NOTES

Antibody to Mouse Alpha/Beta Interferon Abrogates Pichinde Virus-Induced Liver Lesions in Suckling Mice

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Infection of newborn C3HeB/FeJ mice with the arenavirus Pichinde resulted in stunted growth, severe liver cell degeneration, and death. Administration of sheep anti-mouse alpha/beta interferon globulin completely abrogated liver lesions in virus-infected mice, although it did not decrease the incidence of mortality. These results indicate that endogenous interferon may be responsible for some manifestations of viral disease.

Administration of mouse alpha/beta interferon (IFN- α/β) to suckling mice of different strains induces marked alterations in lipid metabolism (22), characteristic tubular aggregates in the endoplasmic reticulum (13), and severe liver cell degeneration (4, 5, 8). The finding that injection of neonatal mice with lymphocytic choriomeningitis (LCM) virus resulted in similar lesions which could be abrogated by administration of a potent antibody to mouse IFN- α/β (14, 17, 18; Y. Riviere and M. B. A. Oldstone, *J. Exp. Med.*, in press) added further evidence that IFN itself was the factor responsible for liver pathology. We have suggested, therefore, that under certain special circumstances some viruses may cause cytopathology by inducing the production of host substances (i.e., IFN) generally believed to be an integral part of the host defense against viral infection (4). As this concept is seemingly paradoxical, it was of interest to extend these observations by determining whether infection of mice with another virus in the family *Arenaviridae* (other than LCM virus) might also induce IFN-mediated liver necrosis. Accordingly, we examined the capacities of several new world arenaviruses (Amapari, Parana, Tacaribe, Tamiami, and Pichinde viruses) to induce liver lesions in newborn C3HeB/FeJ mice. Lipid accumulation was observed in the livers of suckling mice infected with Amapari and Tamiami viruses; focal areas of necrosis in the livers of Tacaribe virus-infected mice and extensive necrosis in the livers of Pichinde virus-infected mice. No lesions were observed in the livers of Parana virus-infected mice. We chose Pichinde virus for further experiments.

Newborn C3HeB/FeJ mice were injected subcutaneously with 10^5 PFU of the AN 3739 strain of Pichinde virus and sacrificed every other day thereafter for 12 days (six mice each time). The livers were prepared for light and electron microscopy as previously described (13). Marked liver lesions were found in all virus-infected mice 8 days after virus infection. The liver lesions identical to those previously observed in IFN-treated or LCM virus-infected suckling mice were characterized by extensive areas of necrosis beginning below the capsule with cytolysis, pyknosis, and

karyorrhexis (Fig. 1A). The remaining parenchyma was edematous, and many hepatocytes contained vacuoles consistent with fatty degenerative change (5). Furthermore, tubular aggregates similar to those we have previously described (13) were also observed in the endoplasmic reticulum of hepatocytes of Pichinde virus-infected mice (Fig. 1B). Pooled sera from litters of mice were assayed for IFN on days 2, 4, 6, 8, and 10 after virus infection. IFN was detected on days 4 and 6 (titers of 1:512 and 1:256, respectively) by using assay techniques previously described (5, 8, 17).

As these results suggested that interferon might be responsible for the development of liver lesions in these mice, we determined the effect of administration of a potent sheep polyclonal antibody to IFN- α/β on the evolution of Pichinde virus disease in suckling mice.

The pooled results of two experiments (Table 1) may be summarized as follows. (i) All virus-infected mice showed some inhibition of weight gain compared with uninfected suckling mice, but this effect was most marked in virus-infected mice injected with antibody to IFN (administration of antibody to IFN- α/β did not affect the growth of uninfected mice). (ii) All virus-infected mice died regardless of treatment. (iii) Whereas all virus-infected mice that were untreated (8 of 8) or treated with normal sheep globulin (10 of 10) showed extensive liver cell degeneration, 0 of 9 virus-infected mice treated with antibody to IFN- α/β had detectable liver lesions. Furthermore, six of eight mice of the former two groups showed typical hepatic cell tubular aggregates, whereas no tubular aggregates were observed in the hepatocytes of four virus-infected mice treated with antibody to IFN. (iv) Although administration of antibody prevented the development of liver lesions in virus-infected mice, it did not inhibit viral multiplication in the livers of these mice.

