LETTER TO THE EDITOR**REPLY TO THE LETTER BY S. H. HORI**

In "Letter to the editor" of Vol. 8, No. 3, Dr. Hori commented on my article (Histochemical demonstration of the activity of Ribose-5-phosphate isomerase in human placenta and bovine heart muscle) which appeared in Vol. 8, No. 2 of this journal. The author also agrees with his comment that ribose-5-phosphate isomerase reaction does not occur in the direction that was shown in the schema. He made an advice for me in his comment that multi-step reaction in histochemistry should always be observed with attention, because a positive reaction was demonstrable in my experiment in spite of theoretical difficulty. Some adequate interpretation was thought to be necessary for Dr. Hori's "something in the reaction", that is, the mechanism which made it possible to reveal the positive reaction. One possibility may exist in the reversible mechanism of 6-PGDH reaction for the formazan-production (B. L. Horecker; *J. Biol. Chem.*, 193, 383, 1951).

FIG. 1.

The author finds, however, more possible mechanism in Fig. 2 (B. L. Horecker and P. Z. Symrniotis; *Method in Enzymology* Vol. 1 Academic Press, New York and London, 1962, p. 371), (Martin Gibbs; *Nature* 170, 164 (1952), *Method in Enzymology* Vol. 1 Academic Press, New York and London, 1962, p. 5411), than the former which needs energy reduction.

FIG. 2. Mechanism of histochemical demonstration of R-5-P isomerase

I believe, therefore, that the positive reaction obtained histochemically in the present experiment is specific for ribose-5-phosphate isomerase. Because negative

result has been observed in no substrate control which means lack of the first step in the multi-step reactions. I am planning further examination on the specificity of this histochemical reaction.

The above mechanism was reported in the Japan Society of Histochemistry and Cytochemistry of the 16th annual meeting (October 31, 1975 Okayama).

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MASAO OHYUMI
Department of Pathology
Kumamoto University, Kumamoto 860