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APPRAISAL OF DENTAL ENAMEL HYPOPLASIA IN THE MIDDLE MIOCENE DEINOTHERIIDAE: IMPLICATIONS OF THE SIWALIK PALEOENVIRONMENT OF PAKISTAN

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ABSTRACT – The paleoecological fluctuations left their impression marks on the tooth enamel of mammals during their tooth development. These marks can be used as stress indicators because they reflect the type of duress faced by the extinct mammalian species during their lives. The enamel hypoplasia (EH) is a common stress marker to trace out the paleoenvironment of the region and the likelihood of species extinction. The material used for current EH analysis belongs to the genus *Deinotherium*, family Deinotheriidae, collected from Middle Miocene (15.2–11.2 Ma) Siwaliks of Pakistan. In this analysis 35 samples consisting of 52 teeth of two species, *Deinotherium pentapotamiae* and *D. indicum* are included. The results indicate that 13/52 (25%) of the analyzed teeth have occurrences of EH giving a prediction that these Siwalik deinotheriids were facing the physiological and/or ecological stresses during the Middle Miocene epoch of Pakistan. The higher frequency of EH in molars (30.30%) compared to premolars (21.05%) express that the individuals experienced a comparatively high stress at the adult stages of their lives. This higher magnitude of EH in molars supports the idea of ecological stressors, i.e., dietary, mating, disease, and predator-prey associations, amplify the likelihood of extinction by dint of EH occurrences.

Keywords: enamel hypoplasia, Siwaliks, Middle Miocene, Proboscidea, *Deinotherium*, root crown junction.

RESUMO – As flutuações paleoecológicas deixaram suas marcas impressas sobre o esmalte dentário durante o desenvolvimento dos dentes dos mamíferos. Essas marcas podem ser usadas como indicadoras de estresse porque refletem o tipo de pressão enfrentado pelas espécies extintas de mamíferos durante suas vidas. A hipoplasia de esmalte (HE) é um marcador de estresse comum para sugerir o paleoambiente e a probabilidade de extinção de espécies. Para análise de HE, o presente estudo avaliou material pertencente ao gênero *Deinotherium*, família Deinotheriidae, proveniente do Mioceno Médio (15,2–11,2 Ma) de Siwalik, Paquistão. Nesta análise foram incluídas 35 amostras, consistindo em 52 dentes de duas espécies, *Deinotherium pentapotamiae* e *D. indicum*. Os resultados indicam que em 13/52 (25%) dos dentes analisados ocorre EH, indicando que estes deinoterídeos de Siwalik estavam enfrentando um estresse fisiológico e/ou ecológico durante o Mioceno Médio no Paquistão. A elevada frequência de HE em molares (30,30%) em relação aos pré-molares (21,05%) expressa que os indivíduos experimentaram um estresse comparativamente elevado na fase adulta de suas vidas. A magnitude maior de HE aqui observada em molares suporta a ideia de estressores ecológicos, ou seja, dieta, acasalamento, doenças e associação presa-predador, aumentando a probabilidade de extinção por causa das ocorrências de HE.

Palavras-chave: hipoplasia de esmalte, Siwaliks, Mioceno Médio, Proboscidea, *Deinotherium,* junção raiz coroa.

INTRODUCTION

The mammalian order Proboscidea includes the living elephants and all their extinct relatives, among them deinotheres, mammoths, gomphotheres, mastodons, barytheres, amebelodonts and stegodonts. The family Elephantidae includes the two living genera (*Elephas* and *Loxodonta*), with restricted distributions in Southeast Asia and Africa respectively, which are remnants of a much wider proboscidean radiation in the geological past (McKenna & Bell, 1997). Proboscideans originated in Africa during the Paleocene epoch or perhaps earlier and during the Miocene occurred their dispersal and diversification to all over the northern continents, eventually reaching the South America at the end of Miocene (Prothero *et al*., 1988). In the Siwalik subgroup of Pakistan four proboscidean families were registered, Deinotheriidae Bonaparte, 1841, Gomphotheridae Hay, 1922, Stegodontidae Osborn, 1918, and Elephantidae Gray, 1821, with ten valid genera, *Deinotherium* Kaup, 1829, *Gomphotherium* Burmeister, 1837, *Choerolophodon* Schlesinger, 1917, *Protanancus* Arambourg, 1945, *Anancus* Aymard, 1855, *Paratetralophodon* Osborn, 1929, *Stegolophodon* Schlesinger, 1917, *Stegodon* Falconer, 1857, *Paleoloxodon* Matsumoto, 1924, and *Elephas* Linnaeus, 1758, with 22 extinct species, from which 18 are valid (Tassy, 1996) and four taxa have uncertain taxonomy.

Among the proboscideans, the representative taxa of the family Deinotheriidae are well known during ~16–10 Ma by their fossil record from the Siwaliks (foothills of Himalayan) in Pakistan. All the fossil remains analyzed in this study were collected from different localities of the Middle Miocene Siwaliks of Pakistan (15.2–11.2 Ma). The Deinotheres are characterized by the presence of vertically erupted all check teeth and downwardly pointed lower incisors (tusks). The family constitutes a monophyletic Siwalik group with only a single genus *Deinotherium* (Sarwar, 1977). The erection of *Antoletherium* and *Prodeinotherium* by Falconer (1868) and Éhik (1930) respectively, was erroneous and they cannot be differentiated from *Deinotherium.* The taxonomic status of the genus is still controversial, being that Tiwari *et al*. (2006) attributed the smaller sized animal to *Prodeinotherium.* This study is based on the two valid species of genus *Deinotherium*, including the smaller and older species *D. pentapotamiae*, and the larger and younger *D. indicum* (Harris, 1973; Poulakakis *et al*., 2005) from the Middle Miocene Siwaliks of Pakistan.

