constantly preceded by a sequence of deflections of low amplitude[10] (figure 1, A), the latencies of which (figure 1, B) were significantly related to the height of subjects (figure 2, A and B). On the opposite, the interwave latencies S wave-N20 (5.6±0.4 msec), S wave-N16 (2.8±0.4 msec) and N16-N20 (2.8±0.4 msec) did not show a significant relationship to the height of subjects (figure 2, C–E), their distribution curves being normal, at least within the age-range investigated.

It has thus been shown that the latency of each SEP component under investigation (i.e. S wave through N20) is significantly related to the height of the subject. Therefore, changes in latency of the SEP components, per se, would be scarcely adequate to detect abnormalities in conduction time of the central segment of the lemniscal pathway, especially if the disease process is so diffuse (or disseminated), as to affect bilaterally conduction of afferent impulses. To this purpose, three interwave latencies seem to be particularly suitable, i.e. S wave-N20, S wave-N16 and N16-N20, all of them bearing no significant relationship to the height of subjects. As to the functional significance of such intervals, it should be recalled that several lines of evidence gathered from both animals[12] and humans[13] support the hypothesis that the sequence of small waves, consistently preceding the primary response, are far-field potentials generated in the cervical posterior columns (S wave), the medial lemniscus and/or the dorsal columns nuclei (N14-P15 complex), the thalamus (N16) and the thalamocortical radiation (N17). Specifically it is postulated that: a) S wave-N20 reflects the time interval between the transit of an afferent volley through the posterior columns of the spinal cord and its arrival at the contralateral somatic area: therefore it represents the central conduction time of the lemniscal pathway; b) S wave-N16 and N16-N20 correspond respectively to the spino-thalamic and thalamocortical conduction times, though further evidence is needed to confirm this assumption. Studies are now in progress to find out to what extent the central conduction time of the lemniscal pathway may be affected by aging. Thereafter, the effective clinical relevance of these data will be verified.

1 Part of these results has been presented at the XX National Congress of the Italian Society of Neurology, held in Rome on November 24–26, 1977.
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Normal liver actually possesses a high vascular plasminogen activator activity

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Summary. Blood vessels isolated from the liver of the rat, guinea-pig, rabbit, dog and pig showed histochemically a more or less high plasminogen activator activity. In whole liver sections, the abundant release and diffusion of inhibitors of fibrinolysis from the liver parenchyma during the histochemical procedure, partially or totally mask this high vascular activity.

Human[1–7] and animal[8–12] liver is regarded as an organ which normally shows a very low vascular fibrinolytic activity[2–3,7–12] or no activity at all[1,4–6,8–9]. This conclusion was drawn by histochemical studies on whole liver sections. Comparative histochemical studies on whole organs and isolated tissue layers have shown, however, that in whole organ sections the release and diffusion of inhibitors of fibrinolysis from layers rich in inhibitors during the histochemical procedure partially or totally mask the fibrinolytic activity in adjacent tissue layers[13]. Normal liver is very rich in plasmin inhibitors[7,12]. Therefore, this study was undertaken to compare the fibrinolytic activity and plasmin inhibition on whole liver sections and sections of blood vessels isolated from corresponding areas of the liver in several species.

Materials and methods. Liver specimens from 18 adult, normal Wistar rats, 15 guinea-pigs, 10 White New Zealand rabbits, 10 dogs and 8 pigs were taken immediately after the animals were killed. From corresponding anatomical areas of each liver, blood vessels of various diameter were isolated as gently as possible. Liver specimens and isolated blood vessels were washed in saline, frozen and stored at a temperature of −20 °C or below. Both tissue preparations were studied simultaneously within 1 week after the initial freezing.

Fibrinolytic activity. Fibrinolytically active sites were located in frozen sections (6 µm) by the histochemical fibrin slide technique as described before[14], using bovine plasminogen-rich and plasminogen-free fibrinogen (Poviet, Organon-Teknika, Oss, The Netherlands). An average of 50 sections were examined from each specimen. The evaluation of the fibrinolytic activity was made as described before[14].

