OBSERVATIONS ON THE UNUSUAL CYTOPLASMIC LACUNAE
OF THE PORCINE HEPATOCYTE

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INTRODUCTION

The most unusual and characteristic feature of the fine structure of the porcine hepatocyte is the presence of large lacunae in the cytoplasm (1). The lacunae are only observed in electron microscope preparations of hepatic tissue, and they appear as large spaces enclosed by a single membrane. There is no documented account of similar structures in other mammalian hepatocytes (2-4), and they appear to be unique to the porcine hepatocyte. No function has been ascribed to them. Their specialized association with other cytoplasmic organelles has not been established, and it has been suggested (1) that they may be derived from or be in continuity with either the bile canaliculus or the space of Disse. The present paper describes further observations which show
Figure 1  Low power electron micrograph of a typical porcine hepatocyte showing the general arrangement of the cell contents and the cytoplasmic lacuna (L). Its large size can be judged by comparison with the nucleus (N). Note the cytoplasmic blebs protruding into the lacunar space (arrows). A faceted microbody (mb), which is characteristic of the porcine hepatocyte, is shown. × 18,000.
Figure 2: Porcine hepatic cytoplasm. A highly complex network of lacunae (L) divided by cytoplasmic bridges is present throughout the cytoplasm. $\times 13,600$.

Figure 3: Detail of cytoplasmic lacunae showing the cytoplasmic bridges (arrows). The lacunae are seen to contain a finely granular, homogeneous, electron-opaque material (*). $\times 16,000$. 
that the cytoplasmic lacunae are in continuity with the space of Disse, and that they have a more complicated structure than first reported.

**MATERIALS AND METHODS**

Hepatic tissue from young adult pigs (Sus domestica) was removed immediately after they had been killed by stunning. It was finely minced in ice-cold cacodylate-buffered 4% glutaraldehyde, pH 7.2 (5) and fixed for 4 hr. The tissue was washed for 16 hr at 0°-4°C in cacodylate-buffered 0.25 M sucrose and postfixed for 2 hr in phosphate-buffered 1% osmium tetroxide (6). The fixed tissue was dehydrated in ethanol and embedded in Epon 812, essentially by the method of Luft (7). Thin sections were cut on a Servall MT-1 Porter-Blum ultramicrotome with glass knives or Ge-Fe-Ri diamond knives. They were mounted on washed copper grids, stained with lead tartate (8), and examined in either a Philips EM 300 or Hitachi HS7 electron microscope.

**OBSERVATIONS**

The cytoplasm of the young adult porcine hepatocyte commonly contains lacunae which are present as one or more large, irregular, or oval spaces. Their position in the cytoplasm varies but they are frequently observed beneath the perisinusoidal hepatic cell surface. The limiting membrane does not possess microvilli but cytoplasmic blebs may protrude into the lacunar space (Fig. 1).

A highly complex network of lacunae extends throughout the cytoplasm. The lacunae are divided by thin cytoplasmic bridges (Figs 2, 3) which lack cytoplasmic organelles apart from the occasional glycogen rosette. They contain a finely granular, homogeneous, electron-opaque material (Fig. 3). In some cells the network extends to the hepatic plasma membrane at the space of Disse on more than one side of the hepatocyte.

In favorably orientated sections a direct connection between the cytoplasmic lacunae and the space of Disse by a narrow channel has been demonstrated (Fig. 4). The limiting membrane of the channel does not appear to possess specialized regions. The lumen appears to communicate freely with the space of Disse, although the cellular debris,
which is present in the space of Disse, is never observed within the lacunae.

**DISCUSSION**

The lacunae of the young adult porcine hepatocyte form a complex cytoplasmic network which had not been observed or reported in other species. This unusual network of lacunae, which is present throughout the hepatic cytoplasm and is divided by thin cytoplasmic bridges, extends to the cell surface at the space of Disse. Direct continuity between the lacunae and the space of Disse is effected by a narrow channel. Although the narrow connection is freely open and its limiting membrane does not appear to possess specialized regions, cellular debris which is commonly contained in the space of Disse is apparently prevented from entering the lacunae. The nature of the finely granular, homogeneous, electron-opaque material in the lacunae is not known.

The pig is also unusual in possessing a circulating autoantibody-like factor in its plasma, which is reactive with a membrane-located antigenic determinant on autologous disaggregated hepatocytes. However, the porcine hepatocyte showed no evidence of having reacted with this autoantibody in vivo. Warr et al. suggested that the sinusoidal endothelial lining forms a perfect membrane so that the hepatocytes are not in free contact with the circulating plasma. The endothelial lining, however, is clearly discontinuous, so that the hepatocyte cell surface is necessarily directly bathed by the plasma. It is possible that the membrane-located antigen may be sited within the limiting membrane of the cytoplasmic lacunae. The narrow channel which is apparently effective in preventing cellular debris from entering the lacunae may also prevent the access of the autoantibody.

The large cytoplasmic lacunae, on the evidence presented, appear to represent a highly specialized structure which is not merely an invagination of the cell surface at the space of Disse. The lacunae are not derived from or in continuity with the hepatic bile canaliculi. The functional significance of this unusual network, which may extend throughout the hepatic cytoplasm to more than one cell surface at the space of Disse, is not known.

**SUMMARY**

An extensive network of lacunae divided by thin cytoplasmic bridges is described within the cytoplasm of the porcine hepatocyte. Direct continuity between the lacunae and the hepatic cell surface at the space of Disse by a narrow channel has been demonstrated. It is suggested that the lacunar membrane is a possible site of the membrane-located antigen reactive with an autoantibody-like factor present in the porcine serum.

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