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Histomorphometric analysis of unilateral condylar hyperplasia in the temporomandibular joint: the value of the condylar layer and cartilage island

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Abstract. This study aimed to describe the condylar layer and cartilage island in subjects with unilateral condylar hyperplasia (UCH). Five individuals (15–18 years old) with a diagnosis of UCH, treated in a university hospital in Temuco, Chile, were included. The analysis examined the presence, extension, and thickness of the layers on the condylar surface, the number, depth, and area of the cartilage islands, and the argyrophilic proteins of the nucleolar organizer region (AgNOR) score. Statistical significance was set at $P < 0.05$. The fibrocartilaginous layer was thickest (0.13 ± 0.05 mm) and the joint layer was thinnest (0.07 ± 0.01 mm) ($P < 0.05$). With respect to the number, depth, and area of the islands, case 1 presented the highest values, followed by case 2; the cartilage island was related to the fibrocartilaginous layer ($P < 0.05$). All cases had AgNOR proteins in the proliferative and fibrocartilaginous layers, as well as the islands with the greatest presence of chondrocytes ($P = 0.245$). A relationship was observed between the histopathological alterations in the different layers on the condylar surface and the thickness of the fibrocartilaginous layer, as well as the thickness of the latter and the number, depth, and area of the cartilage islands in the trabecular bone.

Key words: unilateral condylar hyperplasia; facial asymmetry; condylectomy.

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Unilateral condylar hyperplasia (UCH) of the temporomandibular joint (TMJ) is a disease of low prevalence that can cause significant facial changes associated with facial asymmetry. The most important sequelae of this condition are deviation of the mandible towards the non-hyperplastic side, the presence of a unilateral posterior crossbite, and complications related to normal functioning of the oral cavity[1,2].

There are clinical classifications for UCH-related facial asymmetry and the options for treatment[2,3]; however, consensus has not yet been reached on the histological classification. Slootweg and Müller presented one of the first histological classifications[4], but in recent years Saridin et al. have shown the limitations

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of that initial classification[5]. Vásquez et al. broadened the discussion, reporting on the divergences in the histological results and the clinical behaviour of the disease[6].

In some previous studies, the histological description of hyperplastic and non-hyperplastic condyles was not able to establish a clear difference[7], whereas in other studies, greater connective tissue has been observed in hyperplastic condyles compared to healthy condyles[8]. In addition, the presence of cartilage islands at different levels of the trabecular bone in the condylar head may be one of the important variables indicating the growth and aggressiveness of the disease[6,9], although there appear to have been no specific studies on this issue.

The aim of the present study was to describe UCH histomorphometrically in a series of cases, characterizing the presence of cartilage islands as a new method to evaluate the extension of this disease.

Materials and methods

A study was conducted on a series of cases assessed in the Division of Oral and Maxillofacial Surgery and the Centre for Morphological and Surgical Studies (CEMYQ) at the Universidad de La Frontera. All patients agreed to participate in the study. The study was performed in compliance with the Declaration of Helsinki and was approved by the Ethics Committee of the Universidad de La Frontera.

Diagnosis and surgical procedure

Five individuals diagnosed with UCH, aged between 15 and 18 years, were included; three were female and two were male. The methodology of diagnosis and treatment has been reported previously and followed the sequence described by Olate et al.[10], initially using clinical studies of facial and dental analyses to identify the asymmetry, including a chin deviation greater than 5 mm from the midline and the presence of a unilateral crossbite. All subjects were classified clinically as 1 B according to Wolford et al.[2]: unilateral accelerated asymmetric growth and deviated mandibular prognathism with an ipsilateral class III occlusion.

The imaging study utilized cone beam computed tomography (PlanMeca, Helsinki, Finland, Piezotome 2), with a photo capture protocol performed in centric occlusion. The analysis was performed using Ez3D Viewer Plus software (Vatech Co., Hwaseong-si, Gyeonggi-do, 445-170, Korea); differences in condylar size and shape were identified in this analysis. Finally, single photon emission computed tomography (SPECT) was conducted, as described by Saridin et al.[11].

