renal damage is probably reflected in the weight loss shown by the animals exposed to the higher dose levels. While the hypertrophic response of the kidney following uninephrectomy may not be affected by rather large amounts of radiation, there is evidence of latent injury in that mean survival time of such locally irradiated mice is reduced. This effect, however, is seen also in control mice in which both kidneys have undergone local irradiation: with 40 kr both groups showed a mean survival time of about 10 days; with 30 kr about a month; with 20 kr almost 3 months; with 10 kr about 3½ months.

The possibility exists that a kidney irradiated immediately after removal of the contralateral organ might be more or less sensitive than one undergoing hypertrophy at the time of exposure. To test this, mice were uninephrectomized, subjected to local kidney irradiation 5 days later, and sacrificed at 14 days. The picture is similar to that seen when irradiation occurs immediately after nephrectomy; the hypertrophic response shows a significant decrease at about the 20 kr level (Table II). It has been reported(6) that sublethal whole body X-irradiation 3 hours after unilateral nephrectomy suppresses mitosis and inhibits kidney enlargement in LAF_1 mice. However, if irradiation is delayed 48 hours, until mitosis has occurred, enlargement is essentially similar to that seen in nonirradiated controls. There are obvious differences of local vs whole body radiation effects as well as possible strain differences, but the discrepancy between these results and ours may also be attributable to the age-dependency noted previously.

Summary. In adult female CF no. 1 mice, kidney enlargement after unilateral nephrectomy is a consequence mainly of hypertrophy. New cell formation as evidenced by an increase in total DNA content of the kidney may be a contributing factor in young animals but this becomes negligible with increasing age. The compensatory response occurring after uninephrectomy in the adult mouse is quite radioresistant, whether irradiation occurs immediately or 5 days post-operatively. This contrasts with the well-known radiosensitivity of cell renewal systems.

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Metabolic Fate of Injected C^{14}-Phytosterols.*‡ (27076)

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It has been demonstrated(1-4) that plant sterols are absorbed by animals and man to a slight extent. The mechanism of phytosterol absorption has been investigated and shown to be similar to that of cholesterol(4). Several reports(1,2,5) have shown that following administration of H^3-β-sitosterol or C^{14}-ergosterol by feeding or intravenous injection labeled sterols could be detected in a number of tissues, the highest levels of activity being in the serum and liver. There is also evidence that fed H^3-β-sitosterol disappears more rapidly from the tissues and blood than cholesterol (1). Regarding the metabolic degradation products of the absorbed plant sterols, it has

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been shown that H^3-β-sitosterol is converted to H^3-cortisol in the guinea pig(5) and that C^{14}-ergosterol and H^3-β-sitosterol may be converted to bile acids(2,5). Thus far, it does not appear that fed or injected labeled plant sterols(1,2,5) are converted to cholesterol. In continuation of our studies(4) on plant sterols we have determined the comparative tissue distribution of injected C^{14}-phytosterols and cholesterol-4-C^{14}, their conversion to the esterified form in blood and tissues, and their conversion to bile acids.

Methods and Materials. Male rats weighing 225-250 g, of the Carworth strain, were divided into 4 groups; 2 groups were normal fasted rats which received intravenously (femoral vein) either 0.5 ml of an emulsion containing 0.34 mg cholesterol-4-C^{14} or C^{14}-phytosterols. The other 2 groups had bile fistulas and received similar emulsions. The source of the C^{14}-phytosterols was a crude lipid extract from the leaves of tobacco plants grown in a $C^{14}O_2$ atmosphere. The sterols were isolated and identified as described earlier(4). The labeled sterol emulsions were prepared by dispersing 0.34 mg of the labeled sterol in 0.5 ml of a solution containing 3 mg soybean lecithin in 0.05 M phosphate buffer pH 7.3(2). The animals were maintained under light anesthesia during the intravenous injection period (about 2 minutes). Bile was collected during the ensuing 24-hour period in the animals with bile fistulae. Twenty-four hours after injection of the labeled sterols the animals were sacrificed, and blood, liver, small intestine and adrenals removed. The small intestine was washed with saline and these washings were pooled with the feces of each animal. Lipid extracts of the tissues were prepared according to procedures described earlier(6). The bile samples (10-15 ml) were extracted with 10 volumes of ethanol and the sterols and bile acids separated(7). Free and total sterols were determined by the method of Sperry and Webb(8). The C^{14}-activities of the free and esterified tissue sterol fractions were determined as previously described(6). To ascertain if the injected C^{14}-phytosterols were converted to C^{14}-cholesterol, the sterols isolated from the several tissues were subjected to repeated passage through the dibromide and crystallization of the dibromides from 5:1 ether-acetic acid. This procedure has been shown to eliminate H^3-β-sitosterol from a sterol mixture containing cholesterol(5); the plant sterols do not form insoluble dibromides.

