Observations and interpretations on past microbial activities at Eppawala Phosphate Deposit

1 author:

Nalaka Deepal Subasinghe
National Institute of Fundamental Studies - Sri Lanka

OBSERVATIONS AND INTERPRETATIONS ON PAST MICROBIAL ACTIVITIES AT EPPAWALA PHOSPHATE DEPOSIT

N. DEEPAL SUBASINGHE

Institute of Fundamental Studies, Hantana Road, Kandy, Sri Lanka

E-mail: *deepal@ifs.ac.lk*

ABSTRACT

An extensive, economically viable phosphate deposit exists at Eppawala in the north central Sri Lanka. Textures and certain microscopic features found in this ore body had led to the speculation of a sedimentary origin for this deposit, although the parent rock is of igneous origin. Detailed investigations reveal that this deposit is a result of weathering followed by in-situ diagenesis. This phoscrete-type phosphate deposit has been developed on an apatite-rich carbonatite body. In certain sections of the ore, microbial activities were detected under the scanning electron microscope as well as the light microscope. These microbial activities might have started and ceased during the formation of secondary phosphates. Currently micro-organisms are preserved in cavities and silicified areas of the secondary phosphatic matrix. Morphology and the dimension of different microbial components point towards the presence of more than one group of microbes. This paper attempts to categorise some of the observed microbes as well as understand the role played by them, using photomicrographic and mineralogical evidences.

Keywords: *Eppawala, apatite, carbonatite, microbial activity, Sri Lanka*

INTRODUCTION

Eppawala phosphate deposit, located in the Anuradhapura district of Sri Lanka, about 200 km from Colombo, was discovered in 1971 (Jayawardana, 1976). This deposit exists as a dense weathering profile underlain by an apatite containing carbonate rock. The ore bodies at Eppawala contain up to 42% P_2O_5, and citric acid solubility of different components varies from 4 to 6%. Owing to intense weathering of the apatite-bearing carbonate rock, a phosphate deposit has been formed by relative accumulation of the primary apatite crystals, together with the secondary products derived from them (Dahanayake and Subasinghe, 1988; 1989a; Subasinghe 1998).

The Eppawala phosphate deposit, which is a secondary deposit formed over a parent carbonatite rock display some features of a sedimentary phosphate deposit. The term "*Phosphorite*" is generally used for sedimentary phosphates of marine origin. However, the phosphate deposits created by terrestrial processes, such as *in-situ* weathering and diagenesis can also be referred to as phosphorites (*sensu-lato*) despite their original source. Dahanayake and Subasinghe (1988, 1989a, 1990a, 1991) used the term phosphorite to describe the phosphate deposit at Eppawala in this context. Eppawala deposit was formed by relative accumulation of primary apatite and diagenesis of secondary phosphates. However, since those processes took place in terrestrial settings rather than marine or lacustrine, it is more appropriate to call the deposit as a *phoscrete-type phosphate deposit* (Dahanayake and Subasinghe, 1991; Subasinghe, 1998).

In many phosphate deposits in the world, especially in sedimentary 'phosphorite' deposits, microbial processes have played a significant role. According to Hirsch *et al.* (1995), microbial rock degradation is independent of climate, or location, or annual seasons. Dahanayake and Krumbein, (1985) studied the microbial mats and emphasised their importance in the formation of phosphorites. Soudry and Champetier (1983), reported the microbial processes in the Negev phosphorites in southern Israel. Soudry (1987) presented a detailed account on the same deposit, and discussed the ultra-fine structures and possible role of biological activities in the genesis of phosphorite. Comparing the structures presented by the above workers, Dahanayake and Subasinghe (1988; 1989b) also suggested some possible influence of microbial activities in the formation of the terrestrial phosphate deposit at Eppawala, Sri Lanka. Soudry (1992) presented further analogy on phosphorites from Negev, Israel. The numbers, growth rates and productivity of heterotropic bacteria in sediments of the continental-margin of Eastern Australia were determined and discussed by Moriarty *et al.* (1991). Reimers *et al.* (1990) also presented evidence for phosphate mineralisation by microbial mat formation. They studied and compared the present day microbial mats with those found fossilised in Monetary Formation, California. Working on phosphatic hardgrounds and stromatolites in Central Greece, Pomonipapaioannou and Solakius (1991) considered that phosphate was supplied to the pelagic sediment by a bacterially controlled precipitation from interstitially circulated solutions.

On the other hand, there are many studies confirming that microbes, especially those found in terrestrial environments, can solubilise the phosphate, thus increasing the availability of phosphate to the plants (e.g. Panhwar *et al.*, 2012; Khan *et al.*, 2009; Ivanova *et al.*, 2006). In a petrological context, these microbes have a destructive effect on the phosphatic rock, rather than constructive.

Microbial activities were observed in the secondary matrix of the phosphate rock at Eppawala. Morphological information obtained through electron microscopic observations are the main data available on them.

Discovery of microbes in the phosphatic saprolite prompted a question about the role of the microbes. It is necessary to understand whether the texture of the saprolite was significantly affected by the microbes (dissolving or forming phosphate minerals and textures) or, whether microbes are playing a passive role and only inhabit the areas favourable for them without considerably affecting the already existing textures.

In this paper, the author presents strong evidence on the presence of microbes and their activities in the Eppawala Phosphate deposit with the intention of understanding the nature and the roles they would have played in bringing the phosphate deposit to its current state. This is a tribute to Prof. Kapila Dahanayake who put forward the possibility of the existence of microbes in the Eppawala phosphate more than 2 decades ago (Dahanayake and Subasinghe, 1988; 1989b).

GEOLOGICAL SETTING AND PREVIOUS STUDIES

The apatite-bearing carbonate rocks at Eppawala, occur as massive, discontinuous bodies in a Precambrian, high-grade metamorphic terrain. Based on the field observations, mineralogy and petrology, this carbonate rock has been assigned an igneous origin, and thus called *carbonatite* (Jayawardana, 1976). Later, geochemical work including oxygen and carbon isotopic studies confirmed the igneous origin Subasinghe 1998; Weerakoon *et al.*, 2001; Pitawala *et al.*, 2003; Manthilake *et al.*, 2008; Pitawala and Lottermoser, 2012). Therefore, the term 'carbonatite' is used hereafter to indicate the parent carbonate rock with primary apatite crystals. The lithological units within the carbonatite complex could be grouped into those of the basement and carbonatite. The economic phosphate deposit has been developed as a secondary weathering profile over the parent carbonatite rock. Very large primary apatite crystals of up to 1 m in length can be observed occasionally in the parent carbonate rock. These primary apatite grains are large, yellowish green crystals with subhedral to euhedral shape.

