A Note on the Fate of Stibophen in the Body

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By means of a simple and rapid polarographic technique [Goodwin & Page, 1943] we have been able to determine the rate of excretion of Sb after the injection of stibophen (Fouadin; sodium antimonyIII bis-pyrocatechol-3,5-disulphonate) with a higher degree of accuracy than has previously been possible. It was considered that a critical examination of the relative rates of excretion of the Sb and the Na catechol disulphonate portions of the stibophen molecule would throw some light upon the fate of the drug in the body.

Khalil [1936 a, b] made use of the colour reaction given by catechol with iron salts to detect the presence of stibophen in the urine of patients receiving intramuscular injections of the drug. The urine was treated with a solution of FeCl₃ or ferrous ammonium sulphate and made strongly alkaline with NH₄OH. The depth of colour produced corresponded approximately with 'the amount of Sb in the urine as revealed by the Reinsch test'. However, the excretion of Sb continued after the colour test on the urine had become negative. Hassan [1937, 1938], using the Beam & Freak [1919] method for the determination of Sb, found that not more than 20 % of the Sb administered as stibophen was excreted in 24 hr. by schistosomiasis patients. In the present work, the rates of urinary excretion of the Sb and catechol fractions of the stibophen molecule were measured in normal human subjects by accurate methods of assay.

EXPERIMENTAL

Determination of Sb. Sb was determined in the urine by the direct polarographic method previously described [Goodwin & Page, 1943].

Determination of catechol. For the accurate estimation of the catechol portion of the molecule, Khalil’s test had to be modified, as the quality and intensity of the colour was found to be affected by normal urine constituents. When the ferrous tartrate reagent described by Moehll [1928] was used, a clear colour unaffected by normal urine constituents was produced, and this reagent was therefore chosen in preference to FeCl₃ or ferrous ammonium sulphate. The catechol compound in a concentration of 1:10,000 in urine produced a measurable depth of colour, and the colour produced by Na catechol disulphonate was identical qualitatively and quantitatively with that produced by an equivalent amount of stibophen.

General procedure. Young normal male volunteers received intramuscular or intravenous injections of stibophen (6-3 % solution) or Na catechol disulphonate (4-66 % solution). Urine was collected hourly for the first 6 hr. after injection, and subsequently in 6-12, 12-24, and 24-48 hr. fractions.

To 5 ml of urine in a test tube were added 5 ml of a tenfold dilution of concentrated NH₄ solution (sp. gr. 0-880), and the mixture was heated for a few minutes in a boiling water-bath to coagulate any precipitate formed. After centrifuging, 2 ml of the supernatant fluid were mixed with 2 ml of a freshly prepared solution containing 0-1 %, FeSO₄ and 0-5 %, Rochelle salt, diluted to 10 ml, and the colour intensity estimated with a Hilger ‘Spekker’ absorptiometer. A blank in which the reagent was replaced

Table 1. The urinary excretion of Sb and catechol by normal human subjects after the injection of stibophen

<table>
<thead>
<tr>
<th>Injection</th>
<th>% dose excreted (hr.)</th>
<th>Total excretion in 6 hr. %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sb (5 ml. of a 6-3 % solution)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0-1</td>
<td>2-3</td>
<td>3-4</td>
</tr>
<tr>
<td>Sb: %</td>
<td>6:0</td>
<td>1:9</td>
</tr>
<tr>
<td>Range</td>
<td>4:1-9:9</td>
<td>0:9-3:7</td>
</tr>
<tr>
<td>Catechol: %</td>
<td>3:3</td>
<td>2:3</td>
</tr>
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by H₂O was used to compensate for the colour of the original urine. The concentration of catechol was calculated by reference to a standard curve prepared with urinary solutions of stibophen of known concentrations.

![Graph](image)

**Fig. 1.** The excretion of stibophen by subject M.C.
- • antimony, 
- ○ catechol, both from a 5 ml. dose of stibophen (6.3%).
- ▲ catechol, from a dose of sodium catechol disulphonate equivalent to 5 ml. of stibophen (6.3%).

**RESULTS**
The results (Table 1, Fig. 1) show that in the normal subject catechol excretion was almost complete in about 6 hr., whereas the Sb excretion was very much slower, and followed a different course. The rates of excretion were not affected by changes in urine volume. In Khalil's experiments, his colour test failed after 6 hr. because the catechol portion of the molecule had been completely eliminated. The function of the catechol appears to be to keep the circulating Sb in solution in a non-toxic form while it is being absorbed by the liver or excreted by the kidney. Meanwhile, excretion of the catechol takes place independently of that of the Sb and (as shown by the results) at the same rate as that of an equivalent dose of Na catechol disulphonate.

**SUMMARY**

1. Khalil's test for stibophen in urine has been modified for use as a quantitative method for the determination of the catechol fraction of the molecule.
2. The catechol fraction has been shown to be excreted very much more rapidly than the Sb of stibophen, elimination being complete in six hours in the normal subject.

Our thanks are due to the volunteers, especially Mr E. Rogers, who was also responsible for the catechol determination.

**REFERENCES**


The Action of Amino-acids and Proteins on Liver-fat Deposition

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Any explanation of the lipotropic action of proteins must take into account the now well-established facts that while cystine accelerates, methionine partially prevents the deposition of fat in the livers of rats maintained on diets high in fat and low in lipotropic factors. Agreement on the question as to whether the effects of these two amino-acids alone account for the lipotropic behaviour of proteins has, however, yet to be reached. Thus Best & Ridout [1940] and Channon, Manifold & Platt [1940] conclude that these two amino-acids are not the only factors involved; in contrast, Tucker, Treadwell & Eckstein [1940] claim that the cystine and methionine contents of the diets used by them suffice adequately to explain their results. More recently, Treadwell, Groothuis & Eckstein [1942] arrived at similar conclusions but also found that when methionine was present as the free amino-acid in the diet it appeared to be superior to that contained in protein (caseinogen) so far as lipotropic action is