Enamel hypoplasia (**EH**) is a dental anomaly categorized by the deformed enamel resulting from the disruptions during amelogenesis (White, 1978). Enamel of tooth in this regard is unique because of its inability to remodel. On the basis of ease of enamel examination, sensitivity, failure to remodel, and chronological array of its developmental pattern it can be the perfect tissue for recording alterations in physiology of an animal during its tooth development (Kreshover, 1940; Massler *et al*., 1941; Sarnat & Schour, 1941). The indications of EH in fossilized material, especially in mammalian dentition, has been recently unveiled by many paleontologists (Mead, 1999; Franz-Odendaal *et al*., 2003, 2004; Franz-Odendaal, 2004; Byerly, 2007; Roohi *et al*., 2015; Bohmer & Rossner, 2017; Ahmad *et al*., 2018; Barron-Ortiz *et al*., 2019), which add an additional and much reliable evidence to propose the paleoclimatic fluctuations on regional scale. Physical anthropologists frequently study the defects of tooth enamel to assess the frequency of physiological stressors including mineral deficiencies, metabolic disruptions, premature births, and contagious diseases (Pindborg, 1982). Teeth revealing enamel disorders are routinely "scored" and quantitatively compared with standardized tables (Hillson, 1986) to assess the type and severity of the stress.

EH displays a wide array of expressions, comprising of vertical or horizontal grooves, pits, or broad bands of missing or incomplete enamel (Hillson, 1996; Hillson & Bond, 1997). Mellanby (1929) carried out a classic series of experiments on hypoplasia in beagle dogs, grouped these defects involving pitting and the deficient formation of large areas of enamel as gross hypoplasia. Since that time, it has become common to use the term M–hypoplasia (M for Mellanby) for furrow-like defects and G–hypoplasia to describe the gross defects. M–hypoplasia may involve a single furrow around the crown, or a whole group in a wide band (Mellanby's washboard pattern). G–hypoplasia may be just a single line of small pits, a band several pits wide, a band of pits which have coalesced together, or the various types of defects where whole layers of enamel are missing. This classification is really one of convenience and followed despite more than two centuries of epidemiological and experimental study. The study indicates that G–type of EH is more severe than that of M–type, where the animal is greatly affected by the systemic disorders/disease.

There are three broader categories of EH according to FDI (Federation Dentaire International, 1982) that are pits, grooves and areas missing enamel, semicircular enamel hypoplasia (SEH) and linear enamel hypoplasia (LEH). There can be different etiologies for these types of EH, *e.g.* birth trauma, metabolic and nutritional disorders, infections, exposures to toxic chemicals (Seow, 1991), rickets (Nikiforuk & Fraser, 1981), low birth weight (Slayton *et al*., 2001), poor health status of the mother (Armelagos *et al*., 2009), nutritional conditions of an area (El Najjar *et al*., 1978; Ogilvie *et al*., 1989), general physiological stress (Guatelli-Steinberg *et al*., 2004), weaning stress (Mead, 1999), post weaning stress (Moggi-Cecchi *et al*., 1994), nutritional and/or environmental stress (Franz-Odendaal *et al*., 2004). Goodman & Rose (1990) reported the circular and linear types of EH on the basis of appearance. Among the types, LEH is the most common and its marks can macroscopically be observed on the enamel of living as well as fossilized dental remains (Franz-Odendaal *et al*., 2004). This type of EH is normally expressed in permanent dentition in the form of one or multiple horizontal or transverse grooves, or a linear array of pits (Goodman & Rose, 1991; Skinner & Goodman, 1992). The disorders as a result of localized trauma are identified as localized or non-linear EH (Lukacs, 1999, 2001). Normally these forms of defects do not occur on the uninjured side of the mouth (Skinner, 1986; Skinner & Hung, 1986, 1989), whereas the hereditary enamel defects are usually pretentious with other congenital problems (Winter & Brook, 1975; Stewart & Poole, 1982) where the individuals have very rare possibilities of survival (Rushton, 1964). The circular enamel hypoplasia is present in the form of circular array of large or small pits with missing/underdeveloped enamel and may be in the form of single or multiple pits (Skinner & Hung, 1986, 1989).

The perturbations of EH occur as a result of three causal situations: (i) localized trauma, (ii) hereditary anomalies, or (iii) physiological or systematic metabolic stress (parturition, weaning, nutritional, illness and stress during cow-calf separation) in developing teeth at a specific ontogenetic age (Weinmann *et al*., 1945; Shawashy & Yaeger, 1986; Suckling, 1989) recognized as EH (Goodman & Rose, 1990; Neiburger, 1990; Mead, 1999; Dobney & Ervynck, 2000; Lukacs, 2001; Franz-Odendaal *et al*., 2003). The perturbations resulting as hereditary causes are the most severe and usually affect all of an animal's teeth (Weinmann *et al*., 1945; Stewart & Poole, 1982). The defects emerge due to local trauma or other non-systematic components which could also be relatively terrible but can influence only one or a few adjoining teeth (Weinmann *et al*., 1945; Andreasen *et al*., 1971; Ravn, 1975, 1976; Stewart *et al.*, 1982; Shafer *et al.*, 1983; Skinner & Hung, 1989).