As shown in Figure 1, the phosphate deposit at Eppawala is located close to the boundary between Wanni and Highland complexes. Granitic gniesses, granites, quartzites, charnockitic gneiss, hornblend-biotite gneiss and pegmatitic rocks can be observed in the surrounding area.

In the carbonate rock (the parent rock), calcite, dolomite and apatite are major minerals, whereas magnetite, spinal, pyrite, and ilmenite occur as important accessories. In addition, forsterite, phlogopite, enstatite, magnesite, martite, diopside, tremolite and few other minerals also occur in smaller amounts.

Secondary material mainly consists of carbonate- and iron-phosphate varieties (Dahanayake and Subasinghe, 1989; Subasinghe 1998) with relict primary apatite, magnetite, mica and few other minerals. Formation of secondary phosphate minerals and the observed textures have been studied in detail by previous researchers (Dahanayake and Subasinghe, 1988a; 1988b; 1989a; 1989b; 1989c; 1990a; 1990b; Subasinghe 1998). Further, Subasinghe (1998) stressed the importance of iron in the process of formation of secondary phosphates and discussed the potential use of this natural phenomenon in material science applications (Subasinghe, 2012).

Field observations indicate that the phosphatic saprolite profile at Eppawala more or less resembles a supergene weathering profile, having loose lateritic material on the top and more consolidated material towards the bottom. It also indicates that the phosphatic saprolite is more resistant to chemical weathering than its parent rock or the siliceous saprolite. Borehole data show that the phosphatic saprolite is always

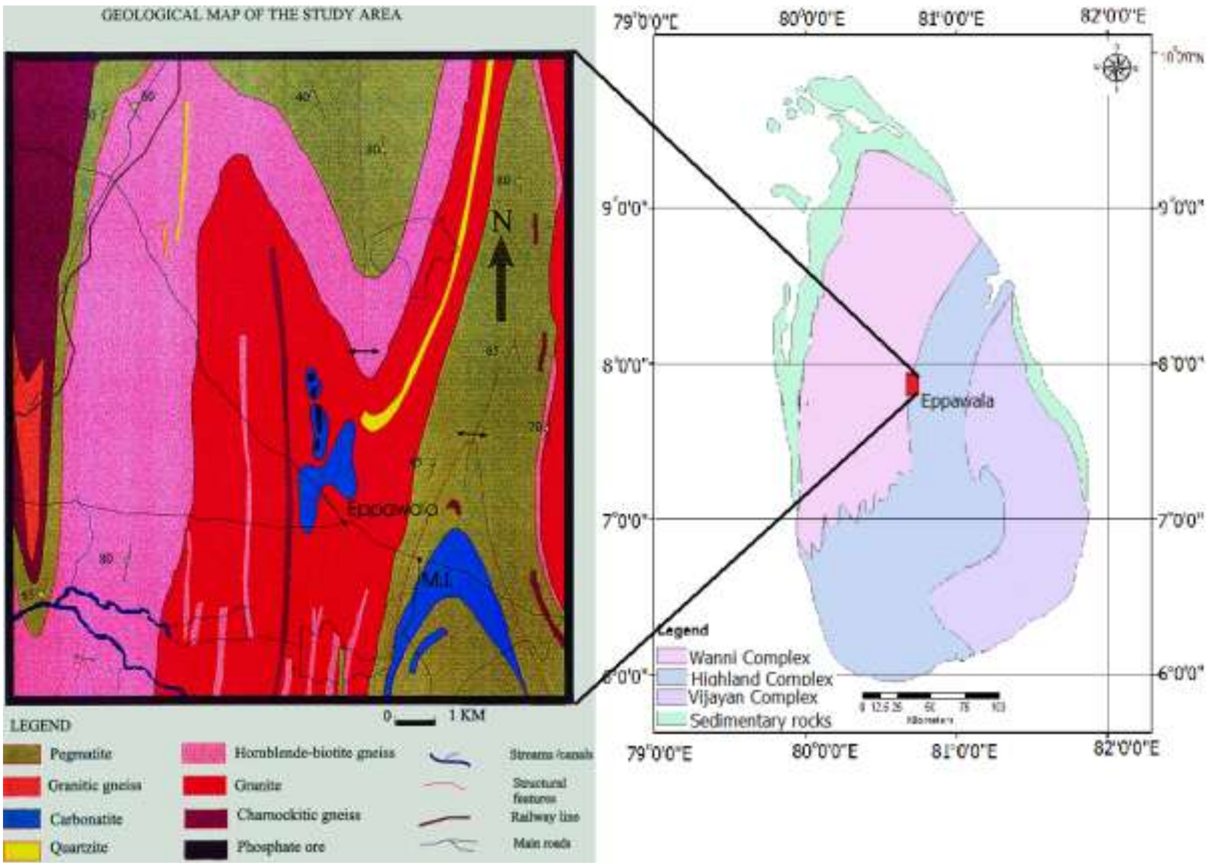

Figure1. Location and Geology of Eppawala Phosphate deposit (Subasinghe, 1998, modified after Jayawardena, 1976).

overlain by weathered material; either phosphatic lateritic soil or siliceous lateritic soil, or, siliceous saprolite. Also, at no place was siliceous saprolite or laterite found underneath the phosphatic saprolite cover. These observations are explained if supergene weathering is considered.

MATERIALS AND METHODS

Samples were collected from different regions of the Eppawala phosphate deposit to represent different weathering stages and varying mineralogy. Partially weathered carbonatite samples as well as secondary phosphate with detrital primary apatite crystals were included in the sampling. More interesting secondary textures with well preserved and relatively less disintegrated samples were found towards the southern end of the deposit. Fresh samples were stored in sample bags and brought to the laboratory. Samples were cleaned, dried, surface sterilised using UV light, and stored in a cold room to avoid any biological contamination, until they were used for analysis. Sub samples were selected from well consolidated samples for SEM studies.

Handspecimen Studies

Handspecimens were inspected using a hand lens. Certain samples were cut and polished to show specific features photographically whereas some samples were observed with their naturally broken surfaces.

Scanning Electron Microscopy (SEM)

Selected samples were broken in to small pieces using gentle fracturing so that natural features will be clearly seen. These samples were mounted on metal sample stubs and coated with either gold or carbon. Samples were examined in JEOL JXA-840 scanning microanalyser and JEOL JSM5300 scanning electron microscope at 20KV accelerating voltage and usual settings. Magnification range used for the analyses is from 10 x to 10,000 x. Qualitative elemental analysis was carried out at certain points of interest, using energy dispersive X-ray analysis. This was mainly used to identify the major element composition to confirm the identity of a suspected mineral grain or feature.

Some samples were prepared as polished sections so that they could be observed under SEM as well as the light microscope, consequently.

