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PATHOLOGIC DINOSAURIAN EGG SHELLS FROM KHEDA DISTRICT, GUJARAT

D. M. MOHABEY
11-B, Sudershan Society, Naranpura, Ahmedabad 380 013, India.

WELL preserved dinosaurian egg nests have recently been discovered in infratrappean limestone of Kheda district, Gujarat[1]. In the area under review, the total thickness of the infratrappean sequence is about 6 m. The sequence starts with about 1 m thick conglomerate at the base, overlying granitoids and pegmatoids. This conglomerate which is very rich in dinosaurian bones, grades upward into calcareous pebbly sandstone which grades upward into siliceous limestone and further into pure limestone. The thickness of the limestone is about 3 m and the egg clutches are confined to the upper part of this limestone (figure 1).

The SEM study carried out by the author[2] has revealed the presence of two types of egg shells comparable to tubocanaliculate and angusticanaliculate types[3]. The normal egg shells show only one complete egg shell layer of tubercles (spheroliths).

Two types of pathologic tendencies are known so far in dinosaurian eggs. One is reduction in thickness

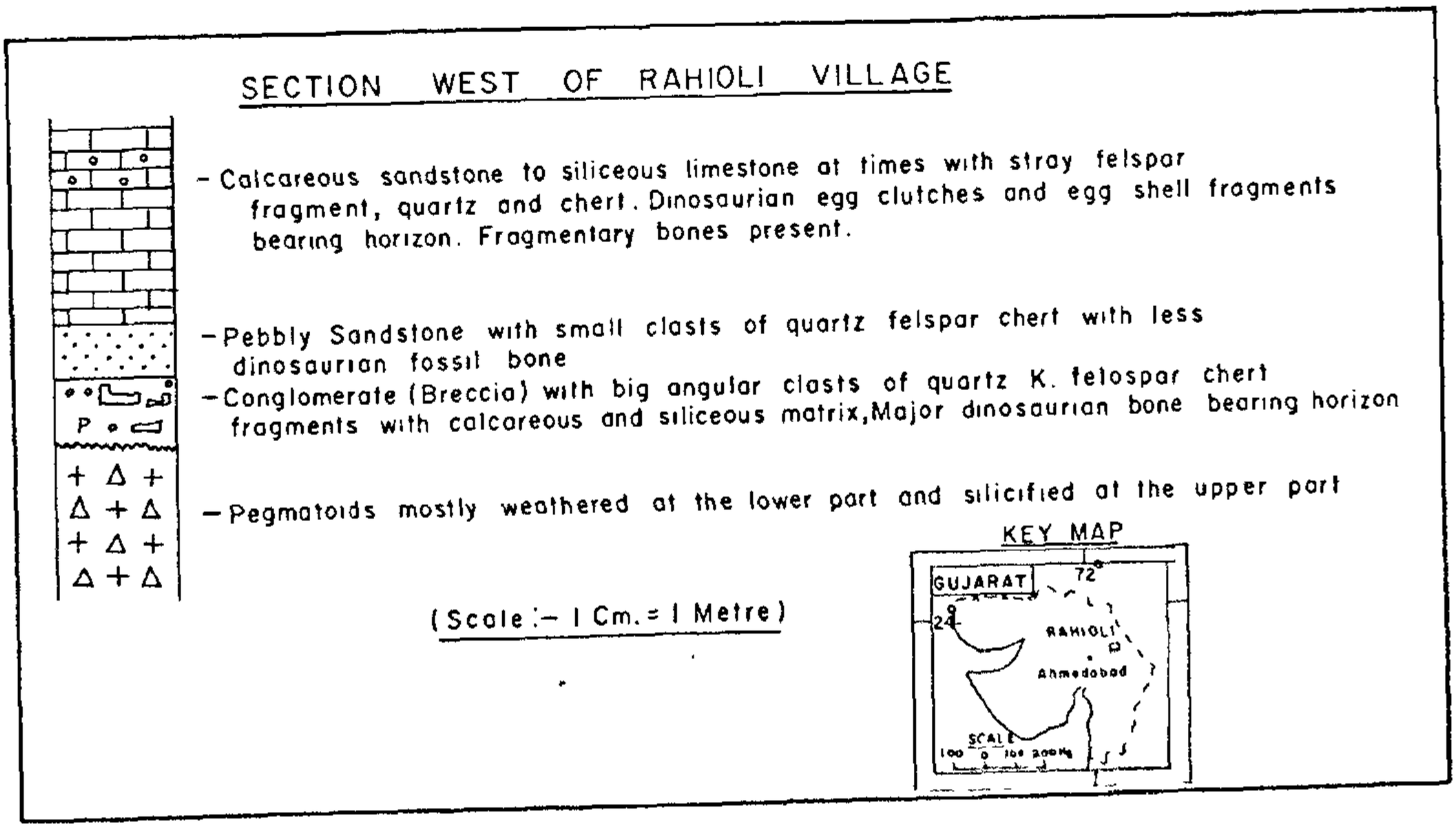

Figure 1. Section west of Rahioli village.

