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RESEARCH ARTICLE

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Does nutrition have an effect on the timing of tooth formation?

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Abstract

Background: The effect of nutritional status on the timing of permanent tooth formation is not well understood, despite clear evidence that systemic stresses result in enamel defects during tooth formation.

Aim: This study investigated the effect of nutritional status (measured as BMI, height, weight, mid-upper arm circumference, and head circumference) on permanent tooth formation.

Method: This was a prospective cross-sectional study involving 642 (270 males, 372 females) healthy Black South African participants aged 5–20 years. The height and BMI were converted to z-scores using WHO z-score for age tables. The participants were grouped into underweight/short for age (<−2), normal (≥ − 2 to 2.0) and overweight/obese/tall for age (>2). Panoramic radiographs of the children were assessed using Demirjian's eight stages of permanent tooth formation and age was estimated using Demirjian's method. Probit regression analysis was used to calculate the mean age of attainment of the developmental stages for the left mandibular teeth (I2–M2).

Results: There was significant advancement in the age of attainment of the final (H) stage for the majority of permanent teeth in the overweight (mean male BMI 22.9; female 27.94) group compared to the underweight ($p < .05$). Negative binomial regression indicated that age, height, and BMI were significant predictors of the dental maturity score for males ($p < .05$), while age, height, weight, BMI and HC were significant predictors for females ($p < .05$).

Conclusion: Nutritional status had a significant effect on the timing of tooth formation in males and females in the study population. The effect was mainly noticeable for children in the extremes of the spectrum of BMI z-scores.

KEYWORDS

BMI, height, nutrition, tooth formation, weight

1 | INTRODUCTION

Tooth formation is generally regarded as highly heritable and immune from environmental stresses relative to other aspects of human growth and development (Demirjian, 1986; Demirjian, Buschang, Tanguay, & Patterson, 1985). The timing of tooth formation and emergence were found to be highly correlated within monozygotic twins compared to dizygotic twins (Garn, Lewis, & Polacheck, 1960). Consequently, the timing of dental development events is considered to be similar across populations and dental atlases are used globally for anthropological and forensic purposes.

There is abundant evidence that systemic stresses during the period of tooth formation lead to enamel hypoplasias and molar incisor hypomineralization (Goodman, Armelagos, & Rose, 1980). The

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severity of enamel hypoplasia is dose- and time-dependent. Populations exposed to high levels of undernutrition and disease have higher rates of linear enamel hypoplasia caused by perturbation of ameloblast functions (Goodman et al., 1980; Goodman & Rose, 1990). If an extreme stressor can lead to a complete disruption of the secretory phase of ameloblasts and a quantitative enamel defect, it logically follows that lower stressors could potentially limit the secretory ability of ameloblasts leading to a delay in the attainment of a tooth formation stage. The specific relationship of these stressors to changes in the process of tooth formation is not known.

Malnutrition is a common stressor affecting growth and development. It can result in stunting and delayed maturity in situations of nutritional shortages, or advanced sexual and skeletal maturity in cases of obesity. The effect of nutrition on tooth formation remains controversial, with some studies finding no effect (Cameriere, Flores-Mir, Mauricio, & Ferrante, 2007; Eid, Simi, Friggi, & Fisberg, 2002; Elamin & Liversidge, 2013) while others observed a delay in tooth formation (Hilgers, Akridge, Scheetz, & Kinane, 2006; Mani, Naing, John, & Samsudin, 2008). Well-designed studies on severely malnourished children are lacking and may be difficult to undertake because of ethical reasons. These studies focused on the relationship between general proxies of nutritional status (height, weight, and body mass index [BMI]) and tooth formation, but none investigated this relationship using other somatic variables to evaluate growth under varying nutritional statuses. Hence, this study investigates the influence of nutritional status on tooth formation using general proxies of nutrition along with two measures (head circumference and mid-upper arm circumference) that are documented to correlate with nutritional stress.

Studies from Peru, Brazil, Iran, and Northern Sudan that used BMI as a proxy for nutritional status found no significant influence of BMI on tooth formation (Bagherian & Sadeghi, 2011; Cameriere et al., 2007; Eid et al., 2002; Elamin & Liversidge, 2013). In contrast, other studies (DuPlessis, Araujo, Behrents, & Kim, 2016; Hilgers et al., 2006; Mack, Phillips, Jain, & Koroluk, 2013; Zangouei-Booshehri, Ezoddini-Ardakani, Aghili, & Sharifi, 2011) showed dental development was significantly accelerated with higher BMI, even after adjusting for age and sex. This developmental advancement was attributed to accelerated linear growth and early sexual maturation, which is usually associated with obesity (Sánchez-Pérez, Irigoyen, & Zepeda, 2010; Slyper, 1998). One reason for these differing results regarding BMI may be the imprecision of the measures of nutritional status (Frisancho, 1990; Myatt, Duffield, Seal, & Pasteur, 2009). For example, standardized BMI in children varies with body shape (Garn, Leonard, & Hawthorne, 1986) and ethnicity (Lear, Humphries, Kohli, & Birmingham, 2007) and may overestimate the prevalence of acute malnutrition in some populations (Frisancho, 1990; Garn et al., 1986; Lin, Wang, & Wang, 2004). BMI is poor in sensitivity and specificity and does not necessarily reflect the changes that occur with age (Rothman, 2008). Additionally, the relationship between BMI and percentage of body fat is not linear and differs for males and females (Rothman, 2008).

