Results. The data in the Table show that ethanol had no obvious effect on the basal release of glycerol from isolated fat cells. However, when lipolysis was provoked by rat serum, there was a linear correlation between the degree of inhibition of glycerol release and the concentration of ethanol in the incubation medium. The calculated regression line in the Figure clearly shows this.

Discussion. The lack of pharmacologic effect of ethanol (10^{-7} M–1.0 M) on the basal lipolysis of free fat cells obtained from fasted rats (Table) is in keeping with the observations of BIZZI and CARLSON[5]. The inhibition of hormone (i.e. serum) stimulated lipolysis, by ethanol, in isolated fat cells obtained from fasted rats over the concentration range of alcohol of 10^{-7} M–1.0 M, contrasts with the promotion of lipolysis, observed in fed rats, by ethanol at appromimately 5×10^{-2} M–5×10^{-1} M. However, it is known that adipose cells from fasted rats are more sensitive to lipolytic hormones than are cells from fed rats. This dual effect of ethanol, dependent upon the nutritional status of the fat cell, could be explained in the following terms: in lipocytes from fed rats there is present a quantity of dietry lipid from the circulation not as yet incorporated into the storage pool of neutral fat. If this is subsequently esterified with glyceride-glycerol formed from ethanol rather than glucose/alpha glycerophosphate (the alcohol would enter the fat cell at a faster rate than glucose owing to higher lipid solubility and smaller molecular mass) then glycerol formed via the α-glycerphosphate pathway is surplus to requirements and effuses from the fat cells. In the fasting state, lipogenesis is minimal, ethanol still penetrates the fat cells and becomes partitioned between the water soluble compartment and the lipid soluble compartment of the fat cell thus reducing available interface where the lipase(s) act(s), in effect blocking the enzyme from the substrate so inhibiting lipolysis[16].

Résumé. Il y a une connexion linéaire entre la concentration progressive de l'éthanol et l'inhibition de la lipolyse stimulée par le sérum dans les cellules libres du tissu adipeux de rats affamés. L'éthanol n'a pas affecté la décharge basale du glycerol.

P. B. CURTIS-PRIOR

Department of Medicine, Guy's Hospital, London S.E.1 (England), 29 May 1972.

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Comparison of Sterol Composition and Transformation in Chicken and Pigeon Intestines

During our studies using the spontaneously atherosclerosis-susceptible White Carneau pigeon, we noted that coprostanol and other 5β-stanols were absent from the feces of this species while chicken feces contained a significant amount of coprostanol[1,2]. This stimulated us to compare the nature of the sterols and their transformation products in the intestine and intestinal contents of these 2 species. Specifically, 2 aspects of sterol transformation in the intestine were examined: 1. formation of coprostanol and 2. the nature of plant sterols in the intestinal tissue and their relation to the plant sterols in the diet. The uptake of plant sterols by the avian intestines has previously been demonstrated[3,4].

Materials and methods. Adult White Carneau pigeons were obtained from the Palmetto Pigeon Plant (Sumter, S.C.) and were fed a mixed grain diet (Purina Pigeon Chow, Ralston Purina Co., St. Louis, Mo.). Roosters and hens obtained locally were fed chicken feed (Laudon Bros. Feed and Seed Co., Dover, Minn.). Cholesterol-4-^{14}C (specific activity, 60.9 mCi/mole) and reference sterols were obtained commercially. Intestinal tissue segments and their contents were obtained after the birds were killed. The intestinal tissues were rinsed with saline and 1 mM sodium taurodeoxycholate solution and weighed.

Sterols and steryl esters were extracted from the tissue samples asaportion of the total lipid extract, as described by FOLCH et al.[5] Free sterols and steryl esters were then separated from each other and other lipids by thin layer chromatography (TLC) on silica gel G with the solvent system, heptane-isopropyl ether-acetic acid (65:40:4, v/v/v)[6]. The sterols of the intestinal contents were extracted and purified as described previously for the fecal steroids[1,2].

After TLC, the trimethyl silyl ether derivatives of the sterols were identified by gas-liquid chromatography (GLC) in an F & M 402 high-efficiency gas chromatograph equipped with a flame ionization detector[7]. The sterols were chromatographed on glass columns packed with 3.8% W-98 on Diatoport (80 to 120 mesh)[1,2]. The sterols were quantitated by gas chromatography with 5α-cholestane as an internal standard[1,2,7].

Results and discussion. Sterols of the intestinal contents represent a mixture of sterols derived from the bile, intestinal wall, and diet and their bacterial products. On TLC, chicken intestinal contents contained a band corresponding to coprostanol, but no such band was found in pigeon intestinal contents. There was no band corresponding to coprostanone in either species. The presence of coprostanol derivatives in the chicken but not in the pigeon is in agreement with our previous studies on the fecal sterols of these 2 species[1,2].

