

POLYPHOSPHATE BODY FORMATION AND DEGRADATION IN *PLECTONEMA BORYANUM* (CYANOPHYCEAE)

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A survey of stations in Saginaw Bay of Lake Huron during the period 1974-1976 demonstrated that: 1.) certain phytoplankton populations were exiting the bay and surviving transport to southern Lake Huron, 2.) many of these populations in the bay were found to have polyphosphate accumulations in them, and 3.) several of the populations of blue-greens and diatoms in the bay had lead sequestered in the polyphosphate bodies. Moreover, it has been demonstrated with both cultured blue-greens and diatoms that heavy metals may be incorporated into polyphosphate bodies during phosphate uptake.

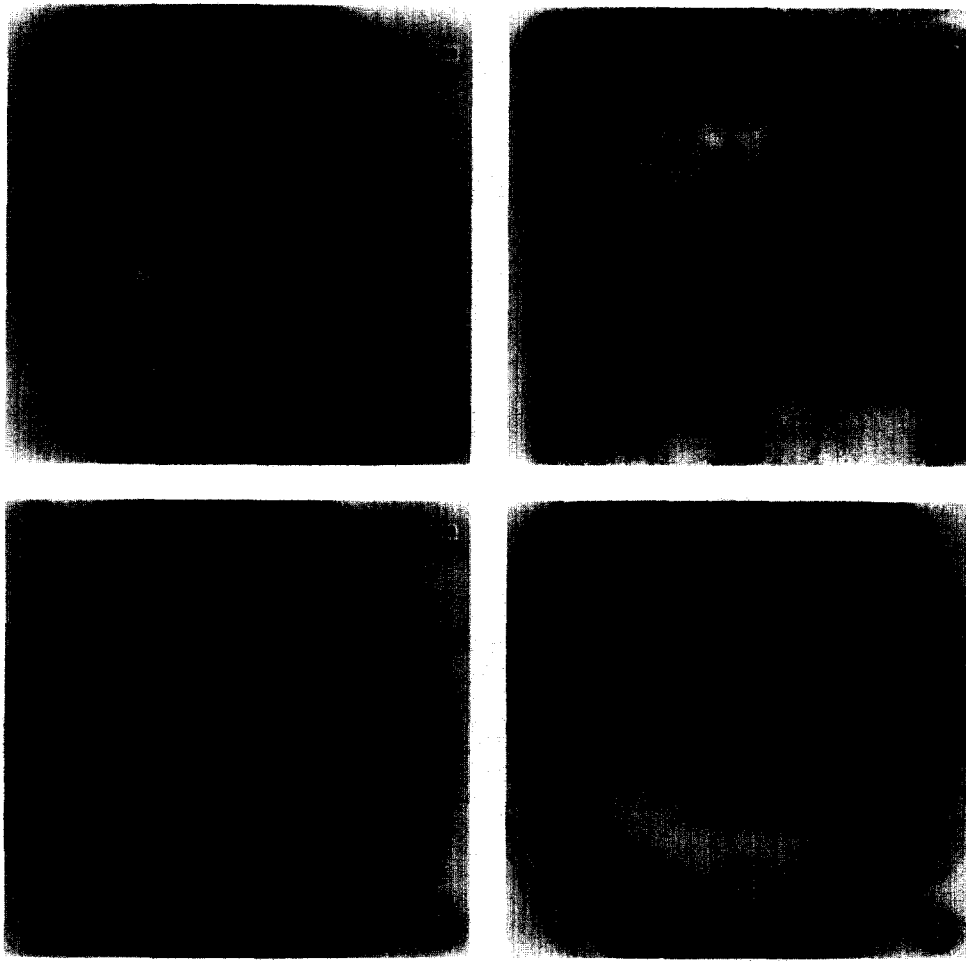
The eventual fate and ecological significance of polyphosphate is not well known. If polyphosphate serves as a phosphorus storage form that can be degraded under conditions of limiting phosphorus, then phosphorus limitation should make polyphosphate (and heavy metals if they are so bound) available to intracellular sites.

Laboratory experiments were conducted to determine the time sequence and consequences of polyphosphate degradation in cultured algae as well as the effects of heavy metal exposure at the time of luxury phosphorus uptake. Preliminary results are presented here.

P. boryanum (Cyanophyceae) was grown to logarithmic phase at 20°C on a 12/12 hour light-dark cycle in modified Fitzgerald's medium. The cells were washed three times in P-free medium prior to transfer to this medium for P starvation. After three days of starvation, phosphate was added to a final concentration of approximately 8 mg PO₄/liter (twice the normal level in the growth medium) and lead was added with thorough mixing in the Pb treatment flasks to a final concentration of 20 ppb. Cells were incubated under these conditions in a growth chamber for three hours, then washed in phosphate-free medium three times prior to transfer to phosphate-free medium. Subsequent samples for electron microscopy were withdrawn at 2, 4, and 7 days after transfer to P deprivation medium. They were fixed in a 1% solution of paraformaldehyde-glutaraldehyde in cacodylate buffer, post-fixed in 1% OsO₄, dehydrated in a graded ethanol-propylene oxide series and embedded in Epon for transmission electron microscopy.

Detailed ultrastructural examination of control, phosphate starved, and phosphate uptake cells of *P. boryanum* have been published elsewhere. In general, polyphosphate body number and relative volume increase during phosphate uptake and subsequently decrease during P deprivation. During the uptake process, with or without lead, polyphosphate bodies are relatively small and scattered throughout the cell (Figs. 1-2). However, fewer numbers are present during P deprivation and they are both larger and located in areas of medium electron density (Fig. 3). Images of what we interpret as polyphosphate degradation are present as early as 2 days in the P deprivation medium. Polyphosphate bodies appear to be less electron dense; they are more prone to volatilization in the electron beam, and often only a small dense portion is left at the periphery of the body (Figs. 3-4). There appears to be a degradation sequence which essentially is a reverse of the formation sequence described for blue-greens. We observed increased numbers of porous structures as well as images of bodies where there was only a remnant of electron dense polyphosphate at the periphery as the length of time in deprivation medium increases. Cells exposed to Pb during P uptake did not survive longer than 4 days. Cells with polyP but no lead survived longer than one week.

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FIGS. 1-4. *Plectonema boryanum*. Intrathylakoidal space (IT), Thylakoids (T), PolyP (P), Polyhedral bodies (PH).

- FIG. 1. Phosphate uptake cell (3 hours). Note light areas of intrathylakoidal space. Numerous polyphosphate bodies are scattered throughout the cell. Thylakoids are also evident.
- FIG. 2. Phosphate + lead uptake (3 hours). Greater numbers of polyphosphate bodies are present with this treatment.
- FIG. 3. P uptake cells incubated 2 days in P deprivation medium. Note fewer numbers of polyP and altered structure (arrows).
- FIG. 4. Day 2 deprivation medium of cells not previously incubated in starvation medium. PolyP bodies are larger and more numerous than in other P deprivation treatments.