Frequency of Occurrence and Degree of Expression of the Parastyle in Several Modern Human Populations

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ABSTRACT The aims of this study are to describe the frequency of occurrence and degree of expression of the parastyle in six different ethnic groups; to assess inter- and intra-observer errors when scoring the feature; and to compare the expression of the feature in a small number of twin pairs. Dental casts were examined for evidence of the parastyle from samples available in the Adelaide Dental School. A dental plaque developed by Katich & Turner was used to standardize scoring. The highest percentage frequency of parastyle occurrence was found in a sample of European twins with a value of 1.7%. The buccal aspect of the mesiobuccal cusp of the permanent maxillary right second molar was the most common site for the parastyle. Inter-observer reliability in scoring was lower than intra-observer reliability. In 10 pairs of twins (seven pairs of monozygotic [MZ] twins and three pairs of dizygotic [DZ] twins) only two pairs of MZ twins showed concordance for presence of the parastyle. The expression of parastyles likely results from a complex interaction of genetic, epigenetic, and environmental influences during dental crown development. There may be a relationship between parastyles and supernumerary teeth that are occasionally located buccally to the maxillary molar teeth.

Since Louis Bolk (1916) first described an unusual projection on the buccal surfaces of maxillary molar crowns, researchers have had trouble explaining its etiology and giving the feature an appropriate name. The trait has been termed a maxillary paramolar cusp, paramolar tubercle, stylar anomalous cusp, supernumerary inclusion, and a parastyle. There are few more enigmatic features in the human dentition. Albert Dahlberg was aware that some system of identification needed to be established to standardise methodology and he devised his own descriptive method (Dahlberg, 1945). The importance of Dahlberg's terminology was that he considered the feature in terms of its location on the mesiobuccal cusp of the permanent maxillary second and third molars (that is, on the paracone or cusp 2) and, by applying his paleontological knowledge, he referred to it as a parastyle (Dahlberg, 1945). A similar feature was noted in a corresponding position on the protoconid of the lower molars and Dahlberg called it a protostylid.

Another difficulty encountered by researchers when studying parastyles has been how to describe their size objectively. These traits are difficult to assess with only limited guidelines (e.g., a single standardised plaster reference plaque) for comparison. Many previous references to parastyles have been case reports or linked to descriptions of the management of supernumerary teeth that have included photographs and radiographs as illustrations (Nagaveni et al., 2010; Parolia et al., 2011; Duddu et al., 2012; Nabeel et al., 2012; Jain et al., 2014; Shuangshuang et al., 2016).

A smaller number of more detailed studies have provided insights into the frequency of occurrence and variation in expression of the parastyle in different human populations. The frequency of occurrence of the parastyle has been estimated to vary from zero to 0.1% in first molars, 0.4 to 2.8% in second molars and 0 to 4.7% in third molars (Kustaloglu, 1962). These data were obtained from analyses of material representing recent *Homo sapiens* (Whites, American Blacks, Melanesians, Filipinos, Hawaiians, Middle Easterners [Kish], and Native Americans [Southwest Indians, Northwest Coast Indians, Peruvians]). Kustaloglu (1962) found that the parastyle was more common in Native Americans than the other population groups (2.6%).

A retrospective study of the parastyle in children

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aged two and eight years looked at factors relating to parastyle expression, including unilateral versus bilateral positioning in the dental arch, its frequency in both males and females, and its occurrence in primary and secondary dentitions (Nagaveni et al., 2017). In conclusion, the authors noted the parastyle was extremely rare in the primary dentition and that findings were inconclusive as to the other areas under review. Importantly, the difficulty in scoring the parastyle and the need to provide a more accurate method of measuring the feature were emphasized (Nagaveni et al., 2017).