Although the BMI cutoff point for malnourished children is still being debated, it is suggested that to be able to identify high-risk malnourished children, the cut-off point of standardized BMI should not be less than −3 (Briend, Maire, Fontaine, & Garenne, 2012). Frisancho (1990) reported that the assessment of nutritional status based only on BMI, especially when the degree of undernutrition or overweight is moderate, is unlikely to be useful in distinguishing the truly wasted or truly overweight individual from a normal or low weight child. This may have influenced the interpretations of previous studies that used the cutoff point of −2 to categorize the study population into two groups without exploring the extremes of the BMI z-scores.

The method of analysis might also be responsible for conflicting results regarding the role of BMI in tooth formation timing. Eid et al. (2002), utilizing the extremes of BMI z-scores (<−2 and > 2), generated mean ages of attainment and used correlational analysis to determine the relationship between dental development and nutritional status. Growth curves are not always linear since acceleration, deceleration, and stunting occur at different stages, resulting in considerable variation in growth velocity (Pinhasi & Mays, 2008). Thus, conclusions should not be solely derived from a linear perspective. Besides, the use of bivariate analysis may not account for potential confounders, especially in growth studies. One of the limitations of bivariate analysis is that it does not factor in how variables influence each other. Bivariate analysis, therefore, cannot explain the relationship between two variables; it only provides a description. An explanatory analysis is needed to infer cause (Spicer, 2005). In summary, tooth formation studies utilizing at least one extreme of BMI classification and employing robust explanatory statistics are needed.

The influence of height and weight on permanent tooth formation is complex. Short height for age reflects a failure of linear growth due to diseases or malnutrition (WHO, 1997). Vallejo-Bolanos and Espana-Lopez (1997) found a delay in the dental development in familial short stature children, while Keller, Sather, and Hayles (1970) found similar results among children with metabolic and endocrine disorders. All these findings point to a relationship between growth hormones and dental development. Malnutrition is a known cause of growth hormone deficiency.

Conversely, some studies suggest that dental development is buffered from environmental stresses. Sarnat, Kaplan, Pertzelan, and Laron (1988) did not find accelerated dental development, but only enhanced bone development, in children being treated with growth hormones. Similarly, Takano, Ogiuchi, Hizuka, Sangu, and Shizume (1986) found an insignificant delay in the dental age but a significant delay in the skeletal age of children with growth hormone deficiency compared to normal children. Body weight is one of the most critical variables for nutritional assessment. It is very useful in predicting macronutrient and fluid requirements and also acute malnutrition (ADSA, 2009). The limitation is that body weight is a poor variable for distinguishing between fat, protein, bone and water levels. It can also be influenced by fluid status, organomegaly, and tumor growth. Currently, there is no information on the direct relationship between weight and permanent tooth formation.

Mid-upper arm circumference (MUAC) is a better index of nutritional status or malnutrition than BMI in children and adolescents (Briend & Zimicki, 1986; Fernández, Delchevalerie, & Van Herp, 2010; Jelliffe & Jelliffe, 1969; Shakir, 1975; Velzeboer, Selwyn, Sargent,

Pollitt, & Delgado, 1983; WHO, 1986). It is less affected by the localized accumulation of excess fluid (pedal edema, periorbital edema, and ascites) commonly seen in famine. MUAC is likely to be a more sensitive index of tissue atrophy than low body weight, and it is relatively independent of height (Olukoya, 1990). MUAC correlates well with BMI in adult populations, yet a globally recognized cutoff point has not been established to classify malnutrition among adolescents and adults (Chakraborty, Bose, & Bisai, 2009; Mazıcıoğlu et al., 2010). Though many countries have established cutoff points for their populations, there is no evidence to support using any particular value. There are no published studies on the relationship between MUAC and tooth formation. Hence, there is a need to investigate the efficacy of MUAC versus BMI when comparing somatic growth with dental development.

