Potential of Geophagous Earthworms In Ecofriendly And Sustainable Biomining Of Gold From Auriferous Soils

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Abstract:

Earthworms are the major contributors of soil invertebrates. Earthworms play a major role in soil amendments, nutrient turnover and participate in the aeration as well as in mixing of various soil horizons. Earthworms are mentioned as one of the Uparasa in Indian Ayurvedic medicine due of its ability to uptake heavy metals. They are soil feeders and hyperaccumulators of heavy metals. There are studies on the bioavailability of Au nanoparticles from soil and biodistribution within earthworms. Goa is situated in the north western part of the metallogenic archean Western Dharwar Craton (WDC) and there are various reports on presence of gold in the WDC. This paper reports how geophagous earthworms can be used in ecofriendly and sustainable biomining of gold from auriferous soils of Goa. Exploration and collection of vermicasts from mining and non-mining areas of Goa was carried out. Detection of gold in the vermicasts samples was done, morphological and typological classification of vermiform gold and fractal analysis of the vermicasts of Goa was carried out. The gold in ppm ranged from 0.66 to 1.77 ppm. We also present the future roadmap of ecofriendly and sustainable biomining of vermiform gold from auriferous soils of Goa.

***Keyword*: Geophagous Earthworms, Vermicasts, auriferous soils, Western Dharwar Craton (WDC), Gold, Biomining**

I.INTRODUCTION

Earthworms are mentioned as one of the Uparasa in Indian Ayurvedic medicine due of their ability to take up heavy metals. In Rasagranthas reference about Bhunaga satwa bhasma is explained as substitution to Tamra (Copper) bhasma (Patel et al., 2018), and different methods of Bhunag satwapatana are explained. Earthworms growing in copper mines are to be rubbed with guda, guggulu, laksha, una (wool), matsya, oil cakes (tilkhali) and tankan and made into a ball. This is to be dried to be heated, resulting in the discharge of a copper like essence. Earthworms have the properties as that of Tamra bhasma and can be used as its substitute (Chandrashekhar et al., 2013). Ancient Indian alchemists knew about the properties of earthworms known in Sanskrit as Bhunag as soil feeders and hyperaccumulators of heavy metals. Probably they knew that the ash of the earthworms yield metals and metal oxides with some interesting and useful properties (Manojkumar, 2013). Physiographically diverse India with hundreds of soil subtypes sports a rich diversity of earthworms but there is comparatively very less work on soil feeding or geophagous earthworms and their unique behaviour. The earthworm fauna of India is well reported as compared to other Asian Countries. Presently, 451 valid species / subspecies of earthworms under 71 genera are known from the Indian territory, including the islands of Andaman, Nicobar and Lakshadweep (http://earthwormsofindia.com/). The ancient Greeks regarded the earthworms to play an important role in improving the quality of the soil. The Greek philosopher Aristotle (384-322 B.C) regarded worms as “the intestine of the earth”. Egyptians were the first to recognize the beneficial status of the earthworms. Cleopatra (69-30 B.C) recognized the earthworms contribution to Egyptian agriculture and declared them to be sanctified. In Egypt, removal of earthworms was punishable by death. Egyptian farmers were not even allowed to touch an earthworms for fear of offending the god of fertility. The excessive fertility of the soil in the Nile valley was large part due to the work of earthworms. The ancient Chinese also considered earthworms as “angels of the earth”. Earthworms are the major contributors of soil invertebrates and play a major role in soil amendments. Earthworms participate in nutrient turnover by processing the organic matter and play a role in aeration and mixing of various soil horizons (Kiyasudeen et al., 2014). The drilosphere of geophagous earthworms involved in production of earthworms casts by dwelling inside soil and taking up of the organic and inorganic matters. The organic and mineral fraction bound to the organic matter are further subjected to hydrolytic decomposition by microbial and enzymatic actions inside the digestive track. The humification of organic matter occurs and ions are released which are available for plants whereas the mineralization occurs releasing the minerals which are further absorbed by tissue or further chelation and immobilization of metals. Earthworms in auriferous or heavy metal rich areas are likely to take in soil rich is secondary gold which may pass through their gut and get deposited in the vermicasts and the whole cycle can get repeated several times. However, we report the presence of gold particles in vermicasts in Goa for the first time on basis of our field collection and laboratory analysis. Previous studies have provided evidence for bioavailability of Au nanoparticles from Soil and biodistribution within Earthworms (*Eisenia fetida*) and also trophic transfer of Au nanoparticles from soil along a simulated terrestrial food chain (Urine et al., 2012). Potentiality of earthworms as bioremediating agent for nanoparticles has been reported earlier (Yadav, 2017). Goa forms the part of Western Dharwad Craton (WDC) which is Asia’s major metallogenic province. There are various reports on presence of gold in WDC (Radhakrishna & Curtis, 1991). The study was aimed at exploration and collection of vermicasts from mining and non-mining areas of Goa, carry out fractal dimension analysis of the vermicasts of Goa, detection of Gold in the vermicast samples and morphological and typological classification vermiform Gold.

