ess, do not involve chromosomal rearrangements. It is possible that similar viral agents accidentally introduced *via* serum or tissue extracts could produce similar results in established cell lines.

In conclusion, it should be realized that morphological changes can occur in cell populations, and that they are not necessarily due to contamination by foreign cells. Such changes offer opportunities for study of the population dynamics of *in vitro* cell populations.

Summary. Morphological transformations occurred in 2 clone populations of pig kidney cells. Both populations carried distinct chromosomal markers whose incidence and type did not alter during transformation. Thus, the alteration of morphology was intrinsic and could not be due to cell contamination. In one clone, a change in chromosome number seemed to be related to the appearance of a new morphological type.

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Relationship of Serotonin and Reserpine in the Chicken. (27121)

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Numerous effects have been ascribed to the use of reserpine as a poultry feed additive. Reports on average daily gain and feed efficiency have been inconsistent(1-5). Experiments have been conducted to determine the sedative action of reserpine in the hope of obtaining a drug that will reduce economic losses caused by undue excitement of the poultry flock. The results of such tests were inconclusive(2,4,5). Effects of orally administered reserpine on egg production and quality have been reported(6,7). Low levels of reserpine in the diet increased egg weight and shell quality during periods of high environmental temperatures. No effect on albumen quality was seen.

Inconstant results obtained in feeding tests may be related to many factors, *e.g.*, dose, duration of intake, age of animal, and diet. Carlson(8) suggested that poultry rations low in tryptophan, a precursor of serotonin (5-hydroxytryptamine), might result in growth depression as a result of adding reserpine to the feed. Although many reserpine-feed additive tests have been conducted, the pharmacodynamics of reserpine in poultry has not been extensively studied.

Sturkie *et al.*(9) investigated the physiologic effects of reserpine on the chicken and observed that it produced a significant hypothermia, bradycardia and a decrease in mean arterial pressure when administered intramuscularly at levels of 0.1 to 0.2 mg/kg. This level of reserpine produced diarrhea.

Premachandra *et al.*(10) investigated the relationship of parenterally administered reserpine and thyroid activity in chickens. No effect on thyroid uptake of I^{131} was observed.

TABLE I. Serotonin in Tissues (in μg/g of Tissue).

Tissue	No. of chickens	Mean and S.E.
Normal chickens		
Duodenum	10	12.4 $\pm$ 1.13
Proventriculus	6	6.3 $\pm$ 1.04
Liver	6	4.4 $\pm$ 1.03
Crop	6	0
Reserpine-treated chickens		
Duodenum	5	0
Proventriculus	5	0
Liver	5	0

Similarly, a normal rate of release of I^{131} from the thyroid gland resulted.

It has been demonstrated that reserpine impairs the ability of tissues to maintain serotonin in a bound form. Thus, serotonin is released and metabolized. Reserpine may be considered to be an inhibitor of the serotonin-storing mechanism of the tissues(11). The physiologic disposition of reserpine and serotonin following reserpine administration indicates that, although reserpine disappears rapidly, its effects on serotonin and its pharmacologic effects persist(12). Degree of sedation is directly related to the reduction in tissue serotonin content.

This communication reports the serotonin content of avian gastrointestinal tissues before and after parenteral administration of reserpine.

Materials and methods. Adult Leghorn hens, weighing 1.5-2.5 kg, individually housed and fed a commercial laying ration, were used in this study. Portions of the crop, proventriculus, duodenum and liver were obtained for assay.

Serotonin was extracted from the tissues and assayed colorimetrically following the procedure outlined by Udenfriend *et al.*(13) which utilizes 1-nitroso-2-naphthol.

Reserpine* was administered at a level of 4 mg/kg subcutaneously 12 hours before the tissues were obtained at necropsy.

Results and discussion. Tissues from 15 birds were assayed for serotonin. Results are presented in Table I. The serotonin content of the duodenum of normal chickens was the highest of the tissues tested, with an average of 12.4 μg per g of fresh tissue. The proventriculus, a small glandular organ which secretes gastric juice, contained an average of 6.3 μg serotonin per g of fresh tissue. The liver contained an average of 4.4 μg serotonin per g of fresh tissue. Analyses of normal crops failed to demonstrate the presence of serotonin in this organ.