Is there a link between dental development and brain development? In a study by Godfrey, Samonds, Jungers, and Sutherland (2001) on a broad range of living primates, brain development proved to be a better predictor of dental development than somatic development. The correspondence of head circumference and the emergence of deciduous teeth has been investigated in humans. Vejdani, Heidarzadeh, and Ebrahimi (2015) found a relationship between primary tooth emergence and head circumference. An earlier study also demonstrated significant associations between the number of primary teeth present and head circumference in males but not in females (Infante & Owen, 1973). These studies demonstrate that a relationship exists between brain development and aspects of primary tooth development in the very young. Head circumference is a proven surrogate for brain development, and it is proportional to brain weight and volume in infants and children. Hence, head circumference is used to monitor cognitive function in postnatal brain growth (Thureen, 2012). Its usefulness as a measure of nutrition is, however, limited. It is poorly sensitive to malnutrition since the brain is spared at marginal calorie intakes that do not support skeletal growth and weight gain. Therefore, it is important to investigate if a relationship exists between head circumference, other nutritional status indicators, and permanent tooth formation, particularly in the early years of dental development.

2 | METHODS

This is a prospective cross-sectional study of 642 clinically healthy Black Southern African participants aged 5–20 years, whose parents and grandparents are indigenous Southern Africans. The participants were selected randomly from primary and secondary schools in Johannesburg during the operation of the Community Oral Health Outreach Program (COHOP) of the Department of Community Dentistry of the School of Oral Health Sciences, University of the Witwatersrand. Children with systemic diseases, mandibular hypodontia (except third molars), and those who had lost teeth on both sides of the mandible were excluded. Ethical approval (NO: M141001) was obtained from the Human Research Ethics Committee (Medical) of the University of the Witwatersrand. Permission to carry out the study was obtained from the local education authority and respective school heads. Consent was obtained from parents while assent was obtained from the children.

2.1 | Sample size

A total of 14 age cohorts (all 1 year except for a combined 18–20-year cohort) were sampled from ages 5 to 20 years. The formula used to determine the minimum sample size was $N = 4z_{\alpha}^{2}S^{2}\text{-}W^{2}$, where S = *SD*, W = desired total width and z_{α} is the standard normal deviate for the 95% confidence level. Using values determined in a similar study by Cameriere, Ferrante, Liversidge, Prieto, and Brkic (2008) (mean of 1.076 and a standard deviation of 0.824 derived from a standard error of 0.030) and a width of 0.2, the minimum total sample size required for the 14 cohorts in our study is 280. However, to improve the power of the study we elected to recruit more children. A total of 642 children were ultimately found to fit the inclusion criteria.

Information collected from the school records included the date of birth and sex. Selected participants were examined on a dental chair in a mobile dental van and intraoral examination was done with a sterile wooden spatula. Panoramic radiographs were taken, and those showing gross pathology or poor image quality were excluded.

Participants were weighed in the standing position on a platform scale (Hana Power), calibrated to a precision of 100 g. Height was measured with a stadiometer (Weylux model 424). The height was recorded to the nearest 0.1 cm. Mid-upper arm circumference (MUAC) of the left upper arm was obtained with a cm tape measure at the midpoint between the tip of the shoulder and the tip of the elbow. Head circumference (HC) was measured by placing a tape measure across the forehead, and the greatest circumference was recorded. All circumferences were recorded to the nearest 0.1 cm. The anthropometric portion of the study was pilot tested on 40 randomly selected students. Intra-examiner reliability of measurement was calculated using Lin's Concordance Correlation Coefficient (height = 0.99, MUAC = 0.96, HC = 0.92).

2.2 | Dental maturity score and age assessment

Dental age assessment was performed according to the original version of Demirjian's method (Demirjian, Goldstein, & Tanner, 1973). The investigator did not have access to the chronological age of the participants. The panoramic radiographs of each child were enhanced using Microsoft Office Picture Manager, labeled with a unique identification number and digitally archived. Each radiograph was assessed for the development of the left seven permanent mandibular teeth rated on an 8-stage scale from A to H, based on the stages of tooth formation proposed by Demirjian et al. (1973) with stage 0 for nonappearance. Each stage was allocated a sex-specific biologically weighted score and the sum of the scores for each participant was used to determine the dental maturity measured on a scale of 0 to

100. The dental maturity score of each child was converted to dental age using standard tables and percentiles curves for both sexes (Demirjian et al., 1973). The mean age of attainment of the H stage of tooth formation was calculated using probit regression analysis (Hayes & Mantel, 1958).

