50% higher than the value obtained from the classical goiter-prevention assay without radioiodine in the young chicken. The value derived from the classical goiter-prevention assay may be closer to normal secretion rate, because in this way thyroxine substitution usually starts on the same day as the thiouracil feeding, and the increase in TSH production by the pituitary by thiouracil is inhibited in chickens receiving the successive excess dose of thyroxine.

Summary. The effects of thiouracil on turnover of I^{131} by the thyroid in immature and mature chickens were studied. Rate of uptake of I^{131} by the thyroid was significantly reduced and release rate of I^{131} from the thyroid was significantly augmented in thiouracil-fed birds. Rate of recycling of I^{131} from the metabolized thyroid hormone measured by accumulation of I^{131} by the thyroid from I^{131}-labelled thyroxine injected was always a little less than rate of uptake of I^{131} by the thyroid. Both uptake and recycling of thyroidal I^{131} decreased with length of the period of thiouracil feeding. Rate of release of I^{131} from the thyroid increased with the lengthened time of thiouracil feeding. The thyroxine secretion rate expressed either per 100 g body weight or per bird measured by the thyroxine substitution significantly increased in thiouracil-fed birds compared with normal diet-fed birds. It also increased with length of thiouracil feeding. Use of thiouracil is not adequate for precise determination of thyroxine secretion rate in the chicken.

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Incorporation of Tritiated Thymidine into DNA after Oral Administration.* (26234)

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Recent studies of cellular proliferation utilizing parenteral tritiated thymidine, H^3TDR, have demonstrated widespread incorporation of this DNA precursor into proliferating cells(1,2,3). Metabolic studies with H^3TDR in man have shown prompt utilization of this compound for DNA synthesis with degradation of approximately one-third of the injected dose to tritiated water and excretion in the urine of only small amounts as non-volatile tritiated products (4). No information is available on the fate of ingested H^3TDR. Such studies are needed for evaluation of the potential hazard of this material in respect to its laboratory use. In this paper autoradiographic (ARG) studies will demonstrate absorption following oral administration and subsequent incorporation

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TABLE I. H^3 Activities of Body Fluids Following H^3 Thymidine Ingestion.

		Ingested H^3TDR, μc	Sacrificed after	Catabolized to tritiated water, %	Urine non-volatile H^3 activity excretion, %
Guinea pig	1	500	2 hr	15.8	.35
	2	100	2	16.5	.32
Rat	1	100	3	8.1	.36
	2	100	3	8.1	.23
	3	100	6	8.4	.32
	4	100	6	10.9	.41

of H^3TDR into DNA of widespread body tissues.

Methods and materials. Adult male guinea pigs, rats, and mice were maintained on a stock diet. H^3TDR of high specific activity, 1.9C/mM,† was used for all studies. Solutions of H^3TDR were introduced into the stomachs of animals by intubation with a curved blunt needle. The animals were killed after varying times. ARG's were prepared from formalin fixed tissues by methods previously described(2,3). In some animals the penile urethras were ligated at the time of H^3TDR ingestion to study the appearance of tritium activity in urine as well as plasma and bile. Total tritium activity and distilled volatile H^3 activity, (tritiated water) were assayed by liquid scintillation spectrometry(4). Non-volatile H^3 activity was estimated from the difference between total H^3 activity and tritiated water activity of the same sample.

In one guinea pig, tissues were obtained 2 hours after H^3TDR ingestion along with gastric contents. These were lyophilized to remove tritiated water. Aliquots were dissolved in Hyamine and assayed for non-volatile H^3 activity using liquid scintillation counting(5).

Results. Results of H^3 counting are shown in Table I. Two hours after H^3TDR ingestion in guinea pigs, plasma activity was predominantly tritiated water. Assuming uniform distribution in total body water, approximately 15% of ingested H^3TDR had been catabolized to tritiated water and significant plasma non-volatile H^3 activity was not present. Tritiated water activity of the bile was similar to plasma; non-volatile H^3 was not detected. Urine from the obstructed bladders contained non-volatile H^3 activity that accounted for 0.35 and 0.32% of the ingested dose.

