NONNEOPLASTIC LESIONS

Cytomegalovirus Infection, Salivary Glands, Mouse, Rat, Hamster

Stephen W. Barthold

Synonyms. Salivary gland virus infection, cytomegalic inclusion disease.

Gross Appearance

Under most circumstances there is no grossly visible lesion. During acute, generalized infections in mice, the liver may be enlarged and pale with hemorrhagic foci. Intestinal serosa can be irregularly reddened and mucosa congested (McCordock and Smith 1936). Mineralization of skeletal muscle and brown fat (Lussier 1975) and blood-tinged ascitic fluid (Olding et al. 1976) have also been observed. Mice infected as neonates are runted with thymic and splenic involution (Schwartz et al. 1975).

Microscopic Features

Acute generalized infections in susceptible (especially if immunosuppressed) mice are characterized by focal necrosis, cytomegaly, intracytoplasmic and intranuclear (type A) inclusion bodies, and nonsuppurative inflammation of multiple organs. Lesions can be found in salivary and lacrimal glands, brain, liver, spleen, thymus, lymph nodes, paritoneal connective and adipose tissue, lungs, skin, renal glomeruli, bowel, pancreas, adrenals, skeletal and cardiac muscle, cartilage, and brown fat (Brody and Craighead 1974; Gardner et al. 1974; Jordan 1979; Lussier 1975; McCordock and Smith 1936; Mims and Gould 1979; Olding et al. 1976; Schwartz et al. 1975). Lesions may be restricted to salivary glands in natural infections of mice, rats, and hamsters (Gardner et al. 1974; Lussier 1975; Lyon et al. 1959; Priscott and Tyrrell 1982). Submaxillary salivary glands are preferentially infected, the sublingual glands less so and the parotids least of all (Mims and Gould 1979; Ruebner et al. 1966). Eosinophilic intracytoplasmic and intranuclear inclusions are present in acinar and ductal epithelial cells. Intranuclear inclusions are Feulgen positive and intracytoplasmic inclusions are PAS and Feulgen positive. Infected cells can become atypically large (cytomegaly). This is accompanied by infiltration of the surrounding interstitium with lymphocytes and plasma cells (Fig. 172). In the acute phase of generalized infections, other organs, particularly liver (Fig. 173), are more apt to possess lesions than slivary glands, which develop lesions later in the course of infection (Brodsky and Rowe 1958; Gardner et al. 1974; Henson and Strano 1972; Ruebner et al. 1966).

