

September 27, 1949.

Dr. Sol Spiegelman,
Dept. Bacteriology,
University of Illinois,
Urbana, Illinois.

Dear Sol:

Enclosed herewith is about 50 mg. of *o*-nitrophenyl- β -D-galactopyranoside which should suffice for a considerable number of galactosidase determinations. I have been using it at M/2000 (.15 mg/ml), developing the color either at pH 7.5 in the buffered reaction mixture, or by adding Na_2CO_3 to M/10 after the reaction is completed. M/50,000 nitrophenol has an optical density of 0.1 in an 18. mm. testtube at 4200 \AA , so that you can easily work in a range of negligible change in substrate concentration. If K_s is not too high, you could probably even use the substrate as low as 10^{-4} M or lower.

I will have some detailed directions on the synthesis for you in a couple of days. It involves the preparation of aceto-brom-galactose, and coupling it with *o*-nitrophenol in alkaline solution. The nitrophenyl-galactoside pentacetate is then hydrolysed with $\text{Ba}(\text{OH})_2$ in alcohol. The prep. is supposed to be quite smooth, giving readily crystallized products in high yield.

Sincerely,

Joshua Lederberg