Autoradiographs of guinea pig tissues demonstrated extensive labeling. Heaviest label was seen in the small intestinal mucosa. Labeling in other tissues examined was scant but definite in bone marrow, spleen, lymph node, liver and kidney.

Guinea pig No. 1 received 500 μc H^3TDR *per os* and was sacrificed 2 hours later. Tissues and stomach contents were lyophilized and dissolved in Hyamine. Non-volatile H^3 activity of these tissues are recorded in Table II.

Similar results were obtained in 4 rats which received 100 μc of H^3TDR and were sacrificed at 3 and 6 hours after ingestion. Tritiated water activity was present in bile, plasma, and urine and significant but only small amounts of non-volatile H^3 activity were present in bladder urine (Table I).

To demonstrate better intranuclear labeling, a rat (200 g) and a mouse (30 g) were similarly intubated and given 100 μc and 25 μc H^3TDR respectively. The animals were killed 7½ hours later. After 5 weeks

TABLE II. Non-volatile H^3 Activity of Guinea Pig Tissues 2 Hours after H^3TDR *per os.*

	Non-volatile H^3 activity, cpm/mg dry wt
Stomach contents	442
Spleen	44
Liver	28
Bone marrow	28
Kidney	15
Ilium	11
Iliopsoas	5

† Schwarz BioResearch Inc., Mt. Vernon, N. Y.

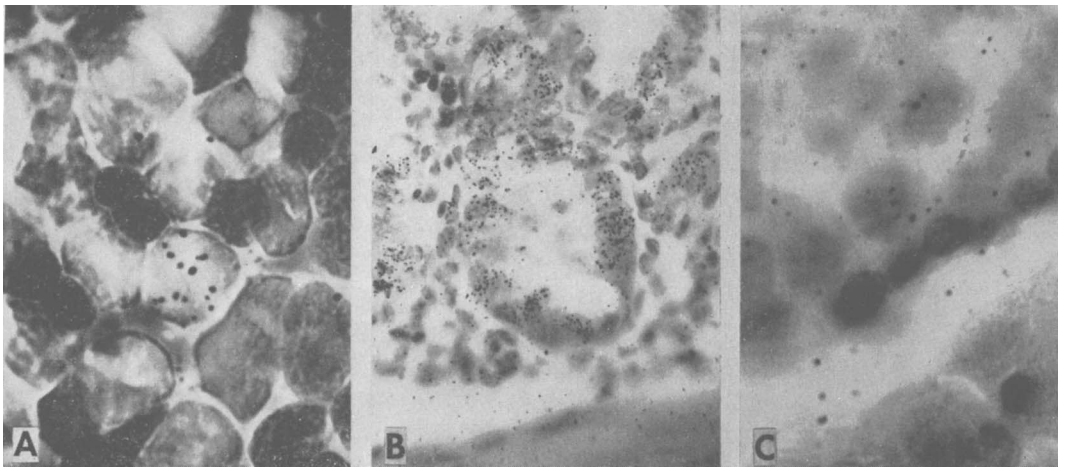

FIG. 1. ARG of rat and mouse tissues 7½ hr after H^3 thymidine ingestion. A = mouse bone marrow; B = rat ilium; C = mouse testes.

exposure, radioautographs (Fig. 1, A, B, C), were obtained.

Discussion. The widespread labeling of proliferating body tissues after H^3TDR ingestion strongly suggests that absorption of H^3TDR through the intestinal mucosa occurs. The extensive labeling of gastrointestinal mucosa seen here is indistinguishable from mucosa labeled with H^3TDR following parenteral injection(6) and after incubation of gastrointestinal mucosa *in vitro* with H^3TDR(7).

Wilson and Wilson(8) using an *in vitro* system have demonstrated biochemically the ability of thymidine to pass intact through guinea pig intestinal mucosa.

