PROFESSOR R. A. FISHER, Sc.D., F.R.S.
MISS M. F. I. SPEYER, B.Sc.
Research Assistant and Secretary

UNIVERSITY OF CAMBRIDGE
DEPARTMENT OF GENETICS

WHITTINGEHAME LODGE
44 STOREY'S WAY
CAMBRIDGE
Tel. 55822

23rd. June 1949.

Dear Dr. Lederberg,

Thank you for your letters of June 9th. and ~~April~~ 28th. I hope to receive soon your strains, which will certainly be all very useful to me.

My time has been devoted mainly, in the last two months, to the new stock of which I wrote you in my last letter. Your questions are all answered by the fact that the stock is a derivative of 58-161, and has kept the same physiological and biochemical characteristics. I agree as to the kinetical difficulties you put forward. As a matter of fact, the frequency of prototrophs is increased only 2-300 times when the mixture of 58-161 with high frequency of recombination (Hfr) and W 583 is plated in agar depth, in respect of the usual cross 58-161 x W 583. But if the same mixture is plated on the surface of agar, the increase is nearly 10^4 times, and when few (100-200) cells of one of the two strains (either of 50-161 Hfr or W 583) are plated with an excess (10^8-10^9) of the other strain, the frequency of prototrophs in presence of aneurin varies between 10% and 100% according to conditions.

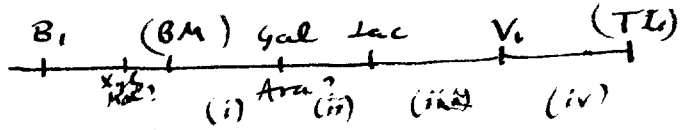
The Hfr stock was obtained through selection for nitrogen mustard resistance, but probably the two phenomena, nitrogen mustard resistance (Ny^R) and Hfr are uncorrelated, for (i) further independent Ny^R mutants do not show the same behaviour,

(ii) after recombination the two characters Ny^R and Hfr seem somewhat independent in segregation. Incidentally, resistance to nitrogen mustard seems to be due to several genes with grossly additive action.

The cross 58-161 Hfr x W 583 shows many discrepancies in the gene frequencies observed in the prototrophs. With the exception of Gal+, the frequency of which is very markedly reduced, the frequency of Lac+, Mal+, Xyl+, V_1^R and B_1^+ are significantly increased. Thus I wondered whether the prototrophs which arise in this way might not be mixed, and show the phenotype which would dominate in the mixture. I have found that most prototrophs are mixed with respect to one or two characters. Diploid formation does not seem to play a role. Of the two major explanations: a) more than one zygote taking part in the formation of the prototrophic colony b) 4-strand c.o. with survival of ~~all 4~~ two meiotic products, I cannot yet favour ~~any~~ either.

Microscopic examination has not yet given great help, though serial photographs of a developing plate show that some bacteria which are at the beginning clearly separated lie closer together after some time. Unfortunately the examination is made difficult by the fact that microcolonies up to 100-200 bacteria are often formed, probably due to traces of growth factors in Difco agar.

I was very interested in your communication of branching at the Maltose locus. I was realising that something went wrong with the Mal locus. In order to make a comparison with



	(iv)	(iii)	(ii)	''' iv	'' iv	' iii	(i)	''' iv
Lac	−	±	+	+	−	−	+	+
V_1	R	S	S	R	R	S	S	R
Gal	−	−	−	−	+	+	+	+
With B_1	58	95	44	6	6	9	19	2
Without B_1	42	96	36	2	1	2	27	2

~~247~~ 239

208

447

One might assume that the epistatic action of Gal+ on Lac has not been fully eliminated by scoring on lactose at 3%, and that therefore the 18 Lac- Gal+ observed are really gal+ lac+; but, even so, one should assume too many triple crossovers to allow for the segregation of V_1^R. It is also to be noticed that the Gal segregation is different from any other in the Hfr strain. Unfortunately I found Ara-, which is closely linked to Gal and might have helped to explain, very difficult to score in a consistent way.

No!

A disturbing phenomenon which I noticed is the appearance of mixed prototroph colonies in about 2% of the cases, and especially from uncrowded plates (supplemented with threonin, or with no supplements). The mixture can be easily observed from the fact that, on EMB plates with T_1, some mixed colonies made of $Lac^+V_1^S$ and $Lac^-V_1^R$, for instance, give easily recognisable peculiar streaks.

I am very anxious to know more of your results. The situation seems rather complex, but no doubt very interesting.

As concerns EMB, it works now quite properly, the difficulty lay in methylene blue.

the data of 58-161 Hfr, I have collected a certain number of data on 58-161, and I have prepared a summary of them, should they be of any use to you. A comparison with your data will, at any rate, be of great help to me.

Xyl and Mal seem to be (in W 583) rather closely linked, on the left of BM, at about 15% of the distance between BM and the B_1 locus. Their mutual relationships are, however, not fully clear. These are the data:

	Prototrophs in presence of vitamin B_1	Prototrophs in absence of vitamin B_1
Xyl^+Mal^+	2	15
+ −	0	8
− +	3	6
− −	234	179

It is possible that there are two chromosomes with a reciprocal translocation. Another possible way of explaining the results is, of course, that of assuming a higher average number of chiasmata than apparent at first sight.

I was thinking of a "branch" in the case of Galactose, to which it is very difficult to assign a locus on the main chromosome on the theory of random segregation. Gal+ segregation is not influenced by the presence of aneurin; Gal+ is linked with BM and should therefore lie close to Lac, but even so the data given below do not favour a linear order.

I shall be grateful if you will send me a reprint of your very interesting paper on heterozygous strains.

Yours very sincerely,

Luigi Cavalli