II. Methodology

Goa is situated in the north western part of the metallogenic archean Western Dharwar Craton. The Dharwar Craton is divided into Eastern and Western Cratons wherein Goa is situated in the north western part of the WDC which includes Sanvordem, Bicholim, and Vagheri Formations (Dessai, 2011). Exploration and collection of vermicasts was carried out from mining and non-mining areas of Goa (**Fig.1**). The collected vermicasts samples were dried and classified based on size and morphology. Fractal dimension analysis of the vermicasts was carried out using Jfrad software which analyses the fractal geometry of complex biofilm architect in landscape images (Center For Microbial Ecology Image Analysis System-CMEIAS). Image analysis of fractal geometry can be used to gain deeper insights into complex ecophysiological patterns and processes occurring within natural biofilm landscapes, including the scale-dependent heterogeneities of their spatial architecture, biomass and cell-cell interactions of colonization behaviour, all driven by the ecological theory of optimal spatial positioning of organisms to maximize their efficiency in utilization of allocated nutrient resources. Quantification of gold by ICP-AES method (Cera Laboratories Mumbai) was carried out. Dried samples were sieved and powdered into different fractions. Morphological classification of vermiform Gold was carried out by direct DPX mount, Light and phase contrast microscopy. SEM-EDX characterization was carried out using Carl-Zeiess Scanning electron microscope (SEM) (USIC Goa University) for detection and typological classification of vermiform gold.

