The influence of the sagittal split osteotomy on the condylar cartilage structure and the subchondral vascularization of the temporomandibular joint: A preliminary study in goats
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Objective. The concern that a sagittal split osteotomy of the mandibular ramus could compromise the subchondral vascularization and especially the venous outflow in the condylar area, which in turn could influence the structure of the condylar cartilage, prompted this preliminary study on goats.

Study design. A sagittal split osteotomy was performed in the mandibular ramus at 1 side in each of 6 young adult goats. The contralateral side served as control. The animals were killed after different postoperative periods, ranging from 1 to 15 days. Histomorphometric analyses were performed after perfusion of the vascular system with India ink.

Conclusion. Based on the observed cartilage thickening, the sagittal split osteotomy may influence the condylar cartilage in the first days postoperatively through a disturbance of the vascular supply.


Bilateral sagittal split osteotomy (BSSO) is widely used in maxillofacial surgery, but little is known about the consequences of this operation for the vascularization of the bony fragments. Experimental studies performed in the 1970s1,2 suggest that reduction of the blood flow in the proximal segment above the osteotomy site might be sufficient to cause necrosis of the ramal cortex. The consequences of osteotomies for the blood supply of the temporomandibular joint (TMJ), however, are still unknown. Yet, it has been suggested that impairment of condylar vascularization may play a role in the etiology of progressive condylar resorption (PCR)3; a condition that has been observed in 4% of the patients who undergo BSSO.4 Whether or not this idea holds true depends on the vascular supply in the condylar area, which is rather complicated.

The main blood supply of the body of the mandible (the teeth, the periodontium, and related regions in humans, monkeys, and other mammals) is via the inferior alveolar artery.5,6 The mandibular angle, the coronoid process, and the condyle each has a regional blood supply.2,7-10 The head and neck of the condylar process are supplied through the lateral pterygoid muscle,1,8,11 and additional arterial branches from the medial pterygoid and masseter muscles provide blood supply to the vertical ramus. The TMJ itself is supplied through the superficial temporal artery with the rami parotidei and the transversal facial artery. The anterior tympanic artery, which is a branch of the maxillary artery, is responsible for the dorsal supply of the TMJ. Another branch of the maxillary artery, the posterior profound temporal artery, supplies the anterior part.12 The venae articularis temporomandibularis maintain the venous outflow directly into the retromandibular vein or pterygoid plexus.

The intraosseous subchondral vascularization of the condylar head is probably through vascular anastomoses between the vascularization of the TMJ and the inferior alveolar artery and vene.8,12-16 The role of this part of the vascularization in the development of PCR is unclear up to now. Its importance was recognized by Grammer et al.,2 who concluded from a study on monkeys that the proximal segment of the mandibular ramus after sagittal split osteotomy (SSO) suffers from a reduction in blood flow. In some cases, that was sufficient to cause its devitalization after SSO. Therefore, it was recommended to detach the mucoperiosteum and the pterygomasseteric sling minimally in order to reduce intraosseous ischemia and necrosis.1,17 It is still not known what the consequences are of the SSO, including its horizontal bone cut above the mandibular foramen, for the vascularization of the condylar head and neck. It is obvious that the vascularization in the proximal segment is cut off from the inferior alveolar artery and vein while stripping of the mucoperiosteum causes at least temporary impairment of the blood circulation. Therefore, it can be hypothesized that the BSSO can compromise the

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Received for publication Jun 1, 2004; accepted for publication Nov 20, 2004.
Available online 19 February 2005.
1079-2104/5 - see front matter
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doi:10.1016/j.tripleo.2004.11.007
subchondral venous outflow in the condyle, which might influence the structure of the condylar cartilage.

Only a few studies have focused on the changes in the condylar cartilage under controlled conditions. Advancement surgery in the mandibles of rhesus monkeys using BSSO showed an effect of the type of fixation of the segments on the morphology of the condylar cartilage. Intermaxillary fixation led to thicker fixation of the segments on the morphology of the monkeys using BSSO showed an effect of the type of advancement surgery in the mandibles of rhesus the condylar cartilage under controlled conditions.