Plasmin inhibition. An average of 50 sections from the same specimen were examined with Noordhoek Hegt and Brakman's[7] fibrin slide 'sandwich' technique as follows: Frozen sections (16 µm), collected on precleaned microscope slides, were covered with a layer of fibrin by mixing and spreading 100 µl of a solution of plasminogen-free bovine fibrinogen and 20 µl of a solution of bovine thrombin (Leo pharmaceuticals, Denmark) in saline (20

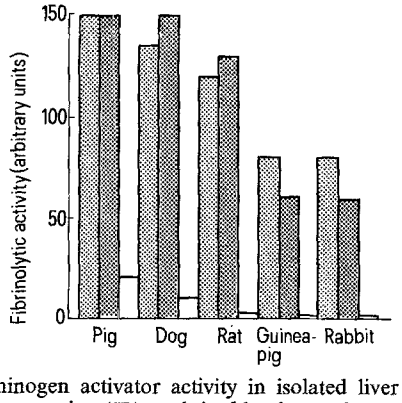
NIH units/ml) over an area of 2.5 × 4 cm. The slides were then left for 30 min in a horizontal position in a refrigerator with a moist atmosphere for diffusion of inhibitor into the fibrin layer. A section (10 µm) of a frozen solution of swine plasmin (Novo, Copenhagen, Denmark) in saline with 15% gelatin in a concentration equivalent to 1.5 MDH caseinolytic units/ml was placed on the top of each fibrin slide. These 'sandwich' slides were then incubated at 37 °C in a moist incubator for 120–240 min. The concentration of the plasmin solution used produced complete lysis of the fibrin layer after incubation for 120 min in the absence of tissue sections. Unlysed fibrin remaining after incubation for 120 min or longer in slides with tissue sections indicates the presence of plasmin inhibitors.

Results. Fibrinolytic activity. Whole liver sections showed a relatively low or very low vascular fibrinolytic activity (depending on the species), while sections of blood vessels isolated from corresponding anatomical areas of the liver and studied under the same experimental conditions showed a more or less high activity (figure). Small blood vessels in the perivascular connective tissue – of isolated vessels – showed also a varying fibrinolytic activity. Nerve sections occasionally seen in the perivascular tissue were very active.

The fibrinolytic activity was varying among the species studied. The species variation in the activity of the isolated blood vessels was not so marked as the species variation in the vascular activity revealed on whole liver sections (figure). Sections of isolated blood vessels or whole liver sections covered with plasminogen-free fibrin showed no lysis, indicating that the activity reported above was caused by a plasminogen activator.

Plasmin inhibition. Fibrin overlying the wall of the isolated arteries or veins was partially lysed after 120 min of incubation, while after 150 min of incubation the lysis was almost complete or complete. Fibrin overlying whole liver sections was unlysed even after 240 min of incubation, in all the species studied.

Discussion. Fibrinolytic studies on whole liver by using histochemical techniques[1–12] or extraction procedures[15–24] have shown that human[1–7,16,19,21,24] and animal[8–12,15,17,18,20,22,23] liver exhibits normally a very low activity (plasminogen activator activity)[2–3,7–12,19,24] or it is fibrinolytically inactive[1,4–6,8,9,15–18,20–23]. Studied with the same techniques, human cirrhotic liver[1,19] and the liver after haemorrhagic shock in humans[21], rat[25] and pig[26] showed, however, an increased fibrinolytic activity. Some cases of high fibrinolytic activity in apparently pathological human liver were also reported[27,28].



Mean plasminogen activator activity in isolated liver veins (▨), isolated liver arteries (▩) and in blood vessels of whole liver sections (□) of pig, dog, rat, guinea-pig and rabbit.

In the present histochemical study on normal liver, the fibrinolytic activity (plasminogen activator activity) was always much higher in isolated blood vessels than in vessels of the same type and diameter in whole liver sections. In whole liver sections, the abundant release and diffusion of inhibitors of fibrinolysis from the liver parenchyma during the whole histochemical procedure (damage of the tissue during sectioning, etc.) partially or totally mask the high fibrinolytic activity of the blood vessels. Liver parenchyma was found to be very rich in plasmin inhibitors, in agreement with previous studies[7,12], while the presence of inhibitors was low in the wall of the isolated blood vessels as well as in isolated bile ducts (unpublished). Therefore, in situ the blood vessels of the liver normally should show a more or less high plasminogen activator activity inspite (and independently) of the high concentration of plasmin inhibitors in the surrounding parenchyma. However, the situation might be different after an acute massive damage in situ with abundant release and diffusion of inhibitors throughout the damaged area.

In conclusion, from the results obtained on isolated blood vessels, normal liver should no longer be regarded as an organ fibrinolytically inactive or with very low fibrinolytic activity.

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