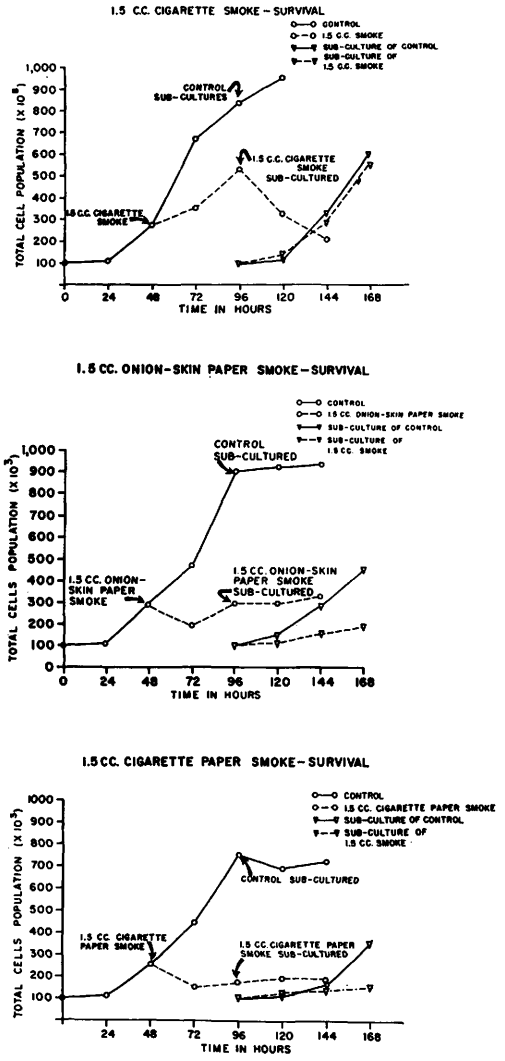

FIG. 1. Survival studies on Strain L cells exposed to 1.5 cc of cigarette smoke, 1.5 cc of onion skin paper smoke, or 1.5 cc of cigarette paper smoke.

It was also noted that particulate matter found in smoke varied per unit amount according to the source and volume of the smoke. Smoke from paper contained more particulate matter than smoke from cigarette tobacco or from the whole cigarette. The experimental changes appeared to be related in part to the amount of particulate matter in the medium after exposure. In recent investigations of the effect of cigarette smoke on ciliated epithelium(3), the results indicated impedance of mucus flow and ciliary action. This work suggested that the particulate phase of cigarette smoke may be of significance.

Perfusion chamber studies of the immediate effect of smoke and non-smoke gases on cell morphology are being reported(4).

Summary. Under the conditions of this study it appears that there is a "toxic" effect of smoke gases on strain L cells in culture. This results both in morphological changes in the cells and in alterations in growth curves as indicated by the reduction of cell populations. The smoke from the combustion of either cigarette paper or onion skin paper appears to be more "toxic" than that from the combustion of a cigarette or cigarette tobacco alone. The effect of cigarette or other smoke gases may result in part, at least, from the particulate matter present in smoke gases.

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Excretion of Urinary 5-HIAA in Dog Pellagra.* (27406)

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It is generally accepted that serotonin synthesis is related to tryptophan intake. The major pathway of tryptophan metabolism leads to kynurenin → 3-OH-kynurenin → 3-OH-anthranilic acid and → nicotinic acid (1,2). The minor pathway leads to 5-OH-

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TABLE I. Dog-Food Composition.*

	Component	Control diet	Tryptophan deficient diet	Nicotinic acid deficient diet
1	Sucrose	66 g	66 g	66 g
2	Cottonseed oil	11 g	11 g	11 g
3	Casein (vitamin-free)	19 g	—	—
4	Corn-starch	—	19 g	—
5	Grounded-corn (zea-maize)	—	—	19 g
6	Salt mixture (NB standard)	4 g	4 g	4 g
7	Nicotinamide	2 mg	—	—
8	Thiamine	.2 "	.2 mg	.2 mg
9	Riboflavin	.3 "	.3 "	.3 "
10	Pyridoxine	.2 "	.2 "	.2 "
11	Calcium pantothenate	1.6 "	1.6 "	1.6 "
12	Choline chloride	100.0 "	100.0 "	100.0 "
13	Vit-A	10,000 I.U.	10,000 I.U.	10,000 I.U.
14	Vit-D	10,000 "	10,000 "	10,000 "
15	Vit-E			
16	Vit-K	.1 mg	.1 mg	.1 mg