Once active UCH was confirmed, the next step was a proportional condylectomy[12]. This was done through a partial resection of the mandibular condylar head using an ultrasonic system (Piezotome 2; Satelec Acteon, Acteon Group, Paris, France Leica Microsystems, Heerbrugg, Switzerland) Histosec; following the technique described by Olate et al.[13]. A short endaural approach to the TMJ and minor retraction were used, and a sequence of two tips (10 mm and 18 mm long) from the piezoelectric system was used for the condylar osteotomy.

Histomorphometric study

The hyperplastic condyles were fixed in 10% buffered formalin for 48 h, decalcified in 10% ethylenediaminetetraacetic acid (EDTA) buffer solution for a period of 2 months, dehydrated in a series of ascending alcohol solutions, and embedded in paraffin (Histosec; Merck Millipore, Darmstadt, Germany). Sections 3 μm thick were obtained using a Microm HM 325 Microtome, Microm International, Walldorf, Germany) and these were then stained with haematoxylin and eosin (H&E) and stained to observe argyrophilic proteins in the nucleolar organizer region (AgNOR), as described by Ploton et al.[14]. The slides were analyzed under a Leica DM 750 microscope and photographed with a Leica ICC50 HD camera (Leica Microsystems, Heerbrugg, Switzerland, Histosec).

The presence, extension, and thickness of the tissue layers comprising the condylar surface were analyzed according to the description of Wurgaft and Montenegro[15] (Fig. 1). The thickness of the joint, proliferative, and fibrocartilaginous layers, as well as their total thickness, were analyzed on five histological slides per sample at 100 × magnification (H&E). The number, depth, and area of the cartilage islands were determined in the middle zone of each mandibular condyle on five slides at 400 × magnification (H&E). The middle area of the condyle was obtained after the coronal measurement of the condyle, obtaining the middle point in the upper and lower areas; then, a line for cutting the condyle was made following the microtome technique. The images were analyzed using Image J2 software (National Institutes of Health, Bethesda, MD, USA).

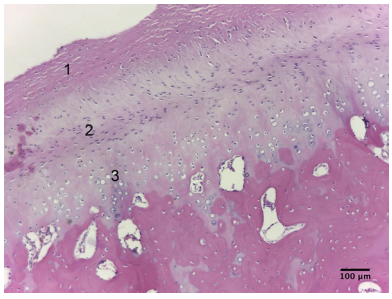

Fig. 1. Joint layer (1), proliferative layer (2), and fibrocartilaginous layer (3) in a hyperplastic condyle. Cartilage islands dispersed in the trabecular bone. H&E staining.

The average number of nucleolar organizer regions, visualized as black or dark brown points at the nuclear level, was quantified using conventional light microscopy, evaluating 100 cells in each condyle at 1000 × magnification (AgNOR)[16].

Statistical analysis

The statistical analysis was performed using IBM SPSS Statistics version 21.0 software (IBM Corp., Armonk, NY, USA) and the assumptions were verified with the Shapiro–Wilk test (data normality test) and Levene's test (homoscedasticity analysis). For the analysis of the differences between groups, one-way analysis of variance (ANOVA) was used, as well as Tukey's post hoc honestly significant difference (HSD) test and Dunnett's T3 test (as appropriate). For each condyle, five slices were included in this analysis, considering differences in layer size and cartilage island count and position, and observing differences in the histological conditions in each slide. *P*-values were considered significant at $P < 0.05$ and very significant at $P < 0.025$, when comparing the five condyles.

Results

The clinical characteristics of the study patients are given in Table 1. The histological analysis revealed different types of condylar architecture. In the areas of greatest pressure (superior and anterior), all condyles presented defined joint, proliferative, and fibrocartilaginous layers. However, their extension and thickness varied across the condylar surface.

The mean total thickness of the layers in the condyles affected by UCH was 0.30 ± 0.03 mm, with no significant differences among the cases ($P = 0.106$). The thickest layer was the fibrocartilage layer (0.13 ± 0.05 mm) and the thinnest layer was the joint layer (0.07 ± 0.04 mm). There were significant differences in the

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