A radiologic-pathologic study of the histopathologic variants of ameloblastomas and their proliferation indices

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Objectives. This study aimed to analyze the clinicoradiologic features and Ki-67 proliferation indices between the histopathologic variants of ameloblastomas (ABs) for possible associations.

Study design. The diagnosis and histopathologic variant were confirmed for all cases by experienced Oral and Maxillofacial Pathologists. Immunohistochemistry for Ki-67 was performed on the most representative formalin-fixed paraffin-embedded tissue block. Demographic, clinical data and radiologic features were analyzed from patient records and available radiographic examinations. The investigators were blinded to the histopathologic variant and proliferation index when the clinicoradiologic features were assessed.

Results. The current study included 116 cases of AB in the final sample. The indolent behavior of the unicystic variant was supported by their low proliferation index and slow growth paired with low frequencies of cortical destruction, loss of teeth, root resorption, and encroachment on anatomical structures. In contrast, the comparatively high proliferation index of the plexiform variant correlated with their fast growth and pain. Furthermore, high radiologic frequencies of cortical destruction, loss of teeth, and encroachment of surrounding anatomical structures supported their more aggressive clinical course.

Conclusion. Statistically significant differences were noted between certain variants and Ki-67, location, borders, locularity, and cortical destruction, providing better insight into their biological behavior. (Oral Surg Oral Med Oral Pathol Oral Radiol 2024;138:403−413)

Ameloblastoma (AB) is a benign epithelial odontogenic tumor, originally termed adamantinoma, with Ivy and Churchill coining the term ameloblastoma in 1930.[1,2] In 1970, Vickers and Gorlin established criteria outlining the cellular features associated with the histopathologic diagnosis of AB.[3] These included ameloblast-like columnar cells with palisading, hyperchromatic nuclei exhibiting reverse polarization away from the basement membrane with subnuclear vacuolization. ABs are classified into conventional, unicystic, extra-osseous/peripheral, and metastasizing variants.[4,5]

Intra-osseous ABs are broadly divided into unicystic and conventional subtypes.[4] Unicystic ABs are subdivided into 3 histopathologic variants, where the neoplastic cells remain confined to the wall/lining of the cystic cavity (luminal variant) or proliferate into the cystic lumen (intraluminal variant). The third type, or mural variant, is the most prevalent and is characterized by neoplastic epithelial islands within the wall of the cystic neoplasm.[6] It was proposed that the mural variant be better classified under the conventional AB category due to reports of similar biological behavior and treatment approaches.[7] However, significant differences have been found between conventional and mural unicystic ABs concerning age, sex, radiologic locularity, borders, and the association with an impacted tooth.[7] Therefore, the mural variant was still retained under the unicystic ABs category in the latest 5th edition of the World Health Organisation (WHO) Classification of Head and Neck Tumors.[5]

Conventional ABs have different histopathologic variants based on their overall morphology. These include follicular (Fo), plexiform (Pl), acanthomatous

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Statement of Clinical Relevance

Significant differences were noted between the histopathologic variants of ameloblastomas regarding their proliferation indices, clinical presentation and radiologic features. Indolent versus aggressive radiologic signs correlated with their proliferation indices, better elucidating their biological behavior and aiding patient management.

(AA), granular cell (Gr), basaloid/basal cell (Ba), and desmoplastic (De).[4] The distribution and prevalence of these variants differ among different population groups.[8] Exceedingly rare variants, including keratoameloblastoma (KA) and papilliferous KA, have also been described in the literature.[9] The Fo variant presents histopathologically with islands of ameloblast-like cells surrounding a core resembling the stellate reticulum.[4] The Pl variant is characterized by a network of interconnecting strands of neoplastic epithelium.[4] In the AA variant, the stellate reticulum-like areas undergo squamous metaplasia with varying degrees of keratin pearl formation. The Gr variant is characterized by groups of central granular cells containing eosinophilic granules resembling lysosomes that typically replace the stellate reticulum-like area. The Ba variant presents as densely packed nests and strands of basaloid cells with minimal to absent stellate reticulum-like zones. The De variant has a dense hyalinized and hypocellular stroma that compresses the neoplastic epithelium into thin strands.[4] Keratoameloblastoma and papilliferous KA are rare variants of ABs that exhibit excessive keratinization in unique arrangements.[9]

Radiologic analysis of lesions affecting the maxillofacial region gives insight into their extent and biological behavior.[10] Hunter and Niklander emphasized the importance of a thorough radiologic evaluation in assisting the diagnostic process.[11] Moreover, it is a critical step in managing patients as it assists the surgeon with surgical planning and appropriate resection margin determination. Several key radiologic features allude to the biological behavior of a lesion.[10] Well-demarcated margins, bony expansion, and tooth displacement often point to the benign behavior of a lesion. In contrast, poorly demarcated margins with cortical destruction are features better suited to aggressive growth.[10,12]

The proliferation index of a tumor simply implies the percentage of cells in a tumor undergoing cell division. This gives insight into a tumor's growth rate, aggressiveness, and biological behavior. Ki-67 is a reliable marker in measuring proliferation indices, especially in odontogenic tumors.[13] Studies have found a significant association between Ki-67 immunohistochemical expression and the recurrence rates of ameloblastomas,[14,15] with higher expression associated with a higher risk of recurrence.[14-16] The mean recurrence-free survival has been reported at 8 years in tumors with a Ki-67 index below 10%, decreasing to 5 years in tumors with an index above 10%.[15]

Currently, all histopathologic variants of conventional ABs are managed similarly, as no conclusive evidence supports differences in biological behavior. This study aims to analyze the histopathologic variants of ABs and assess the radiologic features and Ki-67 proliferation indices for possible associations or differences. This will provide better insight into their biological behavior and clinical presentation.

