at 250nm and/or bound cadmium. Thus the presence of cadmium-metallothionein in the liver cytoplasm of these animals seems unlikely.

When PM 10-concentrated cytosol fractions were dialysed against 10mM-sodium phosphate buffer, pH 7.4, none of the cadmium passed through the dialysis membrane. By contrast, after attempts to fractionate the material with (NH₄)₂SO₄, dialysis of the redissolved pellets resulted in the loss of much of the cadmium through the dialysis membrane. Similarly, adjustment of the PM 10-concentrated fraction to a final concentration of 10% (v/v) trichloroacetic acid also caused the cadmium to become freely soluble in the aqueous supernatant. After removal of the trichloroacetic acid-insoluble material, these supernatant fractions were neutralized. When they were subjected to ultrafiltration with a PM 10 membrane, none of the cadmium was retained by the membrane. These results are consistent with the formation of a complex between cadmium and one or more components of high molecular weight, distinct in character from the lower-molecular-weight protein, metallothionein.

A similar distribution of cadmium was observed when gill tissue from the same animals was fractionated in an analogous manner to that described above for liver tissue.

The concentration of cadmium administered to the fish in these experiments was considerably lower than those used in other investigations in which a metallothionein response has been observed (Overnell et al., 1977; Marafante, 1975).


### Effect of 5-Fluorouracil on Liver and Plasma Vitamin A in Female Rats

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Several vitamins play an important role in hepatic oxidative metabolism. Vitamin A deficiency in rats significantly decreases microsomal protein and oxidative enzymes. Vitamin A decreases, and deficiency enhances, the carcinogenic properties of certain chemicals. Large doses of vitamin A are toxic and tend to destabilize membranes.

5-Fluorouracil is used either alone, or in combination with other drugs, in the treatment of metastatic cancer, but as with all cytotoxic drugs the margin between therapeutic and toxic doses is small.

In the present study female Wistar albino rats (200g body wt.) were given an intraperitoneal injection of 5-fluorouracil on 3 consecutive days. Blood and liver samples were taken on day 4.

Plasma vitamin A was measured by the method of Hansen & Warwick (1969) with the fluorescence technique of Van Steveninck & De Goeij (1973); liver vitamin A was measured by the method of Thompson et al. (1971).

Table 1 shows that the human therapeutic dose of 15mg/kg body wt. decreased the haematocrit and caused a slight decrease in liver and plasma vitamin A concentrations.

At the higher doses both liver weight and terminal body weight were greatly decreased. The animals had diarrhoea, and the haematocrit increased with the loss of body fluid.

Both liver and plasma vitamin A concentrations were elevated, but in each case the increase was greater than could be accounted for by the decrease in liver weight or loss of body fluid. The amount of vitamin A per whole liver was decreased significantly in 15mg/kg-dose group and significantly increased in the other groups.
Table 1. Effects of 5-fluorouracil (5fUra) on vitamin A concentration in liver and plasma of female rats

Values shown are means ± S.E.M. Significant differences from control values determined by t-test are shown: †P<0.1; ‡P<0.05; *P<0.02; **P<0.01; ***P<0.001.

<table>
<thead>
<tr>
<th>5-flUra dose (mg/kg)</th>
<th>Body weight change (g)</th>
<th>Liver weight (g)</th>
<th>Liver [vitamin A] (µg/g)</th>
<th>Haematocrit (%)</th>
<th>Liver Plasma [vitamin A] (µg/100ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>+3.0 ± 1.1</td>
<td>8.94 ± 0.23</td>
<td>161.3 ± 11.8</td>
<td>38.5 ± 0.7</td>
<td>25.6 ± 1.8</td>
</tr>
<tr>
<td>15</td>
<td>-0.5 ± 0.5††</td>
<td>8.83 ± 0.23</td>
<td>154.0 ± 2.4</td>
<td>36.0 ± 0.0*</td>
<td>20.1 ± 2.1</td>
</tr>
<tr>
<td>30</td>
<td>-9.0 ± 0.0***</td>
<td>8.02 ± 0.40†</td>
<td>224.2 ± 16.1**</td>
<td>41.6 ± 1.2†</td>
<td>38.5 ± 3.3**</td>
</tr>
<tr>
<td>45</td>
<td>-20.0 ± 2.0***</td>
<td>7.53 ± 0.34**</td>
<td>220.5 ± 20.8††</td>
<td>43.7 ± 1.7***</td>
<td>35.8 ± 1.3***</td>
</tr>
<tr>
<td>90</td>
<td>-28.8 ± 2.4***</td>
<td>6.75 ± 0.18***</td>
<td>212.7 ± 3.3***</td>
<td>52.0 ± 0.8***</td>
<td>—</td>
</tr>
</tbody>
</table>

We conclude that 5-fluorouracil affects vitamin A concentrations in plasma and liver by some action that is not directly related to its cytotoxic action.

Evidence is accumulating that the nutritional status of the patient may be important in the control and treatment of cancer. A cytotoxic drug such as 5-fluorouracil may upset this balance in a way that diminishes the effectiveness of treatment. Thus it may prove necessary to monitor vitamin concentrations and dietary status during cancer treatment in order to obtain the best possible response.


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Phenol is largely excreted as glucuronide and sulphate conjugates in mammalian species (Capel et al., 1972; Mehta et al., 1978). Few studies on glucuronidation in vitro use phenol as an acceptor substrate, as no simple assay procedure has been available. The procedure presented here is an adaptation of that described by Lucier et al. (1971) for measuring the glucuronidation of 1-naphthol.

The standard incubation medium consisted of: 0.2mM-[14C]phenol (diluted with unlabelled phenol to approx. 0.9mCi/mmoll before addition), 2.7mM-UDP-glucuronic acid, 66mM-Tris/HCl buffer, pH7.4, 10mM-Mg2+ and up to 0.05ml of microsomal fraction; total volume 0.3ml. Microsomal fraction was prepared from a 30% (w/v) rat liver homogenate in 0.25M-sucrose/0.05M-Tris/HCl (pH7.4) by differential centrifugation (Illing et al., 1977). The reaction was stopped after 5 or 10min by addition of 10ml of 1.5% (w/v) 5-(biphenyl-4-yl)-2-(4-t-butylphenyl)-1-oxa-3,4-diazole in toluene. The mixture was shaken and transferred to a scintillation vial for radioactivity counting in a liquid-scintillation counter. An external-standard channels-ratio technique (Johnson et al., 1972) was used for quench correction.