Before data collection, a reliability study was conducted to assess the magnitude of the intra-observer error of dental stage scoring. Twenty-five radiographs (with 175 individual tooth ratings) were randomly selected and assessed at day one and day three. The investigator (TAE) was the only rater for the developmental stages of the teeth. Intra-examiner reliability of dental age assessment for the Demirjian et al. (1973) method was calculated using Cohen's Kappa (Landis & Koch, 1977) and was found to be 0.97.

2.3 | Data analysis

The data were analyzed with Stata 12 for Windows. The analyses included frequencies and cross-tabulations. Association between categorical variables was tested with chi-square while those between continuous variables were tested with the Student's *t* test. Nonparametric equivalents were used as appropriate. BMI was calculated from the height in meters and weight in kilograms. The height and BMI were converted to z-scores using the WHO z-scores for age tables (WHO, 1995). A cutoff z-score of <−2 for both BMI and Height for Age (HAZ) was used to place children into the categories of underweight/short for age, > − 2 to 2.0 for normal weight/height, and > 2 for overweight/obese/tall for age. The mean age at the time of attainment of tooth formation stages and standard deviations were computed using probit analysis after Liversidge (2003). Analysis of variance (ANOVA) was used to determine if any variation exists between the subdivisions of the data. Games-Howell post hoc tests for multiple comparisons of means were then employed to compare the mean age of attainment of H status for the nutritional status subgroups with one another. Student's *t* tests were used where necessary to compare the mean age of attainment of the H stage.

Since there are no WHO standard categories of mid-upper arm circumference or head circumference for older child age cohorts, they were modeled with other predictor variables in a regression analysis. A Shapiro–Wilk test showed that the dependent variable (dental maturity score) and the predictor variables were not normally distributed. Therefore, a negative binomial regression model was used with the dental maturity scores modeled as the dependent variable and the anthropometric variables as predictors. Adequacy of fit was checked using the deviance residuals as recommended by McCullagh and Nelder (1989). The deviance residuals showed normal distribution and the plot of the residuals against each of the covariates also showed model fit. As expected, the collinearity test showed that BMI, height, and weight were significantly collinear. When these variables were excluded from the model, there was no difference in the values of the output. Hence, the variables were included in the final model for generalized linear regression analysis. The model was built using forward selection. Statistical significance was inferred at $p < .05$.

3 | RESULTS

3.1 | Comparison of mean values of anthropometric variables and nutritional status

The basic age and sex distribution of the sample are provided in Tables 1 and 2. The majority of the children fall within the normal category for both height and weight. Only two of the 372 females (0.5%) are categorized as underweight while 22 (8.2%) of the 270 males fall in the underweight category. Notable sex differences are seen in the proportion of overweight children (23 or 8.5% of males vs. 45 or 12.1% of females) and short children (32 or 11.9% of males vs. 19 or 5.1% of females). Very few children of either sex are in the tall category.

Table 3 provides results of ANOVA analyses comparing the anthropometric variables and standardized height and weight scores for males and females in the three BMI categories. The mean age and height of the underweight group are significantly higher than the normal and overweight groups for both males and females. Although the children in the underweight categories are older and taller, their weights are significantly lower compared to the overweight children ($p < .05$ for both sexes). Post hoc analysis by Games Howell multiple comparison of means showed significant differences between the underweight and overweight cohorts only in males, while in girls the differences were significant between the underweight and normal weight and between the normal weight and overweight children ($p < .05$). Furthermore, the BMI values are significantly lower in the underweight compared to normal and overweight categories, and also between the normal and overweight categories for both sexes. Similarly, the z-scores for height and BMI are significantly lower in the underweight group compared to the overweight children for males and females. The MUAC values are significantly lower in the underweight compared to the normal and overweight boys, but the same pattern does not hold for the girls as the small underweight sample ($N = 2$) has a larger mean MUAC than the normal weight girls. In contrast, there are no significant differences in the mean head circumference between the underweight children and the overweight children ($p > .05$ for both sexes).

3.2 | Age of attainment of H stage by nutritional status

In males, the age of attainment of the H developmental stage was calculated for each of the BMI z-score (underweight, normal and overweight) categories using probit analysis after dichotomizing the appearance of the H developmental stage as present or absent. Other developmental stages were not considered because of the small sample size of the underweight group (Table 4). ANOVA showed significant variations in the means of the BMI z-score categories for all the teeth except the lateral incisors among males (Table 4). Further analysis using a Games-Howell post hoc test of multiple comparisons of means revealed significant advancement in the age of attainment of H