* Items No. 1-6 are calculated per 100 g of food. Items No. 7-16 are calculated per average daily food intake.

tryptophan → 5-hydroxytryptamine (serotonin) → 5-hydroxyindole acetic acid (5-HIAA). Both pathways depend on the effect of pyridoxal phosphate(3). Thus, tryptophan is the precursor of both nicotinic acid and serotonin.

Pellagra is known to be caused by a deficiency in tryptophan, nicotinic acid or both. Limiting intake of these essential amino acids and thereby producing the deficiency disease could lead to a decrease in the products of the above metabolism, including serotonin. It should be noted that in pellagra mental symptoms occur in the final stage and might, therefore, be caused by general depletion of serotonin reserves in both brain and blood platelets. To test this hypothesis, we studied urinary excretion of 5-HIAA in pellagrin dogs, from the beginning of the deficiency disease until its final stages.

Animals and methods. We used 7 five-week-old puppies of both sexes, from one litter. Immediately after weaning, 4 dogs were placed in one metabolic cage forming the experimental group and 3 in a second cage serving as controls. They were not kept in individual cages so as to enable us to follow their "social" behaviour. We also knew from previous experiments that isolation in individual cages might act as a physical and mental stress factor which we tried to eliminate.

The experiment was divided into 2 parts: the first stage (53 d) included creation of the deficiency disease by a tryptophan deficient diet and measuring 5-HIAA output. During the second stage (13 d) urinary 5-HIAA was measured under a nicotinic acid deficient diet, as nicotinic acid synthesis can originate from tryptophan(4). The composition of the 2 diets and of the control-diet is given in Table I(5,8).

The dogs were observed daily for 66 days, at the same hour, by the same person searching for clinical symptoms of the suspected deficiency disease, as: glossitis, salivation, vomiting, diarrhea, skin defects with alopecia, lacrimation, photophobia, scleritis, conjunctivitis, nystagmus, appearance of "black tongue".

Body weight was checked once weekly; 24-hour urine was collected daily in a jar containing 1/30 of the expected urine volume of a mixture of 28 ml glacial acetic acid and 2 ml toluene. The 5-HIAA level was determined by the method of Macfarlane *et al.* (6), 2 samples of 5 ml each being examined of every specimen. EEG tracings were recorded at beginning of experiment on both groups and were repeated on appearance of the first clinical symptoms. The tracings were taken in 2 areas of the skull—frontal and occipital—with 2 pairs of electrodes; they were recorded by a penwriter "Med-

TABLE II. Pellagrin Symptoms in Dogs on a Diet Deficient in Tryptophan and Nicotinic Acid.

Symptoms	Experimental group	Day of appearance	Control group
Anorexia	+	30	—
Glossitis, magenta tongue	+	35–38	—
Mucoid diarrhea	+	10–15	—
Salivation	+	25–30	—
Vomiting	+	40–42	—
Conjunctivitis, lacrimation, scleritis	+	40–50	—
Photophobia	—	—	—
Skin lesions and alopecia	+	50–52	—
Nystagmus	—	—	—
Black tongue	—	—	—
EEG (voltage frequency pattern)	normal	—	normal

TABLE III. Average Values (and Standard Error) of Diuresis and 5-HIAA Excretion in Pellagrin Dogs.

		Diuresis per Dog ml/d			5-HIAA per Dog mg/d	
Days of exp	Excretion	1-10	11-25	26-35	36-50	51-66
Control dogs	Diuresis	60 (± 3%)	96 (±4%)	100 (± 3%)	70 (±5%)	63 (± 6%)
	5-HIAA	.580 (± 8%)	.620 (±7%)	.435 (± 9%)	.370 (±8%)	.380 (±10%)
Tryptophan deficient dogs	Diuresis	68 (± 5%)	102 (±4%)	80 (± 5%)	—	—
	5-HIAA	.480 (±10%)	.700 (±6%)	.642 (±12%)	—	—
Nicotinic acid deficient dogs	Diuresis	—	—	—	67 (±6%)	58 (± 4%)
	5-HIAA	—	—	—	.413 (±6%)	.450 (± 5%)

craft" apparatus.