MATERIALS AND METHODS

The study was conducted following approval by the University of Pretoria, Faculty of Health Sciences Research Ethics Committee (Reference number: 723/2022). All procedures followed the ethical standards of the Helsinki Declaration of 1975, as revised in 2008.

Confirmed cases of ABs were retrospectively reviewed from the histopathologic archives of the Department of Oral and Maxillofacial Pathology, University of Pretoria. The first author (CS) and 2 experienced Oral and Maxillofacial pathologists (LR and WvH) confirmed the diagnosis of all included cases from available histopathologic slides. The histopathologic variant was classified based on the predominant pattern visible in representative tumor sections from all available specimens (biopsy and resection, when available) (Figure 1). The variants were classified based on the latest WHO classification of head and neck tumors.[5] Cases where insufficient tissue was available to confirm the histopathologic variant were excluded. Additionally, cases of ABs that transformed into ameloblastic carcinomas and adenoid ABs were excluded from this study. Adenoid AB cases were excluded from this study as this entity has not been fully elucidated. The overlapping features with other odontogenic tumors and lack of *BRAF* mutations, frequently present in unicystic and conventional ABs, further support the exclusion at this stage. Further research on this entity is required before considering it a variant of AB.[5] Only cases with available radiographic examinations were included.

Immunohistochemical analysis for Ki-67 was performed on freshly cut 3 μm sections from the representative formalin-fixed paraffin-embedded (FFPE) tissue block for each included case. The most representative specimen (biopsy or resection) per case was used for immunohistochemical staining. In cases where the resection specimen was decalcified, the largest biopsy specimen was used to ensure accuracy. Staining was performed on a Ventana Benchmark GX Automated System (Ventana Medical Systems Inc.). Heat-induced epitope retrieval was performed with Ventana high pH CC1 retrieval solution, followed by incubation with ready-to-use Ki-67 (MIB1) monoclonal mouse anti-human antibody (Dako, Agilent Technologies) according to the manufacturer's recommendations. Detection was performed using the Ventana OptiView DAB Detection kit according to the manufacturer's instructions. Following which sections were counterstained with hematoxylin and mounted with a permanent mounting media. The first author (CS) evaluated and

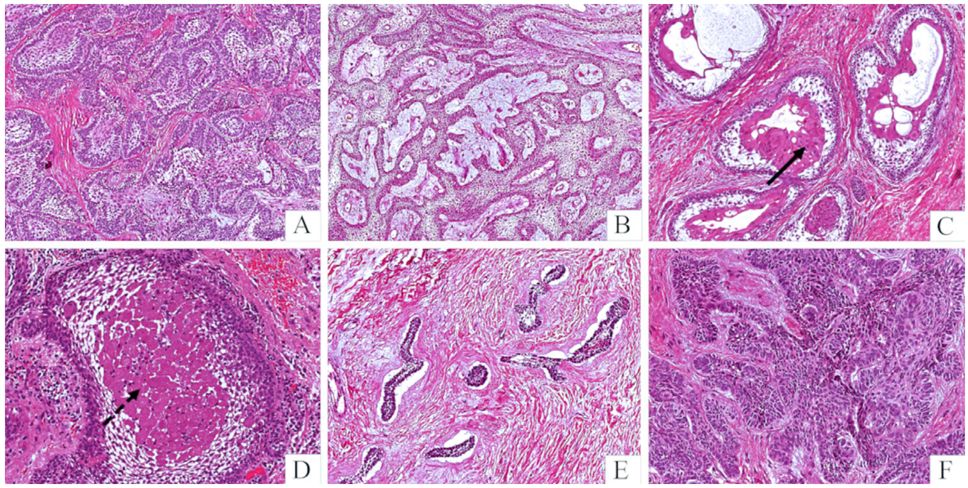

Fig. 1. Examples of the histopathologic variants of conventional ABs. (A) Follicular, (B) Plexiform, (C) Acanthomatous with extensive squamous differentiation (solid arrow), (D) Granular with central eosinophilic granular cells (interrupted arrow), (E) Desmoplastic and (F) Basaloid/basal cell.

scored the proliferation index under the guidance of 2 experienced Oral and Maxillofacial pathologists (LR and WvH). The most stained areas (hot spots) in each section were evaluated and expressed as a percentage of cells counted. A graticule was used to prevent the double-counting of positive cells. A minimum of 100 cells was counted on each representative section of the tumor. Appropriate positive controls were included in all cases (Figure 2).

Demographic and clinical data were retrieved from the patient's records. The clinical signs and duration of the lesion were recorded as reported by the patient. Conventional and digital radiographs, including panoramic (PR) and skull radiographs, and specialized imaging, consisting of computerized tomography (CT) and cone-beam CT (CBCT), were utilized for radiologic evaluation. The investigators were blinded to the histopathologic variant and proliferation index when the radiographs were assessed. Radiologic information, including location, borders, radiodensity, locularity, and effects on the surrounding structures, were analyzed from the available radiographic examinations. Lesions located from the incisor to the canine region were classified as anterior, whereas lesions located posterior to the canine region were classified as posterior. The radiologic features were analyzed by the 2 authors (CS and AU), who have experience in the field of Maxillofacial Radiology, with any disagreements resolved by consensus and/or consultation with a third author (WvH).