The aims of this study are to describe the frequency of occurrence and degree of expression of the parastyle within and between six different ethnic groups; to assess inter- and intra-observer errors when scoring the feature; and to compare the expression of the feature in a small number of twin pairs. Our specific objectives are as follows: (1) make comparisons of the frequency of occurrence and degree of expression of the parastyle between primary and permanent dentitions of the same individuals (based on a study sample of twins); (2) compare expression on the first, second and third molars; and, (3) make comparisons between maxillary and mandibular molar teeth, as well as between right and left sides and between males and females. Another objective is to assess inter- and intra-observer errors when scoring the feature to determine the usefulness of the plaque developed by Katich and Turner when examining the parastyle. A final objective is to explore the possible roles of genetic, epigenetic and environmental influences on observed variation of the parastyle by comparing the expression of the feature in a small number of twin pairs. Finally, some thoughts about the etiology of the feature and its relationship to supernumerary molar teeth are provided, based on a threshold model of dental expression (Brook, 1984; Brook et al., 2014a, b).

Materials and Methods

Dental casts representing both sexes were examined for evidence of the parastyle from samples of six different ethnic groups available in the Murray Barrett Laboratory, Adelaide Dental School, The University of Adelaide, by a single trained observer (GS). The frequency of occurrence and degree of expression of the trait were calculated for each of the ethnic groups. Casts showing the presence of a parastyle were selected for further study.

The dental plaque developed by Katich & Turner in 1974 (Turner et al., 1991) was used to standardize scoring of the feature and to determine degrees of expression (Fig. 1). Reference was also made to written descriptions of the appearance of the parastyle, ranging from a small pit to a large cusp-like structure, provided by Turner et al. (1991). The reference plaque includes the following categories:

0 - The buccal surfaces of cusps 2 and 3 are smooth.
1 - A pit is present in or near the buccal groove between cusps 2 and 3.
2 - A small cusp with an attached apex is present.
3 - A medium-sized cusp with a free apex is present.
4 - A large cusp with a free apex is present.
5 - A very large cusp with a free apex is present. This form usually involves the buccal surface of both cusps 2 and 3.
6 - An effectively free peg-shaped crown attached to the root of the third molar is present. This rare condition is not shown on the plaque.

Whilst reference to the plaque was useful in providing a standard for scoring, it only showed five variations of parastyle expression and was therefore limited in the information it provided.

The method used in the production of the dental casts was uniform for all the groups studied. Alginate impressions were obtained of the subjects' dentitions

Figure 1. The dental plaque developed by Katich and Turner in 1974, used to standardize scoring of the parastyle and to delineate its degrees of expression.

and these impressions were poured using dental stone according to the manufacturer's specifications. After the dental stone had set, the impressions were removed from the casts and trimmed prior to examination.

The Australian Aboriginal dental casts were obtained from a longitudinal growth study of Central Australian Aboriginals conducted at Yuendumu Settlement in the Northern Territory of Australia, between the years 1951-1972 (Brown et al., 2009). The ages of the subjects ranged between 5 to 77 years, with most being teenagers. The sample totalled 405 subjects.

The same method of dental cast production was followed for the following ethnic groups: Malay Malaysians, 293; Chinese Malaysians 196; Indian Malaysians, 253; and Orang Asli, 71. These casts were collected as part of a PhD study that investigated dental variation in Malaysian schoolchildren with application to human identification (Khamis, 2005). The ages of the subjects ranged between 12 and 51 years, and the sample totalled 813 subjects.

Dental casts of twins of European ancestry and known zygosity, who are involved in an ongoing study of orofacial morphology and growth and oral health at the Adelaide Dental School, The University of Adelaide, were also included in this study (Hughes et al., 2014; Townsend et al., 2015). A total of 620 subjects from Cohort 1 were scored for parastyles with the casts of twin pairs sorted randomly and the operator blinded to zygosity. The twins were all of European ancestry and ranged in age from 6 to 63 years. The total number of dental casts examined in all of the samples was 1838.

Using the parastyle plaque of Katich and Turner, the question of intra- and inter-observer reliability was assessed with three experienced observers scoring the feature twice. The period between each observer score was at least two weeks enabling an assessment of concordance/discordance to be performed. It also gave the observers the ability to assess the associated criteria for determining different degrees of parastyle expression. Deliberately, the three operators who scored the parastyle, although being experienced dental anthropologists, had no training together prior to each scoring the feature independently, so that the value of the plaque as a means of standardising across different observers could be investigated.

