

August 27, 1956

Professor H. Kikkawa
Department of Genetics
Faculty of Medicine
~~Osaka~~ Osaka, Japan

Dear Professor Kikkawa:

Thank you for your letter of May 3, and for Mr. Hirota's ms. We
Have also seen the publication in NATURE.

Mr. Richter

We are still interested in trying to reproduce this result here, and I am sorry to say still unsuccessfully. He has, for example, recently done the following experiment:

A 24 hr. nutrient broth culture of M-F⁺ was spun down, washed with saline and resuspended in saline. The culture was then starved 1 hr. at 37^o. Co⁺⁺ was then added to give .02M. After 3 hrs., at 37^o, the culture was plated on nutrient agar containing .04M citrate. The survival was 50%. Of 40 colonies tested all were F⁺ still. In a repetition with various Co conc., the following results were obtained:

Co ⁺⁺ ,M	Survival	F tests
.02	.03	18/18 F ⁺
.04	10 ⁻⁴	18/18 F ⁺
.06	10 ⁻⁵	18/18 F ⁺

It seems very likely that our strain(s) have deviated from yours. For that reason, we would greatly appreciate getting back the 58-161 or Y-40 strain that Mr. Hirota has currently and successfully been using for these experiments. If you can offer any other suggests that would help us to confirm your results we would appreciate that very much too. It might also be helpful if we could have one of the derived F- strains (if reinfected with F⁺, this derivative might be more readily amenable to the exptl. effect).

We are also greatly concerned about the extent of killing which occurs in Mr. Hirota's experiments, which is not specifically mentioned in the paper. Can you get the formation of F- under conditions of high survivorship?

In discussing the matter further with Mr. Richter, we notice that he had added citrate to the nutrient agar in the 3-hour treatment-expts. just summarized, since this was described for the earlier 24-hour treatments. However, your letter does not mention the addition of citrate here. Richter found that plating, after 3 hours, on medium without citrate, led to quite low survivals. The question of extent of survival may be the key to our problem.

I am very sorry that I will not be able after all to discuss these matters with you in person at the Tokyo symposium. My good friend and colleague, Professor Crow will, however, be there and also intends to visit Osaka afterwards. May I name him as intermediary, both to facilitate the exchange of the cultures requested, and to discuss the questions I have raised here. It would probably be helpful if you show him this letter.

Some years ago, Dr. Skaar discovered a method for obtaining F- from F+, namely by passage of motile cultures through "motility agar" (See Nelson & Lederberg, 1954, Proc. Nat. Acad. Sci. U.S. 40:419, footnote 10.) We have not published this in detail because we were waiting for definite evidence whether the treatment selected or induced the F- variants. We were especially hopeful of the latter, and were thinking in terms of the dilution of a cytoplasmic factor (like kappa in Paramecium) during prolonged rapid fission of the bacteria. Recent experiments by Richter ~~and others~~ now indicate, however, that it is a selective effect after all, the F+ parents being slightly more sluggish than F- variants from them. The problem was complicated because the rate of motility is also conditioned by a polygenic background.

Yours sincerely,

Joshua Lederberg
Professor of Genetics

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