The results of the radiologic examinations were recorded using Microsoft Excel (Version 2016), with subsequent statistical analysis of the categorical data performed using Stata 18.0 SE (StataCorp LLC). The distribution of data was confirmed using the Shapiro-Wilk test. Descriptive statistics were reported using median and interquartile ranges. Categorical data was analyzed using the Chi-square test along with Fisher's Exact test as a reasonability check when the underlying assumptions of Chi-squared were violated. Dunn's test using rank sums with Bonferroni correction was used for multiple comparisons. Asymptotic Significance

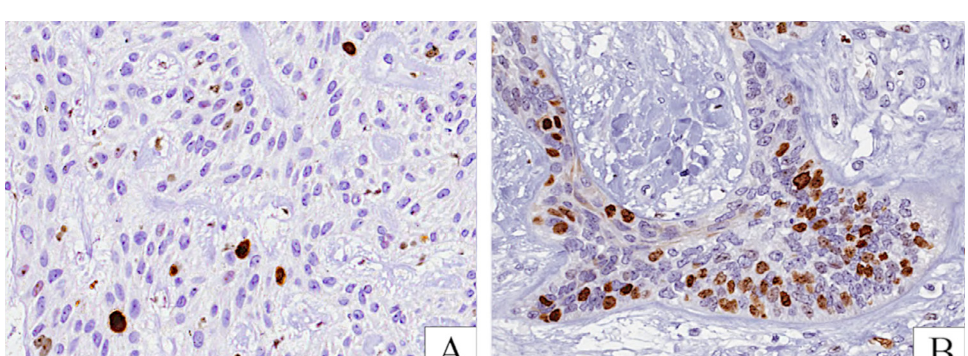

Fig. 2. Examples of (A) low (~2%) and (B) high (~20%) Ki-67 proliferation indices.

(*P*-value) of less than .05 were considered statistically significant.

RESULTS

A total of 156 cases of ABs with available radiographs were collected during the study period. Forty cases were excluded due to insufficient tissue. In these cases, the available tissue was adequate to reaffirm the diagnosis of AB, however, the limited tissue precluded accurate subtyping of the histopathologic variant and were therefore excluded from this study. Therefore, 116 cases were included in the study as the final sample, from which 116 biopsy samples and 64 resection samples were available for analysis. The radiologic analysis was primarily done on panoramic radiographs, with 18 cases having additional CBCT/CT imaging available. The AA variant made up the majority of the sample, followed by Fo, Pl, Gr, and unicystic variants. The other variants, including KA, De, and Ba, were rare and only seen in 3, 2, and 1 cases, respectively. Due to the overall low prevalence of these variants in the current study, their results should be interpreted with caution.

The demographic and clinical features of included cases are summarized in Table I. The Fo and Pl growth patterns presented at a similar median age to the overall sample. Unicystic ABs presented at a younger median age, and the remaining variants presented at a significantly later age than the overall median (Figure 3). When comparing unicystic to conventional ABs, a statistically significant younger age was noted (Supplementary Table S1). A near-equal male-to-female distribution was noted, with a slight male predominance in AA and unicystic cases. In the current sample, ABs had an overall long reported duration before treatment was sought (median of 12 months). The duration of AA and Fo cases was longer than the overall average, with unicystic and Pl variants having shorter overall durations (Figure 4). The mean Ki-67 proliferation index of all included ABs was 9.4% (median = 5%), with higher values seen in the Pl variant (10%). In contrast, the unicystic variant had the lowest proliferation values (2%) (Figure 5). Dunn's pairwise comparison showed that a statistically significant difference existed among the Ki-67 values between unicystic and Pl ($P = .0191$) and unicystic and KA ($P = .0080$) variants. Furthermore, a statistically significant difference in Ki-67 values were noted between unicystic and conventional ABs.

Nearly all cases presented as painless swellings, with associated pain reported in less than one-third of cases. The reported pain was lowest in the unicystic variant and highest in the Pl variant. Most lesions were reported as slow-growing. In rare instances, associated bleeding, fistula formation (2 cases), and trismus (1 case) were reported. There was no statistically significant difference in clinical signs between the histopathologic variants of AB.

The location of included cases is summarised in Table II. In this study, maxillary lesions were significantly rarer (5 cases, 4.3%). All maxillary cases involved the posterior region, and all but one extended to the anterior region. No maxillary lesions crossed the midline. All variants of AB had a posterior mandibular predilection. Statistically significant differences existed between the variants and their location. The AA variant had higher frequencies of extension to involve the