RESULTS AND DISCUSSION

Parent material, the apatite-bearing carbonatite rock, is shown in Figure 2. Note the white coloured unweathered carbonatite matrix at the bottom, and the development of brown coloured secondary material at the top part that is exposed to meteoric weathering. Some meteoric water penetrates into the rock through the fissures and fractures of the rock. They leave a brownish stain in the rock. Primary apatite crystals are more resistant to weathering than the carbonate matrix, thus getting accumulated when the carbonate matrix is partially removed and converted to secondary matrix, which is usually light to dark brown in colour.

Figure 2. Partially weathered carbonatite rock with small primary apatite crystals (pale green), dissolution cavities and partially developed secondary material (brown colour). Width of the photograph is about 11 cm.

Secondary materials appear as a fine-grained matrix to the naked eye. However, under the SEM, they display crypto-crystalline hexagonal apatite crystals, clay minerals, detrital silicate minerals and apatite grains and most interestingly, some microbial activities at certain places, as shown in Figure 3.

Close observations show that microbial activities are in several stages of development. As shown in Fig. 4, at initial stages of microbial activities, *bacillus* shaped connected chains of microbes are observed on the apatite crystals. At this stage, apatite crystals show little or no deterioration. However, microbes are firmly attached to the apatite crystals and spread their filamentous material across them indicating their affinity for phosphate.

At advanced stages of microbial activities, apatite crystals had been dissolved from all directions, as shown in Figures 5-7. Diameter of the *bacillus* material is in the order of 1 µm on average, while that of the filaments is less than 0.5 µ.

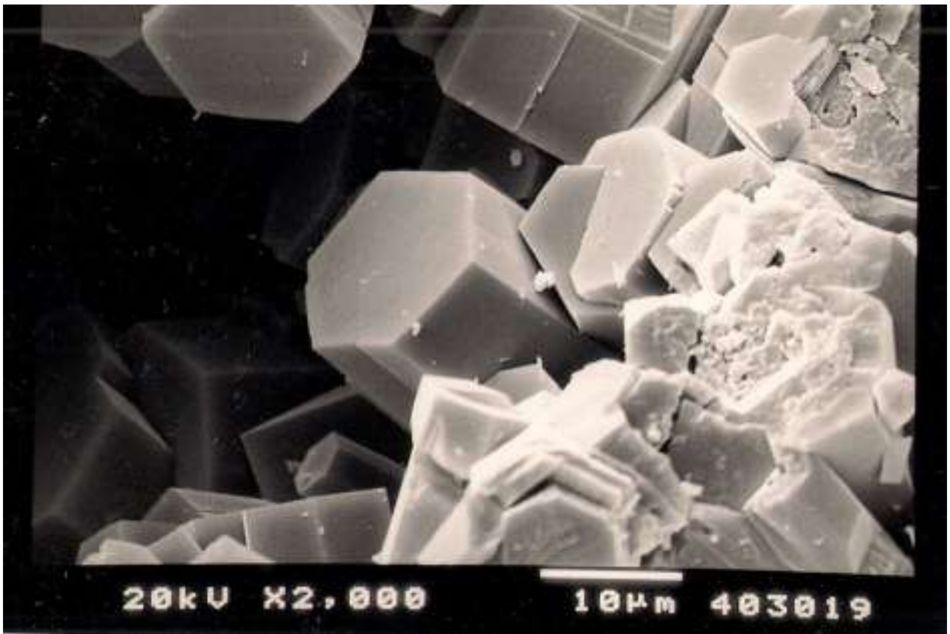

Figure 3. Microscopic apatite crystals under SEM. No microbes are seen in this sample and apatite crystals are intact with no indication of dissolution

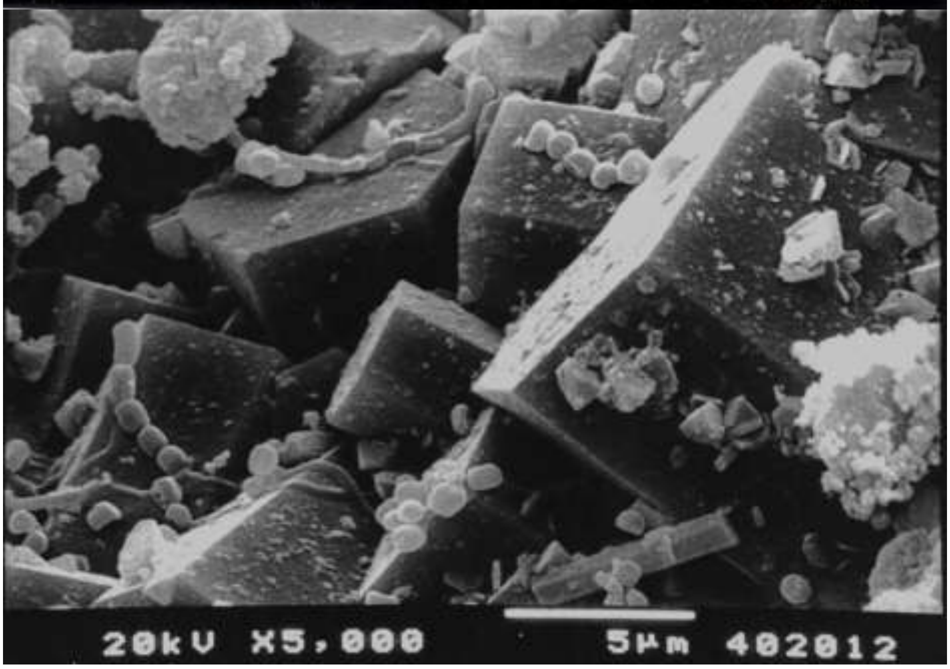

Figure 4. Initial stages of some microbial activities in the secondary matrix of Eppawala phosphate. Note the bacterial material and their filaments attached to the hexagonal apatite crystals

