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charcoal (etc.) been tested with model inorganic polyphosphate compounds?

### Microbial Inorganic Polyphosphates: Factors Influencing Their Accumulation in the Soil

I was very interested in the article on microbial inorganic polyphosphates by I. L. Pepper, R. H. Miller, and C. P. Chonsikar (*Soil Sci. Soc. Am. J.* 40:872-74.) as my own work involves the study of the phosphorus cycle in connection with the transport of phosphorus from animal slurry in soils. On reading the article some questions arose:

- (1) Could not the determination and identification of poly-P be subject to error due to the fact that the assumption is made that all organic phosphorus compounds are adsorbed by activated charcoal?

From our experiments I know that inositolhexaphosphate and glycerophosphate are not adsorbed by charcoal. This may also be the case for other organic (sugar) phosphates. The adsorption by charcoal is also pH dependent, with usually an optimum at pH 4-6 for organic compounds. The possibility thus exists that organic phosphates are mistaken for inorganic polyphosphates. In the case of inositolhexaphosphate the error is negligible as even after boiling one hour in 1*N* HCl only about 0.1% (as P) is decomposed. Other organic phosphates can be more labile. Organic phosphates can also give barium-precipitates, of which the IR spectra can be analogous to IR spectra of inorganic polyphosphates. Organic phosphates are also more or less excluded from Sephadex G-25 gel (Zabransky, 1975; Capek and Stanek, 1975). This exclusion of phosphates is mainly due to ionic (repulsion) effects and much less to molecular size. Another complication is possible owing to the fact that inorganic polyphosphates may occur complexed with proteins and polynucleotides (O'Kelley, 1973) and thus (if stably complexed) escape analysis when adsorbed in this form on charcoal.

- (2) Is perchlorate removed?

Assuming 100 g of soil to be extracted with 500 ml 0.5*N* HClO<sub>4</sub> and the extract condensed to 100 ml, a final concentration of about 2.5*N* HClO<sub>4</sub> in the extract can be expected. If this extract is filtered on charcoal poor adsorption of organic compounds will occur. When the extract is made 1*N* with respect to HCl and heated, evolution of chlorine and oxidation of organic compounds can take place. In the case of organic phosphates additional phosphate may then be released. The oxidizing nature of perchlorate and chlorine must also be taken into account when determining P colorimetrically as with some methods oxidizing substances can either enhance or inhibit color development.

- (3) To what extent can the enormous excess of added glucose, of which the decomposition will be slowed down by the relative shortage of nitrogen, lead to an accumulation of glucose-derived phosphates in the microbial cells after addition of orthophosphate? (The possible error due to sugar phosphates is covered by question 1).

- (4) Has the entire analysis procedure (extraction, evaporation,

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