The findings that inoculation of neonatal mice with Pichinde virus results in liver cell degeneration and cytoplasmic tubular aggregates characteristic of the pathology induced by IFN administration (4, 5, 8, 13) or infection with LCM virus (17, 18; Riviere and Oldstone, in press) and that this liver pathology can be abrogated by administration of

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FIG. 1. (A) Liver from an 8-day-old mouse infected with Pichinde virus, showing extensive areas of subcapsular necrosis. An area of normal hepatocytes with islands of haemopoietic tissue is present (lower left corner). (B) A tubular aggregate composed of a network of small tubules within an hepatocyte from a 5-day-old mouse infected with Pichinde virus.

potent antibody to IFN- α/β support our contention that IFN is the factor responsible for these lesions. Whereas administration of antibody to IFN- α/β to neonatal mice infected with LCM virus also inhibited the effect of viral infection on weight gain, decreased the incidence of mortality (18; Riviere and Oldstone, in press), and inhibited the development of glomerulonephritis (6) in some strains of mice infected with some strains of LCM virus, under other experimental conditions only the liver pathology induced by LCM virus was blocked by administration of antibody (14). In the experiments reported herein, only the liver pathology

induced by infection of mice with Pichinde virus can be attributed to IFN, as antibody to IFN- α/β did not inhibit the failure to gain weight (in fact, mice in this group showed the least weight gain) nor did it reduce the incidence of mortality.

The interest of this work may be twofold. First, it emphasizes the concept that viral infection may result in severe disease, either because the virus can perturb certain cellular functions in the absence of a viral cytopathic effect, as reported by Oldstone and co-workers (14, 15), or because virus-induced host products such as IFN can under some

TABLE 1. Effect of antibody to IFN- α/β on the evolution of disease in suckling mice infected with Pichinde virus^a

Group, treatment	Mean cumulative weight increase (g) $\pm$ SD in 10 days	Mean day of death $\pm$ SD (no. of mice)	No. of mice with liver lesions/no. of mice examined	No. of mice with tubular aggregates in liver/no. of mice examined	Virus titers in livers ($\log_{10} \pm$ SD)	
					Day 5	Day 10
Uninfected (None)	5.7 $\pm$ 0.6		0/10	0/4		
Infected						
None	4.5 $\pm$ 0.6	15.6 $\pm$ 3.1 (11)	8/8	4/4	6.5 $\pm$ 0.5	5.9 $\pm$ 0.1
Normal sheep globulin	4.6 $\pm$ 0.2	12.9 $\pm$ 3.8 (9)	10/10	2/4	5.2 $\pm$ 0.2	5.9 $\pm$ 0.1
Antibody to IFN- α/β	1.8 $\pm$ 0.5	12.4 $\pm$ 1.1 (12)	0/9	0/4	7.9 $\pm$ 0.8	6.7 $\pm$ 0.5

^a Five to six litters of C3HeB/FeJ mice were used for each of the above experimental groups. All mice were less than 24 h old when inoculated subcutaneously with 10^5 PFU of Pichinde virus, with virus and normal sheep globulin, or with virus and 10^6 neutralizing units of hyperimmune anti-mouse IFN- α/β globulin prepared and assayed as previously described (7). Globulin preparations were injected 2 1/2 h before viral injection and 72 h thereafter. Mice were weighed daily. At 8 days after injection, two litters in each group were sacrificed for histologic examination of the livers. At 5 and 10 days after injections, two litters in each experimental group were sacrificed, and the livers were assayed for virus as previously described (12). The mean cumulative weight increase for uninfected mice injected with antibody to IFN- α/β was 5.3 $\pm$ 0.3 g after 10 days.

conditions cause cellular injury (4). Second, the finding that the endogenous IFN induced by two arenaviruses (LCM and Pichinde) can cause severe liver lesions suggests the possibility that endogenous IFN may also contribute to the severe liver lesions (1–3, 11, 16, 19–21) observed in humans infected with other highly pathogenic arenaviruses such as Junin, Lassa Fever, and Machupo viruses. In this regard, very high levels of IFN- α have been detected in the serum of patients dying of Argentine hemorrhagic fever due to Junin virus infection (9). Levis et al. suggested that these high levels of IFN may not only be a marker of the degree of infection but also have deleterious effects (10).

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