



The width of the fontanels shifted from intermediate to narrow, but this seemed more related to the more favourable conditions for development in the smaller litters than to thalidomide doses.

For the peculiar pattern demonstrated by the cervical vertebrae another explanation should be taken into consideration. Since the number of mature young decreased definitely with increasing doses of thalidomide it is conceivable that with certain doses minor anomalies occur that do not cause death in early foetal life. Larger doses inducing additional damage might cause more animals to

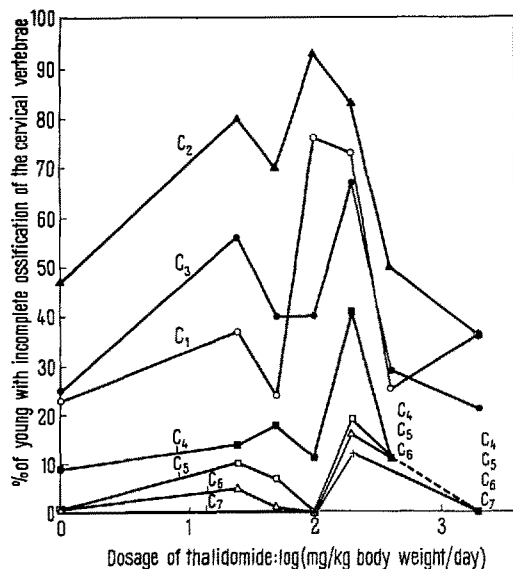
die that would have been susceptible already to smaller doses and a number of fairly normal animals to survive in the highest dosage groups. It might be important that the vertebral anomalies are in the region where a disturbance of the nerve supply of the upper extremities might be possible as well, but this cannot be investigated at the same time as the skeleton staining.

**Conclusion.** It has to be concluded that not only anomalies are induced by the drug given but that also fairly wide variations around the normal or average occur that are not related to the drug given. From the data of the control animals it appears that those bones that show already many variations under normal circumstances are the most susceptible to this drug. For future investigations it is important to know that more abnormalities might be found when giving small doses than giving large doses, since in the latter too many young will not survive until birth<sup>2</sup>.

**Zusammenfassung.** Nach Thalidomidverabreichung an trächtigen Ratten fanden wir Fruchtresorption und Anomalien am fünften Brustbeinkern der Föten. Weitere ergänzende Skelettuntersuchungen ergaben, besonders bei mittelgrossen Thalidomidgaben, Verknöcherungsstörungen an den Halswirbelkörpern. Andere Skeletteile zeigten keine Thalidomid-abhängigen Entwicklungsvariationen.

H. J. KLEIN OBBINK and L. M. DALDERUP

Laboratory of Physiological Chemistry, University of Amsterdam and the Netherlands Institute of Nutrition, Wageningen (The Netherlands), December 28, 1963.



Relation between anomalies of the cervical vertebrae in new-born rats and the dosage of thalidomide in the mother-animals during pregnancy.

<sup>2</sup> **Acknowledgment.** We wish to express our thanks to the Organization for Health Research TNO for financial support, to Prof. E. C. SLATER for providing hospitality in his laboratory for this work, to Chemie Grünenthal for supplying thalidomide, and to Miss M. VAN UFFELEN for technical assistance.

## Effect of Histamine on the Diameter of Skeletal Muscle Fibers in Mice<sup>1</sup>

The occurrence of abnormally large numbers of tissue mast cells in skin of mice with hereditary muscular dystrophy<sup>2</sup> and the association of these cells with tissue content of histamine<sup>3</sup> and 5-hydroxytryptamine<sup>4</sup> suggests a possible interrelationship between these two substances and the muscle abnormality. Reports in the literature do not indicate that either compound affects skeletal muscle of any species. This study was undertaken to determine if repeated injections of different concentrations of histamine would modify the skeletal muscle fibers of mice. Selection of specific doses of the drug has been based on biological assays of histamine in normal mouse skin and in skin from mice with hereditary muscular dystrophy<sup>5</sup>. In addition, the effect of injecting an extremely high concentration of histamine has been included.

**Materials and Methods.** Experimental groups of mice were given intraperitoneal injections of histamine dihy-

drochloride (Calbiochem.) in 0.25 ml physiological saline twice daily for 21 days. Control animals were injected with equal volumes of saline on the same schedule. All mice were weighed daily. At 28 days after the initial injection the mice were anesthetized with ether and fixed *in toto* in Helly's or Bouin's solution for 24 h. Fixation by this method controlled the degree of muscle contraction in each animal. After fixation the tissues were washed, and the gastrocnemius and gracilis major muscles were removed, embedded, and sectioned at 7  $\mu$ . Sections were

<sup>1</sup> Supported by grant HD-00221, National Institutes of Health, U.S. Public Health Service. I wish to gratefully acknowledge the technical assistance of Miss F. VAUGHAN.

<sup>2</sup> W. K. O'STEEN and J. J. HRACHOVY, Texas Rept. Biol. Med. 20, 70 (1962).

<sup>3</sup> J. F. RILEY and G. B. WEST, J. Physiol. (Lond.) 120, 528 (1953).

<sup>4</sup> E. P. BENDITT, R. L. WONG, M. ARASE, and E. ROEPER, Proc. Soc. exp. Biol. Med. 90, 303 (1955).

<sup>5</sup> W. K. O'STEEN and J. J. HRACHOVY, unpublished results (1963).