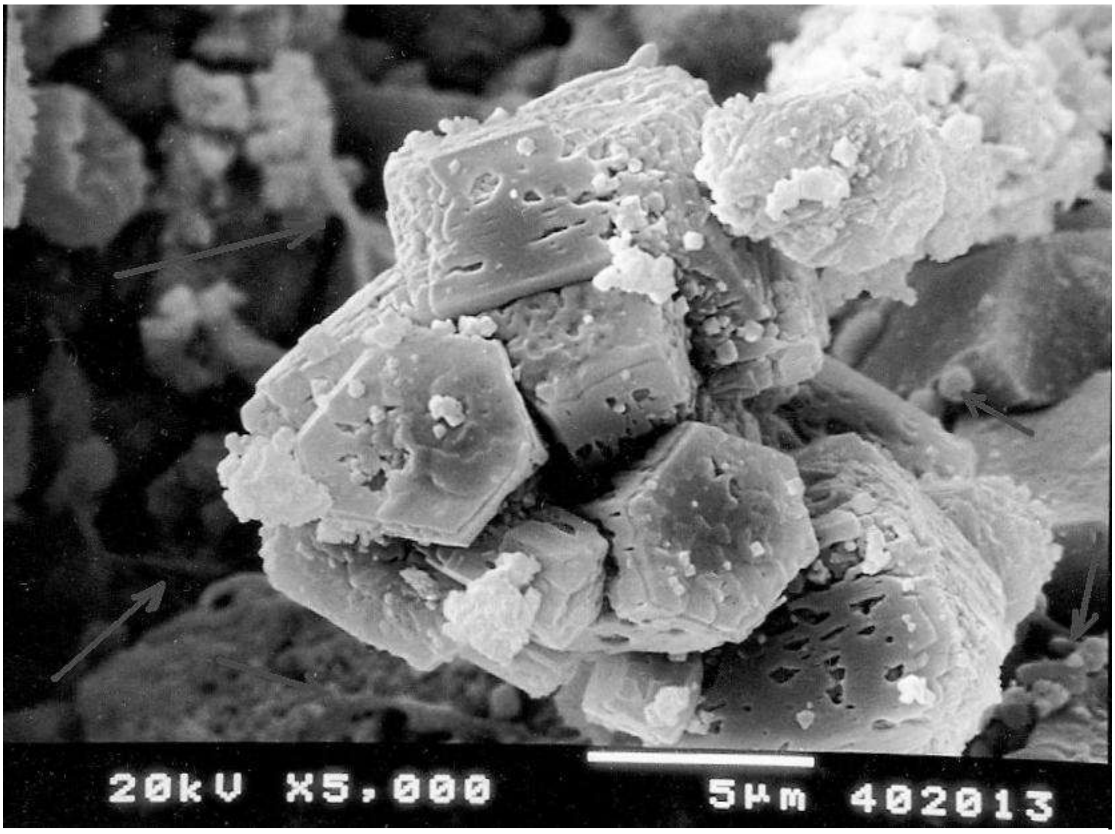

Figure 5. Microbial activities (arrows) cause apatite crystals to dissolve from sideways, i.e. parallel to prism faces. Compare with Figure 3

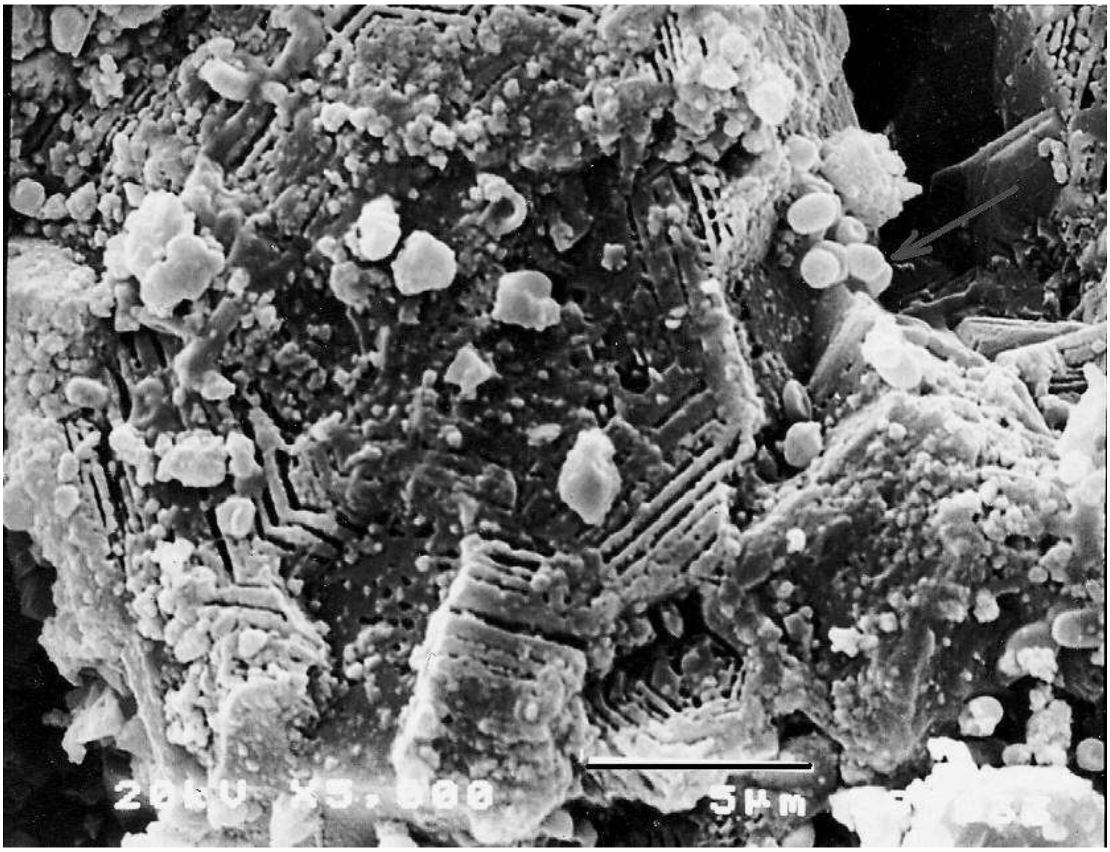

Figure 6. Hexagonal cross-sectional view of apatite crystals showing the inter-planar dissolution, probably due to secretions resulting from microbial activities (arrows)