Results

The frequencies of occurrence (prevalence) of the parastyle in the six different ethnic groups are presented in Table 1. Table 1 shows that the highest percentage frequency of parastyle occurrence was in the sample of European twins with a value of 1.6%. This was followed by the Australian Aboriginal sample with a frequency of 1.5%. The Malay Malaysians and Indian Malaysian groups showed fewer parastyles with only 0.3-0.4% of individuals displaying the trait, while no Chinese Malaysians were found to display the feature. The Orang Asli group result was 1.4% but the sample size was relatively small.

TABLE 1. Prevalence of parastyles in different ethnic groups

Ethnic group	n	present	%
Australian Aboriginal	405	6	1.5
Malay Malaysian	293	1	0.3
Chinese Malaysian	196	0	0.0
Indian Malaysian	253	1	0.4
Orang Asli	71	1	1.4
European Twins	620	10	1.6

The degrees of expression of the parastyle in the different study samples are shown in Table 2 and some examples of the expression of the feature are provided in Figure 2.

Only one individual (T234A) showed evidence of the parastyle in the primary dentition, with the feature being displayed on the primary maxillary right first molar. Across all ethnic groups, the buccal aspect of the mesiobuccal cusp of the permanent maxillary right second molar was the most common site for the parastyle, with 13 observations. Other teeth that showed the trait were the permanent maxillary right first molar (score: 1), permanent maxillary left second molar (score: 6), permanent maxillary right third molar (score: 1), and the deciduous maxillary right first molar (score: 1). Given that most of the subjects included in this study were children or young adults there were few cases where third molars could be scored.

Using the Katich and Turner plaque, degrees of expression of the parastyle ranging from scores of 1 to 6 were recorded. There were similar numbers of males and females displaying evidence of the parastyle (11 males and 8 females). Scores for parastyle expression that highlight intra- and inter-observer reliability are given in Table 3.

Within observers, there was no difference that was greater than one grade between the first and second scores. The difference in scoring between observers was never greater than two grades. In 17 cases, there were scoring differences noted between the three observers. This greater difference between observers' scores occurred seven times. Scorer 1 displayed four

TABLE 2. Frequency of occurrence and degree of expression of parastyles in different ethnic groups

Ethnic Group	Tooth affected	Degree	Sex
Australian Aboriginal			
Δ9	17	2	M
Δ17	17	5	M
Δ72	17, 18	3, 3	F
Δ119	17	2	M
Δ338	17	5	F
Δ578	17	2	M
Malay Malaysian			
MM 165	17, 27	2	M
Orang Asli			
OA 3	17	2	M
Indian Malaysian			
IM 105	17	2	F
European Twins			
T67B	17	3	M
T85B	17, 27	5, 5	M
T136B	27	5	M
T163B	17	2	M
T192B	27	4	F
T226A	27	4	F
T234A	54	3	F
T256A	16, 26	1	F
T262B	27	5	M
T304B	17	2	M

different scores from scorers 2 and 3, scorer 2 displayed four different scores from scorers 1 and 3, and scorer 3 displayed nine different scores from scorers I and 2.

With regard to the sample of twins of European ancestry, after scoring one member at random from each pair of twins, the co-twins of all those twins who displayed the parastyle were examined. Table 4 shows that in the 10 pairs examined (seven pairs of MZ twins and three pairs of DZ twins), only two pairs of MZ twins showed concordance for presence of the parastyle. In one of these pairs (T256), both members of the pair not only showed evidence of parastyles in their maxillary molars but also displayed protostylids on their permanent lower first molars.