The position, width, and depth of EH can give information about the age of occurrence of anomaly, the period of stress event, and the severity of the stressors, respectively (Moggi-Cecchi *et al*., 1994). EH is associated with the lower fitness of the individuals and an increased possibility of death prior to sexual maturity in humans (White, 1978). However, dental imperfections are known to occur in many other extinct mammalian taxa and their data have been potentially used in paleoecological explanations to understand the possibilities of extinction and evolution (Hillson, 1986). The organisms respond individualistically to the climatic changes (Graham *et al.*, 1996; Stewart, 2009). This individualistic response of plant species may result in large-scale restructuring of vegetation, causing a disruption of coevolutionary interactions between plants and animals (Graham & Lundelius, 1984). These changes are postulated to have reduced niche differentiation among large herbivorous mammals, leading to competition for dietary resources and causing nutritional stress in some animal species (Graham & Lundelius, 1984; Barrón-Ortiz *et al.*, 2019).

The study of EH on the Siwalik mammals is known by Roohi *et al*. (2015) on rhinocerotids and Ahmad *et al*. (2018, 2020) on giraffes and tragulids. The current research is on the assessment of the Siwalik Middle Miocene paleoenvironment through the study of dental EH in the Family Deinotheriidae of the order Proboscidea.

The present article aimed to provide information about: (i) the impact of physiological and ecological stresses on the Siwalik proboscideans by using EH as a stress marker; (ii) inter family comparison of EH in the Siwalik proboscideans and (iii) to trace out the ecological role of the proboscidean in the Middle Miocene Siwalik ecosystem and the possible dietary effects that can be associated with the disappearance of these giant herbivores by the terminal of Middle Miocene.

MATERIAL AND METHODS

A total of 52 teeth belonging to 35 individuals of the genus *Deinotherium* were analyzed for the study of dental EH (the details of studied material are given in Table 1). The paleontological collection was taken from Dr. Abu Bakar Fossil Display and Research Centre, University of the Punjab, Lahore, Pakistan and from the sub-campus, Jhelum. Among the analysis of EH the fossil remains were first scrutinized as "readable teeth" or "unreadable teeth", and only the readable teeth were used to determine the frequency of EH. Among unreadable teeth were included those that: (i) extremely fragmented or have weathered enamel; (ii) completely covered with dental calculus or cement; (iii) or in case the teeth were erupting and concealed in alveolus to that extent where the surfaces found impossible to examine. Before the examination of EH, the readable dental material was prepared (if broken at some parts), washed, and cleaned.

The macroscopic examination of EH was carried out following the methods outlined by Lukacs (1989), Goodman & Rose (1990), Berti & Mahaney (1995), and Hillson & Bond (1997). Different tools which were used for the examination and recording of EH are (i) digital Vernier caliper with 100 mm measuring capacity; (ii) digital handheld HDR camera for photography; (iii) large dissecting trays with cotton bed to shift the samples; (iv) 60–100 watt incandescent light with variable power was used for laboratory examination of samples and (v) 10X hand lens used for the magnification of the sample to properly identify and determinate the occurrence of EH.

All the samples were examined separately for the macroscopic observation and then the presence or absence of EH was recorded for the interpretation of results.

Two different light sources (natural sunlight and 60–100 watt variable incandescent light) were also oriented obliquely to the tooth specimen following Lukacs (1989). The amount of artificial light should also be controlled at certain limit to precise the identification of enamel hypoplastic mark. However, several times a variable amount of light was used for each individual sample to identify either EH was present or not.

Macroscopic direct observations also were done by the second rater to obtain more accurate results. The repeatability and reliability are an indispensable exercise during the inspection of EH especially in proboscidean teeth because of rugosity of enamel and the presence of perikymata. Perikymata are incremental growth lines that express on a tooth surface as a series of linear furrows. The difference in opinion between the two researchers was documented separately to minimize the chance of errors and for the validity of results. The photographic results of EH given in different publications were used to differentiate the observed marks of LEH on the analyzed remains from the other possible observable irregularities and normal findings (such as growth lines) on the surface of a tooth enamel (Mead, 1999; Franz-Odendaal *et al*., 2004; Lacruz *et al*., 2005; Teegen & Kyselý, 2016; Lyman, 2018).

During examination, position of EH was recorded starting from the root crown junction to the enamel defect. The distance was measured to the center of the enamel defect and scored. For each enamel defect, distance from root crown junction to the defect indicated the timing of EH formation. An average of three repeated readings was used to collect the exact distance of defect in relation to root crown junction. A Canon EOS-350D digital professional series was used to snapshot EH position.

The tooth terminology of Harris (1976) and Tassy (1996) was followed for the study of EH in deinotheriid dental material. The occurrence of LEH was counted both in upper and lower teeth as well as from both labial and lingual surfaces. The measurements to the EH were calculated in millimeters (mm) during examination of entire fossil material of the family Deinotheriidae relied on the procedure of Mead (1999). The studied material of *Deinotherium pentapotamiae* was found from Bhilomar (extinct at 13.8 Ma), Chinji (extinct at 13 Ma), Dhok Ban Ameer Khatoon (extinct at 11.8 Ma), and Chabbar Syedan (extinct at 14.2 Ma) localities of the Siwaliks of Pakistan. Whereas *D. indicum* belongs to Lava (extinct at 12 Ma), Dial (extinct at 13.5 Ma), Chinji (extinct at 13 Ma), and Bhilomar (extinct at 13.8 Ma) localities.