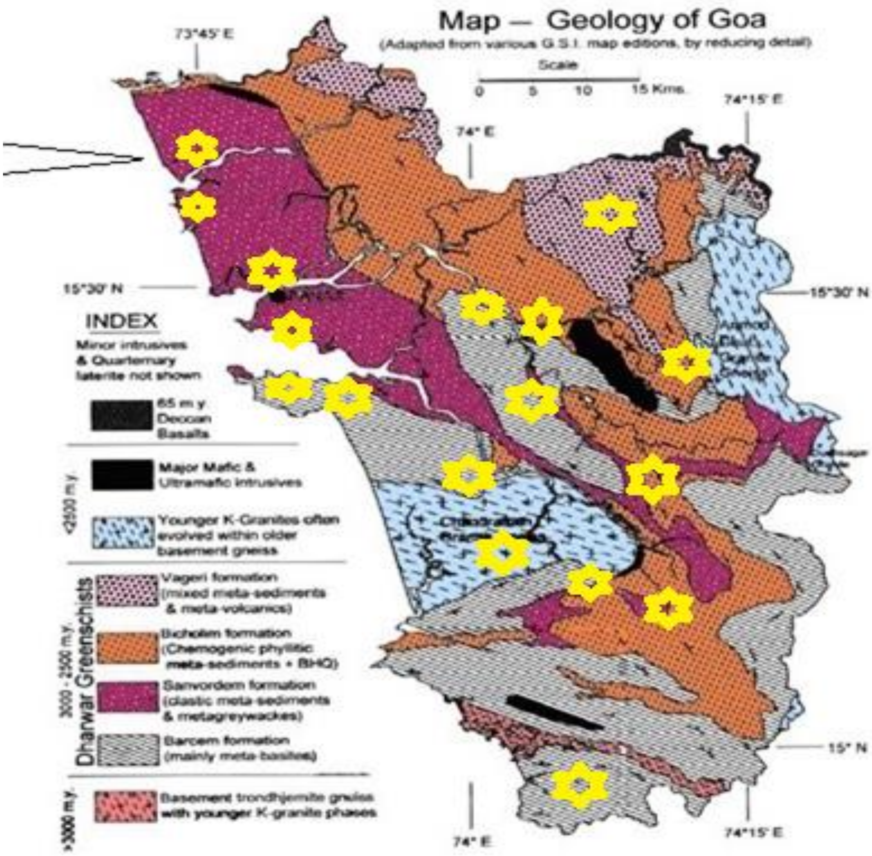


Figure 1 Map of Goa showing the sampling sites ( ) (Adapted from Sreenivasa et al., 2014)

Figure 2 Earthworm habitat teeming with vermicasts

III. RESULTS AND DISCUSSION

Vermicasts samples were collected from mining and non mining areas of Goa and were classified based on the location and morphology as shown in **fig 3(a-d)**. The size of the vermicasts ranged from few mm to 5cms, having irregular and globular shapes. Table 1 gives the detail characteristics of the vermicasts. The fractal dimension analysis of the vermicasts revealed fractal dimension measured as Euclidean distance ranging from 1.22 to 1.26 (Table 2). Samples from mining areas showed high fractality index as compared to samples from non-mining areas. This is the first report on the fractal dimension studies of the vermicasts. It was found that vermicasts samples from mining areas contained gold ranging from 0.76 to 1.77 ppm whereas in case of non mining areas the gold value ranged from 0.66 to 1.03 ppm. The vermiform gold ranged from circular to irregular and exhibited lemon yellow, yellow to golden colour (Table 3). SEM EDX studies revealed the presence of globular, circular form of gold and presence of gold was indicated by specific count (1.3-11.7). All these results indicate that earthworms are involved in hyperaccumulation of heavy metals, similar studies earlier provided evidence for bioavailability of Au nanoparticles from Soil and biodistribution within earthworms (**Eisenia fetida**) and also trophic transfer of Au nanoparticles from soil along a simulated terrestrial food chain (Urine et al., 2012).