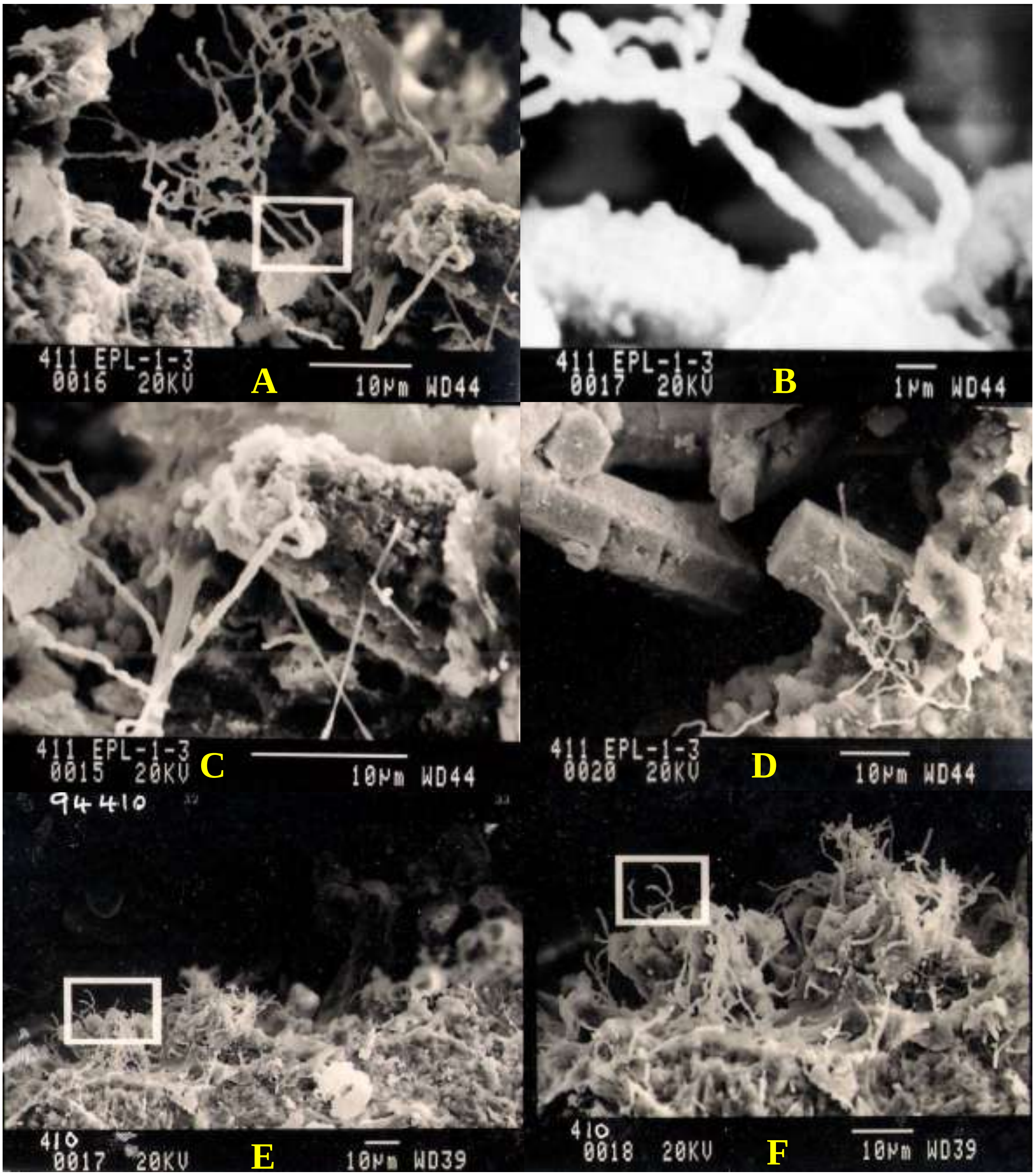

Figure 7. Filamentous bacterial colonies in advanced stages of microbial activity. B and C shows enlarged views of the photo A. D-Partially dissolved apatite crystals and some microbes. E and F show thick microbial colonies. Note the size of the microbes and their filaments

Figure 7 shows a collection of different views of microbial activities in the secondary phosphatic matrix. It is clear that the size of the *bacillus*-shaped interconnected microbial material have diameters smaller than 1 µm whereas filaments are also show similar dimensions (Fig. 7 A and B). Such dimensions are typical of bacteria. Usually, fungal materials are several times larger in dimension. The presence of similar size filaments suggests that bacterial origin and not fungal origin. This indicates that the microbial material commonly observed as secondary material at Eppawala phosphate deposit are filamentous bacteria (actinobacteria).

Figure 8 shows a special feature observed in the secondary matrix. However, this feature was observed in an area rich in siliceous material. The morphological features of Figure 8 may suggest a detached fungal spore. However, for a detached fungal spore to survive the sample preparation procedure as well as the high vacuum and high voltage in the SEM, it should be a hard material firmly attached to the substrate, the siliceous host mineral. Further, it should be fossilised, since the fungal spores do not have hard cell walls. Also, the fungal spores do not have perfectly circular shapes or long, relatively thin stems between each spore. Further, no fungal spores are observed in any of the samples despite the fact that some of the samples are densely populated with different types of microbes. Significant morphological and dimensional differences, as well as other factors as mentioned above strongly suggest that the microbes shown in Figure 8 cannot be displaced conidiophores.

In addition, microprobe analysis of the host mineral as well as the microbial feature reveals high silica contents, suggesting that they are made of quartz or other silica rich mineral. Therefore, it is unlikely that Figure 8 is an image of a detached and fossilised fungal spore. In fact, perfect geometrical shape and regular distance between circular grains are characteristics of diatoms (Barber and Hayworth, 1981; Mann, 1999). The existence of this material in a silica rich mineral is another positive factor to support that this could be a valve view of interconnected silica-secreting diatoms**.**

When the role of the microbes is considered, there are two possibilities to consider: Whether (a) microbes are playing an active role in producing secondary phosphates, as in certain marine deposits, or (b) if they are solubilising and consuming some phosphate. The extent of the microbial activities and the overall effect on the phosphate deposit is the other factor to be considered.

Extensive work carried out by the author as well as other researchers on the deposit helped establishing the present state of the phosphate deposit (Dahanayake and Subasinghe, 1988, 1989a, b, 1990b, 1991a; Subasinghe, 1998; Weerakone *et al.*, 2001; Pitawala *et al.*, 2003). Geochemical, mineralogical and sedimentological evidence supported by isotope

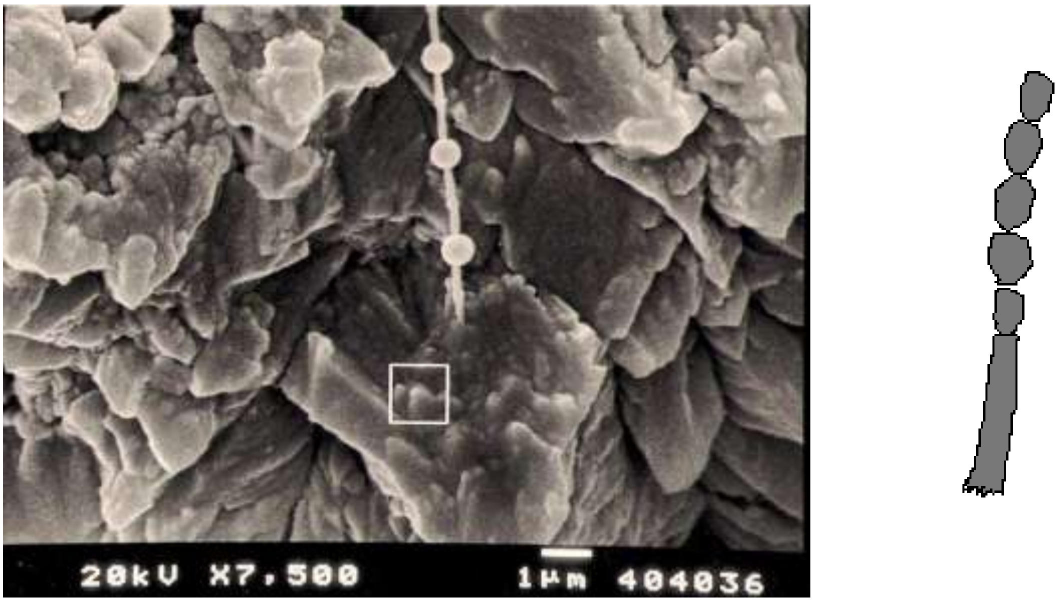

Figure 8. Another type of microbial material observed in the matrix. Note the perfect circular shape and regular distances maintained between each circular feature. Area marked with a square was analysed for elemental composition and found that it is predominantly silicon. A sketch of the typical conidiophores is shown on the right for comparison. Significant morphological differences are evident.

results strongly suggest that the secondary phosphate material had formed as a result of meteoric weathering acting on the parent carbonatite rock containing primary apatite crystals.

