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Tipping Points: A Statistical Comparison between Humans and Conodonts

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Abstract

We analyzed Conodonts and rocks using Inductively Coupled Plasma-Optical Emission Spectroscopy (ICP-OES) using statistical methods to compare the tipping points for arsenic (As) and lead (Pb) in conodonts and contemporaneous human tissues. We found that ancient seas contained high levels of As which also occurred in Conodont teeth and which was leached from rocks; a biogenic enrichment. We show that there is a significant decline in reaching the tipping point with time (P=0.01) implying that the sources of these neurotoxic metals in modern teeth have decreased such that Pb and As no longer accumulate in human apatite as it did in the apatite of Conodont teeth. The probability of exceeding the tipping point increases significantly with increasing As concentration (P=0.01) and increasing Pb concentration (P=0.04). This suggests that the toxic effects of these metals may be additive.

Keywords: Conodont; Biogenic enrichment; Neurotoxic metals; Apatite; Logistic regression

Introduction

Fossil marine bio-apatites from conodonts teeth; small animals that lived in ancient seas, can store a record of the content of paleoseawater of arsenic and lead [1]. This can then be compared statistically to the arsenic and lead contents in contemporaneous human teeth [2] and to the amounts of these metals found in rocks [3]. A tipping point is a statistically determined amount reached at which small changes become big enough to cause a large change. In biology the term tipping point refers to oscillating biological systems which cease to oscillate and consequently the organism dies. Here we use statistical methods to compare the tipping points for arsenic and lead in conodonts and contemporaneous human tissues [2].

Materials and Methods

Sample preparation

Samples of rocks and Conodonts teeth from New Mexico weighing ~1.000 to ~2.000 grams were dried crushed manually using a pestle and mortar. The samples were then pulverized using a SPEX mixer mill with carbide vessel and mixing balls. One thousand to 2.000 grams of the sample were weighed and digested using a mixture of 3 ml concentrated nitric acid (HNO₃) and 2 ml concentrated hydrochloric acid (HCl). Samples were digested using DigiPrep heating block equipped with temperature controller at 95°C for about two hours. After digestion was completed, samples were filtered (0.45 micron) and brought to final volume "25 ml".

CP-OES analysis

Digested samples were transferred into 15 ml glass test tubes and were setup on the Inductively Coupled Plasma- Optical Emission Spectroscopy (ICP-OES) (PerkinElmer, Optima 5300DV ICP-OES and for Hg FIMS) autosampler. The system was optimized using mercury optical alignment and manganese (Mn) view touch alignment.

Statistical methods

The tipping point in terms of arsenic (As) and lead (Pb) defines the concentrations of each which would compromise the natural biological rhythms of an animal beyond recovery as described in the introduction. The Pb cut score is defined [2] represents the amount Pb found in our previous study in human subjects residing in the Four Corner Region of New Mexico, the As cut score is facilitated by the scatter plot of As concentrations versus the Pb concentrations in the samples of this study. The extreme As concentrations include that of a human brain from Chile; the effect of As in Chilean subjects is well documented [4]. We selected the samples as best representing values for analysis in the model used. With this binary, a statistical method that is a data type which describes binary variables, which can take only two possible values, here either extreme As or Pb concentrations, we use logistic regression that is a statistical method for analysing a dataset in which there are one or more independent variables here As or Pb, that determine an outcome, to obtain the probabilities of exceeding the tipping point versus geological time and versus the concentrations of As and Pb separately. P-values £ 0.05 are considered statistically significant for the relationships of the tipping point with period, here As concentration and Pb concentration.

Results

In the present study we present a statistical analysis as defined in the method section to show the relationship between period that is a time span of approximately 300 million years and the concentration of As and Pb in the environment, the result of geochemical enrichment, and in teeth the result of biological enrichment in New Mexico USA, (Figure 1). The time (period) in years is shown in Figure 2. Tipping point defined as exceeding Pb or As cut points. Periods are ~300 million years for Conodonts to -50 years for humans expressed on a log10 scale. There is significant decline in the probability of exceeding the tipping point over geological time (P=0.01) and the probability of

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Figure 1: Scatter plot of log₁₀ of As versus log₁₀ of Pb for Conodonts (blue circle), Biological samples (green circle) and Rock samples (grey triangle). Tipping point for Pb (dashed red line) in reference [3] and tipping point for As (dashed areen line) determined herein.



reaching the tipping point for As and Pb is given in Figure 3. Tipping point defined as exceeding Pb or As cut points. Concentrations of As and Pb range approximately from 0.01 to 100 mg/Kg expressed on a log 10 scale. The probability of exceeding the tipping point significantly increases with increasing As concentration (P=0.01) and increasing Pb concentration (P=0.04). The concentrations of a 50% probability of exceeding the tipping point are 1.6 mg/Kg for As (dashed green line) and 3.2 mg/Kg for Pb (dashed red line).

Discussion

The Conodonts in our study were obtained from the Pennsylvanian Atrasado Formation of central new Mexico, about 305 million years old. Conodonts were small animals that had teeth containing the same chemicals as those of modern humans [1]. Such biogenic apatite dating to ~300 million years ago provides insights into the composition of metals in ancient teeth, gleaned from sea water that can be compared to that found in modern human teeth accumulated from food



and modern environments. Here we used the tipping point for As determined in Figure 1 and the known effect of As on humans in Chile [4] to model the tipping point for As. Four rock samples, 1 Conodont sample and one other biological sample were above the tipping point for As. This implies that ancient seas contained high levels of As which also occurred in Conodont teeth and which was leached from rocks; a biogenic enrichment as determined using statistical modeling. Our results shown in Figure 2 give the probability of reaching the tipping point for Pb or As over geological time periods. We show that there is a significant decline in reaching the tipping point with time (P=0.01) implying that the sources of these neurotoxic metals in modern teeth have decreased such that Pb and As no longer accumulate in human apatite as it did in the apatite of Conodont teeth. Figure 3 shows the probability of exceeding the tipping point for As and Pb concentrations in teeth. The probability of exceeding the tipping point increases significantly with increasing As concentration (P=0.01) and increasing Pb concentration (P=0.04). This suggests that the toxic effects of these metals may be additive that is accumulation of both together is worse than accumulation of one only. However, tipping points are statistical models only which allow the estimation of eventual harm to humans arising from environmental neurotoxins as has been demonstrated previously [2,3]. Their usefulness is limited by the geographical area from which the material for analysis was obtained and time period of observation; herein 300 million years.

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