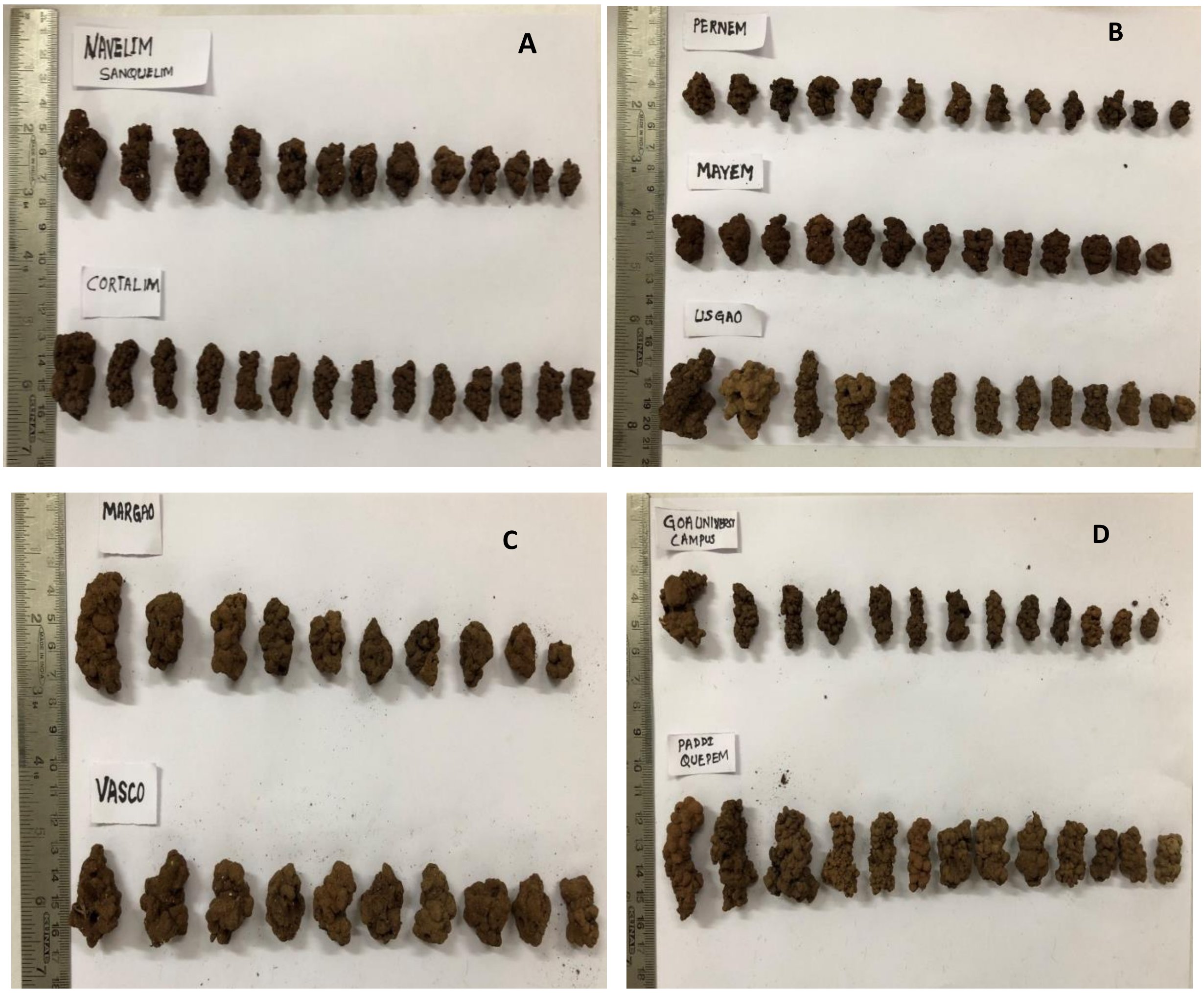


Figure 3 Classification of the vermicasts samples based on location and morphology

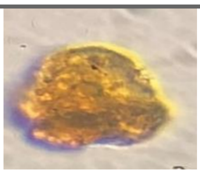
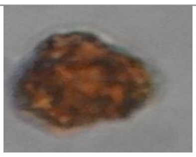
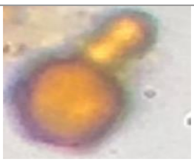
Table 1 Characteristics of vermicasts

	Characteristics of vermicasts
Colour	Black, brown, reddish
Smell	Muddy
Shape	Irregular, Globular
Size	Few mm to 5cms
Weight	0.5 grams to 15 grams

Table 2 Fractal analysis of the vermicasts of Goa

Sr.no	Sample location	Fractal Dimension of vermicasts (Euclidean distance)	Fractality Index
1	Codli (Mining area South Goa)	1.23	1230
2	Assonora (Mining area, North Goa)	1.26	1260
3	Siolim (Non-mining area, North Goa)	1.22	1220
4	Taleigao (Non-mining area, North Goa)	1.23	1230
5	Taleigao (Non-mining area, North Goa)	1.23	1230
6	Chapora (Non-mining area, North Goa)	1.22	1220

Table 3 Vermiform Gold concentration and morphology

Sr.no	Sample location	Gold in ppm	Vermiform Gold morphology
1	Codli (Mining area South Goa)	0.76	
2	Assonora (Mining area, North Goa)	1.77	
3	Siolim (Mining area, North Goa)	0.87	
4.	Taleigao (Non-mining area, North Goa)	1.03	
5.	Taleigao (Non-mining area, North Goa)	0.76	
6.	Chapora (Non-mining area, North Goa)	0.66	

