Biliary Atresia Associated with Prenatal Infection by *Listeria Monocytogenes*

D. M. O. BECROFT

From the National Women's Hospital and the Princess Mary Hospital for Children, Auckland, New Zealand

Becroft, D. M. O. (1972). *Archives of Disease in Childhood*, **47**, 656. **Biliary atresia associated with prenatal infection by *Listeria monocytogenes*.** Recent disorganization of extrahepatic bile ducts was shown in a premature infant dying at 3 hours of age of an intrauterine pneumonia associated with an amniotic infection by *Listeria monocytogenes*. The alimentary canal was heavily infected with *Listeria* and listerial abscesses were located in the periportal zones of the liver. An ascending listerial cholangitis was considered to have caused both the abscesses and the disruptive changes in the biliary tract. Heavy infection of the alimentary tract from the amniotic cavity is usual in prenatal listeriosis, and the infection may resolve postnatally without severe systemic involvement. *Listeria monocytogenes* should be added to the list of prenatal infections which might cause 'idiopathic' biliary atresia.

Prenatal infection by the Gram-positive bacillus, *Listeria monocytogenes*, is an established cause of perinatal morbidity and mortality with a world-wide distribution (Seeliger, 1961; Gray, 1963; *Proceedings of the Third International Symposium on Listeriosis, Utrecht, 1966*; Maguire and Riley, 1967; Meerbach and Wöckel, 1969). Nevertheless, little is known of the usual sources and portals of entry of the maternal infection, the route by which this infection is transmitted to the fetus, and the reasons for the particular susceptibility of the conceptus. In 1969, 13 cases of neonatal listeriosis were diagnosed within 3 months at the National Women's Hospital in Auckland where the disease was rare previously: 7 infants died. The clinical and pathological features of this epidemic have been described previously (Becroft *et al.*, 1971) but the unusual changes found in the biliary tract of one infant have implications which will be considered in more detail in this report.

Case Report

A female Maori infant aged 2 hours was admitted to hospital because of episodes of apnoea associated with prematurity. The 18-year-old mother had had two previous pregnancies. The first child had been delivered at term, but died aged 3 hours, and pulmonary atelectasis was the only abnormality recorded at necropsy. The second pregnancy aborted at 12 weeks' gestation. The mother had been well during the third pregnancy.

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Her blood groups were A, D positive, and there were no abnormal blood group antibodies present in her serum at 21 weeks' gestation. The Wassermann reaction and VDRL were negative.

Labour began spontaneously at 36 weeks' gestation without earlier rupture of the membranes. There was no pyrexia during the 7 hours of normal labour. The delivery of a 1910 g infant was uneventful, but the baby was limp and cyanosed from birth, and though spontaneous respirations were established, apnoeic attacks began within 10 minutes. On admission the infant was collapsed, cyanosed, and hypothermic, and despite resuscitative measures she died 3 hours after delivery.

The following investigations were performed shortly before death. *Capillary blood*: Hb 13.3 g/100 ml; film normal for age; glucose 364 mg/100 ml; pH 6.78; PCO₂ 62 mmHg; standard bicarbonate 6.6 mEq/l. *Urine* (obtained by bladder puncture): no cells, no growth on culture. *CSF*: 11,500 red cells/mm³; 40 leucocytes/mm³, of which 60% were neutrophils and 40% lymphocytes; protein 265 mg/100 ml; sugar 169 mg/100 ml. *Bacteriology*: (methods described previously by Becroft *et al.*, 1971). Gram-positive bacilli were seen in direct smears of a swab taken from an external auditory canal; *Listeria monocytogenes* type 4B was isolated from this swab and also from the throat, conjunctiva, and umbilical cord, but not from the nose or from the CSF. *Listeria monocytogenes* was also isolated from a vaginal swab taken from the mother 3 hours *post partum*. X-rays: the lungs were poorly aerated; the skeleton was normal.