TABLE 1 BMI and height distribution of males

	BMI z-score category				Height z-score category			
Age cohort	Under weight	Normal	Over weight	Total	Short	Normal	Tall	Total
5–5.99	0	10	0	10	0	10	0	10
6–6.99	0	24	2	26	1	25	0	26
7–7.99	0	6	0	6	1	5	0	6
8–8.99	2	25	6	33	1	32	0	33
9–9.99	1	30	5	36	0	35	1	36
10–10.99	0	16	0	16	2	14	0	16
11–11.99	0	16	1	17	2	15	0	17
12–12.99	1	19	1	21	5	16	0	21
13–13.99	2	12	1	15	4	11	0	15
14–14.99	6	18	4	28	6	22	0	28
15–15.99	4	20	1	25	4	21	0	25
16–16.99	2	9	1	12	1	11	0	12
17–17.99	3	16	1	20	3	17	0	20
18–20.00	1	4	0	5	2	3	0	5
Total	22	225	23	270	32	237	1	270

TABLE 2 BMI and height distribution of females

	BMI z-score category				Height z-score category			
Age cohort	Under weight	Normal	Over weight	Total	Short	Normal	Tall	Total
5–5.99	0	13	0	13	0	13	0	13
6–6.99	0	26	2	28	1	27	0	28
7–7.99	0	8	1	9	1	7	1	9
8–8.99	0	26	1	27	1	24	2	27
9–9.99	0	27	3	30	2	28	0	30
10–10.99	0	21	1	22	3	19	0	22
11–11.99	1	32	3	36	1	35	0	36
12–12.99	0	36	8	44	2	42	0	44
13–13.99	0	36	3	39	0	39	0	39
14–14.99	0	30	2	32	1	31	0	32
15–15.99	0	20	7	27	0	27	0	27
16–16.99	0	22	6	28	1	27	0	28
17–17.99	1	18	6	25	3	22	0	25
18–20.00	0	10	2	12	3	9	0	12
Total	2	325	45	372	19	350	3	372

stage for all the permanent teeth in the overweight males compared to the underweight group ($p < .05$).

For females, probit analysis was used to calculate the mean age of attainment of the H developmental stage in the normal and overweight groups only, due to very few participants ($N = 2$) in the underweight subgroup. The mean ages of attainment of H stage of all the teeth, except for the canine, were significantly advanced in the overweight group compared to the normal BMI group ($p < .05$) (Table 4).

3.3 | Age of attainment of the H stage by height for age

The Height for Age (z-scores) (HAZ) was dichotomized into "short for age" and "normal" groups in the males. The only male in the tall category was removed from the analysis. The age of attainment of the H developmental stage was calculated using probit regression analysis and the mean ages of attainment of H stage were compared using

TABLE 3 Analysis of variance of anthropometric variables by BMI z-score categories in males and females

	MALES (*N*=270)							FEMALES (*N*=372)						
	Underweight (*N*=22)		Normal (*N*=225)		Overweight (*N*=23)			Underweight (*N*=2)		Normal (*N*=325)		Overweight (*N*=45)		
Variable	Mean	*SD*	Mean	*SD*	Mean	*SD*	*p*	Mean	*SD*	Mean	*SD*	Mean	SD	*p*
Age (years)	14.40	2.79	11.40	3.70	11.03	3.45	**.00**	14.92	4.24	11.99	3.52	13.75	3.17	**.00**
Height (cm)	153.63	15.70	140.68	19.33	139.47	16.98	**.01**	153.25	25.81	143.26	16.36	149.89	12.86	**.03**
Weight (kg)	34.90	9.72	35.14	13.66	46.24	18.46	**.00**	27.00	19.80	38.44	13.41	63.93	16.16	**.00**
BMI	14.41	1.50	17.02	2.43	22.90	3.45	**.00**	10.54	4.81	18.03	3.18	27.94	4.15	**.00**
MUAC	18.11	1.96	18.60	1.86	20.59	3.04	**.00**	19.35	0.92	19.14	1.81	23.29	4.07	**.00**
HC	50.11	2.15	50.67	2.46	51.30	3.22	.28	51.40	1.27	50.28	2.58	50.77	2.07	.48
Height z-score	1.72	0.46	1.90	0.31	1.87	0.34	**.05**	-2.39	0.29	-0.39	0.83	2.52	0.15	**.00**
BMI z-score	-2.48	0.30	-0.31	0.95	3.12	1.03	**.00**	-3.47	1.10	-0.04	0.92	3.43	1.18	**.00**

TABLE 4 Analysis of variance comparison of the age of attainment of H stage of mandibular teeth by BMI z-score category for males and females