Upon ingestion of H^3TDR, absorption from the stomach is slow. Most of the H^3TDR remained within the stomach contents of guinea pig I which was sacrificed 2 hours after ingestion (Table II).

The gastrointestinal mucosa was labeled most heavily in these experiments. Other tissues received only small amounts of the label. The lyophilized tissues did contain non-volatile H^3 activity (Table II). These activities are based on mg dry weight of tissues and fail to take into account the varying cellularity and extent of cell renewal populations of the tissues studied. In spite of these difficulties, the data show non-volatile H^3 was present in the spleen and other tissues with very little present in muscle. These results are similar to those seen in rat tissues after parenteral H^3TDR(9). We know of no evidence indicating storage of H^3TDR in tissues other than in DNA of proliferating cells.

Some H^3TDR is catabolized to tritiated water; the site of this degradation is not known. Small amounts of readily detectable non-volatile H^2 activity are excreted in the urine after H^3TDR ingestion. Some of this may be H^3 beta-aminoisobutyric acid as occurs after intravenous H^3TDR injection in man(4).

Bile and plasma obtained from these animals contained tritiated water but failed to reveal the presence of any detectable non-volatile H^3 activity. The absence of non-volatile H^3 activity in bile is suggestive of the absence of any enterohepatic circulation of H^3TDR or its degradation products. The absence of plasma non-volatile H^3 activity was expected in view of the rapid disappearance curve of injected H^3TDR in man(4).

Hazards from H^3TDR have been postulated(2,12) and radiation effects described (10,11,13). The labeling of spermatogonia following ingestion of H^3TDR shown here further urges reasonable caution in handling this compound in any significant amounts.

Summary. 1. H^3TDR was administered *via* stomach tube to rats, mice, and guinea pigs. 2. Some ingested H^3TDR was absorbed and labeled proliferating cells of gas-

trointestinal mucosa as early as 2 hours after ingestion in guinea pigs. 3. Labeling of diverse tissues including proliferative cells of the testes was seen in rats and mice 7½ hours after ingestion. 4. Some of the H^3TDR was degraded to tritiated water while small amounts of non-volatile H^3 activity were excreted in the urine. 5. The significance of absorption of H^3TDR through intestinal mucosa is discussed as related to potential hazard.

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Staining of Malarial Parasites by the Fluorescent Antibody Technic. (26235)

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The fluorescent antibody technic has been used successfully by a number of workers to stain a variety of organisms(1, and others). The ease with which serums may be conjugated with the isothiocyanate derivative of fluoerscein and the stability of the labeled preparation has rapidly advanced the method from one of a purely research tool to one which is developing into a diganostic instrument as well. Our study was made to bring this useful and proven procedure to the study of malariology where its research and diagnostic potential appear promising. The following experiments were undertaken: (1) to determine whether natural infections produce antibodies which could be tagged with the dye to detect the organisms in the host; (2) whether antibodies could be produced in rabbits by a series of injections with malarial organisms and (3) whether one stage of the parasite, *e.g.*, the sporozoite which develops in the mosquito, could be used to produce antibodies which would react with the erythrocytic stage in the vertebrate host.

Materials and methods. Serum was collected from 2 *Macaca mulatta* monkeys which had been inoculated originally with blood containing about 2.0×10^8 parasites of *P. cynomolgi bastianellii.* Each of these animals developed a high parasitemia (average 2.0×10^5 parasites/mm^3 of blood). They apparently had a spontaneous recovery from the disease. About 7 months later both animals were reinoculated with 2.0×10^8 parasites and again developed a fairly high parasitemia (about 5.0×10^4 parasites/mm^3). After approximately 6 weeks the monkeys were inoculated for the third time with the usual inoculum of parasites. Neither animal developed a parasitemia. One of the animals was given a fourth inoculation one week later. No parasites were detected by blood smear examination for one week. At this time the animals were bled. The serums were pooled and the gamma globulin