However, both studies mentioned above used an osteotomy in combination with a relative displacement of the bony segments. This displacement inevitably leads to changes in the mechanical loading of the condylar cartilage, and the subsequent changes in the condylar cartilage or its subchondral vascularization are not necessarily due to the osteotomy itself. For a better understanding of the development of PCR and the possible measures that can be taken to prevent or to overcome this problem, it is necessary to conduct a study into the effects of an isolated sagittal split osteotomy without displacement of the bony fragments. However, such studies have never been documented in the literature.

Therefore, the aim of this preliminary study was to develop an experimental animal study into the changes that occur over time in the subchondral vascularization and the condylar cartilage after a sagittal split osteotomy without repositioning of the bony fragments.

**MATERIAL AND METHODS**

Six adult white goats (5 females, 1 hermaphrodite) were used in this study. These Saanen goats, weighing about 60 kg each, were 2-2.5 years of age and were housed according to the national guidelines for the care and use of laboratory animals. The Animal Ethical Committee of the University of Nijmegen granted permission for this research.

The operations were performed under general anesthesia, which was induced by an intravenous injection of pentobarbital and was maintained by 2%-3% isoflurane through a constant volume ventilator, administered through an endotracheal tube. The heart rate was constantly monitored. Prophylactic antibiotics were given to reduce the risk of perioperative infection. The goats received 3 doses of ampicillin: during the operation (Albipen 15%, Intervet Int., Boxmeer, the Netherlands) and on the first and third postoperative days (Albipen LA).

At the operation site the skin was shaved and disinfected with povidine iodine solution. A submandibular skin incision was made, while the facial artery and vein were kept intact. The masseter muscle and the medial pterygoid muscle were dissected from the mandibular angle to obtain sufficient access to the osteotomy site just cranial of the mandibular foramen. The neurovascular bundle, entering the mandible, was kept intact as well. The buccal osteotomy line was made just behind the last molar (Fig 1).

A 5.4-mm preshaped osteosynthesis titanium plate (Martin, Tuttlingen, Germany) was adapted to the local morphology. The sagittal split osteotomy was performed using burrs and osteotomes. The segments were mobilized and subsequently placed in the original position and fixed, using the preshaped plates and 2.3-mm monocortical center-drive screws (Martin). The soft tissues were closed in layers with resorbable Vicryl 3.0 sutures. The contralateral side was not operated on and served as control. Postoperative pain was treated by Flunixin 1.5 mg/kg (Finadyne, Schering-Plough, Segre, France).

At 1 (2 goats), 3, 8 (2 goats) and 15 days post-operatively, the 6 animals were killed for histological evaluation. Prior to killing them, the animals were brought under general anesthesia as described above. Then 0.5 mg/kg heparin (Thromboliquine; Organon, Boxtel, The Netherlands) was given intravenously, followed by a lethal dose of pentobarbital after a few minutes. The thorax was opened and the vascular system was perfused with physiologic saline through the aortic arch, followed by a freshly prepared India ink solution. The India ink solution was prepared as a 1:1 dilution of filtrated India ink (Talens, Apeldoorn, The Netherlands) in physiologic saline.

Next, the right and left halves of the mandibles were dissected and immersed in 4% buffered formaldehyde for 2 weeks. Smaller blocks, containing the mandibular ramus and the condyle, were prepared and decalcified in 20% formic acid and 5% sodium citrate. After decalcification the tissue blocks were split sagittally, resulting in specimens containing either the buccal or the lingual part of the condyle and the cranial part of the ramus. These specimens were dehydrated in a graded series of ethanol and embedded in paraffin. Sagittal serial sections of 7 μm were cut and 2 series (1:25) were mounted on glass slides. One series was stained with hematoxylin and eosin (H&E), the other series with 1% toluidine blue in water for 5 minutes.

Morphometric analysis was performed on 5 sections, 175 μm apart, per tissue block. This means that per condyle 10 sections were used in total. The H&E-stained series was used for quantification of the relative blood vessel volume, which was facilitated by the perfused India ink. Per section, 5 microscopic fields were randomly selected at a final magnification of 100×. An ocular point-grid was used to analyze the
area adjacent to the erosion zone underneath the cartilage. The relative number of grid points inside blood vessels was determined as a measure for the relative vessel volume. The toluidine blue-stained series was used for the analysis of the relative thickness of the different cartilage layers. Again, 10 sections per condyle were used, and the relative thickness of the germinative, the hypertrofying, and the hypertrophic layers were determined, using an ocular micrometer at a final magnification of 160×. Per section, 3 randomly chosen microscopic fields in the ventral and 3 in the dorsal area of the condylar cartilage were evaluated.