	MALES								FEMALES					
	BMI z-score								BMI z-score					
	Underweight (*N*=22)		Normal (*N*=225)		Overweight (*N*=23)				Normal (*N*=325)		Overweight (*N*=45)			
Tooth type	Mean age (years)	*SD*	Mean age (years)	*SD*	Mean age (years)	*SD*	F	*p*	Mean age (years)	*SD*	Mean age (years)	*SD*	F	*p*
I2	9.05	0.66	8.65	1.33	8.38	0.85	1.69	.20	8.44	1.14	6.58	1.16	10.24	**.00**
C	13.06	0.48	12.64	0.77	12.54	1.08	3.22	**.04**	11.59	1.23	11.46	0.82	0.69	.49
P1	14.17	1.69	12.86	0.91	11.17	0.89	52.34	**.00**	12.21	1.30	11.75	1.72	2.13	**.03**
P2	14.87	0.81	13.63	1.11	13.37	1.37	13.59	**.00**	12.58	1.09	11.99	0.50	3.58	**.00**
M1	9.05	0.66	8.73	1.06	8.16	0.17	4.90	**.01**	8.12	1.26	6.58	1.27	7.68	**.00**
M2	15.39	0.62	14.83	0.79	14.77	0.41	5.76	**.00**	14.11	1.14	13.07	0.53	6.02	**.00**

Note: Ages are chronological ages. All individuals had attained the H stage of development for the central incisor.

TABLE 5 Comparison of age of attainment of dental stage H by height for age (HAZ) for mandibular teeth for males and females

	MALES						FEMALES					
	HAZ						HAZ					
	Short for age (*N*=22)		Normal (*N*=237)				Short for age (*N*=19)		Normal (*N*=350)			
Tooth type	Mean age (years)	*SD*	Mean age (years)	*SD*	t	*p*	Mean age (years)	*SD*	Mean age (years)	*SD*	t	*p*
I2	–	–	–	–	–	–	8.75	0.95	8.37	1.16	1.40	.16
C	13.30	1.10	12.89	0.84	1.88	**.03**	11.95	0.44	11.63	1.23	1.13	.26
P1	13.27	1.96	12.90	1.06	1.64	.15	–	–	12.17	1.28	–	–
P2	14.08	0.81	13.89	1.19	0.88	.19	–	–	12.91	1.05	–	–
M1	8.17	0.66	8.06	1.06	0.48	.63	8.75	1.00	8.01	1.27	2.50	**.01**
M2	15.17	0.24	14.84	0.84	2.21	**.01**	14.17	–	14.05	1.11	–	–

Note: Ages are chronological ages. Dash indicates when the probit analysis did not return values.

TABLE 6 Spearman's rho correlation between dental maturity score and anthropometric variables

	Male (*N*=270)		Female (*N*=372)	
	Maturity score		Maturity score	
Variable	r	*p*	r	*p*
Age (years)	.96	.00	.94	.00
Height (cm)	.92	.00	.86	.00
Weight (kg)	.88	.00	.83	.00
HC (cm)	.19	.00	.34	.00
MUAC (cm)	.63	.00	.60	.00
BMI	.49	.00	.65	.00

TABLE 7 Negative binomial regression model of predictors of dental maturity in males

Maturity score	Odds ratio	*SE*	z	*p*	(95% CI)
Age (years)	1.49	0.04	16.94	**.000**	1.42, 1.56
Height (cm)	1.03	0.01	4.24	**.000**	1.02. 1.05
Weight (kg)	0.97	0.01	-1.87	.062	0.94, 1.00
HC (cm)	1.01	0.01	1.32	.188	0.99, 1.03
MUAC (cm)	0.99	0.02	-0.21	.834	0.96, 1.03
BMI	1.06	0.03	2.17	**.030**	1.01, 1.12
Constant	0.00	0.00	-7.30	.000	0.00, 0.01

Student's *t* tests. Significant differences were found for the canine (p = .03) and the second molar (p = .01) in males (Table 5). In comparison, a significant difference was only found between the mean ages of attainment of H stage of the first molars (p = .01) in females when the three tall females were excluded (Table 5). The probit analysis did not return values for P1, P2, and M2 in females.

3.4 | Exploring relationships between the dental maturity scores and the anthropometric variables

Spearman's rho correlational analysis was done to determine if linear relationships exist between the predictor variables and the dental maturity score (Table 6). A significant strong correlation was observed between the dental maturity score and height (r = .92) and weight (r = .88) in males and height (r = .86) and weight (r = .83) in females. A significant moderate correlation was found between the dental maturity score and MUAC and BMI in both males (MUAC r = .63, BMI r = .49) and females (MUAC r = .60, BMI r = .65). The relationship between HC and dental maturity score was significant but weak in both sexes (males [r = .19] and females [r = .34]).

Multivariate analysis was done to determine the relationship between the predictor variables (age, height, weight, BMI, HC, and MUAC) and the outcome variable (dental maturity score). Negative binomial regression analysis was the preferred model because it had the best fit compared to Poisson or generalized linear models. Modeling was done separately for males and females. The results show that age, height, and BMI were significant predictors of the dental maturity for males with all having p values <.05 (Table 7). Interestingly, age, height, BMI and also weight and HC were significant predictors of the dental maturity score for females (p < .05) (Table 8). Thus, MUAC was not a predictor of dental maturity for either sex in these models.

