References and Notes

1. J. F. Jackson, *Exptl. Cell Res.*, in press.
2. P. S. Moorhead *et al.*, *ibid.* **20**, 613 (1960).
3. Generously supplied as the sodium salt by B. H. Sörbo, Karolinska Inst., Stockholm.
4. Y. Ohnuki, A. Awa, C. M. Pomerat, *Ann. N.Y. Acad. Sci.* **95**, 882 (1961).
5. N. L. R. Bucher and D. Mazia, *J. Biophys. Biochem. Cytol.* **7**, 651 (1960).
6. R. Cecil and J. R. McPhee, in *Advances in Protein Chemistry*, C. B. Anfinson, Jr., M. L. Anson, K. Bailey, J. T. Edsall, Eds. (Academic Press, New York, 1959), vol. 14, p. 255.
7. A. Meister, P. E. Fraser, S. V. Tice, *J. Biol. Chem.* **206**, 561 (1954).
8. E. Kun, *Biochim. Biophys. Acta* **25**, 135 (1957).
9. B. H. Sörbo, in *Methods in Enzymology*, S. P. Colowick and N. O. Kaplan, Eds. (Academic Press, New York, 1962), vol. 5, p. 990.
10. J. W. Hylin, *ibid.*, p. 987.
11. E. Kun and H. G. Williams-Ashman, *Experientia* **18**, 261 (1962).
12. A. White, P. Handler, E. L. Smith, D. Stetten, *Principles of Biochemistry*, (McGraw-Hill, New York, ed. 2, 1959), p. 850.
13. A. M. Rutenberg, R. Goffstein, A. M. Seligman, *Cancer Res.* **10**, 113 (1950).
14. M. Cerny, *Casopis Lekaru Ceskych* **100**, 709 (1961).
15. T. C. Hsu and O. Klatt, *J. Natl. Cancer Inst.* **22**, 313 (1959).
16. C. Bottura and I. Ferrari, *Blood* **21**, 207 (1963).
17. A. Levan and J. J. Biesele, *Ann. N.Y. Acad. Sci.* **71**, 1022 (1958).
18. T. C. Hsu, in *International Review of Cytology*, G. H. Bourne and J. F. Danielli, Eds. (Academic Press, New York, 1961), vol. 12, p. 69.

19. Supported by a grant (to K.L.-K.) from the Swedish Cancer Society. Part of a research program on human cytogenetics under the direction of Professor J. A. Böök, aided by grants from the Swedish Medical Research Council and the National Institutes of Health (grant B 4320). On leave from Tulane University, J.F.J. received a U.S. Public Health Service training stipend in human genetics at the Institute for Medical Genetics, Uppsala, Sweden, for 1962–63.

* Present address: Department of Medicine. 1430 Tulane Ave., New Orleans.

3 June 1963

Congenital Malformations in Hamster Embryos after Treatment with Vinblastine and Vincristine

Abstract. *Intravenous injection of vinblastine or vincristine, two antitumor chemotherapeutic agents used in humans, into pregnant golden hamsters on the 8th day of gestation, causes an increase in the fetal mortality rate and the appearance of a significant number of congenital malformations in the surviving fetuses.*

The teratogenic effect of colchicine in the pregnant hamster has been reported (*1*). In a survey of the permeability of the hamster placenta to colchicine and other possible mitotic inhibitors, two more compounds, vinblastine (vincaleukoblastine, VLB) and vincristine (VCR), showed a profound embryocidal and teratogenic effect (*2*). Since these compounds, which are obtained from the common periwinkle plant (*Vinca rosea* Linn.) (*3*), have been used very effectively as chemotherapeutic agents in human tumors (*4*), it seems advisable to mention the possibility of their teratogenic effect in man. Both compounds have similar chemical structures (*5*), yet they appear to produce definite differences in their effects on tumors (*3*). The antimitotic activity of VLB in the hamster has been demonstrated by Cardinali *et al.* (*6*) and VCR has exhibited an antimitotic effect in the bone marrow of the mouse (*7*). Sokal and Lessmann (*8*) have reviewed the literature on the effects of cancer chemotherapeutic agents during human pregnancy and have concluded that aminopterin and the combined therapy of busulfan-6-mercaptopurine have been the only agents thus far studied in this class of compounds that have any known relationship to human congenital malformations.

