

especially pronounced in the relative number of embryos surviving for 11 days or longer. For example, in 1954 4 of 44 or 9.0% of the embryos attained a size equivalent to that of 21-28-day normal turkey embryos, whereas in 1960 87 of 277 embryos or 31.4% were listed in this age category. Increased viability of parthenogenetic embryos is also reflected in the number of embryos completing their incubation period. Since 1956, 67 poults have been hatched, many of which lived for some time, a few surviving to maturity. Viable spermatozoa have been observed in semen samples obtained from each of 3 parthenogenetic males and offspring have been sired by one male, Olsen(11). In 1960, 15 poults were hatched, 4 of which are still alive, ranging in age from 71 to 136 days. These evidences of increased embryonic viability can be attributed in large part to the selective breeding program initiated in 1954.

Summary. A breeding program for increasing parthenogenetic development, based on the progeny test method, was initiated in 1954. More than 42,000 eggs from 926 Beltsville Small White virgin turkey hens were involved in this 9-year study. A marked increase in incidence of parthenogenesis, par-

ticularly of the more highly organized type, has resulted. Parthenogenetic development was observed in 16.7% of the eggs tested in 1952, in 41.7% in 1959. In 1952, embryos were encountered in 0.2% of all eggs whereas in 1959 11.7% of the eggs were so classified. Of 44 embryos found in 1954, 4 or 9% survived until 21-28 days of incubation; in 1960, 87 of 277 or 31.4% did so. Since 1956 a total of 67 embryos, all males, have survived to hatching, a few reaching maturity. Three males produced spermatozoa and one sired offspring.

1. Olsen, M. W., Marsden, S. J., *Proc. Soc. Exp. Biol. and Med.*, 1953, v82, 638.
2. ———, *J. Exp. Zool.*, 1954, v26, 337.
3. ———, *Science*, 1954, v120, 545.
4. ———, *Poultry Sci.*, 1956, v35, 674.
5. Kosin, I. L., Nagra, H., *Proc. Soc. Exp. Biol. and Med.*, 1956, v93, 605.
6. Sato, I., Kosin, I. L., *Proc. 10th Internat. Congr. of Genetics*, 1958, v2, 249.
7. Kosin, I. L., Sato, I., *Jour. of Morphology*, 1960, v103, 263-276.
8. Yao, T. S., Olsen, M. W., *J. Heredity*, 1955, v46, 133.
9. Poole, H. K., *ibid.*, 1959, v50, 150.
10. Poole, H. K., Olsen, M. W., *ibid.*, 1957, v48, 217.
11. Olsen, M. W., *Science*, 1960, v132 (in press).

Received August 1, 1960. P.S.E.B.M., 1960, v105.

Suppression of Human Choriocarcinoma Maintained in the Hamster Cheek-Pouch by Extracts and Alkaloids of *Vinca rosea*. (26083)

ROY HERTZ

Endocrinology Branch, National Cancer Institute, Bethesda, Md.

Noble, Beer and Cutts have reported their pioneer studies on the chemical and biological properties of crude extracts of *Vinca rosea*,* and of the alkaloid, Vincalukoblastine, initially isolated by them from these crude plant extracts(1,2). They noted the marked marrow depressant action of these preparations

in rats and described an interference with estrogen-induced uterine growth in the immature rat as well as carcinostatic effects produced by certain of the active plant fractions. The marrow-depressant, carcinostatic, and uterus-inhibiting properties appeared to resemble the known biological effects of such folic acid antagonists as Methotrexate (4-amino - N10 - methyl - pteroylglutamic acid). Since we had previously observed marked regression of choriocarcinoma in women following intensive Methotrexate therapy it seemed

* The extracts of *Vinca rosea* as well as the specific alkaloids derived therefrom were kindly furnished by Dr. Irving Johnson and Dr. G. H. Svoboda, Eli Lilly Co. Research Labs., Indianapolis, Ind.

TABLE I. Inhibitory Effect of Extracts and Alkaloids of *Vinca rosea*.*

Preparation	Dose, mg/kg†	% inhibition‡
Crude extract	11.25	30
	22.50	50
	33.75	90
	30.0	90
Vincaleukoblastine	.75	90
	.375	40
	.75	80
	.75	90
	.75	90
	.75	80
	.375	50
Leurosine	75.0	90
	37.5	90
	18.75	60
	9.37	40
	4.68	0
Virosene	3.75	0
Lochnerine	3.75	0
Perivine	3.75	0
Catharanthine	75.0	0
	150.0	0
Vindoline	75.0	0
	150.0	0

* WO strain of Erwin-Turner choriocarcinoma used throughout.

† Dosages indicated are daily subcut. inj. started on day 7 after transplantation and continued for 6 days; 8 to 10 animals/group.

‡ % inhibition estimated from comparison with saline treated controls.

desirable to determine the potential inhibitory effect of extracts of *Vinca rosea* and of its alkaloids(3,4) upon human choriocarcinoma maintained in heterologous transfer in the hamster cheek-pouch(5). We were further encouraged to proceed with these studies by the observations of Johnson concerning the oncolytic effect of these preparations in a number of animal tumors(6).

Materials and methods. In these studies we have employed the WO strain of the Erwin-Turner choriocarcinoma maintained in the untreated female hamster as previously described(5). The crude *Vinca* extracts and the respective alkaloids were prepared and characterized by methods described by Svoboda *et al.* and by Gorman *et al.*(3,4). The drug was started on the 6th day after transplantation and after initial growth of the transplant could be clearly observed. The

materials were given subcutaneously in the dosage regimen detailed in Table I. The growth pattern of the transplanted tumor was determined by direct inspection and recorded by free-hand, life size sketches.

Such records are admittedly suitable for recording only very substantial differences in growth rate. However, they permit an estimation of the extent of inhibitory effects obtained (Table I).

Results. The findings clearly indicate that growth of this tumor is markedly inhibited by Vincaleukoblastine, Leurosine, and by the crude plant extract. However, the remaining alkaloids, Virosine, Lochnerine, Perivine, and Catharanthine and Vindoline, all proved inert in the dosages applied.

The growth-inhibitory effects of Vincaleukoblastine and of Leurosine are not accompanied by significant toxicity as evidenced by effects on host body-weight or upon host survival. It would, therefore, appear that these agents may be expected to have an acceptable margin of safety for clinical trial.

Summary. Crude extract of *Vinca rosea* as well as the alkaloids, Vincaleukoblastine and Leurosine markedly inhibit growth of a heterologously transplanted human choriocarcinoma in the hamster cheek-pouch. The host tolerance to these agents suggests their potential clinical usefulness.

The able technical assistance of Messrs. Howard Erwin, Charles K. Turner, and James Brice is gratefully acknowledged.

1. Noble, R. L., Beer, C. T., Cutts, J. H., *Ann. N. Y. Acad. Sci.*, 1958, v76, 882.
2. Cutts, J. H., Beer, C. T., Noble, R. L., *Rev. Can. Biol.*, 1957, v16, 476.
3. Svoboda, G. H., *J. Am. Pharm. Assn.*, 1958, v47, 834.
4. Gorman, M., Neuss, N., Svoboda, G. H., Barnes, A. J., Cone, N. J., *ibid.*, 1959, v68, 256.
5. Hertz, R., *PROC. SOC. EXP. BIOL. AND MED.*, 1959, v102, 77.
6. Johnson, I. S., Wright, H. F., Svoboda, G. H., *J. Lab. Clin. Med.*, 1959, v54, 830; Johnson, I. S., personal communication.

Received August 30, 1960. P.S.E.B.M., 1960, v105.