Table I. Summarized demographic and clinical features of ABs

	Uni *n = 12* *(10.3%)*	*Fo* *n = 31* *(26.7%)*	*Pl* *n = 21* *(18.1%)*	*AA* *n = 33* *(28.4%)*	*Gr* *n = 13* *(11.2%)*	*KA* *n = 3* *(2.6%)*	*De* *n = 2* *(1.7%)*	*Ba* *n = 1* *(0.9%)*	P-*Value*	*Total* *n = 116*
Age (years), Median	16.86	28.05	26.93	35.45	38.00	45.23	31.18	42.93	.090	28.36[1]
Sex (M:F)	7:5 (1.4:1)	14:17 (1:1.2)	10:11 (1:1.1)	19:14 (1.4:1)	7:6 (1.2:1)	2:1	2:0	0:1		61:55 (1.1:1)
Duration (months), Median	4.5	18.00	7.00	18.00	24.00	12.5	24.00	6.00	.278	12.00[2]
Proliferation index %, Median	2%	5%	10%	5%	5%	35%	3.5%	15%	**.006**	5%[3]
Clinical signs	**10**	**28**	**18**	**30**	**13**	**2**	**1**	**1**		**103**
Swelling	10 (100%)	26 (92.9%)	17 (94.4%)	30 (100%)	13 (100%)	2 (100%)	1 (100%)	-	.092	99 (96.1%)
Pain	1 (10%)	7 (25%)	6 (33.3%)	9 (30%)	4 (30.8%)	2 (100%)	-	1 (100%)	.322	30 (29.1%)
Slow growing	3 (30%)	7 (25%)	5 (27.8%)	10 (33.3%)	4 (30.8%)	-	1 (100%)	-	.931	30 (29.1%)
Fast growing	-	1 (3.6%)	1 (5.6%)	1 (3.3%)	-	-	-	-	1.000	3 (2.9%)
Mobile teeth	-	5 (17.9%)	2 (11.1%)	2 (6.7%)	3 (23.1%)	-	-	-	.522	12 (11.7%)
Paraesthesia	-	2 (7.1%)	2 (11.1%)	-	-	-	-	-	.457	4 (3.9%)
Ulcer	-	4 (14.3%)	1 (5.6%)	1 (3.3%)	1 (7.7%)	-	-	-	.684	7 (6.8%)
Pus	-	2 (7.1%)	-	1 (3.3%)	-	1 (50%)	-	-	.276	4 (3.9%)

Uni, Unicystic; *Fo*, Follicular; *Pl*, Plexiform; *AA*, Acanthomatous; *Gr*, Granular cell; *KA*, Keratoameloblastoma; *De*, Desmoplastic; *Ba*, Basal cell/Basaloid.
Data available for [1]114 patients, [2]82 and [3]113 patients, respectively.

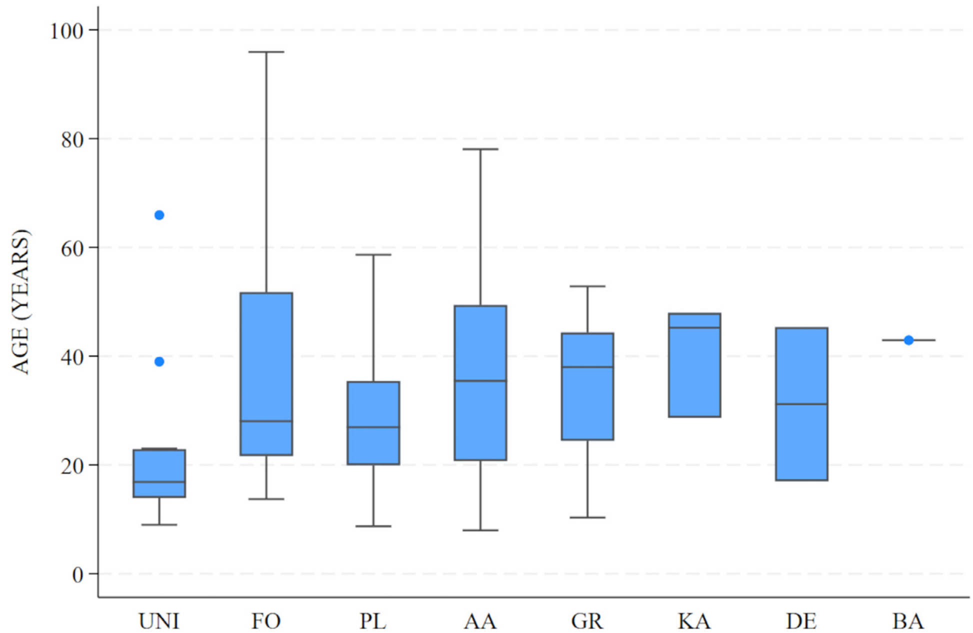

Fig. 3. Box and whisker plot for the age of ABs at presentation over variants.

anterior regions and cases crossing the midline. Ramus involvement was statistically more common in the Pl variant. Unicystic cases rarely extended to involve the ramus, coronoid, or condylar complex. When analyzing the conventional variants excluding unicystic cases, De cases had a statistically significant affinity for the condyle (P = .037). No significant differences were noted between unicystic and conventional ABs (Supplementary Table S2).

The radiologic features of included cases are summarized in Table III. Radiologically, ABs presented as well-demarcated multilocular radiolucent lesions in the majority of cases. The borders frequently exhibited focal loss of demarcation. There was a statistically significant difference between border demarcation and the histopathologic variant of AB. Unicystic and Gr cases were commonly well-demarcated, with Fo cases often exhibiting a loss of demarcation. Internal calcifications

