

Sep. 25, 1961.

Dear Maxine:

I want first of all to tell you how deeply I enjoyed working with you and that I am deeply indebted to you for all the time and attention you gave me. My stay there was so profitable to me materially and spiritually that I wish I could return there. Of course, I also hope that I would be able sometime to have you and/or members of the group here. Thanks again for so much.

I got your note and the survey of my ^{preparation} ~~of~~ it was all due to your expert supervision that that it came out so well.

Fritz Lipmann wanted to try out Polyl and asked me about the preparation of the phosphorylase and I felt that the best I could do was to refer him to the the originator and I hope this did not cause you any inconvenience. I am sure your procedure is going to be heavily in demand.

As regards the use of pyridinium Dowex 50 for removing KOH and all K^+ ions from hydrolyzate, this is very simple. We first clean the commercial Dowex 50 (H^+) by NaOH then excess of HCl cycle - till no NaCl comes off - wash thoroughly with H_2O & keep it in this acidic form. With passage of time this resin releases some low mol. wt colored polymers & when I came to using it for making Py^+ form we pack some mls. of wet resin in a little column & put 10-20% Pyridine through it & wash it for $\frac{1}{2}$ hour or so with water. Russell did it while I was there. This freshly prepared ~~Py⁺~~ resin pyridinium (Py^+) resin can be used either in drained form by adding it to the KOH hydrolyzate

or by passing the KOH hydrolysate through a column of the resin & washing the column with 5% aq. Pyridine. For 1 mmole of KOH ca 2-3 ml. of resin is more than enough. Excess of resin doesn't matter of course. The eluate from column now contains the nucleotides as pyridine salts and pyridine. If you wish to concentrate it by evaporation, pyridine goes off first. pH is likely to drop down to about 3. Use this aq. solution for any purpose you want.

For small scale, as I suggested to Sean, one could add Py^{H} resin to the aliquots of alkaline digests & spin down the resin - (it packs quite nicely) and use supernate for analysis. If there are any questions let me know.

Separately I am sending a copy of my little book. Hope you find something of interest in it.

With all the best of regards to you and Sean & love to children.

Gobind.