TABLE 8 Negative binomial regression model of predictors of dental maturity in females

Maturity score	Odds ratio	*SE*	z	*p*	(95% CI)
Age (years)	1.65	0.04	20.77	**.000**	1.58, 1.73
Height (cm)	1.04	0.01	5.29	**.000**	1.03, 1.06
Weight (kg)	0.96	0.01	-2.67	**.008**	0.93, 0.99
HC (cm)	0.98	0.01	-2.21	**.027**	0.97, 0.99
MUAC (cm)	0.98	0.02	-1.13	.257	0.94, 1.02
BMI	1.08	0.03	2.65	**.008**	1.02, 1.15
Constant	0.00	0.00	-7.00	.000	0.00, 0.01

4 | DISCUSSION

Investigation of the assumed relative immunity of the timing of tooth formation to environmental perturbations is very important for anthropological and forensic purposes. The general discrepancy between skeletal age and chronological age, even in unstressed individuals, makes the use of aging standards based on somatic growth very unreliable (Demirjian et al., 1985). If the immunity of tooth formation can be verified, the timing of tooth formation could be used as a global standard for the assessment of growth and development. The major environmental stressors that can potentially alter tooth formation are childhood diseases/infections and malnutrition. These factors have synergistic effects as diseases and infections often suppress appetite or reduce absorption of nutrients—creating or augmenting existing malnutrition. Thus, by studying proxies for nutritional status in this study, the two major sources of potential dental developmental perturbations were evaluated.

4.1 | The relationship between BMI and tooth formation

The results of this study confirm a significant influence of nutritional status on the timing of tooth formation. In both males and females, lower BMI is associated with a delay in the age of attainment of the final H dental developmental stage. This is in agreement with previous studies that found a significant influence of nutrition on tooth formation in children from the United States (Hilgers et al., 2006; Mack et al., 2013) and Iran (Zangouei-Booshehri et al., 2011). The findings of the present study are at variance with three previous reports of no significant difference in the dental development of underweight or

overweight children (Bagherian & Sadeghi, 2011; Eid et al., 2002; Elamin & Liversidge, 2013). Careful consideration of these studies raises some methodological questions that may impact their conclusions. Although Elamin and Liversidge (2013) interpreted their results as showing no difference between underweight and normal weight groups, they did document a uniform and consistent trend of advancement in all stages of tooth formation in the normal weight children compared to the malnourished children from Sudan. Aside from using a combined sex sample that might obscure detection of the magnitude of the variation (as would have been the case for our sample), a more important reason for the lack of significant difference may be that Elamin and Liversidge did not investigate the extremes of the BMI z-scores. Pertaining to this point, Garn, Lewis, and Kerewsky (1965) found nutritional status was only slightly related to tooth formation timing in the Fels Growth Study, but they concluded that the relationship was expected to be more marked if the nutritional range was broader and in situations where protein deficits were the key underlying nutritional difference. Bagherian and Sadeghi (2011) also reported a trend of advancing dental maturity from their underweight group to their overweight group in their study of Iranian children, but the correlational analysis results were not statistically significant. Correlation analysis usually shows a linear relationship even though growth patterns are not always linear, especially across a full range of age cohorts (Pinhasi & Mays, 2008). Any pattern should be further explored with a robust multivariate analysis. Eid et al. (2002) used Demirjian's age estimation method for their study of 6–14-year-old Brazilian children, but the age of entry into the developmental stages was not calculated, and the BMI scores were not standardized for age. Therefore, the individual ages of the participants might have skewed their results.

A limitation of this study was the comparatively small number of participants, especially females, in the underweight (low BMI) category. Studies using BMI cohorts to investigate dental caries patterns typically report that the majority of children fall within the normal weight category (cf. the systematic review by Hooley, Skouteris, Boganin, Satur, & Kilpatrick, 2012), as was the case for this study. A larger sample will not remedy this limitation if our sample is representative of the population. The similarity of our sample's characteristics to other studies of Black South African children (Kimani-Murage et al., 2011; Richter, Norris, Pettifor, Yach, & Cameron, 2007) substantiates our view that it is a representative sample. However, the possible effects stemming from the low numbers of underweight children would be that our results and conclusions, particularly with respect to the females, might not be generalizable for all Black South African children. Theoretically, one way to approach this potential limitation in future work would be to conduct a case–control study of low BMI and normal BMI children's tooth formation in a population where sufficient numbers of low BMI children are represented.

