

September 1, 1951.

The Editors,
Archives of Biochemistry and Biophysics.

Gentlemen:

I am pleased to return the ms. by Landman and Bonner, with my comments.

I have no objection to your forwarding all of these, including the first page, to the authors at your discretion. The latter two pages are, of course, addressed entirely to them. This work should be accepted for publication without undue delay; the writers should be able to give due consideration to the comments in a fairly short time.

Sincerely,

Joshua Lederberg,
Associate Professor of Genetics

Landman and Benzer- Neurospora Lactase. I. Properties of Lactase Preparations from a lactose utilizing and a lactose non-utilizing strain.

This paper is an introductory contribution of considerable merit on a subject of great interest and importance. In general, this reviewer has no doubt as to its acceptability. However, it has not been carefully edited by the writers, and should be revised in order to make it understandable and conformable to proper standards of scientific expression.

The reviewer finds two points in the argument inadequately supported. The evidence on pages 9 and 10 does not seem to have very critical bearing on the conclusion that "hydrolytic cleavage represents the major way in which lactose is utilized", for one might easily imagine that within the mycelium the lactase system functioned differently than in extracts, or alternatively that the major pathway of lactose utilization was mediated by another enzyme whose adaptive responses parallel those of the hydrolytic lactase. Precedents for both types of action can be cited: e.g., transphosphatase (Axelrod), and α -glucosidase in relation to maltase in yeast, respectively. On the other hand, the reviewer regards this issue as overemphasized in relation to the problem of gene-enzyme control, and the writers' general conclusions are not greatly affected by this point.

The second issue is more important. Although some interesting details are given, this paper does not present an exhaustive study of the lactase as an enzyme. Its main interest is in the comparison of the lactases of the standard and a "lactoseless" mutant. From this point of view, the behavior of the mutant, especially its growth and adaptive enzyme responses to lactose, are not adequately documented. One may infer that this is taken up in a companion paper "in press". Unless this paper, reference 16, and to some extent 21, is to adjoin the present paper in the pages of this journal, the reviewer questions the utility of splitting the information. At any rate, he is, to a large extent prevented from an adequate critical view of the present paper by a lack of information on the details of the other.

The discussion is rather long, but well-taken and should not be shortened. The recent papers of Cohn and Monod (*Acta Biochim Biophys*, 1951) and of Lederberg and Beadle (both in *Genetics in the 20th Century*, Macmillan, 1951) are all, however, extremely pertinent and should be cited.

The following remarks concern the form rather than the content of the paper and are addressed to authors primarily for their own consideration:

1. Capitalization is inconsistent and often incorrect in expressions such as Ethanol, Minimal Medium, Standard Strain, Lactose. The entire paper should be carefully edited for orthographic errors. The same for abbreviations and contractions, especially mg / mg., ml / l., and so forth. Volume numbers are inconsistently treated in the References. Reference 18 cites *Adv. in Enzym.*; 19 shows *Adv. Enzym.*

2. "B", used repeatedly before "-galactosidase" should be read (beta)

3. The datum "rpm", pp. 6, 11 is not useful. "RCF" should be given, or mention of the type of centrifuge.

4. "lactoseless", p. 2, and throughout. This expression is bound to be confusing and should be discouraged if the authors agree. It is inconsistent with the usage in, for example, "methionineless", which implies an organism lacking the ability to synthesize methionine. Some organisms are known which require tryptophane; others cannot metabolize it. To use the "-less" terminology for both cases would be very unfortunate. Suggested alternatives: lactase-deficient, alactatic, or lactose-negative.

Specific (mis?) constructions:

p. 1. 8: which/ that. result and resulted ?

L. 14 Is vitamin formation a mutant character?

The second sentence of this paragraph is awkward.

p. 3 Ref. (13) should be moved ~~forward~~ back one phrase. Emerson's description of his strains' behavior on lactose should be cited (Fed. Proc. 1945-6??)

p. 4 P. 2 2% by weight etc./ 20 g of carbon source was added per l. carbohydrate
of medium.

p. 6 L. 8-9. Cation in buffer is not specified, but presumably K.

pH5 / pH 5 or pH 5.

L. 19 incomprehensible. Do writers mean: "Activities of different preparations are expressed in terms of constant dry weights of mycelium per unit volume per unit time" ?

7 L. 5 3.3 for three and a third.

L. 11 implies/ requires

p. 8 L. 1 Concentrations/ levels

11 Confusing! Insert: "activity in mycelia grown on"

p. 9 L. 1 alternate/ alternative

10 "dealing" dangles.

10 9 Two ideas in one sentence confuse!

12 5 ff. One paragraph only

12 14, and 13,7 calories/mole.

12 7 The expression "enzyme-ONPG reaction" is vague. It might, but apparently does not, refer to the initial reaction of adsorption of substrate for which K_s is given later. Since no indication of extrapolation for V_{max} is given, it is difficult to determine whether the temperature effects concern K_s or V_{max} , or both, in view of the non-linearity.

- p. 14 L. 1 Same comment. (See Cohn and Monod for effects of cations on lactose/ONPG activity.
- 14 4 E. coli: Escherichia or Entamoeba ?
- 15 15 Why not document this? The inhibitions themselves, especially by xylose, would be of interest.

Legends for figures:

5. This figure is a plot of $1/k$ against $1/S$, not a "Michaelis constant". The abscissa is incorrectly labelled. If the calculations for p.13 are correct, it should read $1/S \times 10^4$. Most of the S concentrations are too low to be useful in the precise estimation of K_m . It must have been rather difficult to make an accurate determination of the first-order rate constants for an initial substrate concentration of 2.5×10^{-6} M (the rightmost points).

NOTE: If K_m is 4×10^{-4} , and the assay system 2×10^{-3} M, the statement on p. 7 that the reaction is first order under these conditions is not quite correct. When half the substrate has been used up, the rate would have decreased only about 15% from initial. Comments above on p. 12, 7, followed your conclusion on this, but the enzyme is actually about 80% saturated.

Summary:

Neurospora crassa (should be underlined). Note: the specific name is never given in the text- why in the summary?