Results. Among clinical signs, characteristic for pellagra, we found 3 main symptoms in the experimental dogs: glossitis (magenta tongue); anorexia, expressed by reduced food intake; and watery, mucoid diarrhea. The exact incidence of these changes is given in Table II. Mental symptoms were absent and there was no change in the EEG. There were no significant changes in the volume of excreted urine during the experiment in either group, nor in level of 5-HIAA in either of the 2 groups until the pellagrin dogs died (Table III).

Discusion. Our results demonstrate that serotonin production and 5-HIAA excretion do not depend on supply of tryptophan or nicotinic acid. Even when the pellagrin dogs died from their deficiency they still showed normal 5-HIAA levels in the urine. We cannot exclude the possibility that pellagrin dogs do not metabolize serotonin to 5-HIAA alone, but excrete other metabolites in the urine. Thus, *e.g.*, tryptamine has been found in the urine of human pellagrins(7,8). However, since up to 60% of daily tryptophan intake is excreted in man as 5-HIAA(9) and since a similar proportion should be assumed for dogs(10), we feel that urinary 5-HIAA excretion in dogs reflects mainly intestinal serotonin and is not sensitive enough to discover serotonin deficits of the hypothalamus in its early stages. Moreover, dogs die too early to show mental symptoms due to hypothalamic serotonin deficiency. This has been shown by earlier investigations of canine black tongue(11,12). It is also well known that the black tongue is only the outcome of a necrotic secondary infection similar to noma, whereas the beet-red magenta tongue is typical of dog pellagra.

Summary. Dogs kept on a diet deficient in tryptophan and nicotinic acid, developed typical pellagra, but their 5-HIAA urinary excretion did not differ from that of control dogs. The reasons for this discrepancy lie mainly in the typical course of canine pellagra which invariably results in early death due to intestinal or buccal infection (black tongue) before mental symptoms are likely to develop.

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Induction of Runt Disease in Adult Mice by Pre-sensitized Homologous Lymphoid Cells.* (27407)

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The condition of runt disease has now been generally accepted as an immunologic reaction of graft against host tissue(1). This syndrome, often referred to as either homologous, wasting or secondary disease, has been described as the consequence of introducing immunologically competent adult cells into host animals incapable of rejecting the donor cells. The unresponsive hosts have been either fetal or neonatal animals, or adult animals which have been made tolerant at birth or have been subjected to lethal irradiation. Another form of immunological unresponsiveness has been seen in F_1 hybrids which were genetically incapable of rejecting their parental tissues.

Injection of homologous lymphoid cells into immunologically competent adults has not usually resulted in runt disease presumably because of the rapid destruction of the injected cells by the host. Castermans(2), however, in an attempt to produce Felton's type of immune paralysis to tissue antigens, used repeated injections of homologous splenic cells and was able to show a partial abrogation of the host's immune response to homografted skin. In addition, he observed lymphoid hypoplasia, loss of body weight and other characteristics of runt disease in some of the injected animals. More recently, Shapiro *et al.*(3) were able to achieve in parental animals "tissue tolerance" to their F_1 hybrids after repeated injections of F_1 splenic cells.

The purpose of this study was to investigate the possibility of inducing runt disease in normal adult mice with a single injection of homologous lymphoid cells from donors sensitized to the prospective host's tissues. It was also desired to learn whether, by injection of a proper number of homologous cells, a nice balance could be achieved between host and foreign cells so that a permanent chimera might be created without the complication of runt disease.

Materials and methods. An outline of the method is illustrated in Fig. 1. Two inbred strains of mice, A and CBA, were used throughout the study. Seventy-five mice of each strain were sensitized to the homologous strain by cross skin grafts. To harvest sensitized cells from regional cervical and axillary lymph nodes, the skin grafts were cut into triangles and placed on recipients with the apex of the graft in the nuchal region and the base crossing the scapulae. To insure a maximal sensitization, all the animals were injected intraperitoneally with 10^7 homolo-

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