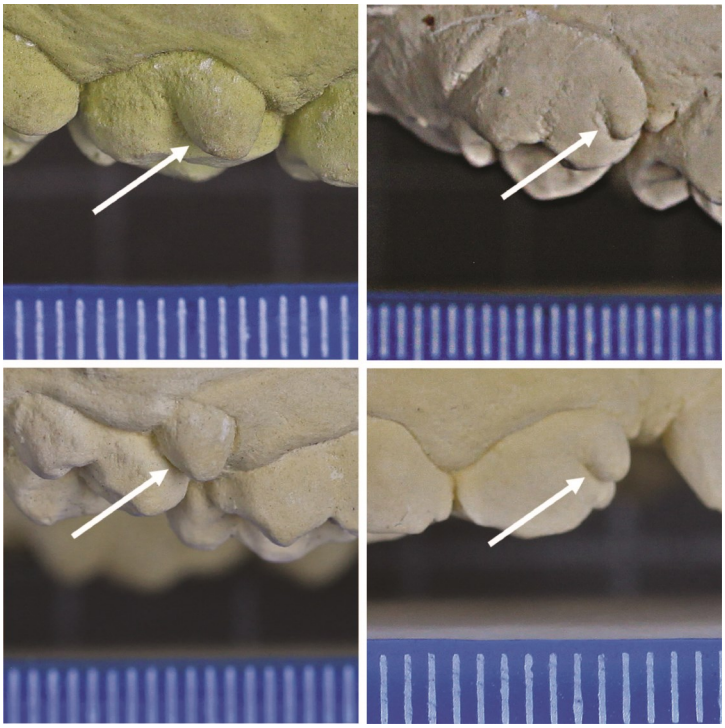

Figure 2. Some examples of the expression of the parastyle. Upper left - Australian Aboriginal male, Δ17, grade 5 on tooth 17; Upper right - Indian Malaysian female, IM 105, grade 2 on tooth 17; Lower left - Twin T85B male, grade 5 on tooth 17; Lower right - Twin T234A female, grade 3 on tooth 54.

Discussion

When comparing the prevalence of the parastyle between ethnic groups under investigation it is interesting to note that two distinct groupings were evident. The Malaysian groups showed fewer parastyles than either the Australian Aboriginals or the European twins, although the sample sizes of the Malaysians were smaller. It is acknowledged that the inclusion of both members of twin pairs could have increased the prevalence estimates in this group if there was evidence of concordance between MZ co-twins for parastyle occurrence. However, only two pairs of MZ twins showed concordance for the feature. The Malaysian groups conformed reasonably closely with the values for parastyle occurrence on permanent molars reported by Kustaloglu (1962) that ranged from 0.1% - 4.7%. Scott et al. (2018) provide a table of paramolar tubercle frequencies compiled from the C.G. Turner II database for over 9000 individuals distributed across 23 geographic groupings. The world average for all

TABLE 3. Intra- and inter-observer reliability for scoring parastyles

Ethnic Group	Cast ID	Location	Scorer 1		Scorer 2		Scorer 3	
			1st	2nd	1st	2nd	1st	2nd
Australian Aboriginal	Δ9	17-MB	2	2	2	2	2	2
	Δ17	17-MB	5	5	5	5	5	5
	Δ72	17-MB	3	3	3	3	2	1
		18-MB	3	3	3	3	5	5
	Δ119	17-MB	2	2	2	2	1	2
	Δ338	17-MB	5	5	5	5	5	5
	Δ578	17-MB	2	2	2	3	2	1
Malay Malaysian	MM 165	17-MB	2	2	2	2	2	2
		27-MB	2	2	3	3	3	3
Orang Asli	OA 3	17-MB	3	3	2	3	1	1
Indian Malaysian	IM 105	17-MB	2	2	2	2	2	2
European Twins	T67B	17-MB	3	3	4	5	3	3
	T85B	17-MB	6	6	5	5	5	5
		27-MB	6	6	5	5	5	5
	T136B	27-MB	4	5	5	5	5	4
	T163B	17-MB	2	2	3	4	2	2
	T192B	27-MB	3	3	4	4	4	4
	T226A	27-MB	4	4	4	4	4	4
	T234A	54-MB	3	4	3	3	2	2
	T256A	16-MB	1	1	1	1	2	2
	T262B	27-MB	4	4	5	5	4	5
	T304B	17-MB	2	2	2	2	1	1

groups was 1.58%. Relative to the groups in this study, Australians had incidence prevalence of 1.8%, Europeans 2.17% (Western) and 0.63% (Eastern), East Asians 1.6%, and Southeast Asians 2.27% and 1.75%, for early and recent groups respectively. These values are in general accord with our findings on recent populations. As the trait generally varies between one and three percent, further studies of large samples are needed to determine whether there are significant differences in parastyle frequency and degrees of expression between different human populations.