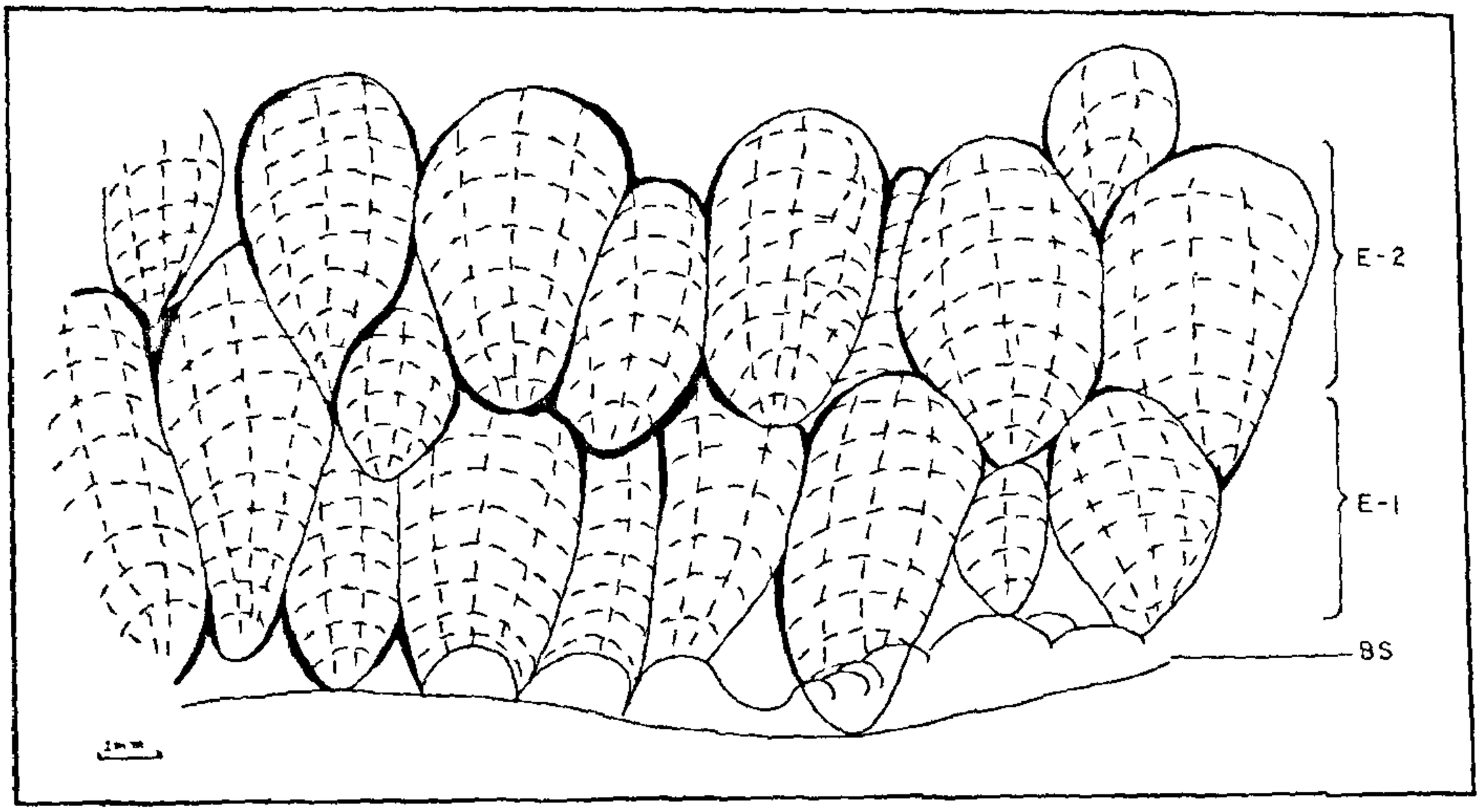

Figure 2. Pathologic ovum-in-ovo condition in dinosaur egg shell under the microscope showing repetition of two egg shell layers. (Magnification about 50 ×) E–1. Egg shell layer 1., E–2. Egg shell layer 2., BS. Basal zone.

of the shell which would lead to collapse of such eggs under the weight of the breeding animal. The abnormally thin egg shell would also lead the excessive evaporation of the liquid content of the eggs leading to dehydration of the embryo. The second pathologic condition is abnormal increase in thickness of the egg shell due to the repetition of the egg shell layers[4]. This condition is also known as ovum-in-ovo condition. The repetition of egg shell layers, would result in blocking of canaliculae of the inner shell with superimposed secondary shell and thereby resulting in the suffocation of the embryo. Thus under both pathologic conditions the effect would be lethal on the embryo resulting in perishing of the embryo, before hatching.