Social factors probably underlie the limited cohort of underweight girls in our sample. The government of South Africa started providing free meals in all public schools in 1994 (Rendall-Mkosi, Wenhold, & Sibanda, 2013), and this practice might account for the low numbers of underweight participants. School attendance was higher for females in this study and access to school meals may vary considerably as a consequence. Additionally, the results of this study support the argument that females are better buffered from environmental stresses than males (Stini, 1975, 1982; Stinson, 1985).

4.2 | Influence of height and weight on the timing of tooth formation

This study found that height influences the timing of tooth formation, similar to what is seen for BMI. Unfortunately, the relationship with height could not be fully explored because of limited sample sizes at the extremes of height (tall for age and short for age) for both sexes. Green (1961) and Demirjian et al. (1985) found a strong correlation between height and dental maturity. It is expected that height and tooth formation are correlated because they are both measures of growth. In our study of Black Southern African children, there was a significant difference in the timing of canine and second molar formation in males that are short-for-age versus those of normal height. For females, a significant difference by height was seen only for the first molars.

Using binomial regression models, this study found no relationship between weight and tooth formation timing in the males whereas a significant relationship was found in females. This may be due to different patterns of weight gain in the two sexes (Geer & Shen, 2009). The weight gain in females occurs earlier and continues until puberty while that of males occurs much later. These differences in the pattern of weight gain characterize Black Southern African children (Kimani-Murage et al., 2011). Although our study found a strong correlation between weight and dental maturity scores in males and females, it is possible that the correlation may be due to weight gain as age increases. Green (1961) found a moderate correlation between weight and dental age although he did not correct for sex. While there is presently no tooth formation study to compare with the result of our multivariate analysis directly, previous studies on tooth emergence found no relationship with weight (Kutesa, Nkamba, Muwazi, Buwembo, & Rwenyonyi, 2013; Oziegbe, Adekoya-Sofowora, Folayan, Esan, & Owotade, 2009).

4.3 | Mid-upper arm circumference and the timing of tooth formation

No significant relationship was found between MUAC and dental maturity in our multivariate models. This is because most of the children in the sample are within the normal range of MUAC. Another consideration is that the distribution of adipose tissue differs from one population to another (Gasperino, 1996), and it changes rapidly during growth. Other measures of body composition, such as waist-to-hip ratios, may be more informative about nutritional status in Black Southern Africans. Furthermore, MUAC is affected by exercise, type of work or household chores, and this may limit its use as a proxy for nutritional status in our study setting. Future studies are needed to explore this research area.

4.4 | Influence of head circumference on dental maturity

This study found a significant relationship between head circumference and tooth formation timing in females but not in males. Although the correlations were low, the relationship among females was significant for the multivariate analysis. The reason for the difference could be explained by a pattern of increase in the head circumference over a more extended period in females compared to males. The males had increased head circumference from the age 5 to 9 year old cohorts while it was from ages 5 to 12 years in females. These findings may be an indicator of a relationship between permanent tooth formation and brain growth. Presently there are no other studies that investigated the correspondence of head circumference with permanent tooth development. However, a study on primary teeth showed that the number of erupted teeth has a positive relationship with head circumference (Vejdani et al., 2015). During perinatal and postnatal development, there is a mobilization of resources to the brain at the expense of other tissues (the Expensive Tissue Hypothesis, Aiello and Wheeler (1995)). It is during this same period that the development of the primary and permanent dentition commences, with the emergence of all primary teeth occurring by ~30 months. The first phase of permanent dental development continues until the emergence of the incisors and first molars at ~6 years, by which time brain size increase is almost completed. The second phase of permanent tooth development is associated with the pubertal growth spurt. Further studies should be done to unravel the precise relationship between tooth formation and brain development to validate this proposition.

5 | CONCLUSION

Although dental development is generally thought to be immune to environmental influences, this study demonstrates that malnutrition does have a significant effect on tooth development in males and females in a Black Southern African sample. The effect was mainly noticeable for children in the extremes of the spectrum of the BMI z-scores. The multivariate modeling results illustrate that the anthropometric variables had different predictive relationships with tooth formation timing for males and females; the MUAC did not show a significant relationship with the timing of tooth formation for either males or females, yet HC was a significant predictor in females.

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DATA AVAILABILITY STATEMENT

The data for this study are freely available and curated with HVIRU, the Human Variation and Identification Research Unit, based in the School of Anatomical Sciences, Faculty of Health Sciences, University of the Witwatersrand (Esan and Schepartz, nd). For access to the data, please contact the authors Lynne.Schepartz@wits.ac.za or esantemitope@yahoo.com or the HVIRU director, Maryna.Steyn@wits.ac.za.

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