Statistical Analysis

Kappa statistics (Cohen, 1960) was applied for the explanation of agreement or disagreement. The calculation of "k" value indicated the inter-rater reliability, and the value of "p" indicated the validity of results, either the difference in rating is significant or non-significant during the analysis of EH in deinotheriids. Chi square test was also run-on data to check either the EH differences are significant or not between the species.

RESULTS AND DISCUSSION

EH occurrence by taxon and tooth

EH prevalence by taxon:

The position of EH on the dental crown, as well as the distance from root crown junction (**RCJ**) of each defect presented varied results in the analyzed material (Tables 1–3). A total of 10 individual specimens out of 35 (28.57%) showed EH in the genus *Deinotherium*. In these 35 specimens of the genus *Deinotherium* 6/15 (40%) samples examined from *D. pentapotamiae* had occurrence of EH and *D. indicum* revealed 20% of EH where four individuals out of 20 were affected. The occurrence of EH is higher in individuals of *D. pentapotamiae* compared to *D. indicum*. The results of data sets possibly indicate that smaller species *D. pentapotamiae* was more susceptible to the environmental stresses compared to *D. indicum*. The occurrence of two EH on a single tooth (Figure 1, PUPC-15/253, m1 and Figure 2, PUPC- D3, PUPC-09/116) indicates more than one systemic stress episodes in the concerned animal's life.

EH prevalence by tooth:

The analyses of isolated teeth from both species showed that *Deinotherium pentapotamiae* comprises of 25 individual teeth out of eight which exhibited 32% occurrence of EH, whereas *D. indicum* presented five defected teeth out of 27 with 18.51% occurrence of EH. The comparative assessment of EH in individual teeth of both species showed much more variable results. The smaller species (*D. pentapotamiae*) was found with higher incidences of EH compared to the larger one (*D. indicum*). Both species of *Deinotherium* were coeval in the Middle Miocene localities from Siwaliks of Pakistan, with almost similar diet and habitat (Tiwari *et al.*, 2006; Sankhyan, 2014). Competition may also be a major role to acquire more diet related environmental stress. The changing ecological conditions from C_3 wet forests to dry deciduous forests and C_4 grasses and the onset of aridity at the terminal of Middle Miocene (Böhme *et al.*, 2007; Gupta *et al.*, 2015; Wu *et al.*, 2018) limits diet for many species. However, the similar diet patterns, possibly between species of a same genus as well as within different taxon, followed the competitive exclusion principle and the competition intensification ultimately lead towards the likelihood of increased environmental stress triggering the events of extinction or migration (dietary, disease, mating, competition, prolong unusual weather, predatory prey interaction etc.) of these animals from the region.

Interpretation of Kappa Results

There was almost substantial agreement between the two diagnoses, K = 0.749 and K = 0.701 for samples of *Deinotherium pentapotamiae* and *D. indicum* analyzed in natural light and artificial lights, respectively, and there were non-significant differences ($p > 0.05$) in opinion between the two raters. The results were significantly ($p < 0.001$) different compared to artificial and natural light (Figure 3).

Interpretation of chi-square test

The chi square results also indicated non-significant differences and both species of the family Deinotheriidae tolerate equal amount of environmental stress. But the count of simple percentages (Table 3) varies prominently and proposed that the *Deinotherium pentapotamiae* was more severely affected compared to *D. indicum*. The occurrence of EH in upper *vs* lower teeth was non-significantly ($p > 0.05$) different. The other comparisons in Table 4 also showed non-significant differences between both species. Non-significantly increased manifestations of EH in molars (27.27%) compared to premolars (21.05%) indicated that the stress related environmental episodes were somehow ascended more vigorously at adult stages.

The diversity of the Siwalik deinotheriids declined during the onset of Late Miocene, perhaps, because of squeezing their habitats and food resources, which is usually attributed to the disease and competition with other herbivores of the region. The deinotheriids became extinct by one of the different unknown reasons contributing to other stress related factors (dietary, disease, mating, competition, prolong unusual weather, predatory prey interaction etc.), where dietary stress may be the primary reason for extinction of this group in the Siwaliks of Pakistan. As the stress was somehow greater in

Table 1. Occurrence of EH in fossil Deinotheriids from the Middle Miocene Siwaliks of Pakistan.

Species	Specimen	locality	Age (Ma)	Enamel Hypoplasia		
				Tooth	Loph/lophid	Location
Deinotherium pentapotamiae	UZ (66/41)	Bhilomar Chinji Horizon	14–12	P3	Labial	One LEH 10.0 mm above the RCJ
	UZ (96)	Chinji village (Jhelum)	14.2–13	P3	Labial	One LEH 10.34 mm above the RCJ
	UZ (67/467)	Chinji village (Jhelum)	14.2–13	M3	1st Loph	One LEH 13.86 mm above RCJ
					2nd Loph	One LEH 12.22 mm above RCJ
	UZ (84/95)	DBAK (Chakwal)	14–11.2	m1	1st Lophid	One LEH 3.15 mm above the RCJ
	PUPC (15/01)	Chabbar Syeddan	14.2–13	p4	2st lophid	One LEH 5.11 mm above the RCJ
				m1	1st lophid	One LEH 15.01 mm above the RCJ
	PUPC (15/253)	Chabbar Syeddan	14.2–13	m1	2nd & 3rd lophid	Two LEH's 6.50 mm above the RCJ 9.80 mm above the RCJ
				m2	2nd lophid	One LEH 19.60 mm above the RCJ
Deinotherium indicum	PUPC (09/116) b	Lava Chinji (Jhelum)	12.2–11.2	m2	2nd loph	Two LEH's 10.40 mm above the RCJ
				m3	1st loph	One LEH 7.21 mm above the RCJ
	UZ-(D_3)	Chinji village (Jhelum)	14.2–13	m1	3rd lophid	Two LEH 10.33 mm above RCJ 26.50 mm above the RCJ
	UZ (66/815)	Dial, Chinji horizon	14–10.5	M2	1st loph	One LEH 10.20 mm above the RCJ
	UZ (66/125)	Bhelomar Upper Chinji	14–12	P3	Labial	One LEH 8.86 mm above the RCJ

Table 2. *Deinotherium pentapotamiae* dental specimens by jaw and type of tooth with percentage prevalence of EH. Modified from Lukacs (2000).