Study of some of the dinosaurian egg shell fragments collected from infratrappean limestone near Rahioli village (73° 21′ 22″:23° 03′ 25″), has indicated the presence of ovum-in-ovo type of pathologic condition. Microscopic study shows the presence of atleast two egg shell layers, one superimposed over the other. Both the layers show complete structure amongst themselves (see figure 2). The mammallary knobs of the superimposed layer, block the canaliculae of the underlying layer. Both the layers consist of individual tubercles with subvertical diverging and concentric lines. The individual tubercles do not show any lateral interlocking.

It is known that pathologic condition in living reptiles and birds, is caused by harmonal imbalance which can be induced by psychic stress due to overcrowding in restricted space, sudden change in the environment etc[4]. The factors like pollution by insecticides and pesticides also result in pathologic condition[5,6]. Thus there are many factors which directly or indirectly result in pathologic condition, but in the present case of dinosaurian pathologic egg, it is difficult to determine the specific activating factor. The pathologic condition must have resulted in the death of embryo before hatching causing the extinction of dinosaurs.

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PSEUDOMONAS FLUORESCENS IS AN ANTAGONIST TO *XANTHOMONAS CITRI* (HASSE) DYE, THE INCITANT OF CITRUS CANKER.

N. UNNAMALAI and S. S. GNANAMANICKAM
Centre of Advanced Studies in Botany,
University of Madras, Madras 600 005, India.

THE terms 'antibiosis' and 'antagonism' refer to the reduction in growth and activities of organisms living in association[1]. Many bacteria and fungi are known antagonistic agents. Several studies have suggested that *Pseudomonas* spp. may play a role in reduction of plant diseases[2–7]. *Pseudomonas fluorescens* produces a siderophore which complexes the environmental iron making it less available to other pathogens and thus reduces disease severity[4].

From cankered citrus leaves collected from three locations of Madras, cultures of *Xanthomonas campestris* pv. *citri* (*X. citri*) were isolated by using nutrient agar plates. The surface-sterilized leaf bits and twigs yielded a fast growing yellowish green fluorescent bacterium which prevented the development of *X. citri*. Subsequently *X. citri* was isolated on NSCA (selective) medium[8].

Biochemical tests were made to characterize the yellowish green fluorescent bacterium abundantly associated with *X. citri* in mature cankers. The results from these biochemical tests (table 1) suggest that the organism is *P. fluorescens*. Further these two strains which were positive for levan production, denitrification, and used sorbitol and ethanol as sole carbon sources were identified as biotype I of *P. fluorescens* as outlined in Bergey's manual[9].

P. fluorescens strains were further characterized and they produced the siderophore (fluorescent pigment) only in low iron containing King's medium B (KB)[10]. When KB was amended with 1 μM of $FeCl_3$, the siderophore production was suppressed. Therefore this bacterium which showed properties of siderophore-producing rhizobacteria[11] was tentatively considered a plant growth-promoting rhizobacterium (PGPR).

In two methods of *in vitro* bioassays (filter paper disc assay and agar well method), *P. fluorescens* cell suspension or a preparation of siderophore inhibited the growth of *X. citri* (figures 1 and 2). The siderophore was prepared following the method of Scher and Baker[10]. In another set of *in vitro* test, *X. oryzae*, the incitant of bacterial leaf blight of rice was also inhibited. Tests were also performed to check if this PGPR like *P. fluorescens* has growth-promoting activity following the method of bacterization of seeds described recently by Scher *et al*[12]. In rice seedlings (cv. IR-20) which were bacterized with *P. fluorescens* (2 strains) and grown in test tubes, shoot measurements were higher (average 5.9 cm) for one of the strains. In non-treated (control) plants, the average shoot length was 5.2 cm. The second strain of *P. fluorescens* did not have the growth-promoting activity and therefore the results are considered inconclusive.

The association of PGPR-like strains of *P. fluorescens* with citrus canker pathogen *X. citri* is reported for the first time from India. While this study was nearing

Table 1 *Biochemical tests for characterising fluorescent pseudomonas strains (of the plant growth promoting rhizobacteria (PGPR) type).*

	Pseudomonas fluorescens	
Test	AHS-PGPR strain*	UC-PGPR strain**
Gram reaction	– ve (small rods)	– ve (small rods)
Levan formation	+	+
Oxidase test	+ (1 sec)	+ (3 sec)
Gelatin liquefaction	+ (2 days)	+ (3–4 days)
Arginine dihydrolase production	+	+
Denitrification	+	+
Utilisation of carbon sources:		
a) Trehalose	+	+
b) Sorbitol	+	+
c) Ethanol	+	+
Antagonism to		
X. citri	+	+
X. oryzae	+	+

+ denotes a positive reaction to the test and – denotes a negative reaction; * AHS refers to strain collected from Agri-Horticultural Society Gardens and ** UC refers to strain collected from University Campus (Madras University).