Results. The recovery of injected cholesterol-4-C^{14} and C^{14}-phytosterols (after 24 hours) in several tissues is shown in Table I. Of the tissues examined, the liver had the largest percentage of injected C^{14}-sterols. The percentage of injected C^{14}-sterols recovered as free C^{14}-sterol in the serum, liver and adrenals was the same for both the cholesterol-4-C^{14} and C^{14}-phytosterol groups. In the small intestine significantly ($P<.01$) less free C^{14}-sterol was recovered in the group injected with C^{14}-phytosterols than in the cholesterol-

TABLE I. Recovery of Injected Cholesterol-4-C^{14} and C^{14}-Phytosterols in Different Tissues after 24 Hours.

Tissue*	% Injected C^{14}-Sterol Recovered as Digitonide							
	Free		Ester		Total		% Ester	
	C^{14}-chol. group	C^{14}-phyt. group	C^{14}-chol. group	C^{14}-phyt. group	C^{14}-chol. group	C^{14}-phyt. group	C^{14}-chol. group	C^{14}-phyt. group
Serum†	2.32 ± .36§	2.50 ± .20	4.52 ±.72	2.74 ±.10	6.84 ±1.04	5.24 ± .30	66.1 ±2.0	52.3 ±3.0
Liver	20.88 ±4.14	20.82 ±2.50	2.12 ±.52	.96 ±.50	23.00 ±4.66	22.00 ±3.00	9.2 ±1.3	4.4 ±2.0
Small Intestine	3.60 ± .54	2.40 ±1.04	.24 ±.10	.08 ±.02	3.84 ± .64	2.52 ±1.01	6.3 ±1.5	3.2 ±1.0
Adrenals	.20 ± .02	.24 ± .06	.60 ±.14	.66 ±.08	.80 ± .16	.90 ± .16	75.0 ±1.6	73.3 ±2.3
Feces and Intestinal Contents	1.22 ± .40	4.04 ± .74	.10 ±.02	1.16 ±.26	1.32 ± .42	5.20 ±1.00	7.6 ±2.0	22.3 ±4.6

* Values represent average of 5 rats per group; each rat was injected with an emulsion containing either C^{14}-phytosterols (.34 mg; 61,000 CPM) or cholesterol-4-C^{14} (.34 mg; 243,000 CPM).
† Serum sterol content was estimated from blood volume and serum sterol concentration.
§ Standard deviation.

4-C^{14} group. The feces and intestinal contents of the C^{14}-phytosterol group had an amount of C^{14}-free sterol that was approximately 3 times greater than the group given cholesterol-4-C^{14}.

Esterified C^{14}-sterol was recovered from the tissues of animals injected with both types of C^{14}-sterols. The adrenal (with both injected C^{14}-sterols) had the highest percentage of esterified C^{14}-sterol of the tissues examined (73.3 and 75.0%). The total serum C^{14}-sterol fraction of both types of animals also contained a high percentage of esterified C^{14}-sterol, but the C^{14}-phytosterol group had significantly less (52.3%) than the cholesterol-4-C^{14} group (66.1%). The liver and small intestine of both types of animals had smaller amounts of esterified C^{14}-sterols than serum: also, less of the injected C^{14}-phytosterols was present in the esterified form in the liver and small intestine than when cholesterol-4-C^{14} was injected. Feces and intestinal contents of the group injected with cholesterol-4-C^{14} contained only a trace of esterified C^{14}-sterol; on the other hand, feces and intestinal contents of the C^{14}-phytosterol group contained 1.16% of the injected C^{14}-sterol as esterified C^{14}-sterol.

Both cholesterol-4-C^{14} and C^{14}-phytosterols were converted to bile acids (Table II): conversion of cholesterol-4-C^{14} to bile acid was twice that for C^{14}-phytosterols (14% vs 7%). The specific activities of the bile and liver free sterol fractions of the group given cholesterol-4-C^{14} were the same. In the C^{14}-phytosterol group a much larger proportion of the injected C^{14}-phytosterols was recovered as C^{14}-sterol in the bile than in the cholesterol-4-C^{14} group. Approximately 44% of the total C^{14}-activity recovered in the bile of the C^{14}-phytosterol group was present as bile sterol and only 9.2% in the group injected with cholesterol-4-C^{14}. The specific activity of the bile sterol of the C^{14}-phytosterol group was considerably higher than the liver free sterol fraction.