Taxon	Jaw	Tooth						Total
		P3	DP4	P4	M1	M2	M3	
D. pentapotamiae (n=25)	Maxillary	3	1	2	0	1	1	8
	Mandible	4	–	2	5	3	3	17
Dentitions with EH		2	–	1	3	1	1	8
% age occurrence of EH		28.57%	–	25%	60%	25%	25%	32%
			25%			38.46%		

Table 3. *Deinotherium indicum* dental specimens by jaw and type of tooth with percentage prevalence of EH. Modified from Lukacs (2000).

Taxon	Jaw	Tooth						Total
		P3	DP4	P4	M1	M2	M3	
D. indicum (n=27)	Maxillary	1	–	2	1	6	0	10
	Mandible	2	–	2	2	5	6	17
Dentitions with EH		1	–	0	1	2	1	5
% age occurrence of EH		33.33%	–	0	33.33%	18.18%	16.67%	18.52%
			14.28%			20%		

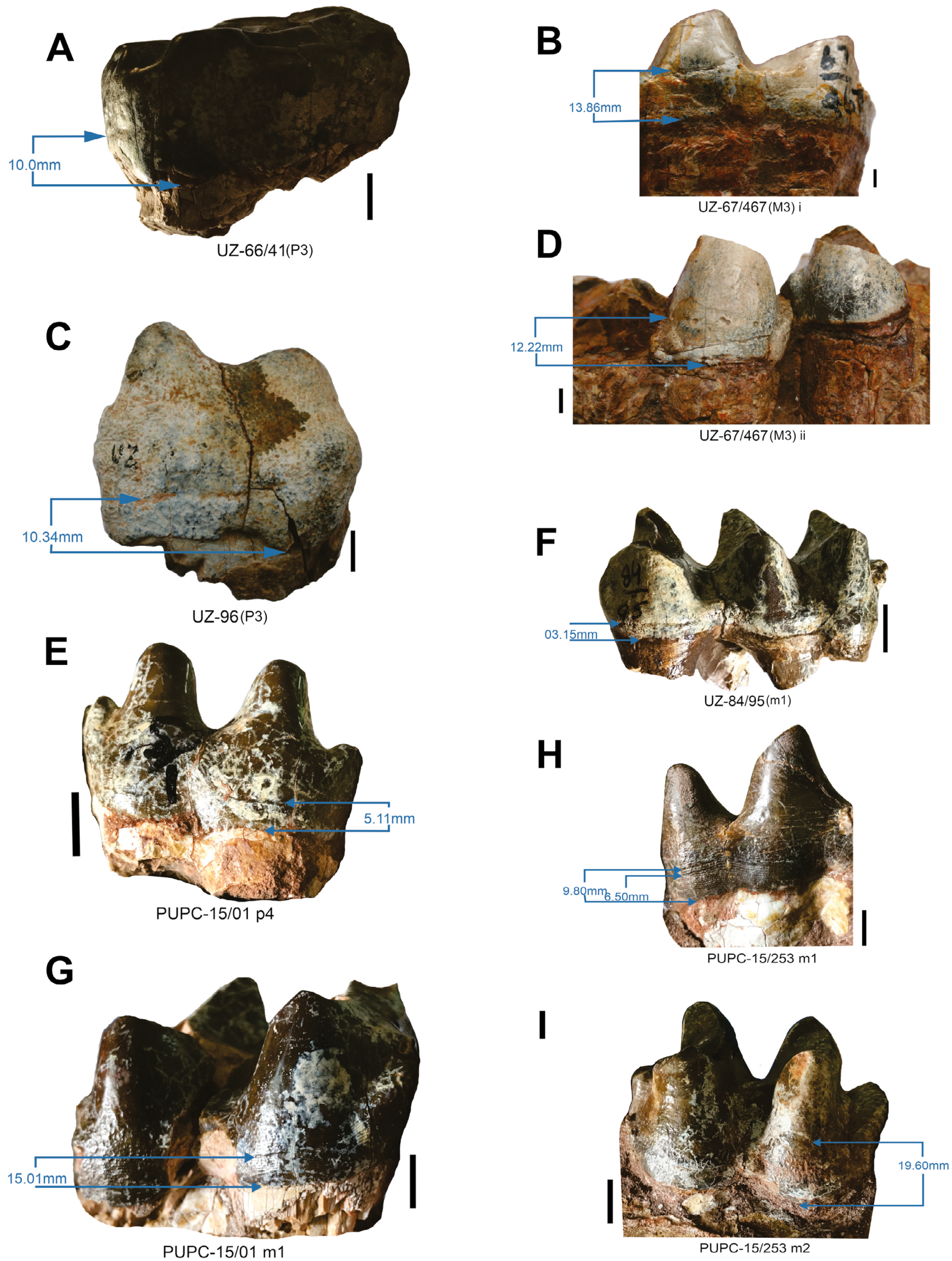

Figure 1. EH in teeth of *Deinotherium pentapotamiae* of the Middle Miocene Siwaliks of Pakistan along with catalogue number, type of tooth, and frequencies of occurrence of EH for different specimens is given. Scale bars = 10 mm.

