots.

On the basis of evaluation of the histological specimens, initial signs of degenerative changes were present in all cases.

Obvious changes were found in the lateral tibial and femoral condyles. No demonstrable histological changes were seen corresponding to the medial tibial and femoral condyles or on the patella. All the specimens originating from the osteotomy sites (E) showed normalized

*Table 1. Macroscopic and microscopic changes.*

| Case no. | Operation | Time (weeks) | Degree of valgus angulation | Macroscopic changes | Microscopic changes |
|---|---|---|---|---|---|
| 10 | left tibial osteotomy with lateral angulation | 12 | 30 | – | B + D |
| 11 | – | 10 | 29 | – | D |
| 12 | – | 10 | 33 | – | B |
| 13 | – | 10 | 36 | cartilage at lateral tibial condyle | B |
| 14 | – | 10 | 33 | contracture of knee and ankle joint | B + D |
| 15 | – | 10 | 30 | – | D |

B: cartilage from left lateral tibial condyle
D: cartilage from left lateral femoral condyle

conditions. Histological investigation disclosed small remnants of the epiphyseal plate on some of the sections from cases Nos. 11 and 12.

Fibrillation, clefts or flaking were seen in the cartilage of the lateral condyles of the operated knees, and furthermore a moderate decrease in the thickness of the cartilage had taken place (Figure 3). Compared with the specimens from the unoperated side, the toluidine blue stained specimens showed loss of metachromasia in the superficial layers, often combined with loss of staining power of the nuclei of the chondrocytes. A clear derangement of the cell columns was found in four of the six cases (Figure 4). Clusters indicating proliferation of the chondrocytes were found in only a few sections. The zone of calcified cartilage and the tidemarks were found to be well preserved. The subchondral bone showed moderate thickening of the bone trabeculae. None of the specimens showed signs of osteoporosis. In two of the specimens (from cases Nos. 12 and 13) vessels were found penetrating into the cartilage (Figure 5).

No signs of osteophyte formation were found, but in some of the specimens changes were observed at the ligament and capsule insertions, where the edges had become frayed in the superficial layer.

## DISCUSSION AND CONCLUSION

Abnormal mechanical stress is considered to be a primary etiological factor in osteoarthritis (Collins 1953), but the degree is not known

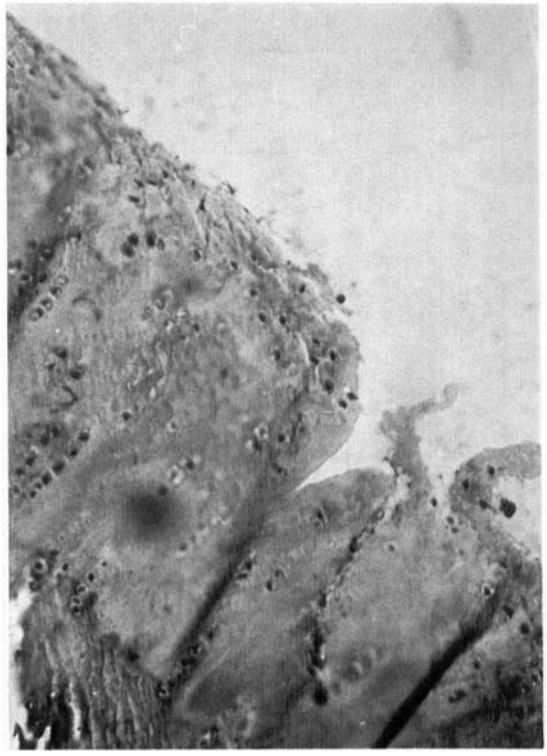

*Figure 3. Articular cartilage from the left lateral femoral condyle (case 15) 10 weeks after the osteotomy. Illustrates fibrillation, flaking and clefts in the superficial layer. Haematoxylin-eosin. Magnification* × *182.*

exactly. Knock-knee and bowleg deformities are reported as static causes of an increased stress on the knee joint, resulting in osteoarthritis on the side to which the knee is turned (Ory 1964).

Even in cases of moderate degrees of valgus or varus angulation, there is a considerable increase of the load on the exposed partial joint cavity and consequently of the compressive force (Marchin et al. 1963). That increased stress induced by compression causes degenerative changes of the cartilage has been proved experimentally (Salter & Field 1960, Thompson & Bassett 1970).

Clinical reports describe osteoarthritic changes occurring after shorter or longer periods of displacement corresponding to the knee condyle which is most stressed (Jackson & Waugh 1961, Coventry 1965). On the basis of a follow-up of 85 knee osteotomies for osteoarthritis with axis displacement, Cauchoix et al. (1968) reported that the radiological

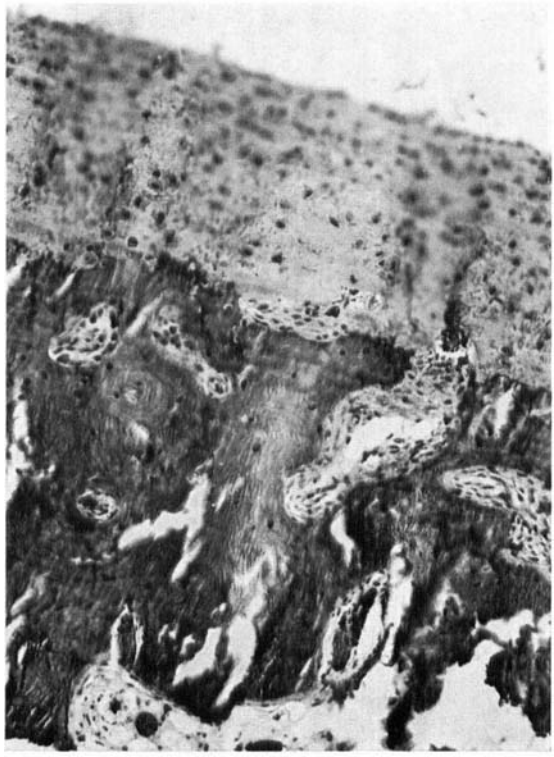

*Figure 4. Slight fibrillation in the superficial layer and derangement of the cell columns of the cartilage from the left lateral tibial condyle 10 weeks after the operation (case 14). Haematoxylin-eosin. Magnification × 140.*

picture had not changed and that the operation seemed to have stopped the degenerative process. This would indicate that abnormal stress must be regarded as an etiological factor.