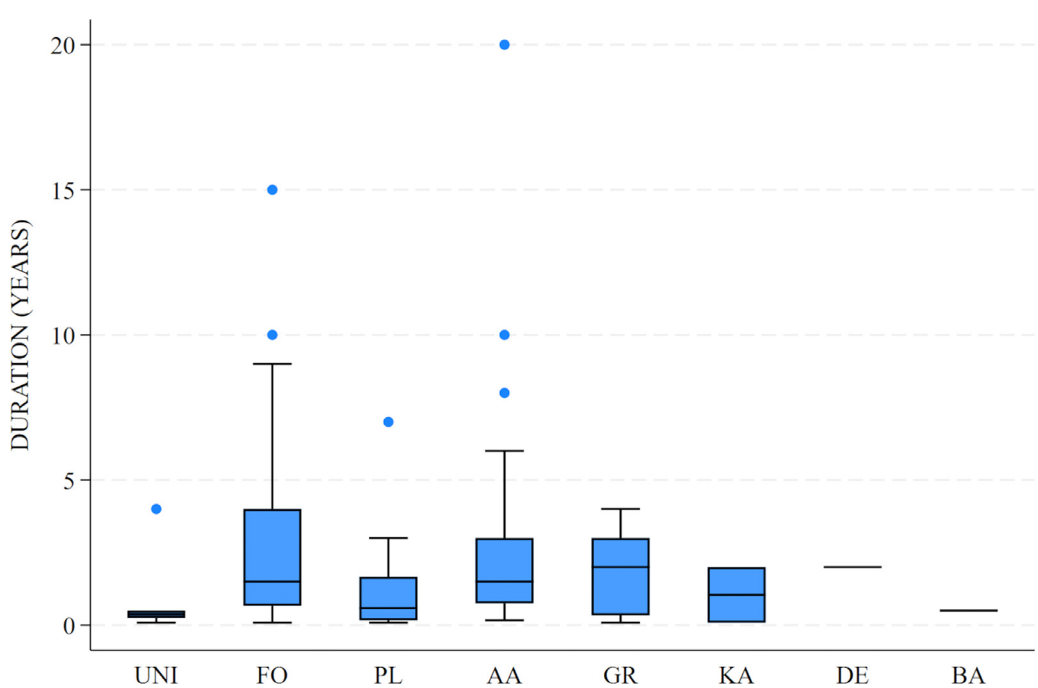

Fig. 4. Box and whisker plot for the duration before presentation over variants.

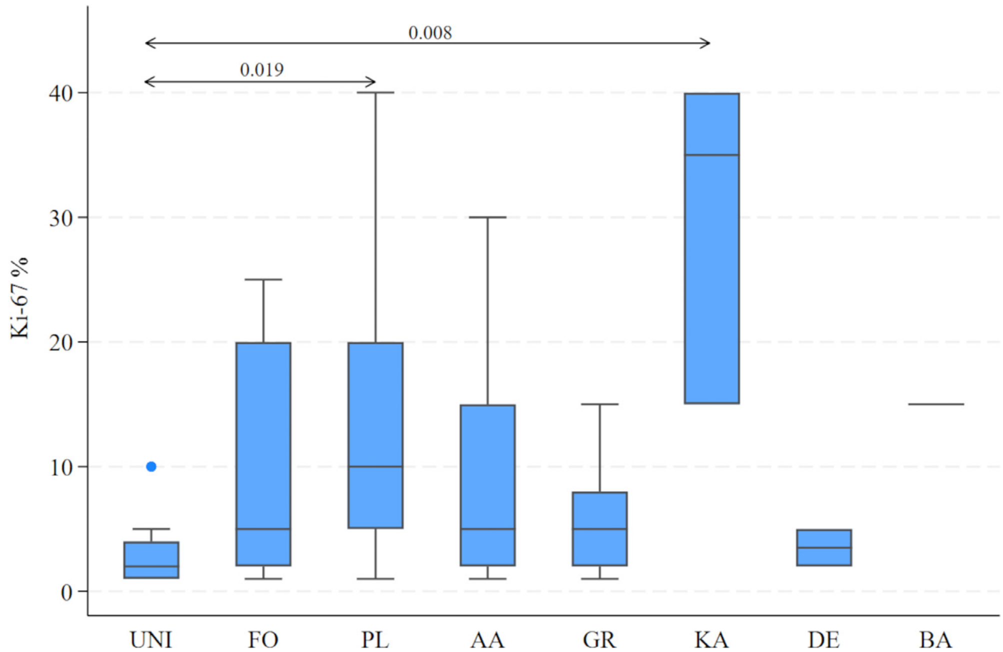

Fig. 5. Box and whisker plot for the Ki-67 proliferation index over variants.

were more common in the KA and Gr variants (Figure 6). There was a statistically significant difference between the locularity and the histopathologic variants. Multilocular lesions were evenly distributed between the conventional variants, with lower frequencies in Pl cases and unseen in unicystic cases. There were low frequencies of unilocular presentations in the Gr and AA variants. Bony expansion was commonly seen in the Gr and Fo variants, with all Gr cases exhibiting cortical thinning. Interestingly, bony expansion and cortical thinning were prominent in the unicystic type, but associated cortical destruction was the lowest of all the variants. Cortical destruction was most common in PL cases, which was statistically significant (Figure 7). In contrast, cortical bone destruction was relatively uncommon in AA cases. Comparing unicystic to all conventional ABs, cortical destruction was statistically significantly higher in conventional ABs (Supplementary Table S3). The De variant showed the highest incidence of associated reactive bony changes, a feature not seen in any of the Gr cases. When the conventional cases of AB were analyzed without unicystic cases, reactive bony changes were statistically significant (P = .037). Loss of teeth associated with the lesion was most common in Pl and Gr cases, and lowest in unicystic cases. In contrast, tooth impaction was most commonly seen in unicystic cases. Commonly impacted teeth in Pl and Fo cases were third molars (100% and 71%, respectively), canines (60%) in AA cases, second molars (57%), and canines (29%) in unicystic cases. In the current study, unicystic cases presented at a mean age of 31.32 years without