Analyses of the 3 different organs from reserpine-treated chickens failed to show the presence of serotonin. Gross sedative effects were observed 4 to 12 hours after administration of reserpine.

Although the normal serotonin tissue concentrations obtained in this study are somewhat higher than those reported by Erspamer (14) and Udenfriend *et al.*(13) the differences in assay procedures, animal breed, sex, diet and environment must be considered. The serotonin content of the crop, proventriculus, and duodenum of the chicken corresponds proportionately to that of their anatomical counterparts in laboratory animals such as the dog and rabbit.

Summary. It appears that in chickens as in other animals, reserpine causes release of bound serotonin which is subsequently metabolized. Sedation was observed when the tissue serotonin content was subnormal.

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Inhibition of Ethanol Metabolism by Oral Antidiabetics.* (27122)

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Ethanol is metabolized in the body by the following processes:

$$\text{Ethanol} \xrightarrow{1} \text{Acetaldehyde} \xrightarrow{2} \text{Acetate} \rightarrow CO_2 + H_2O.$$

Reaction 1 is catalysed by the DPN-dependent enzyme alcohol dehydrogenase (ADH), reaction 2 by the likewise DPN-dependent acetaldehyde dehydrogenase (AcDH).

In vitro studies(17) have shown that tolbutamide inhibits a number of DPN-dependent enzymes, among these ADH. These findings have been confirmed(5) and it was also reported that AcDH is inhibited by tolbutamide. The inhibition seems to be explained by a substrate competition between tolbutamide and DPN(5). This may account for the uncomfortable flushes experienced by some tolbutamide or chlorpropamide treated patients upon exposure to alcoholic beverages(2,3,4,7,9,10,16). During the reaction, which resembles the ethanol-antabuse reaction, high concentrations of acetaldehyde have been found in the blood and the expiratory air(6,8).

As tolbutamide inhibits both ADH and AcDH *in vitro*, it would be expected that the metabolism of ethanol would be diminished after tolbutamide *in vivo*, but in two reports (6,8) this effect could not be demonstrated. However, we considered it pertinent to reinvestigate this question under conditions which permitted a more exact determination of the metabolism of ethanol in the hours preceding and following intravenous injection of tolbutamide. Furthermore, the effect of a tolbutamide metabolite and of carbutamide was tested with the same technic.

Methods. The experiments were performed on intact, nembutal anaesthetized cats in the postabsorptive state. Cannulas were placed in a femoral artery and a saphenous vein for blood sampling and injections. A priming dose of ethanol was injected intravenously. This resulted in a plasma concentration between 50 and 150 mg/l. To maintain this concentration a continuous infusion of ethanol was given at a rate calculated from the average elimination rate of ethanol for cats. The first blood sample was taken one hour after the priming dose when diffusion equilibrium of ethanol had been established. Arterial blood samples were then taken every 30 minutes for 4 hours. Two hours after the sampling was started, the compound to be investigated was injected slowly intravenously in an amount of 100 mg/kg. Ethanol concentration in the plasma was determined enzymatically by the method of Lundquist and Wolthers(12) (coeff. of var. 0.7%); plasma glucose by the Somogyi-Nelson technic(14).

From the weight of the animal, the volume of distribution of ethanol and rise or decline of ethanol concentration, the disappearance rate was determined before and after sulphonylurea injection.

In 8 experiments tolbutamide was given. In one experiment its primary elimination product N - (4-hydroxymethyl-benzolsulphonyl)-N'-n-butylurea (HBBU) and in 3 other experiments carbutamide was given. In 3 control experiments saline was injected instead of sulphonylurea.

Results. The results appear in Table I.

* Tolbutamide (Rastinon) and HBBU were given by the Copenhagen Representative for Farbwerke Hoechst, Lautrup-Larsen A/S. Carbutamide (Nadisan) was provided by C. F. Boehringer & Soehne through A. H. Allesen Holm A/S. The technical assistance of Miss Ulla Bengtsson and Mrs. Inga Almdal is gratefully acknowledged.