The free and esterified tissue sterols isolated from rats injected with C^{14}-phytosterols were subjected to 5 passages through the dibromide and crystallization of the dibromides from 5:1 ether-acetic acid. The specific activity of the tissue sterols of the C^{14}-phytosterol group declined to only about 1% of the original value, whereas, in the cholesterol-4-C^{14} group there was practically no change in the specific activity of the tissue sterols. These results would appear to indicate that very little, if any, conversion of C^{14}-phytosterols to cholesterol occurs.

Discussion. In many respects, C^{14}-phytosterols appear to follow the same metabolic pathways as cholesterol. They are selectively taken up by the liver, esterified by the liver, intestine and other tissues, and are converted to bile acids. However, there were several notable differences with respect to metabolism of the injected C^{14}-phytosterols. The degree of esterification of the injected phytosterols appeared to be less than that for cholesterol. It would also appear that while the C^{14}-phytosterols are removed from the serum by the liver as with cholesterol, they do not readily mix with the liver cholesterol pool and are selectively excreted in the bile. This is borne out by the specific activities of the bile and liver sterols of animals injected with C^{14}-phytosterols and also on the much greater proportion of injected C^{14}-phytosterols recovered as bile sterol. The data on the feces and intestinal contents also indicate that phytosterols are more rapidly excreted than cholesterol; the bile would appear to be the principal route of their excretion. The findings

TABLE II. Distribution of C^{14}-Activity in Bile Following Injection of C^{14}-Phytosterols and Cholesterol-4-C^{14}.

	Bile Sterol		Liver Free Sterol	Bile Acid
Group*	CPM/mg	% dose recovered	CPM/mg	% dose recovered
Cholesterol-4-C^{14}	2355 ±505	1.42 ±0.30	3163 ±450	14.06 ±3.05
C^{14}-Phytosterols	2269 ±647	5.46 ±1.56	615 ±150	6.98 ±2.10

* Values represent average of 5 bile-fistula rats per group; each rat was injected with an emulsion containing either C^{14}-phytosterols (.34 mg; 61,000 CPM) or cholesterol-4-C^{14} (.34 mg; 243,000 CPM).

with respect to conversion of C^{14}-phytosterol to bile acids confirm and extend the observations by Werbin *et al.*(5) and Hanahan and Wakil(2) with H^3-β-sitosterol and C^{14}-ergosterol. Preliminary experiments were carried out in an attempt to identify the bile acids formed. They appeared to be different from those derived from cholesterol.

Summary. Data are presented on the comparative tissue distribution of C^{14}-free and esterified sterols following injection of cholesterol-4-C^{14} and C^{14}-phytosterols to normal and bile-fistula rats. At the end of 24 hours a large proportion of the injected C^{14}-phytosterols and cholesterol-4-C^{14} was recovered in the liver. All tissues examined (serum, liver, small intestine and adrenal) showed the presence of esterified C^{14}-phytosterols, but less was esterified than with cholesterol-4-C^{14}. A much larger proportion of the injected C^{14}-phytosterols was recovered in the feces and intestinal contents than in the cholesterol-4-C^{14} group. Conversion of the injected C^{14}-phytosterols to bile acid was observed; a larger proportion of the injected C^{14}-activity was recovered in the bile as C^{14}-sterol than with injected cholesterol-4-C^{14}. No conversion of C^{14}-phytosterols to cholesterol or related sterols could be demonstrated.

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Destruction of Chimpanzee Kidney Cells by Sera From Patients with Acute Infectious Hepatitis.*† (27077)

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Recent epidemiological evidence(1) has suggested that chimpanzees may act as transient carriers of human hepatitis virus. Since then, recurrent outbreaks of the disease among groups of chimpanzee handlers have further supported this hypothesis(2-4). The outbreaks were sufficiently well documented to lend support to a supposition that the virus may have been propagated in some unknown cellular system within the animals.

Accordingly, chimpanzee liver, omentum, spleen, bone marrow, testis, and kidney were removed from healthy adolescent animals, and standard tissue culture techniques were employed to obtain cellular outgrowths. The various cellular systems were exposed to acute sera from 3 to 12 cases of typical human infectious hepatitis, suspected of containing active virus. Some of the sera were from chimpanzee handlers at Holloman Air Force Base, New Mex.(1), whereas others were derived from outbreaks remote from Holloman and not associated with any known ape contacts.

Initial experiments indicated that some of the sera were capable of destroying outgrowths of chimpanzee macrophages from omental tis-

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† Since this paper was submitted for publication, it has not been possible to maintain the virus past the 4th chimpanzee kidney cell passage.