Table II. Summarized location of ABs

Location	*Uni n = 12 (10.3%)*	*Fo n = 31 (26.7%)[1]*	*Pl n = 21 (18.1%)*	*AA n = 33 (28.4%)*	*Gr n = 13 (11.2%)[2]*	*KA n = 3 (2.6%)[2]*	*De n = 2 (1.7%)*	*Ba n = 1 (0.9%)*	P-*Value*	*Total n = 116*
Anterior	5 (41.7%)	16 (51.6%)	3 (14.3%)	24 (72.7%)	8 (61.5%)	2 (66.7%)	-	-	**.001**	58 (50%)
Posterior	11 (91.7%)	30 (96.8%)	21 (100%)	33 (100%)	13 (100%)	3 (100%)	2 (100%)	1 (100%)	.312	114 (98.3%)
Both anterior and posterior involvement	4 (33.3%)	15 (48.4%)	3 (14.3%)	24 (72.7%)	8 (61.5%)	2 (66.7%)	-	-	**.000**	56 (48.3%)
Ramus	4 (33.3%)	15 (53.6%)	17 (81%)	12 (36.4%)	5 (41.7%)	-	2 (100%)	1 (100%)	**.005**	56 (50.5%)
Coronoid	2 (16.7%)	9 (32.1%)	8 (38.1%)	8 (24.2%)	3 (25%)	-	2 (100%)	1 (100%)	.202	33 (29.7%)
Condyle	1 (8.3%)	5 (17.9%)	1 (4.8%)	3 (9.1%)	-	-	2 (100%)	-	.055	12 (10.8%)
Crosses midline	5 (41.7%)	7 (25%)	2 (9.5%)	19 (57.6%)	5 (41.7%)	1 (50%)	-	-	**.006**	39 (35.1%)

Uni, Unicystic; *Fo*, Follicular; *Pl*, Plexiform; *AA*, Acanthomatous; *Gr*, Granular cell; *KA*, Keratoameloblastoma; *De*, Desmoplastic; *Ba*, Basal cell/Basaloid.
[1]Three and [2]one maxillary cases, respectively.

Table III. Summarized radiologic features of ABs

Radiologic features	*Uni* *n = 12* *(10.3%)*	*Fo* *n = 31* *(26.7%)*	*Pl* *n = 21* *(18.1%)*	*AA* *n = 33* *(28.4%)*	*Gr* *n = 13* *(11.2%)*	*KA* *n = 3* *(2.6%)*	*De* *n = 2* *(1.7%)*	*Ba* *n = 1* *(0.9%)*	P-*Value*	*Total* *n = 116*
Borders										
Well-demarcated	11 (91.7%)	10 (32.3%)	12 (57.1%)	16 (48.5%)	11 (84.6%)	-	-	1 (100%)	**.001**	61 (52.6%)
Loss of demarcation	1 (8.3%)	19 (61.3%)	7 (33.3%)	17 (51.5%)	2 (15.4%)	3 (100%)	2 (100%)	-		51 (44.0%)
Poorly demarcated	-	2 (6.5%)	2 (9.5%)	-	-	-	-	-		4 (3.4%)
Radiodensity										
Osteolytic	11 (91.7%)	27 (87.1%)	18 (85.7%)	27 (81.8%)	9 (69.2%)	1 (33.3%)	1 (50.0%)	1 (100%)	.181	95 (81.9%)
Internal calcifications	1 (8.3%)	4 (12.9%)	2 (9.5%)	6 (18.2%)	4 (30.8%)	2 (66.7%)	1 (50.0%)	-		20 (17.2%)
Mixed	-	-	1 (4.8%)	-	-	-	-	-		1(0.9%)
Locularity[1]										
Multilocular	-	24 (77.4%)	13 (65%)	26 (78.8%)	10 (76.9%)	3 (100%)	2 (100%)	1 (100%)	**.000**	79/115 (68.7%)
Scalloped	4 (33.3%)	2 (6.5%)	1 (5%)	4 (12.1%)	3 (23.1%)	-	-	-		14/115 (12.2%)
Unilocular	8 (66.7%)	5 (16.1%)	6 (30%)	3 (9.1%)	-	-	-	-		22/115 (19.1%)
Effects										
Bony expansion	10 (83.3%)	28 (90.3%)	17 (81%)	26 (78.8%)	12 (92.3%)	2 (66.7%)	2 (100%)	1 (100%)	.738	98 (84.5%)
Cortical thinning	10 (83.3%)	22 (71%)	18 (85.7%)	26 (78.8%)	13 (100%)	1 (33.3%)	2 (100%)	1 (100%)	.168	93 (80.2%)
Cortical destruction	4 (33.3%)	20 (64.5%)	17 (81%)	16 (48.5%)	10 (76.9%)	2 (66.7%)	2 (100%)	-	**.037**	71 (61.2%)
Reactive bone	3 (25.0%)	8 (25.8%)	4 (19.1%)	11 (33.3%)	-	1 (33.3%)	2 (100%)	-	.066	29 (25.0%)
Loss of teeth	3 (25.0%)	14 (45.2%)	13 (61.9%)	16 (48.5%)	8 (61.5%)	2 (66.7%)	1 (50%)	1 (100%)	.458	58 (50.0%)
Tooth displacement	6 (50.0%)	19 (61.3%)	10 (47.6%)	21 (63.6%)	9 (69.2%)	1 (33.3%)	-	-	.430	66 (56.9%)
Tooth impaction	6 (50.0%)	6 (19.4%)	4 (19.1%)	5 (15.2%)	2 (15.4%)	1 (33.3%)	1(50%)	-	.248	25 (21.6%)
Root resorption	8 (66.7%)	22 (71%)	13 (61.9%)	27 (81.8%)	12 (92.3%)	1 (33.3%)	1 (50%)	-	.101	84 (72.4%)
Encroachment of anatomical structures										
Inferior alveolar nerve[2]	5 (41.7%)	16 (50%)	16 (76.2%)	19 (57.6%)	6 (50%)	1 (50%)	2 (100%)	1 (100%)	.302	66 (59.5%)
Maxillary sinus[3]	-	7/7[*4] (100%)	3/3[*3] (100%)	2/2[*2] (100%)	1/1 (100%)	1/1 (100%)	-	-	.285	14 (100%)
Nasal cavity[3]	-	2/3 (66.7%)	-	-	1/1 (100%)	1/1 (100%)	-	-	.116	4 (80.0%)
Zygoma	-	2/5[*2] (40%)	1/1[*1] (100%)	1/1[*1] (100%)	-	-	-	-	.952	4 (44.4%)
Orbit[3]	-	1/3(33.3%)	2/2[*2] (100%)	-	-	1/1 (100%)	-	-	.124	4 (57.1%)
Pterygoid plates[3]	-	3/4[*1] (75%)	1/1[*1] (100%)	-	-	-	-	-	.443	4 (57.1%)

