Dopamine β-Hydroxylase Activity in Five Cases of Phaeochromocytoma

C. BOHUON, F. GUERINOT, PH. TCHERDAKOFF and M. BONNAY

Institut Gustave-Roussy, 94 Villejuif, Val-de-Marne, France

Weinschilboum et al. (1971) have shown that dopamine (3,4-dihydroxyphenethylamine) β-hydroxylase (EC 1.14.2.1) activity in blood correlates well with the activity in the peripheral sympathetic nervous system and that the adrenal medulla plays no significant part in maintaining the enzymic activity in the blood. We have now measured the dopamine β-hydroxylase activity (by a new and sensitive method) in the plasma and tumour of five patients bearing phaeochromocytoma, as this tumour might be considered as an accessory adrenal medulla.

All the tumours had been confirmed by biological, clinical and anatomopathological observations. They were carried in ice from the operating room and frozen at -20°C within 1h. For assay of dopamine β-hydroxylase activity they were thawed and homogenized in 200 vol. of tris buffer containing Triton X-100.

Heparinized blood samples were centrifuged and the plasma was stored at -20°C. The dopamine β-hydroxylase activity was stable for months under these conditions.

For the measurement of dopamine β-hydroxylase activity we used a new sensitive radioassay method based on a double enzymic reaction (Bonnay et al., 1970). In the first step dopamine β-hydroxylase hydroxylates the substrate (phenethylamine for tissue and tyramine for blood; Weinschilboum & Axelrod, 1971; Molinoff et al., 1971), and then the reaction products are submitted to the action of the phenylethanolamine N-methyltransferase in the presence of S-adenosyl[14C]methionine. The labelled N-methylated product is extracted and determined by measurement of its radioactivity by liquid-scintillation counting.

Table 1 shows that a significant decrease in blood dopamine β-hydroxylase activity appears 1 week after surgery. The possible exception (TYP) might be due to the unusual character of this malignant phaeochromocytoma (homovanillate-secretant and hepatic metastasis). However, there was no relation between tumour and plasma dopamine β-hydroxylase activities, nor between dopamine β-hydroxylase activities and catecholamine concentrations.

It is now well established that the adrenal medulla does not participate in maintaining dopamine β-hydroxylase activity in the blood. However, after the ablation of a phaeochromocytoma, which might be considered as an accessory adrenal medulla, the plasma enzyme activity falls significantly (by about 50%).

Several hypotheses may be advanced to solve this apparent contradiction. First, a phaeochromocytoma may have the specific character of releasing the enzyme into the
Table 1. Dopamine $\beta$-hydroxylase activities in plasma and in the tumour in several cases of phaeochromocytoma

<table>
<thead>
<tr>
<th>Patient</th>
<th>Wt. of tumour (g)</th>
<th>In tumour (umol/h per g)</th>
<th>Before surgery</th>
<th>1 week after surgery</th>
</tr>
</thead>
<tbody>
<tr>
<td>JOU</td>
<td>450</td>
<td>5.4</td>
<td>663</td>
<td>375</td>
</tr>
<tr>
<td>GOU</td>
<td>181</td>
<td>0.9</td>
<td>432</td>
<td>180</td>
</tr>
<tr>
<td>FER</td>
<td>?</td>
<td>18.3</td>
<td>393</td>
<td>222</td>
</tr>
<tr>
<td>LEF</td>
<td>55</td>
<td>2.4</td>
<td>327</td>
<td>174</td>
</tr>
<tr>
<td>TYP</td>
<td>Hepatic metastasis</td>
<td>5.4</td>
<td>315</td>
<td>264</td>
</tr>
</tbody>
</table>

general blood stream. The hypothesis of induced synthesis of dopamine $\beta$-hydroxylase by a high concentration of blood amines cannot be discarded a priori, because it is clear that a fall in the concentrations of blood catecholamines and a decrease in dopamine $\beta$-hydroxylase activity are concomitant, even if the mathematical relationship between the two events is not simple. Further, an endogenous inhibitor seems to be present in the tumour itself: when we mixed homogenates of the tumours from patients GOU and FER the dopamine $\beta$-hydroxylase activity was lower than the sum of the two separate values. Attempts are now being made to solve this problem by testing the effect of different concentrations of Cu$^{2+}$.

We conclude that the phenomenon is definitely proved to exist, but its cause remains unexplained.


Epithelial Mucins of the Atlantic Salmon (Salmo salar L.)

JACK HARRIS and STEPHEN HUNT

Department of Biological Sciences, University of Lancaster, Bailrigg, Lancaster, U.K.

Suggestions as to the functional significance of epithelial mucins of fish have included their possible role in defence against pathogenic material, in osmoregulation, in protection against mechanical damage and in enhancing their swimming abilities (Jakowska, 1963; Rosen & Cornford, 1971). Histochemical and biochemical analyses of fish mucus have demonstrated that the components contain both protein and carbohydrate (Lehtonen et al., 1966; Leppi, 1968). A preliminary analysis of the mucous components of salmon has been undertaken as part of an examination of the biology of the epidermis of fish.

Chromatography of dialysed whole mucus on Sephadex G-75 revealed the presence of two major components (Fig. 1). Further separation of the initial peak from G-75 was effected on Sephadex G-200 (Fig. 2) to give two single components, A and B, whose homogeneities were established by polyacrylamide-gel and immuno-electrophoresis. The