**Statistics**

For the measurement of the relative blood volume 5 different microscopic fields were used in each section. Per section, at least 200 ocular grid points were evaluated. The data were added up per section.

Fig 1. **A**, The osteotomy line on the buccal side of the goats mandible. **B**, The osteotomy line above the mandibular foramen just passed this large foramen. The dotted line represents the fracture on the lingual side.

Fig 2. **A**, Anatomical reposition and fixation of the fragments after BSSO. **B**, Control side same goat.
and the relative amount of points within blood vessels was expressed as a percentage of the total number of points counted. The sections at the buccal and lingual areas of each condyle were considered to belong to 1 group. Statistical analysis of the differences between control and experimental condyles for each animal was performed by a Student $t$ test. In a single case the data failed the normality test, and a Mann-Whitney rank-sum test was performed. Differences were considered to be significant after Bonferroni’s correction if $P \leq .0083$.

The difference in mean total thickness of the cartilage in the ventral and the dorsal areas between the control and the experimental side in each animal was analysed by Student $t$ test. Differences were considered to be significant after Bonferroni’s correction if $P \leq .0042$.

The correlation between the mean relative blood vessel volume and the mean cartilage thickness for each condyle was analysed for the control and the experimental sides by linear regression analysis.

RESULTS

All goats had an uneventful recovery and were walking around within a few hours. Within 24 hours all were eating and ruminating again. Their weight remained stable. No wound infections occurred. After killing them, no plate fractures or loose screws were found.

The radiographs taken directly postmortem showed an anatomical reposition and fixation of the fragments after the SSO and good position of the plates and screws in all goats. No abnormalities were seen in the condyles (Fig 2). Histological evaluation of the condylar cartilage was performed on a series of sagittal sections (Fig 3).

The cartilage showed a normal appearance. In most cases, different layers could be recognized: the articular surface layer consisting of fibrocartilage, a germinative and proliferative layer with small chondrocytes, a hypertrophying layer in which chondrocytes increased in size, and a hypertrophic layer. Just below the hypertrophic layer, abundant cartilage resorption was seen and new bone was deposited against cartilage remnants (Fig 4).

In the erosion layer, blood vessels could be recognized by India ink perfusion (Fig 4). No clear morphologic distinction could be made between condyles at the side where the sagittal split was performed and the control side. The results of the measurements on the volume percentage obtained by blood vessels are presented in Table I and illustrated in Fig 5.

**Table I.** Mean volume percentage $\pm$ SD obtained by blood vessels in control and experimental condyles at different time points after surgery

<table>
<thead>
<tr>
<th>Days postsurgery</th>
<th>Control</th>
<th>Experimental</th>
<th>$P$ level</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>16.8 ± 2.2</td>
<td>31.6 ± 6.3</td>
<td>$&lt;.001$</td>
</tr>
<tr>
<td>1</td>
<td>24.2 ± 3.9</td>
<td>19.2 ± 2.5</td>
<td>.003</td>
</tr>
<tr>
<td>3</td>
<td>28.5 ± 5.8</td>
<td>24.8 ± 5.7</td>
<td>.170</td>
</tr>
<tr>
<td>8</td>
<td>23.8 ± 4.8</td>
<td>21.2 ± 3.0</td>
<td>.154</td>
</tr>
<tr>
<td>8</td>
<td>11.1 ± 3.1</td>
<td>21.0 ± 5.9</td>
<td>$&lt;.001$</td>
</tr>
<tr>
<td>15</td>
<td>17.8 ± 6.2</td>
<td>16.9 ± 3.1</td>
<td>.756</td>
</tr>
</tbody>
</table>
In the control condyles the percentage ranged from 11.1% to 28.5% and in the experimental ones from 16.9% to 31.6%. In 3 out of 6 goats a significant ($P < .0083$) difference was found between the experimental and the control sides. The differences, however, do not point in the same direction.