Uni, Unicystic; *Fo*, Follicular; *Pl*, Plexiform; *AA*, Acanthomatous; *Gr*, Granular cell; *KA*, Keratoameloblastoma; *De*, Desmoplastic; *Ba*, Basal cell/Basaloid.
[1]In one-mixed density case, the locularity could not be described.
[2]Only mandibular cases analyzed .
[3]Only maxillary cases analyzed.
*Number of mandibular cases extending to involve this area.

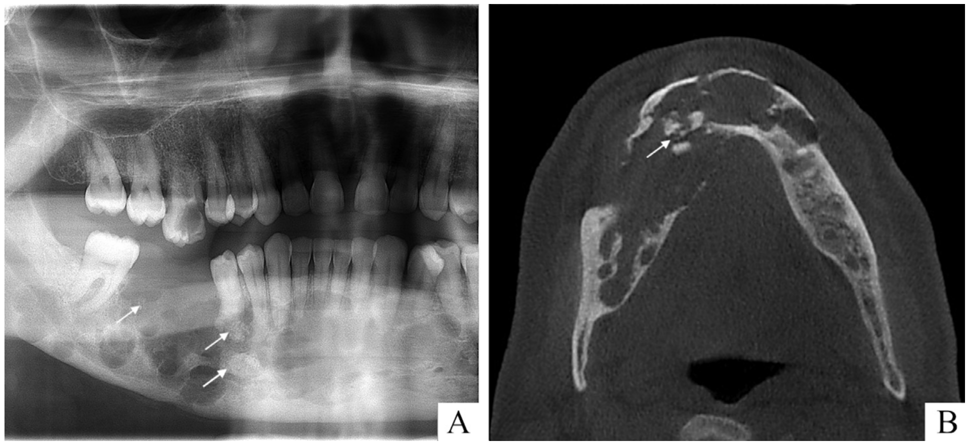

Fig. 6. Panoramic radiograph (A) and axial CBCT (B) images of a KA. Scattered clusters of internal calcifications were noted throughout the lesion (arrows).

an associated tooth impaction, and 13.92 years with an associated impacted tooth. Unicystic ABs had a statistically significant higher rate of tooth impaction compared to conventional cases. Root resorption was most common in Gr cases. Encroachment of surrounding anatomical structures was most common in the Pl variant.

DISCUSSION

Ameloblastomas are considered benign odontogenic neoplasms, despite their locally aggressive behavior.[13,14] Their radiologic signs and proliferation indices (8%-15%) often allude to their aggressive behavior.[13,17] However, there are limited studies analyzing the correlation between the radiologic features and proliferation indices between the different variants of ABs.

The Fo (28%) and Pl (21%) variants of conventional ABs are the most common, followed by the AA (7%), De (4%), Gr (2-4%), Ba (1%) and KA (0.1%).[8,9,18-22] The distribution of variants in the current study largely corresponded with the literature. The higher frequency of the Gr variant may be explained by the reported higher frequency of this variant in black patients.[23] Unicystic ABs were encountered less frequently (10.3%) compared to the reported literature (21%).[8] Unicystic ABs should be diagnosed retrospectively after removal of the entire lesion for histopathologic evaluation to confirm the unicystic nature and accurately subtype the tumor.[24,25] This may have resulted in several unicystic cases being excluded from the current study.

Unicystic ABs typically present at a younger age compared to the conventional subtype.[7,8,17,26] The Fo and Pl variants present at a similar mean age,[18] with the other conventional variants typically presenting later.[4,18,21,27,28] In the current study, however, most variants presented at a younger mean age than reported in literature, but followed a similar age distribution pattern among the variants. This could be explained by

Fig. 7. Axial (A), sagittal (B) and coronal (C) soft tissue window of computer tomography scan showing a Pl AB affecting the left mandible. This lesion shows severe cortical destruction and extends to involve the coronoid and condyle superiorly. Impingement/displacement of the maxillary sinus was also noted.

patients in developing countries presenting with ABs at a younger age.[18,29] In the current study, the long reported duration coupled with younger age at presentation further emphasizes the earlier development of ABs in the current population group.

Conventional ABs present with a shorter duration than the unicystic subtype, possibly due to their more aggressive clinical course.[26] This contrasts the current study, whereby higher than usual frequencies of swelling and bony expansion were reported in unicystic variants, explaining their relatively shorter reported duration prior to treatment.

Overall, ABs have a marked predilection for the posterior mandible.[17,27,30] AA cases have a slightly higher incidence in the anterior region of the jawbones.[18] The AA variant is often absent in the maxilla,[27] and the location findings were mirrored in the current study. Unicystic and Gr cases have a marked predilection for the mandible.[23,30] In contrast, the mandibular predilection of KAs is not as strong, a finding supported by the current cases.[9] The De variant presents with an anterior predilection, especially in the maxilla,[21,31,32] and frequently involves the maxillary sinus.[18] These findings were not seen in the current study, a result interpreted with caution due to their limited representation.