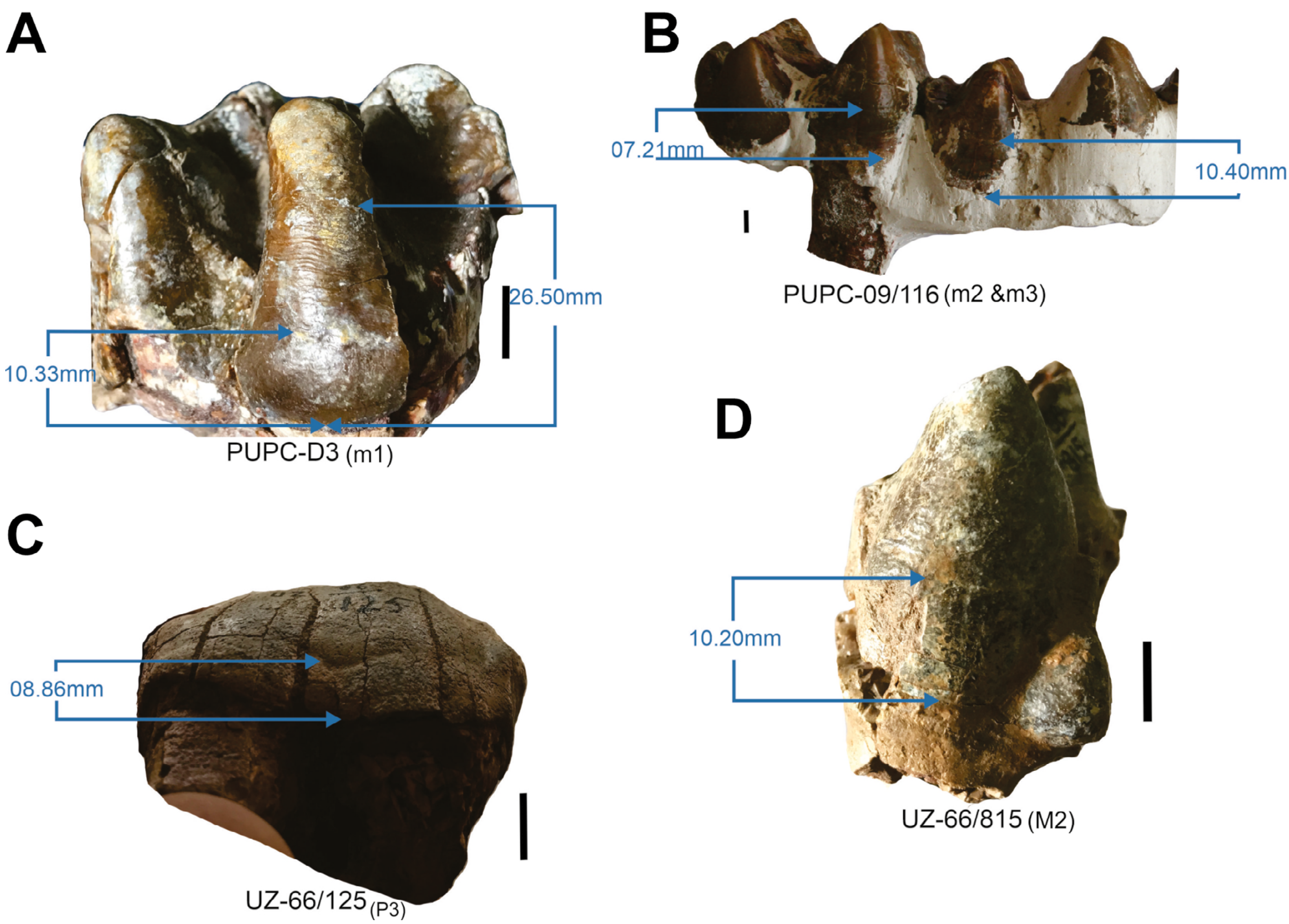

Figure 2. EH in teeth of *D. indicum* of the Middle Miocene Siwaliks of Pakistan along with catalogue number, type of tooth, and frequencies of occurrence of EH for different specimens is given. Scale bars = 10 mm.

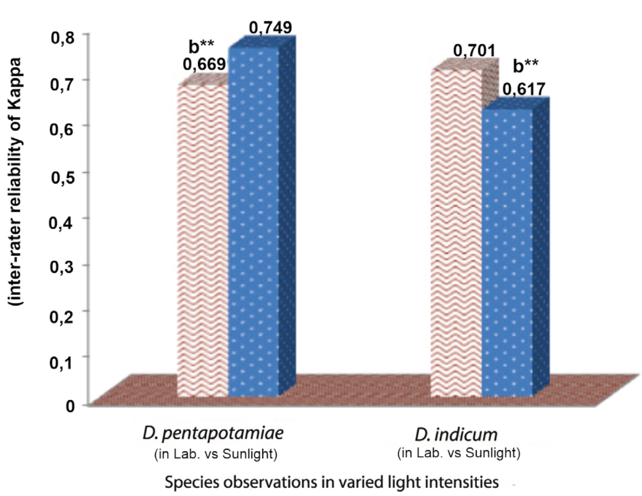

Figure 3. *Deinotherium pentapotamiae* and *D. indicum*, presented along with kappa value: non-significant = p > 0.05; very significan t= p < 0.001=b** (dark red = studied in artificial light, blue = studied in natural light).