As there are good explanations for the formation of secondary matrix through inorganic (non-biological) weathering and *in-situ* diagenetic activities (Subasinghe, 1998; Tazaki *et al.,* 1986; 1987), it can be deduced that the microbial activities do not play a critical role in producing and shaping the phosphate deposit at Eppawala.

However, it is still interesting to study these microbial activities; firstly, this is the first time such activities were recorded in Sri Lankan phosphates. Dahanayake and Subasinghe (1988) suggested microbial involvement in the formation of secondary phosphates, although they could not provide direct strong photomicrographic evidence for their claims at the time, due to technological limitations. Secondly, if the observed microbes are fossilised ones, and if their age can be determined, identifying them would help understand the environmental conditions at the time of their formation. Thirdly, if the role of the microbes is the solubilisation of the phosphate, there is a possibility of producing biofilms utilising the phosphate solubilising microorganisms isolated from this area to increase the availability of phosphate.

Unless specially prepared, biological material such as soft tissues containing water cannot survive under high-vacuum and high voltage electron beams used in the scanning electron microscopes. Any of the observed features did not show deformation or destruction under the electron beam, confirming that they were well-preserved hard material.

This provides two possibilities: Possibility one is that these were bacteria-like soft material that lived, died and fossilised. Since bacteria have no hard cell walls, they can only be preserved as fossils.

Other possibility is that the above may be microbes with hard cell walls. It is a known fact that desmids and diatoms secrete silica into their cell walls.

Remnants of possible microbial colonies were occasionally observed in thin sections, as shown in Figure 9. These thin sections show the hexagonal outlines or 'ghost structures' of previously existing apatite crystals. Currently silica is filled in the outlines, which appear as hexagonal cavities under plane polarised light (PPL), while secondary quartz is observable under crossed polars (Figure 9 B). These observations confirm that the microbes have played an active role in removing phosphates when the silica was in abundance. Silicification process that followed the microbial activities has preserved the visible evidence of microbial colonies that existed.

According to available evidence, it is highly possible that microbes observed in Figure 8 were a diatomaceous species. The microbes might have entered the rock with rain water passing through pores and fractures developed after the initial stages of weathering and settled in the small cavities of the secondary matrix. Konhauser (1997) reported iron mineralisation by microbes. These bacterial colonies can leave dark brown stains in the rock, even after their death. Some dark brown stains found in the silicified areas (e.g. Figure 9) of the Eppawala phosphate could be considered as similar stains left by microbial colonies.

Furthermore, although not commonly encountered, larger filamentous material could also be observed in certain samples. As shown in Figure 10, these materials have diameters of the order of 10 µm (10-30 µm). This dimension is comparable with that of the common fungal filaments. Backscattered images (Figure 10-B) show darker colour in these filaments indicating that they consisted of lighter elements, possibly carbon or silicon, compared with the surrounding phosphatic matrix.

