tonin (7.1 μCi, per mg.), 772.0 mg. of non labeled digitonin and 500 ml. of 50% ethanol, was added to each tube, the volume was adjusted to 0.8 ml., the solution was mixed and the cholesterol digitonide was allowed to crystallize leaving over night at room temperature.

Two-tenth ml aliquot of the supernatant was taken into a counting vial and 15 ml. of scintillator solution was added. The scintillator consists of 750 ml. of toluene, 250 ml. of methyl alcohol, 3 gr. of PPO and 100 mg. of POPOP. The treated samples were counted with a Tri-Carb liquid scintillation spectrometer with an efficiency of 15 to 20%.

A standard curve was drawn by plotting amount of cholesterol versus counts per minute in the supernatant aliquot by employing standard cholesterol samples. The values of cholesterol of the samples were read on the standard curve. The standard curve show linear relationship with a straight line with a bend over the cholesterol level of 520 mg. per dl.

Sixteen serum samples were measured and compared with the Leffler method. The difference was almost under 10%. The reproducibility was examined for 5 serum samples and good results were obtained within a range of standard deviation of 1 to 4% except one with 8.0%. Ester ratio was also measured for 12 serum samples and reasonable results were obtained.

The present method is considered a good device for measuring serum cholesterol with high reproducibility and stability and can be employed in clinical studies which require precise data of serum cholesterol level.

A Clinical Study with Tritiated Digoxin

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The extreme variation in both effective and toxic dosage of digitalis from one patient to another yields difficult problems in the daily practice of clinical medicine. The recent availability of radioactive digitalis has made it possible to investigate the metabolism of the compound more in details. We investigated the serum levels and excretion of digoxin after intravenous administration of tritium-labeled compound in the human subjects with renal insufficiency and diabetes mellitus.

Tritium-labeled digoxin was prepared by the Wilzbach hydrogen-exchange method and purified by column partition chromatography. Assays revealed specific radioactivities of 112 μCi per mg.

Subjects studied are as follows: Three patients hospitalized with renal failure, five patients with diabetes mellitus and three control subjects, who had neither metabolic nor renal diseases.

The dose of tritiated digoxin injected was 0.23 mg., diluted in normal saline solution. The solution was administered intravenously. Specimens of venous blood were obtained from the opposite antecubital vein at 10 and 30 minutes; at 1, 2, 4, 6, 12 and 24 hours after injection. Urine was collected every 24 hours for eight to fourteen days. Stools were collected daily for seven to fourteen days.

Blood and urine specimens were treated with PE-611 scintillator, consisting dioxane, anisole, dimethoxyethane, PPO and POPOP. Stools were treated with Schoeniger combustion method. Counting was performed with a Packard Tri-Carb liquid scintillation spectrometer.

The time course of blood level of radioactivity was varying in each group. When compared control group with the other two groups significant decrease in clearance with higher blood level of radioactivity in 24 hours after injection was observed in the latter two groups. Among these the renal failure groups revealed highest level.

The excretion of radioactivity was further varying in each group. In ten days, urinary excretion of radioactivity was 65% in control group, 16% in the group of renal failure and 59% in the group of diabetes mellitus. Fecal excretion in ten days was 14% in control, 39% in renal failure and 15% in diabetes mellitus...
mellitus.

One patient with renal failure was subjected to blood dialysis and another one was subjected to peritoneal lavage. Turnover study was carried out prior to and during these operations. By these operations only 3% of radioactivity was removed in dialysed fluid and 6% was contained in peritoneal fluid.

The blood level of two and six hours after injection were compared especially. These levels of diabetic group lie between those of renal failure and those of control groups.

The delayed decrease of the levels of this group was examined. It had no correlation to the level of fasting blood sugar, the amount of urinary sugar and the use of drugs just before this study. In contrast to this the blood level, particularly two hours after injection, closely correlated to the results of PSP test and GFR (r=0.79 for PSP 15 min.; r=0.74 for PSP 2 hours; and r=0.82 for GFR). The blood level of digoxin in diabetes mellitus directly reflects the degree of impaired renal functions.

The Effect of Vitamin E on the Pregnants Rats
(Especially Using 14C-α-Tocopherol)

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(Purpose) Following administration of 14C-2-tocopherol (abbreviated, 14C-V.E.) to pregnant rats, incorporation of 14C-V.E. into placentas and fetuses were observed and were studied to solve the effect of V.E treated with progesterone (abbreviated, P) and to clarify the significance of V.E in abortion cases.

(Experimental materials and methods)

Pregnant rats of Wistar strain were divided into following seven groups. Steroids and V.E were administered from the 11th day to the 19th day of gestation. Group A: Control, without treatment. Group B: P (1,0 mg) administered. Group C: P (1 mg) and E (20 mg) administered. Group D: Testosterone propionate (0.5 mg, abbreviated, T.P) and P (5 mg) administered. Group F: P (5.0 mg) administered, after castration on the 11th day of gestation. Group G: P (5 mg) and E (20 mg) administered, after castration.

Following administration of 14C-V.E the rats of seven groups were sacrificed after 24 hours, visceras were all excised and weighed. Particularly the macroscopical findings of placentas and fetuses were observed. By Emmerie-Engel's procedure V.E was extracted and 14C-V.E contents were counted by liquid scintillation counter.

<table>
<thead>
<tr>
<th>(Results)</th>
<th>Weights of Placentas (mg)</th>
<th>Weights of fetuses (mg)</th>
<th>Incorporation of C-V.E into placenta C.P.M/mg</th>
<th>A survival rate of fetuses</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A</td>
<td>280±7</td>
<td>2690±49</td>
<td>4010±62</td>
<td>100%</td>
</tr>
<tr>
<td>Group B</td>
<td>304±5</td>
<td>3050±42</td>
<td>4560±45</td>
<td>100%</td>
</tr>
<tr>
<td>Group C</td>
<td>346±5</td>
<td>3330±38</td>
<td>4240±47</td>
<td>100%</td>
</tr>
<tr>
<td>Group D</td>
<td>315±9</td>
<td>2630±41</td>
<td>3840±57</td>
<td>55%</td>
</tr>
<tr>
<td>Group E</td>
<td>320±7</td>
<td>2760±35</td>
<td>3930±62</td>
<td>69%</td>
</tr>
<tr>
<td>Group F</td>
<td>288±9</td>
<td>2530±86</td>
<td>3260±66</td>
<td>24%</td>
</tr>
<tr>
<td>Group G</td>
<td>302±3</td>
<td>2770±38</td>
<td>3840±41</td>
<td>58%</td>
</tr>
</tbody>
</table>

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