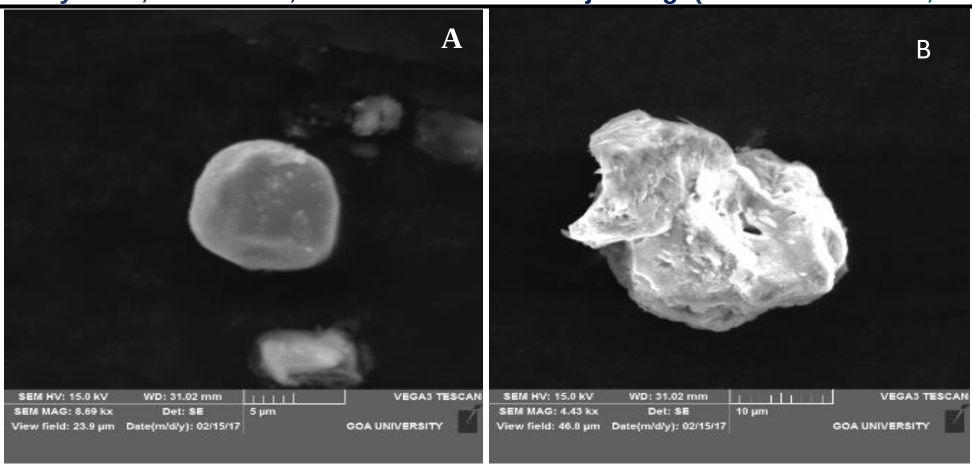


Figure 4(a-b) SEM typology of vermiform Gold

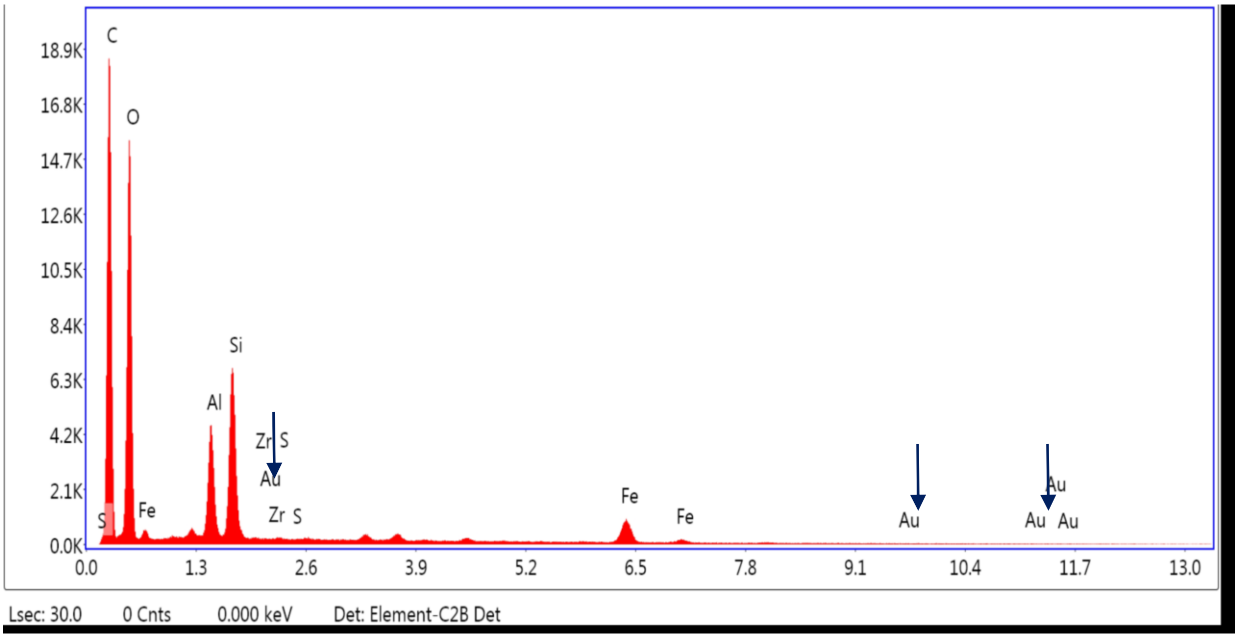


Figure 5 SEM-EDX of vermicast sample indicating Gold

Basically just like age old artisanal alluvial gold mining from auriferous river sands we see the possibility of artisanal extraction of metallic secondary gold by intelligent, systematic and ecofriendly use of vermicasts by rearing local geophagous earthworms with a feedstock blend of organic matter and auriferous soil. This is like reinventing India's lost alchemic heritage because anyone with some patience can rear earthworms and extract small quantities of metallic gold easily. **Fig 6** gives the future roadmap of ecofriendly and sustainable biomining of vermiform gold from auriferous soils. The first step is establishing facility of rearing of earthworms which can feed on auriferous soils which acts as the raw material. Further in the next step mixture of the auriferous soil (which has been analysed for gold) and the feeding materials (such as dry plant litter, humic rich materials, cowdung) need to be prepared. To this mixture, earthworms are to be added, to feed on the mixture and move through the soil and build a organo-mineral structure (vermicasts) with specific physical, chemical and microbiological properties (Stage I). For the enrichment of gold, the vermicasts formed during stage I are mixed with the litter feeding material and to this fresh earthworms are added. Further the vermicasts can be treated by powdering, sieving, magnetic separation, gravity separation/ floatation techniques and finally auriferous material is recovered and purified.