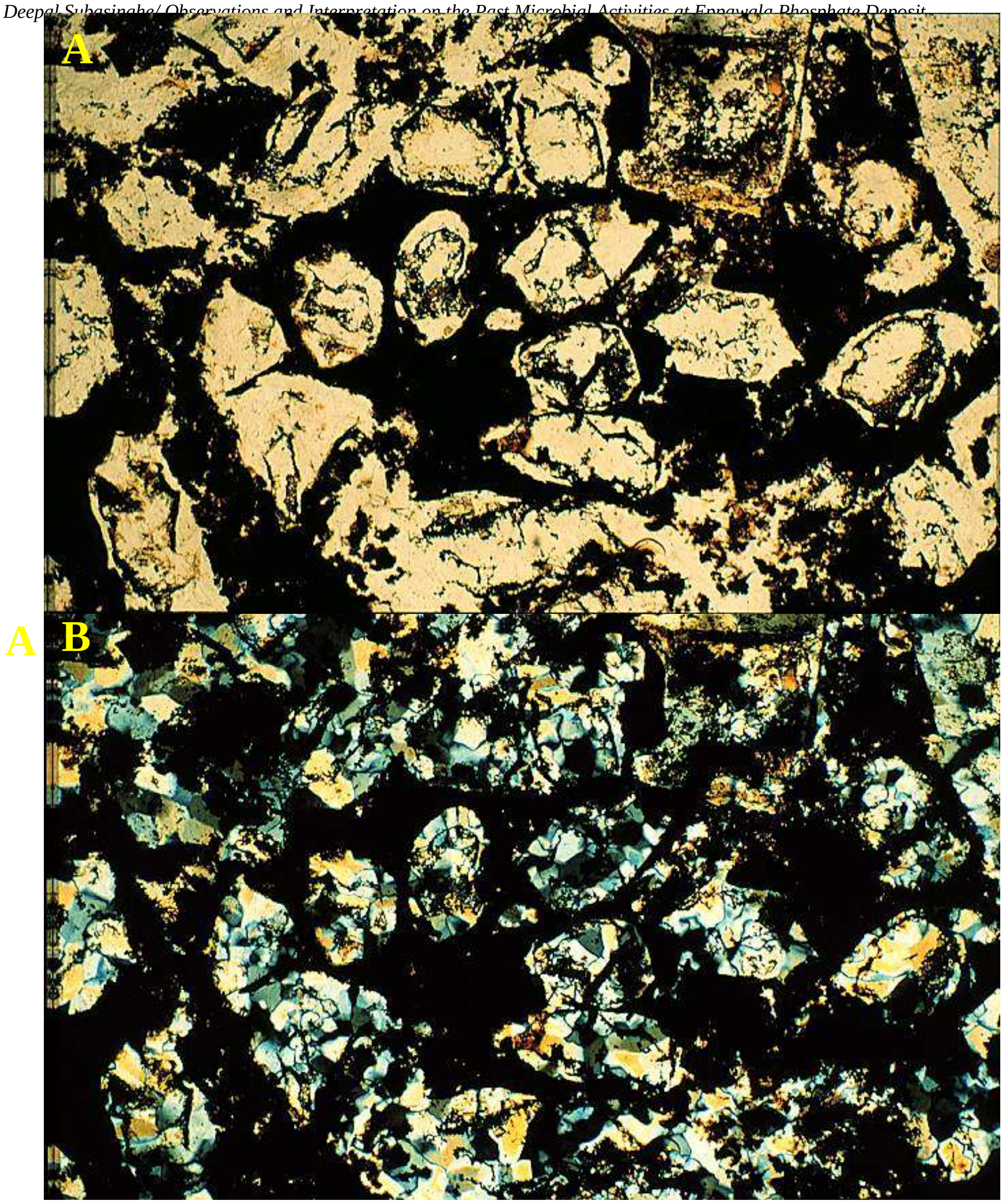

Figure 9. Hexagonal outlines of previously existing apatite crystals preserved in dark secondary material. A.- hexagonal shaped cavities-ghost structures-left by removal of primary apatite crystals (Under PPL). B. Under CPL quartz infillings can be observed. Some dark filamentous materials inside the hexagonal outlines indicate possible existence of microbes during the removal of apatite.

Actino-bacteria were reported in a variety of environments including rocks and building materials. Suihkoa *et al.* (2007) reported hyaline fungal species, diatom algae as well as filamentous bacterial growths on samples taken from the inner and outer surfaces of stone monuments of six historic Scottish buildings and ruins. Morphological features of some of the microbes reported by them have close resemblance to those found at Eppawala samples. SEM images of the microbial mats reported by Reimers *et al.* (1990) also closely resemble some of those found at Eppawala.

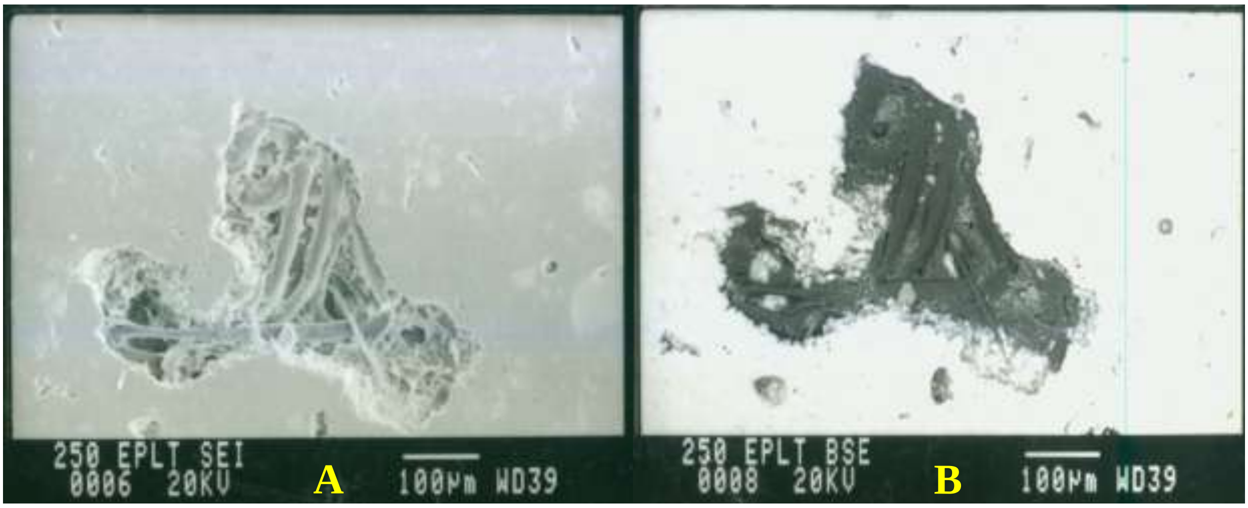

Figure10. Fungal filaments in the phosphatic matrix. A- Secondary emission image; B- under Back Scattered Electrons (BSE), filaments appear darker. Note the scale

Secondary matrix contained detrital primary apatite crystals as well as crypto-crystalline grains. When the conditions were favourable, microbes could have acted to solubilise the apatite crystals while increasing the concentration of silica through their cell wall secretions. It is quite possible that these silica secretions had acted as nucleation material for the silicification process at Eppawala.

Although the shape of the microbial filaments resembles those of fungi, the size factor suggests that these are filamentous bacteria (actionobacteria) rather than fungi. Actino-bacteria (filamentous bacteria) require carbon dioxide for their growth (Round *et al.,* 1990). Weathering of the carbonate matrix releases CO_2, which could facilitate the growth of such microbes. Therefore, it is possible that the microbes observed in Eppawala samples are actinobacteria.

Natesan and Shanmugasundaran (1989) experimented with cultured cyanobacteria under controlled conditions to study their extracellular phosphate solubilisation, especially in the soil environment. Most of the reported microbes in terrestrial environments solubilise the phosphate rather than causing precipitation. Therefore, together with the microscopic evidence, it can be safely concluded that the role of the microbes at the Eppawala phosphate deposit is towards solubilising the phosphates, rather than initiating any precipitation process.

CONCLUSIONS

Microbial activities detected at the Eppawala phosphate deposit are relatively younger and play a passive role when it comes to the formation of new mineral textures. The extent of the activities is not fully evaluated; however, the observations indicate that they are confined to certain sectors of the deposit. Dimensions and morphological characters of the microbes indicate the presence of several species including action-bacteria, diatoms and fungi.

ACKNOWLEDGEMENTS

Postgraduate Research Institute for Sedimentology (PRIS) at University of Reading, UK provided the SEM facilities for this study. Professors K. Dahanayake and V.P. Wright are thanked for their support and encouragements in various ways. Discussions with Prof. S.A. Kulasooriya and Dr. R. Ratnayake, were extremely useful in recognising different types of microbes observed and in understanding their role.

REFERENCES

Barber, H.G. and Hayworth, E.Y. (1981). Guide to the Morphology of the Diatom Frustule. *Freshwater Biological Association*, pp.112.

Dahanayake, K. and Krumbein, W.E. (1985). Ultrastructure of a microbial mat-generated phosphorite, *Mineralium Deposita.* Vol. 20(4), p.260-265.

Dahanayake, K. and Subasinghe, S.M.N.D. (1988). Development of Recent Stromatolite Structures and Phosphatic Enrichment in Precambrian Marble of Sri Lanka. *Economic Geology,* Vol. 83 (7), p. 1468-1474.

Dahanayake, K. and Subasinghe, S.M.N.D. (1989a). A Modern Terrestrial Phosphorite - an Example from Sri Lanka. *Sedimentary Geology*, Vol. 61, p. 311-316.

Dahanayake, K. and Subasinghe, S.M.N.D. (1989b). Secondary Phosphate Mineralization in a Karstic Environment in Central Sri Lanka. *Mineralium Deposita* , Vol. 24, p. 169-175.