Necropsy Findings

Macroscopical. The infant at necropsy weighed 1880 g and had a crown-heel length of 44 cm, crown-

FIG. 1.—*Porta hepatis* showing the portal vein, hepatic artery, and, at right, epithelial remnants along the course of the disrupted hepatic duct. (H. and E. $\times 64$.)

rump length of 30 cm, and head circumference of 31 cm. There were no external features of note. Hepatomegaly was the most obvious visceral abnormality, the liver weighing 125 g. A diffuse mild fibrous expansion of the portal tracts was noted, but otherwise the consistency and colour of the external and cut surfaces were normal. No common hepatic duct could be identified in the porta hepatis though the gall bladder was normally formed and contained faintly bile-stained watery fluid which flowed freely through the common bile duct. The meconium in the large bowel was normally bile stained. The umbilical, portal, and hepatic veins were patent. The weights of other organs were within 1SD of the normal for body weight (Gruenwald and Minh, 1960). The lungs were congested and poorly aerated. There were minor subdural and intraventricular cerebral haemorrhages. The placenta had normal appearance and weighed 475 g.

Microscopical.

Biliary tract. All tissue from the porta hepatis was embedded and serial sections were examined from the course of the extrahepatic bile ducts. The common hepatic duct and its major tributaries were represented by a series of small irregular gland-like spaces lined by cuboidal or flattened cells which lay within a broad mantle of loose fibrous tissue containing scattered inflammatory cells (Fig. 1 and 2). The serial sections revealed that many of these spaces were connected by a very tortuous and narrow lumen, but nowhere was the lumen more than 50 μ wide and there were many

points of occlusion. The residual lumen contained a few neutrophils and macrophages, but no bacteria were identified.

Liver. All portal tracts were irregularly expanded by a cellular fibrous connective tissue containing proliferating bile ductules and a variable number of inflammatory cells, mostly lymphocytic (Fig. 3). The expansion encroached on the periphery of the liver lobules but otherwise the lobular pattern was preserved, the parenchymal cells were normal, and haemopoietic tissue and haemosiderin were present in amounts normal for the gestation. There was little evidence of cholestasis. Focal necrotic lesions were found in the liver, but were infrequent and not present in all sections. These necrotic foci were sharply demarcated and showed a variable but usually slight infiltration by mononuclear cells and degenerate neutrophils. No bacteria were identified. The largest lesion detected was 0.5 cm in diameter and this had many neutrophils centrally, while the periphery of the lesion lay partly within an adjacent portal tract (Fig. 4). A similar periportal localization was established for all necrotic foci encountered in serial sections of the liver.

Alimentary canal. There were masses of Gram-positive bacilli in the intestinal lumen. Similar organisms were seen in the depths of a focus of ulceration encountered in a random section of the upper oesophagus, but no ulcerating lesions were detected in multiple sections of the small and large intestine.

FIG. 2.—Remnants of hepatic duct at higher magnification. Inflammatory cells are seen in the lumen and surrounding fibroblastic tissue. (H. and E. $\times 480$.)

FIG. 3.—Expanded portal tract containing proliferating bile ducts and inflammatory cells. Parenchymal cells and haemopoietic elements are normal. (H. and E. $\times 180$.)

FIG. 4.—Abscess extends from the periphery of a portal tract into the liver lobule. (H. and E. $\times 300$.)

Other organs. In the lungs a majority of alveoli, alveolar ducts, and bronchioles contained neutrophils, macrophages, and a few Gram-positive bacilli. No abscesses or granulomata were detected in the lungs nor in sections which included both adrenals, both kidneys, the spleen, abdominal lymph nodes, bone marrow, myocardium, pancreas, thyroid, thymus, one ovary, and the meninges. The *placenta* and *membranes* showed a severe amnionitis. The chorionic plate and amnion contained Gram-positive bacilli and were heavily infiltrated by neutrophils in a distribution indicating both fetal and maternal cellular reactions. Clumps of Gram-positive bacilli were adhering to the amniotic surfaces. No necrotic or inflammatory lesions were detected in the chorionic villi and decidua.