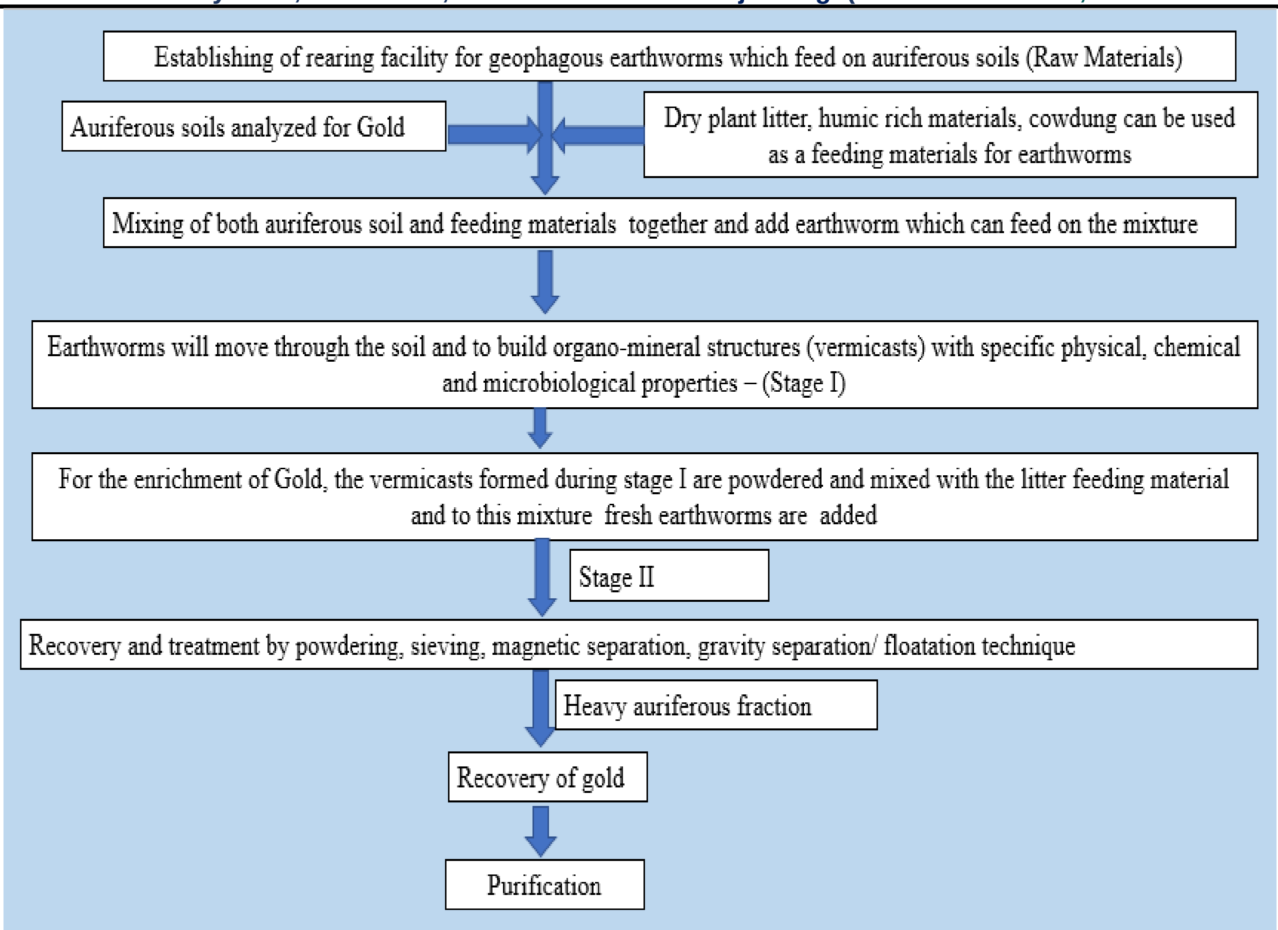


Figure 6 The future roadmap of eco-friendly and sustainable biomining of vermiform gold from auriferous soils

IV. CONCLUSIONS

This is the first report on the fractal studies of the vermicasts from Goa which revealed the fractal dimension value ranging from 1.22 to 1.26. Detection of Gold in the vermicasts samples, morphological and typological classification vermiform Gold has been reported. The Gold in ppm ranged from 0.66 to 1.77 ppm. There is availability of auriferous feedstock to rear earthworms which can feed on auriferous soils. Geophagous earthworms can be used in eco-friendly and sustainable biomining of vermiform gold from auriferous soils of Goa. Their potential for recovery of Gold from auriferous soils is demonstrated.

V.ACKNOWLEDGMENT

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VI.REFERENCES

[1] Chandrashekhar, Y. J., Galib, R., and PK, P. 2013. BRIEF REVIEW OF TAMRA BHASMA (CALCINED COPPER)-A METAL BASED AYURVEDIC FORMULATION. J. Res. Educ. Indian Med, 19(1-2), 17-28.

[2] Dessai, A. G. 2011. The geology of Goa Group: revisited. Journal of the Geological Society of India, 78(3): 233-242.

[3] http://cme.msu.edu/cmeias/fracAnalysis.shtml. Accessed on 28/10/2018.