The areas where cartilage thickness was measured are shown in Fig 6. The measurements of the cartilage thickness are summarized in Table II and illustrated in Figs 7 and 8. At first sight the cartilage seems to be thicker in almost all experimental sides at all times. However, only 4 out of 12 differences turn out to be significant after Bonferroni’s correction for multiple testings. Three of these 4 significant differences were found at 1 day postsurgery.

The regression analysis showed that in the control condyles there was a significant correlation between the relative blood vessel volume and the thickness of the condylar cartilage (explained variance $R^2 = 0.490$). The explained variance in the experimental condyles was far lower ($R^2 = 0.078$).

**DISCUSSION**

The aim of this preliminary study was to develop a model for the study of the effects of a sagittal split osteotomy of the mandibular ramus without
displacement on the condylar cartilage and the subchondral vascularization. To that end, an experimental animal study was performed in goats. The rationale behind the present study is that the few studies dealing with this subject all combined the osteotomy per se with dislocation of the bony segments, thereby also causing confounding changes in the mechanical conditions in the area.

The present study was performed in goats, but to our knowledge no detailed studies are available on the vascular supply and venous outflow of the goat mandibular condyle. Although the dimensions and

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**Fig 7.** Cartilage thickness in the ventral region of control and experimental condyles at different days post surgery. *Indicates significant differences between control and experimental condyles.

**Fig 8.** Cartilage thickness in the dorsal region of control and experimental condyles at different days post surgery. *Indicates significant differences between control and experimental condyles.
the morphology of the mandible in these animals comes close to that of humans, there are fundamental differences. Goats are herbivores with a totally different dental morphology. They have hypsodont premolars and molars, which are continuously erupting teeth and therefore require extensive blood supply. The vascular supply of the mandibular body is also more extensive than in humans, which is reflected in a large mandibular foramen and mandibular canal (Fig 1, B). Since the mastication of goats is also quite different from humans, the vascularization of the ramal and condylar areas is probably different too. Another drawback of the use of goats is that the osteotomy cannot be performed in the same way as in humans because of the long snout and the limited mouth opening. Therefore, an extraoral approach had to be chosen. One has to keep in mind, however, that the vascularization of the ascending ramus and condyle in goats may be somewhat different from human beings. The extraoral approach may also cause a different vascular disturbance. Yet, it comes as close as possible to the clinical situation, given the limitation of the chosen animal. A study on primates would have been more desirable but was not considered because of ethical reasons.

It was expected that the vascular supply and venous outflow would be mostly affected within the first 2 weeks after surgery. For this reason, analyses were performed at 1, 3, 8, and 15 days after surgery. Two extra goats appeared to be available and they were assigned to the 1- and 8-days follow-up periods, respectively. All split osteotomies were uneventful and without any postoperative complications.

The perfusion of the vascular system with India ink was effective, and stained blood vessels could be easily recognized in the sections. The volume percentage of the subchondral bone occupied by blood vessels was variable in the control condyles, which was probably due to individual variations rather than to some time-dependent factor. Two of the animals showed a significantly higher volume percentage in the experimental than in the control condyles. The other 4 animals showed a lower mean value in the experimental condyles than in the controls. This difference, however, was not significant except in 1 case. Therefore the data were not conclusive. This is not surprising, considering the small number of animals involved in the study.

The mean thickness of the condylar cartilage appeared to be larger in all experimental condyles than in the controls. This difference, however, was not significant in most of the cases, and therefore the data only suggest that the normal physiology of the condylar cartilage is disturbed by the osteotomy. In these young adult goats, hypertrophic cartilage is always present in the condyle, suggesting that some growth still is taking place. The increased thickness, therefore, might be caused by a decrease in clastic resorption of the cartilage in the erosion zone. Because clastic cells differentiate from blood-borne monocytes, this suggests that an inadequate blood supply might be involved. This is in accordance with the findings of Kuijpers-Jagtman et al,20 who showed that vascular interferences of the subchondral vascularization in long bones of growing rabbits led to a total or partial thickening of the growth plate due to an accumulation of hypertrophic cells. In these young rabbits, the thickening started to decrease after 5 days, owing to erosion of the hypertrophic cells and the ingrowth of new blood vessels. Assuming that the recovery in adult individuals would take more time, one could speculate that in the present study the cartilage thickening would persist for a longer period of time. If and how this would be related to the onset of progressive condylar resorption remains to be elucidated.

REFERENCES


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