In the present experiment, an attempt was made to induce degenerative changes in the knee joint in rabbits by altering the load-bearing by means of valgus angular osteotomy of the tibia. In view of the time factor, a valgus angulation of 30° was chosen, and 10 to 12 weeks was estimated to be the minimum observation time. After that period no radiographical changes indicating osteoarthritis had occurred. Only in one case were macroscopical changes observed, whereas histological examinition disclosed clearly initial degenerative changes of the cartilage and the underlying bone corresponding to the areas exposed to the biggest stress, i.e. the lateral tibial and femoral condyles.

The degenerative changes found resemble those described in connec-

*Figure 5. From the left lateral tibial condyle 10 weeks after osteotomy (case 13). Vessels, indicated by black, seem to penetrate from the subchondral marrow. Thickening of the bone trabeculae in the subchondral zone. Haematoxylin-eosin. Magnification* × *224.*

tion with human osteoarthritis (Sokoloff 1969). Among the initial degenerative histological changes, the most constant finding was loss of metachromasia in the superficial layers of the cartilage. This is probably due to a reduction in the content of chondroitin sulphate in the cartilage, possibly on an enzymatic basis. Based on this theory, Bentley (1971) experimentally induced degenerative changes of the cartilage by means of intraarticular injection of the enzyme Papain, which is able to liberate the chondroitin sulphate from the protein-polysaccharide complex in the articular cartilage.

Other initial degenerative changes (Sokoloff 1969) consisted in most

cases of changes of the surface, in the form of fibrillation, clefts or flaking. Together with those changes, loss of nucleic staining of the chondrocytes in the superficial layers was often found, indicating necrobiosis (Salter 1960), and furthermore there was derangement of the cell column. Proliferation of the chondrocytes in the form of clusters (Hulth et al. 1970) is reported by Sokoloff (1969) as a regular finding, especially at the bottom of the fissures on the surface. This was also found in some of the sections. The calcified zone and tidemarks were well preserved, which is in accordance with the report by Thompson & Bassett (1970) on the basis of experimental compression tests. Following compression, early thickening of the subchondral bone trabeculae has also been seen (Salter 1960), corresponding to a moderate degree to the lateral condyles subjected to the biggest load.

## SUMMARY

On the basis of the clinical experience that loading of the knee by displacement for a certain period predisposes development of osteoarthritis in the area of the knee exposed to the biggest loading, attempts were made to induce osteoarthritis experimentally on six rabbit knees by means of valgus angular osteotomy of the tibia. Valgus angulation of 30° and an observation time of 10 to 12 weeks were employed. The experiments demonstrated that no radiological signs of osteoarthritis occurred and that slight macroscopical changes of the lateral tibial condyle were found in only one case. However, clear initial changes were found histologically in the cartilage of the lateral tibial and femoral condyles in the form of loss of metachromasia, fibrillation, clefts and flaking in the superficial layers, together with loss of nucleic staining. Moderate sclerosis was found subchondrally.

It is concluded that it would appear to be possible to induce initial degenerative changes analogous to human osteoarthritis by means of an experimentally induced alteration of the load-bearing.

## REFERENCES

Bentley, G. (1971) Papain-induced degenerative arthritis of the hip in rabbits. *J. Bone Jt Surg.* **53**-B, 324–337.

Cauchoix, J., Duparc, J., Lemoine, A. & Deburge, A. (1968) L'ostéotomie dans les gonarthroses avec déviation angulaire dans le plan frontal. *Rev. Chir. orthop.* **54**, 343–360.

Collins, D. H. (1953) Osteoarthritis. *J. Bone Jt Surg.* **35**-B, 518–520.

Coventry, M. B. (1965) Osteotomy of the upper portion of the tibia for degenerative arthritis of the knee. *J. Bone Jt Surg.* **47**-A, 984–990.

Hulth, A., Lindberg, L. & Telhag, H. (1970) Experimental osteoarthritis in rabbits. *Acta orthop. scand.* **41**, 522–530.

Jackson, J. P. & Waugh, W. (1961) Tibial osteotomy for osteoarthritis of the knee. *J. Bone Jt Surg.* **43**-B, 746–751.

Marchin, P. de, Maquet, P. & Simonet, J. (1963) Considérations biomécaniques sur l'arthrose du genou. *Rev. Rhum.* **30**, 775–783.

Ory, M. (1964) Des influences mécaniques dans l'apparition et le développement des manifestations dégénératives du genou. *J. belge Méd. phys. Rhum.* **19**, 103–120.

Salter, R. B. & Field, P. (1960) The effects of continous compression on articular cartilage. *J. Bone Jt Surg.* **42**-A, 31–49.

Sokoloff, L. (1969) *The biology of degenerative joint disease.* The University of Chicago Press.

Thompson, R. C. & Bassett, C. A. L. (1970) Histological observations on experimentally induced degeneration of articular cartilage. *J. Bone Jt Surg.* **52**-A, 535–443.

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