The loss of border demarcation seen in Fo and AA cases was not associated with high levels of cortical destruction in these variants, but rather high levels of reactive bone formation. In most AB variants, the borders are well-demarcated. An exception is the De variant, which often presents with poorly demarcated borders,[18,21,33] due to neoplastic cells infiltrating the marrow spaces, retaining the intervening trabecular bone.[21]

Dystrophic calcifications are more commonly seen in the AA histopathologic variant.[34] In the current study, calcifications were high in the Gr group, likely due to the granular cells being sensitive to hypoxia in the core of the tumor, resulting in dystrophic calcifications. Approximately one-third of KA cases present with mixed/ground-glass density or internal calcifications,[9] due to the extensive keratinization undergoing dystrophic calcification. Radiologically, the De variant often resembles a fibro-osseous or mixed-density lesion, typically containing small calcifications at the periphery[4,21,31] or metaplastic bone deposition within the tumor.[33,35] In the current sample, only 1 case had internal calcifications; however, reactive bony changes were seen in both De cases. Reactive bony changes were commonly seen as peripheral sclerosis in the bone surrounding the lesion or in the central bony septations in the lesion.

Unicystic ABs appear unilocular on radiologic assessment[17,18]; however, conventional ABs present with differing ratios of uni- or multilocularity.[36] In the current study, multilocular lesions predominated in all conventional subtypes, but unilocular lesions were comparatively high in Pl cases.

Unilocular lesions associated with the crown of an impacted tooth are statistically more likely to be unicystic ABs compared to the conventional subtype.[17] In this study, a correlation was observed between impacted teeth and the timing of lesion development. The mean age of unicystic ABs has been reported at 16 years with tooth impaction and 35 years in cases without impaction.[4] The most frequently impacted tooth in the AA variant involved canines, which could not be correlated with the later age of presentation. The longer duration reported in this variant in the current study may imply that the tumor begins earlier, with a slower growth pattern as noted clinically. The De variant has lower associations with impacted teeth than other variants of AB,[21] likely related to their later age of presentation.

When correlating the proliferation indices with clinical and radiologic features, unicystic ABs have a mean proliferation index of approximately 3%,[37] with other reports indicating proliferation indices above 15%.[13] In the current study, the indolent behavior of the unicystic variant was supported by their clinicoradiologic findings of slow growth, low frequencies of loss of border demarcation, cortical destruction, loss of teeth, root resorption and encroachment on anatomical structures, paired with having the lowest proliferation index of all variants. Cortical thinning was high in this subgroup, likely due to the pressure effect during bony expansion. These findings were noted when unicystic cases were compared to individual variants and conventional ABs as a group. The mean proliferation index for the Fo and Pl variants has been reported between 4% and 19%.[13,37] In the study by Ong'uti et al.,[38] the authors found higher proliferation indices in the Fo variant.[38] In the current study, the proliferation index of the Pl subgroup was comparatively high, correlating with the fast growth and pain coupled with a short duration and high radiologic frequencies of cortical destruction and loss of teeth. This variant also had the highest rate of involvement of the ramus complex and encroachment of surrounding anatomical structures. Desmoplastic cases have the lowest Ki-67 proliferation value (2%),[13] likely due to their high stroma-to-epithelium ratio. It was previously believed that the Gr variant behaved more aggressively.[39,40] This study partially supports this notion with high rates of pain, cortical thinning and destruction, loss of teeth, and root resorption seen in this variant. However, the low proliferation index mirrored the incidence of slow growth, high levels of well-demarcated borders, and tooth displacement. The high Ki-67 proliferation index in KA cases correlated with their short duration, clinical signs, and high rate of loss of border demarcation. More studies are needed on this variant to fully understand its biological behavior in comparison to the other variants of AB. The reported proliferation index for basal cell/basaloid ABs is 7%-19%.[13,37] The single case in the current study showed a proliferation index of 15%, marginally higher than the mean value.

This study is novel in that it includes a detailed radiologic analysis of the histopathologic variants of AB. Kim et al.[41] only analyzed locularity as part of the radiologic features of the variants.[41] Adeline et al.[29] found no statistically significant association between radiologic features and histopathologic subtypes of ABs. In contrast, Krishnapillai and Angadi evaluated the radiologic features of the histopathologic variants, albeit in less detail and with a significantly lower sample size (n = 73).[27] MacDonald-Jankowski et al. performed the only detailed radiologic evaluation of ABs to date.[17] The current study differs in that the radiologic investigation was not limited to distinguishing between unicystic and conventional variants, but also detailed the variants under conventional ABs.

In conclusion, the findings of the current study showed that not only is there a distinction between unicystic and conventional ABs in terms of behavior, but also between the different histopathologic variants of conventional ABs. The limitation of the current study includes the reliability of clinical signs, being dependent on the accuracy of clinical record keeping. Multicenter studies using this distinction, especially to obtain larger samples of the rarer histopathologic variants, are needed to substantiate these findings.

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ETHICS APPROVAL

This study was approved by the University of Pretoria, Faculty of Health Sciences Research Ethics Committee (Reference no.: 723/2022). All procedures followed the ethical standards of the Helsinki Declaration of 1975, as revised in 2008.

DECLARATION OF INTEREST

The authors declare that they have no conflict of interest.

CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

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SUPPLEMENTARY MATERIALS

Supplementary material associated with this article can be found in the online version at doi:10.1016/j.oooo.2024.03.007.

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