The hamsters were anesthetized with Nembutal, and a small 1-cm incision was made over the femoral vein into which various concentrations of each compound (Table 1) were injected directly on the 8th day of gestation. Control animals received equal volumes of saline intravenously in a similar manner. The fetuses were recovered on the 14th day of gestation and examined for gross congenital malformations. The mortality rate was calculated by determining the number of resorption sites when the animals were killed. All maternal animals survived the treatment without any evidence of anorexia, weight loss, or diarrhea during the course of the experiment. Ten pregnant hamsters, used as controls, were injected intravenously with equal volumes of normal saline.

Table 1 shows that both compounds have a distinct embryocidal effect in that an increase in dosage causes a progressive rise in the embryonic mortality rate, an effect similar to that of colchicine (*1*). Evidence of gross malformations in the recovered fetuses was recorded and suspicious areas were prepared for microscopic examination. In addition, all fetuses which appeared to have skeletal defects at autopsy were cleared in 1 percent KOH and the skeletons were stained with alizarin red (*9*).

Malformations noted in the group treated with VLB included microphthalmia, anophthalmia, spina bifida, and skeletal defects consisting mainly of rib fusions and vertebral arch deformities. When administered on the 8th day of gestation, 0.25 mg/kg appears to be the most effective teratogenic dose of VLB. The fetuses from those animals receiving VCR showed malformations consisting of microphthalmia, anophthalmia, mild exencephaly, and rib defects. Its most effective teratogenic dose for hamsters is 0.1 mg/kg. It is probable that the incidence of malformations with these two compounds would be greater if a more detailed search by serial histologic sectioning were made.

Table 1. Effect of vinblastine and vincristine on fetal survival and gross congenital malformations when injected intravenously into golden hamsters on the 8th day of gestation.

Dosage (mg/kg)	Litters (No.)	Fetuses			
		Treated (No.)	Surviving (No.)	Mortality (%)	Grossly abnormal (No.)
Vinblastine					
0.1	4	51	42	18	3
0.25	10	134	56	58	16
0.5-2.6	3	38	11	71	2
Vincristine					
0.1	4	51	39	23	6
0.25	3	43	15	65	2
0.5	4	53	7	85	1
0.6-2.3	6	70	9	87	1
Controls					
0 (saline only)	10	119	111	7	0

Colchicine-induced malformations under identical experimental conditions (*1*) showed a great similarity to the malformations described here for VLB and VCR. It would thus appear that one explanation for the common teratogenic action of these compounds might by their mitosis-arresting activity on the developing embryonic tissue. However, in view of the marked difference in the tumor-spectrum activity of these two drugs, other unknown mechanisms may be responsible for their teratogenic action (*10*).

VERGIL H. FERM

Department of Pathology, Dartmouth Medical School, Hanover, New Hampshire

References and Notes

1. V. H. Ferm, *Proc. Soc. Exptl. Biol. Med.* **112**, 775 (1963).

2. Supported by U.S. Public Health Service grant GM 10210-01.

3. I. V. Johnson, G. H. Svoboda, H. F. Wright, *Proc. Am. Assoc. Cancer Res.* **3**, 331 (abstr.) 1962.
4. O. S. Selawry and J. Hananian, *J. Am. Med. Assoc.* **183**, 741 (1963).
5. N. Neuss, M. Gorman, H. E. Boaz, N. J. Cone, *J. Am. Chem. Soc.* **84**, 1509 (1962).
6. G. Cardinali *et al.*, *Proc. Soc. Exptl. Biol. Med.* **107**, 891 (1961).
7. G. Cardinali, G. Cardinali, M. A. Enein, *Blood* **21**, 102 (1963).
8. J. E. Sokal and E. M. Lessmann, *J. Am. Med. Assoc.* **172**, 1765 (1960).
9. A. B. Dawson, *Stain Technol.* **1**, 123 (1926).
10. The VLB and VCR were supplied to Dr. F. G. Ebaugh, Jr., Dartmouth Medical School, by Dr. J. G. Armstrong, Eli Lilly Co., Indianapolis, Ind.

29 April 1963