Deinotherium pentapotamiae, it may due to their shorter size and failure to compete. Whereas some packs of *D. indicum* were successfully migrated from the region and their retreat during the Late Miocene indicate that they were not completely extinct from the Siwaliks, even remains of this species found from the Dhok Pathan Formation (Late Miocene) of India (Colbert, 1935). However, the extinction of the genus *Deinotherium* from the Siwaliks of Pakistan did not follow any retreat in future after Middle Miocene (15.2–11.2 Ma).

Comparison of EH with others Siwalik fossil mammals

The incidence of EH (26.92%) in teeth of the Siwalik deinotheriids (proboscideans) is somewhat closer to the recent study on giraffes (20%) in the Middle Miocene (Ahmad *et al.*, 2018). The rhinocerotids were very less affected. The Figure 4 indicates the comparative occurrence of EH in different Siwalik families. The proboscideans were the most affected animals as they are almost fully reliant on plants at that time where the feed compromising was a challenge.

Table 4. Results of Pearson Chi-square p value, which is greater than 0.05, the proportion of defected teeth in all comparisons are non-significant.

Type of dentition	Lower premolar	Lower molar	Upper premolar	Upper molar
Lower premolar	……..	0.281	0.313	0.527
Lower molar	0.281	……..	0.961	0.690
Upper premolar	0.313	0.961	……..	0.701
Upper molar	0.527	0.690	0.701	…….

The difference in occurrence of EH, most dominant in molars as compared to premolars, inferences that the deinotheriids (proboscideans) were unable to resist effectively to the chapter of changing environmental conditions. The Figures 5 and 6 give information about the rate of occurrence of EH in individual teeth and percentages studied, respectively.

The fossil material evaluated in this study was discovered from the Middle Miocene (15.2–10.8 Ma) of Pakistan. The stable isotopic analysis of carbon and oxygen from paleosols and mammalian tooth enamel documented that the C_4 savannas substituted the C_3 forests and woodlands at 8.5 Ma during the Late Miocene (Quade *et al*., 1992; Quade & Cerling, 1995; Barry *et al*., 2002; Badgley *et al*., 2008). The Siwalik record implies a reduction of annual rainfall with change in seasonality of precipitation between savannas (C_4 grasses) and dry woodlands and monsoon forest (C_3) was maintained by a small difference during Late Miocene (Badgley *et al*., 2008). The disappearance of genus and its diet competitor, *Listriodon pentapotamiae,* with almost similar dental pattern and dietary habits, at the terminal of Middle Miocene indicates somewhat short but severe environmental changes in the region. Hence, it is proposed that the changes in climate pattern during the Late Miocene was not abrupt and overnight, but based on the end of the Middle Miocene and later on accelerated during the Late Miocene. The dental pattern of deinotheriids showed that they were shearing browsers, diggers and scrapers feeding on soft vegetation (Harris, 1975), which indicated that the dietary pattern of the taxon did not remain similar, but with varied and unwanted at the extreme line of their extinction and suffered greatly by narrowing of their habitats. This inconsistent environment affects *Deinotherium* and constrains the genus towards the likelihood of migration as well as extinction from the region.

CONCLUSIONS

The current results of EH analysis indicate that the *Deinotherium* clade (proboscideans), the giant land mammals, were seriously affected at adult stages by the ecological pressures during the Middle Miocene. The presence of *Deinotherium* in the Pliocene of the Indian Siwaliks and even Pleistocene from Europe and Africa supports the idea of migration of some members of this taxon from the Siwaliks (Colbert, 1935), to the related habitats. Even though non-significantly increased amount of EH in *D. pentapotamiae* compared to *D. indicum* supports the hypothesis of their difference in body size and response to the ecological fluctuations. Hence, the data of change in vegetational patterns and feeding activities, presence of simple dental structures specific for browsing activity, vertical arrangement of teeth in the jaw (horizontal in other and descended proboscideans) contributed to these high occurrences of EH, which indicated that ecological stress was occurring more, especially vegetational stress, which can be one of the major reasons for the likelihood of disappearance of these herbivores from the Siwaliks of Pakistan at the end of the Middle Miocene towards the onset of Late Miocene.

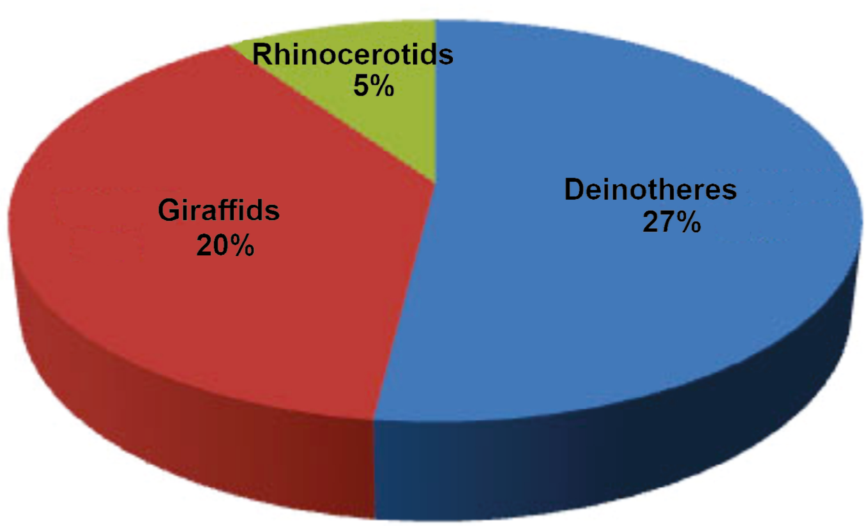

Figure 4. Comparison of the occurrence of EH in the different Siwalik mammalian families.