[4] http://earthwormsofindia.com/about-eathworms-of-india.php. Accessed on 29/10/2018.

[5] Katheem Kiyasudeen, S., Jessy, R. S., and Ibrahim, M. H. 2014. Earthworm's gut as reactor in vermicomposting process: A mini review. International Journal of Scientific and Research Publications, 4(7).

[6] Manojkumar, P. 2013. A REVIEW OF BHUNAG BHUNAG SATW.International Ayurvedic medical journal, 1(4).

[7] Patel, M.K. and Parhate, S., 2018. A Pharmaceutico-Analytical Study of Bhunaga Satva. Journal of Ayurveda and Integrated Medical Sciences (ISSN 2456-3110), 3(3),79-85.

[8] Radhakrishna, B.P. and Curtis, L.C., 1991. Gold-The Indian Scene. GSI Publications, 3(1).

[9] Sreenivasa, A., Jayasheela, H.M., Bejugam, P. and Gujar, A.R., 2014. Heavy mineral studies of beach sands of Vagathor, North Goa, India. Int J Mod Eng Res (IJMER), 4 (70-78).

[10] Unrine, J. M., Shoults-Wilson, W. A., Zhurbich, O., Bertsch, P. M., and Tsyusko, O. V. 2012. Trophic transfer of Au nanoparticles from soil along a simulated terrestrial food chain. *Environmental science & technology*, 46(17), 9753-9760.

[11] Yadav, S. 2017. Potentiality of Earthworms as Bioremediating Agent for Nanoparticles. In Nanoscience and Plant–Soil Systems. pp. 259-278.