Dahanayake, K. and Subasinghe, S.M.N.D. (1990a). Variable Mineralogy and Solubility of a Terrestrial Phosphorite Resulting from Differential Weathering Phenomena - an Example from Sri Lanka. *Chemical Geology*, Vol. 84(1/4), p. 42-44.

Dahanayake, K. and Subasinghe, S.M.N.D. (1990b). Formation of Phosphatic Coated Grains and Peloids by Grain Diminution of Precambrian Apatite Crystals. *J. Geol. Soc. India*., Vol. 35, p.189-197.

Dahanayake, K. and Subasinghe, S.M.N.D. (1991a). Heterogeneity of the Eppawala Phosphate Deposit. *Ceylon Journal of Science.*, Vol. 1, p.1-7.

Dahanayake, K. and Subasinghe, S.M.N.D. (1991b). Mineralogical Petrological and Solubility Variation of the Eppawala Phosphate Deposit in Sri Lanka – a Case for Selective Mining for Fertilizers. *Fertilizer Research*. , Vol. 28, p. 233-238.

Hirsch, P., Eckhardt, F.E.W. and Palmer, R.J. (1995). Methods for the study of rock-inhabiting microorganisms: A mini review. *Journal of Microbiological Methods*, Vol. 23(2), p.143-167.

Ivanova, R., Bojinova, D. and Nedialkova, K. (2006). Rock phosphate solubilisation by soil bacteria. Jour. Univ. Chemical Technology and Metallurgy, Vol. 41(3), p. 297-302.

Jayawardana, D.E. de S. (1976). The Eppawala carbonatite complex in North – West Sri Lanka (Ceylon). *Geological Survey Department, Economic Bulletin,* No. 3.

Khan, A.A., Jilani, G., Akhtar, M.S., Naqvi, S.M.S. and Rasheed, M., (2009). Phosphorus Solubilizing Bacteria: Occurrence, Mechanisms and their Role in Crop Production, *J. Agric. Biol. Sci*., Vol. 1(1), p. 48-58.

Konhauser, K. O. (1997). Bacterial iron biomineralisation in nature. *FEMS Microbiology Reviews*, Vol. 20, p. 315–326.

Mann, D.G. (1999). The species concept in diatoms. *Phycologia,* Vol.38, p. 437-495.

Manthilake, M.A.G.M., Sawada, Y. and Sakai, S. (2008). Genesis and evolution of Eppawala carbonatites, Sri Lanka, *Journal of Asian Earth Sciences*, Vol. 32, p. 66–75.

Moriarty, D.J.W. Skyring, G.W. O'Brien, G.W. and Heggie, D.T. (1991). Heterotrophic bacterial activity and growth rates in sediments of the continental margin of eastern Australia, *Deep Sea Research Part A. Oceanographic Research Papers*, Vol. 38(6), p. 693-712.

Natesan, R. and Shanmugasundaran, S. (1989). Extracellular phosphate solubilization by the cyanobacterium Anabaena ARM310. Biosci., *Vol.* 14(3), p. 203-208.

Panhwar, Q. A., Radziah O., Naher U. A., Zaharah A. R., Sariah M. and Mohd Razi I. (2012). Root colonization and association of phosphate-solubilizing bacteria at various levels of triple supper phosphate in aerobic rice seedlings. African *Journal of Microbiology Research*. Vol. 6(10), p. 2277-2286.

Pitawala, A. and Lottermoser, B.G. (2012). Petrogenesis of the Eppawala carbonatites, Sri Lanka: A cathodoluminescence and electron microprobe study, *Mineralogy and Petrology*, Vol. 105(1-2), p. 57-70.

Pitawala, A., Schidlowski, M., Dahanayake K. and Hofmeister, W. (2003). Geochemical and petrological characteristics of Eppawala phosphate deposit, Sri Lanka. *Mineralium deposita*, Vol.38(4), p. 505-515.

Pomonipapaioannou, F. and Solakius, N. (1991). Phosphatic hardgrounds and stromatolites from the limestone shale boundary section at Prossilion (Maastrichtian-Paleocene) in the Parnassus-Ghiona zone, Central Greece. *Palaeogeography Palaeoclimatology Palaeoecology*, Vol. 86(3-4), p. 243-254.

Reimers, C.E., Kastner, M., and Garrison, R.E. (1990). The role of bacterial mats in phosphate mineralization with particular reference to the Monterey Formation. In: Burnett, W.C., and Riggs, S.R. (eds.), Phosphate Deposits of the World, Vol. III: Neogene to Modern Phosphorites. *Cambridge University Press*, p. 300-311.

Round, F.E., Crawford, R.M. and Mann, D.G. (1990). The diatoms. Biology and morphology of the genera. *Cambridge University Press*, pp. 747.

Soudry, D. and Champetier, Y. (1983). Microbial processes in the Negev phosphorites (Southern Israel). *Sedimentology*., Vol. 30, p. 411-423.

Soudry, D. (1987). Ultra fine structures and genesis of the campanian Negev high-grade phospharites (Southern Israel). *Sedimentalogy*, Vol. 34, p. 641-660.

Soudry, D. (1992). Primary Bedded Phosphorites In The Campanian Mishash Formation, Negev, Southern Israel. *sedimentary Geology*, Vol. 80(1-2), p.77-88.

Subasinghe, N.D. (1998). Formation of a phosphate deposit through weathering and diagenesis – an example from Sri Lanka. *PhD Thesis* (Unpubl.) *University of Reading, UK.*

Subasinghe, N.D. (2012). Electron microscopic studies on phosphate binding processes in the presence of iron. *Advanced Materials Research*, Vol. 343-344, p. 307-311.

Suihkoa, M.L. Alakomia, H.L., Gorbushinab, A., Fortunec, I., Marquardtb, J. and Saarelaa, M. (2007). Characterization of aerobic bacterial and fungal microbiota on surfaces of historic Scottish monuments. *Systematic and Applied Microbiology*, Vol. 30(6), p. 494–508.

Tazaki, K., Fyfe, W.S. and Dissanayake, C.B. (1986). Weathering of Phosphatic Marble to Exploitable Apatite Deposit, Sri Lanka. *Applied Geochemistry* Vol. 1, p. 287-300.

Tazaki, K., Fyfe, W.S. and Dissanayake, C.B (1987). Weathering of Apatite under Extreme conditions of Leaching. *Chemical Geology,* Vol. 60, p. 151-162.

Weerakone, M.W.K., Miyazaki, T. Shuto, K. and Kagami, H. (2001). Rb-Sr and SM-Nd Geochronology of the Eppawala metamorphic rocks and carbonatite, Wanni Complex, Sri Lanka. *Gondwana Research*, Vol. 4(3), p. 409-420.