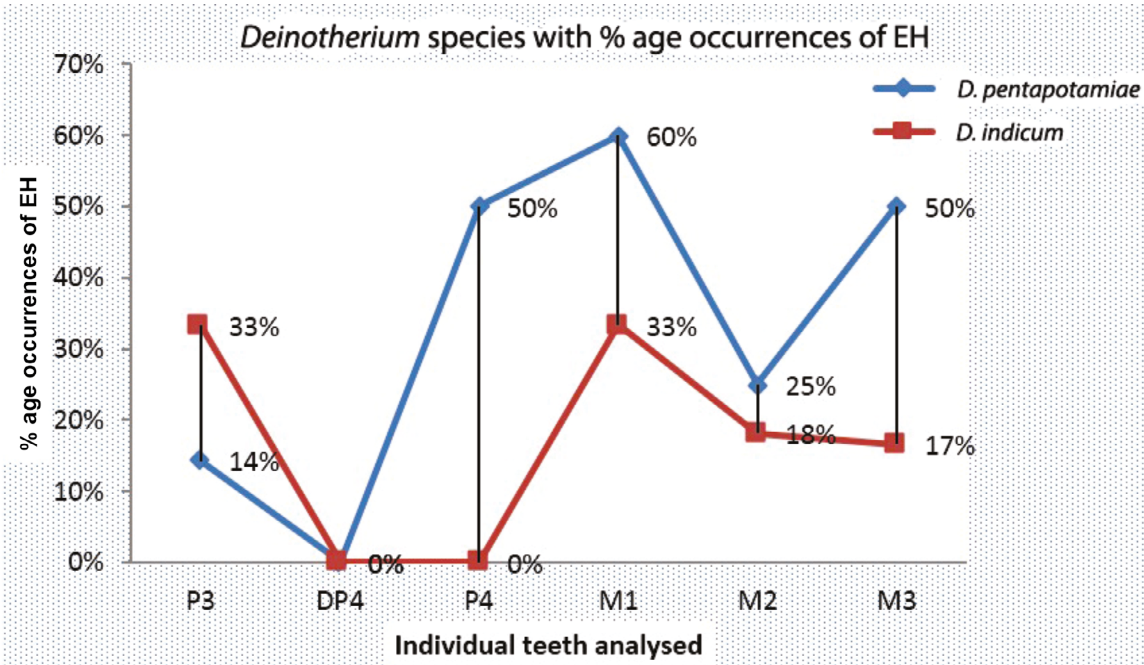

Figure 5. Graph showing the difference of EH in upper *vs* lower teeth of both species, *Deinotherium pentapotamiae* and *D. indicum*, of the Siwalik, Pakistan.

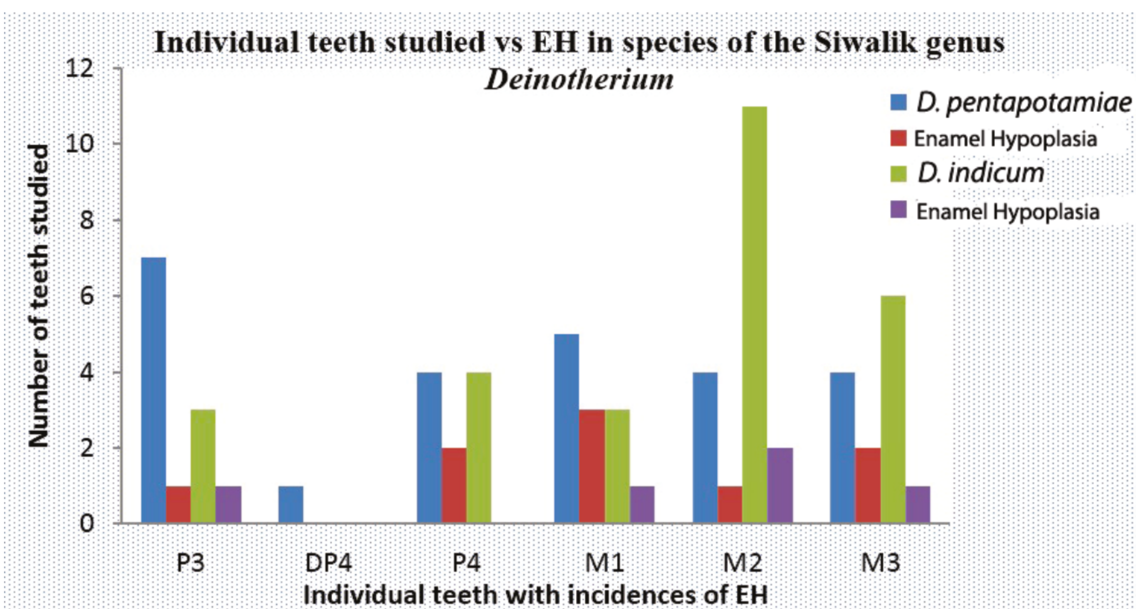

Figure 6. Difference in percentage occurrence of EH in upper and lower dentition from both species, *Deinotherium pentapotamiae* and *D. indicum*, of the Siwalik, Pakistan.

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