On GLC, the band corresponding to coprostanol gave peaks corresponding to coprostanol and the 5β-stanols of

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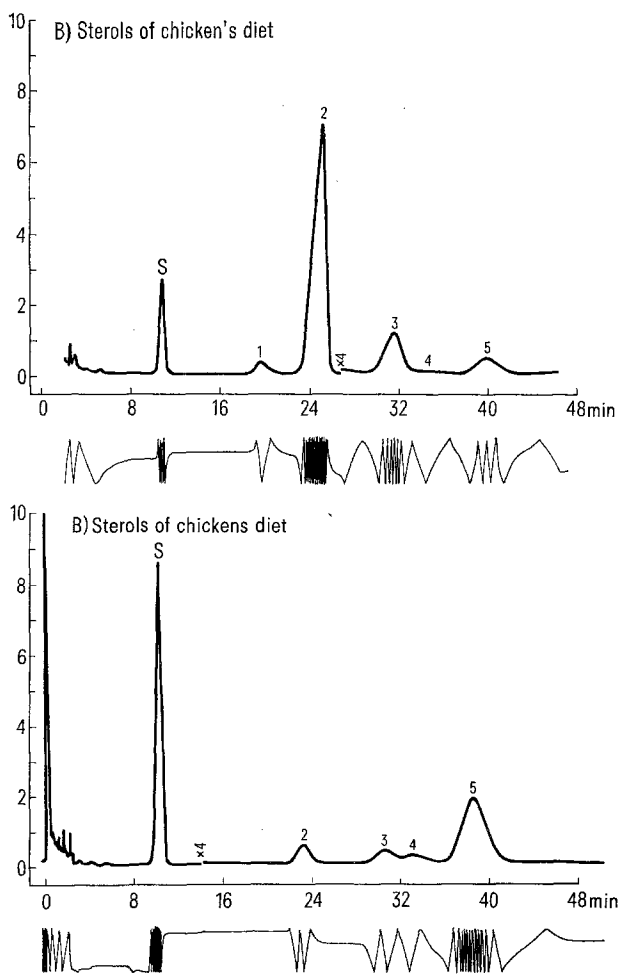
Sterol composition[a] and coprostanol-forming capacity of intestinal contents

	Chicken	Pigeon
Δ^5 Sterols (%)		
Cholesterol	56.4	39.7
Campesterol + Stigmasterol	14.4	17.3
β-Sitosterol	29.2	43.0
5 β-Stanols (%)[b]		
Coprostanol	18.7	Nil
Coprocampestanol + Coprostigmastenol	18.7	Nil
Coprostigmastenol	62.6	Nil
Coprostanol-forming capacity[c]	+	—

[a] Total sterol concentration: chicken, 2.21 mg/g; pigeon, 0.58 mg/g. [b] 5 β-Stanols = 7.6% of total sterols. [c] Determined by incubating cholesterol-4-^{14}C with fecal cultures.



Gas-liquid chromatography of sterols. Sterols were chromatographed as trimethyl silyl ethers; conditions are described in text. A) In small intestine of chicken. B) Sterols of chicken's diet. Peak identification: 1. coprostanol; 2. cholesterol; 3. campesterol; 4. stigmasterol; and 5. β-sitosterol. Peak 'S' represents internal standard (5α-cholestane) in both chromatograms.

the plant sterols. The latter were identified as coprocampestanol, coprostigmasterol, and coprostigmastanol. The area of the gel corresponding to coprostanone, when eluted and analyzed by GLC, gave peaks corresponding to coprostanone, campestanone, stigmastenone, and β-sitosterone. Eluates from the area of the gel corresponding to coprostanol and coprostanone from the pigeon intestinal contents gave no peaks on GLC. Various sterol fractions from chicken and pigeon intestinal contents were quantitated using 5α-cholestane as the internal standard (Table).

Incubation of fecal cultures of these 2 species with cholesterol-4-^{14}C by methods previously used[8,9] indicated that pigeons are unable to form labeled coprostanol. The chickens, however, showed a conversion of 2.13% under similar conditions. This difference in the coprostanol formation between chicken and pigeon may be due to a difference in intestinal bacterial flora[10].

The sterols of small intestinal tissue of the chicken were: cholesterol, 86.6%; coprostanol, 5.9%; and plant sterols 7.5%. Only a small fraction of the sterols were present as esters (3.2%). The major plant sterols present in the intestinal tissue were campesterol, a trace of stigmasterol, and β-sitosterol (Figure 1,A). The proportion of campesterol in the small intestinal tissue was higher compared to its proportion in the diet (Figure 1,B); in the small intestine it accounted for 68.5% of the plant sterols (campesterol/sitosterol [C/S] ratio, 2.16) while the diet contained only 6.8% (C/S ratio, 0.06). This indicates a preferential uptake of campesterol in the chicken intestine. Similar high C/S ratios were found in the large intestine (2.41) and cecum (2.05). C/S ratios in the liver and plasma of the chicken were also high (> 2.0), indicating that the uptake and successive transfer of campesterol to the systemic circulation are higher than those of β-sitosterol. This is consistent with the preferential uptake of campesterol in the pigeon intestine[3].

In the pigeon small intestine, cholesterol contributed 97.9% of the total sterols, the remainder (2.1%) being plant sterols. The C/S ratio was 2.34 in contrast to 0.23 in the diet. Plasma and liver of these pigeons also showed high C/S ratios (> 4.0), indicating the preferential entry of campesterol into the systemic circulation.

These studies indicate a striking difference in the intestinal metabolism of sterols, particularly the formation of coprostanol, by the intestinal bacteria in these 2 species. In addition the studies in the chicken confirm our earlier observation in the pigeon[3] that campesterol is being taken up to a greater extent than β-sitosterol[11].

Zusammenfassung. Es bilden sich 5β-Stanol-Derivate von Sterolen im Verdauungstrakt des Huhns, nicht aber bei der Taube. Die intestinalen Gewebe beider Vögel bevorzugen bei ihrer Resorption Campesterol vor β-Sitosterol.

M. T. RAVI SUBBIAH and B. A. KOTTKE

Mayo Clinic and Mayo Foundation, Rochester (Minnesota, USA), 18 May 1972.

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