Other investigations. *Listeria monocytogenes* was isolated from a lung swab. No cytomegalovirus or other virus was isolated from liver on passage through cultured diploid human fibroblasts.

Discussion

This infant's illness and most of the pathological changes were typical of the majority of cases dying during the epidemic of listerial infection (Becroft *et al.*, 1971). There had been no strong suspicion of maternal infection before labour began prematurely, and an infant was delivered who immediately showed respiratory difficulty and died within the first 12 hours of life. *Listeria monocyto-*

genes was isolated from various external sites and orifices of the infant and from the mother's vagina. A similar pneumonia of intrauterine 'inhalational' type was found in all fatal cases during the epidemic and was considered to be the major factor determining the early clinical and radiological signs and the fatal outcome. A similarly heavy bacterial colonization of the alimentary tract was observed in all fatal cases and minute foci of mucosal ulceration were potential sources of systemic infection. There was histological evidence of amnionitis in all four placentas examined and numerous bacteria on amniotic surfaces. It was assumed that the skin, orifices, and alimentary and respiratory tracts were infected from the amniotic cavity.

The focal necrotic lesions in the liver of this infant were identical in size, general configuration, and cell content to the multiple visceral abscesses found in other cases from the epidemic. Similar lesions, frequently referred to as granulomata, have featured in most descriptions of the pathology of perinatal listeriosis and usually are clearly of septicaemic origin. However, the restriction of the lesions to the liver in the present case and their periportal rather than random distribution in the liver lobules was consistent with a cholangiolitic route of infection. No Gram-positive bacilli were detected in these abscesses, contrasting with the heavy infection found in other cases.

Additional findings, which had no counterpart in other cases in the epidemic, nor as far as could be determined in previous descriptions of neonatal listeriosis, were the disorganization of the extra-hepatic bile ducts and the diffuse changes in the portal tracts resembling those found in biliary atresia. The histological appearances and the bile staining of the meconium are consistent with the bile duct obliteration being recent. This fact, together with the presence of a heavy bacterial infection of the intestinal lumen and the apparent cholangiolitic origin of the liver abscesses, provide strong grounds for considering that the bile duct disruption also was caused by a listerial cholangitis. The alternative was a coincidental occurrence. The bile duct lesions resemble very closely those described in association with rubella embryopathy (Strauss and Bernstein, 1968) and with trisomy 17-18 (Alpert, Strauss, and Hirschhorn, 1969), but there was no evidence of either condition in this case, nor of other virus-induced hepatocellular disease (Tolentino, Braitto, and Tassara, 1971).

A possible sequence of events to explain all lesions in this infant on the basis of listerial infection is as follows. (1) An initial infection of the amniotic cavity. (2) Infection of the alimentary canal from swallowed liquor. (3) An ascending listerial cholangitis with bile duct disruption and periportal abscess formation. (4) Early resolution of the systemic component of the infection in response to combined maternofetal immune reactions. (5) Persistence of infection in the amniotic cavity with eventual onset of premature labour. (6) Neonatal death from intrauterine aspiration pneumonia. The evidence favouring the assumption that the amniotic cavity and the fetal alimentary tract may be infected early in the course of intrauterine listeriosis has been reviewed previously (Becroft *et al.*, 1971). In these circumstances systemic infection may not occur or may be transient. In two prospective studies *Listeria monocytogenes* was isolated from the meconium or placentae of 24 infants of whom 12 remained asymptomatic, the

infections apparently resolving quickly once the infant was removed from the infected amniotic cavity (Ekelund *et al.*, 1962; Alison and Sarrut, 1967). Such occult prenatal infections by *Listeria monocytogenes* might be a rare cause of ascending cholangitis *in utero* and of 'idiopathic' biliary atresia. This possibility would be difficult to confirm retrospectively, but serological studies would be useful for its exclusion.

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Correspondence to Dr. D. M. O. Becroft, Princess Mary Laboratory, P.O. Box 5546, Wellesley Street, Auckland 1, New Zealand.