April 1946.

Dear Francis.

I'm sending you '679-680-Y9' which is a triple mutant, requiring leucine, threchine and methionine. I don't know about the specificity of the lastnamed requirement in re choline or homocysteine, etc. It should be useful as a marker however. I would suggest re-isolating it from a single colony before using using it; I can't give you any information on the quantitative requirements. It was obtained by the new mutant detection techniques from 679-680 irradiated with 65,000 roentgens in the culture medium in a small Petri plate. I may have another triple soon, but am in no hurry about that now that this one has been identified.

In trying to find stable mutations for genetic studies, I made rough tests of the mustard induced mutants, to find that they are grossly not different from the X-Ray ones, some backmutating more than others. No further study is indicated, but the conclusion must be drawn that backmutation must involve some rather unusual chemical events; if the mustard produces mutation by introducing an ethyl group into the gene, backmutation is presumably a reversal of this process, a disruption of what are ordinarily very stable chemical bonds. The premises may not be correct, however.

In re growth tube paper, I have no copy of it, unless perchance the one I sent back was given to me; I have no recollection of when I borrowed it. If you think you can prasw one, I would appreciate it, but it is not vital. However please return a reprint of Roepke's paper which I have loaned you, and forgot about.

We are trying UV for mutations now, and the results look encouraging. Sex, however will be the main problem for a while; Ed has some very encouraging data.

The plate method for identifying mutants (auxanography) is working quite well, but any siggestions on how to cut down diffusion through agar would be appreciated.

In the course of other things, I(ve noted that underlaying a pour plate with another thin layer of agar does help counting, as those troublesome flat things at the glass interface are pretty much eliminated. Colony shape seems to be due to stresses in the agar, if the shape of gas bubbles is any criterion; one of the girls is working on that simple, Matzke-an project

Enclosed are the Stanford comments; also some thoughts of mybown on the implications of coincidental mutations. Please let us know what's cooking in Schermerhorn.

I received word from the Navy Dept. the other day that I am a free man as far as my plans are concerned; they are no longer concerned that I remain in medical school, but you know my sentiments about that already.

Sincerely yours,

Joshua Ledemberg.