The finding that most of the parastyle observations occurred on the right side of the dentition suggests that there could be some directional asymmetry involved, although there is very little evidence to support consistent expression of directional asymmetry in any dental crown morphological features (Scott et al., 2018).

There did not appear to be any significant difference in parastyle frequency of occurrence between the sexes. Table 2 shows that a similar number of males and females displayed the feature, and that the size and shape of the feature did not appear to differ between males or females, so sexual dimorphism was not evident in our study. However, given the very small number of individuals who show parastyles, very large sample sizes are needed to detect systematic differences between the sexes.

It was evident in the scoring of the parastyle that difficulties arose in determining the size of the feature. We suggest a new plaque be developed that has the capacity to improve accuracy in scoring. The present plaque provides only one example of the degree of parastyle formation in the six categories mentioned.

The fact that the teeth used to construct the plaque vary widely in terms of their size and shape makes it difficult to score the relative size of the parastyle when

TABLE 4. Expression of parastyle in twin pairs

Twin ID	MZ/ DZ	Co-Twin ID	P/A	Concordant/ Discordant
T67B	DZ	T67A	A	D
T85B	MZ	T85A	A	D
T136B	MZ	T136A	A	D
T163B	MZ	T163A	A	D
T192B	DZ	T192A	A	D
T226A	DZ	T226B	A	D
T234A	MZ	T234B	P	C
T256A	MZ	T256B	P	C
T262B	MZ	T262A	A	D
T304B	MZ	T304A	A	D

P=present, A=absent

extrapolated to other teeth that are under investigation. We suggest providing more than one example of the categories of the parastyle to highlight the extent of the variation within a category. It would also be helpful to have stereoscopic images of these different variations in digital form that could be viewed from different perspectives using computer technology.

As shown in Table 4 there were seven MZ twin pairs and three DZ pairs examined, with only two pairs of MZ twins showing concordance for parastyle occurrence. Given that MZ twins share all of their genes, while DZ twin pairs, on average, share only 50% of their genes, one would expect a higher concordance of parastyle occurrence within MZ pairs if there was a strong genetic basis to the feature. The lack of concordance suggests that variation in parastyle occurrence and expression is likely to reflect mainly epigenetic and/or environmental influences.

The tendency for the strongest expression of the parastyle to have characteristics similar to supernumerary teeth that form buccally to the permanent molars (i.e., apparently with separate crown formation although possibly fused roots), suggests that the feature may conform to the upper end of the multifactorial unifying aetiological model of dental development (Brook, 1984; Brook et al., 2014a, b) that posits a relationship between tooth size, shape, and presence or absence. It is possible that the strongest expression of the parastyle falls just to one side of a threshold above which a supernumerary molar tooth is formed. Further studies of the associations between tooth size, parastyle expression and supernumerary tooth prevalence within individuals would help to clarify this issue. One of the authors (GRS) has noted structures similar to paramolar tubercles on the lingual surface of the maxillary molars and both buccal and lingual surfaces of the mandibular molars. Buccal manifestations on the lower molars are not protostylids, which are expressed in a constant position on the buccal surface of the protoconid and are less pronounced than lower molar 'pseudo-paramolar' tubercles. These diverse expressions may represent different developmental processes: some of the appearances are compatible with fusion of the molar tooth germ with a buccal supernumerary tooth germ, while others may be additional cusps arising from additional enamel knots.

The presence of both parastyles and protostylids in both members of one of the concordant MZ twin pairs provides some evidence for an association between these two features that may be based on underlying genetic influences. However, this was only one pair, so care is needed in considering the significance of this observation.

Conclusions

This study provides further insights into the frequency of occurrence and degree of expression of the parastyle in several human populations that have not been reported previously. One of the difficulties in scoring parastyles reliably lies in the nature of the system of scoring devised by Katich and Turner, and we have made some suggestions to improve the classification of this feature. It appears that the parastyle, with its varying degrees of expression, results from a complex interaction of genetic, epigenetic, and environmental influences during dental crown development. There may also be a relationship between parastyles and supernumerary teeth that are located buccally to the maxillary molar teeth.

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