Ultrastructure

Initially, nuclei of acinar cells of salivary glands become enlarged with chromatin uniformly dispersed. Nucleoli enlarge and are partially surrounded by fibrillar structures identical to those of the virus membrane. Pleomorphic dense cores become surrounded by these fibrils, followed by the appearance of cores, fibrils, and virions throughout the nucleoplasm. Virus particles acquire a second membrane by passing through the nuclear membrane into the cytoplasm. Particles make contact with membranes of dilated Golgi vesicles and to a lesser extent endoplasmic reticulum, acquiring a third membrane or envelope by the process of invagination. Vesicles enlarge and are filled with virus particles as they approach the cell apex and are extruded into the lumen. Intranuclear inclusions observed by light microscopy correspond to aggregates of granular and fibrillar material intermixed with virus particles. Intracytoplasmic inclusions correspond to virus-filled vesicles (Fig. 174) (Henson and Strano 1972; Ruebner et al. 1966). Similar viral replicative changes have been described in pulmonary macrophages (Brody and Craighead 1974), splenic reticulum cells, and hepatocytes (Ruebner et al. 1964, 1966).
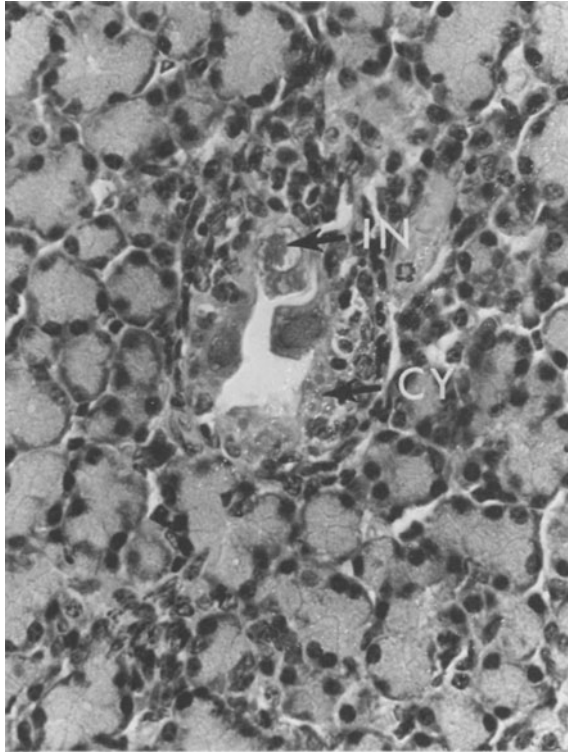
Fig. 172. Salivary gland, mouse. Natural cytomegalovirus infection. Enlarged ductal cells contain large intranuclear (IN) and small intracytoplasmic (CY) inclusions. The surrounding interstitum is infiltrated with a few lymphocytes. H and E, × 440
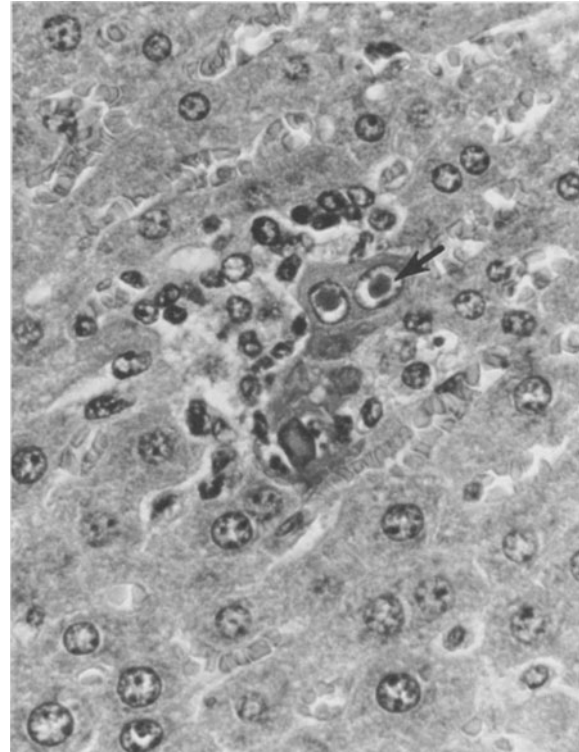

Fig. 173. Focal hepatitis, liver, mouse. Intranuclear inclusions (arrow); experimental cytomegalovirus infection. H and E, × 440

Differential Diagnosis

In mice, sialoadenitis is also caused by polyoma virus and reovirus type 3. Polyoma virus induces intranuclear inclusions, but not cytomegaly or intracytoplamic inclusions, and preferentially infects the parotid salivary gland. Reovirus type 3 can cause necrotizing sialoadenitis without inclusion bodies. Furthermore, mouse salivary gland is a primary target organ for another herpesvirus, mouse thymic virus, but salivary gland lesions have not been described (Cross et al. 1979). Murine mammary tumor virus also replicates in and is shed from salivary glands, but is not associated with light microscopic changes (Bentvelzen and Hilgers 1980).

Biologic Features

Natural History. Infections eith cytomegalovirus in mice under natural conditions are almost invariably subclinical. Virus is transmitted by direct contact through inhalation or ingestion and is ex-

creted in saliva, tears, and urine (Brodsky and Rowe 1958; Lussier 1975). In utero transmission does not play an important role in natural murine infections, but transmission from dam to fetus can occur in latent form under experimental conditions (Chantler et al. 1979). Suckling pups are protected from infection by maternally derived antibody (Mannini and Medearis 1961; Medaris 1964). In susceptible mice, infection is followed by one of four outcomes: death; recovery; chronic localized replication in salivary glands; and latency, in which virus is harbored in tissues in a nonreplicative state (Olding et al. 1976). Considerably less is known about cytomegaloviruses of hamster and rat. In these species, infection appears to be largely restricted to salivary glands (Lussier 1975; Lyon et al. 1959; Priscott and Tyrrell 1982).

Pathogenesis. Most studies on pathogenesis of cytomegaloviruses have been done with eht mouse virus. The course of infection is dependent on route of inoculation, host age and genotype, virulence of the agent, and immune status of the host
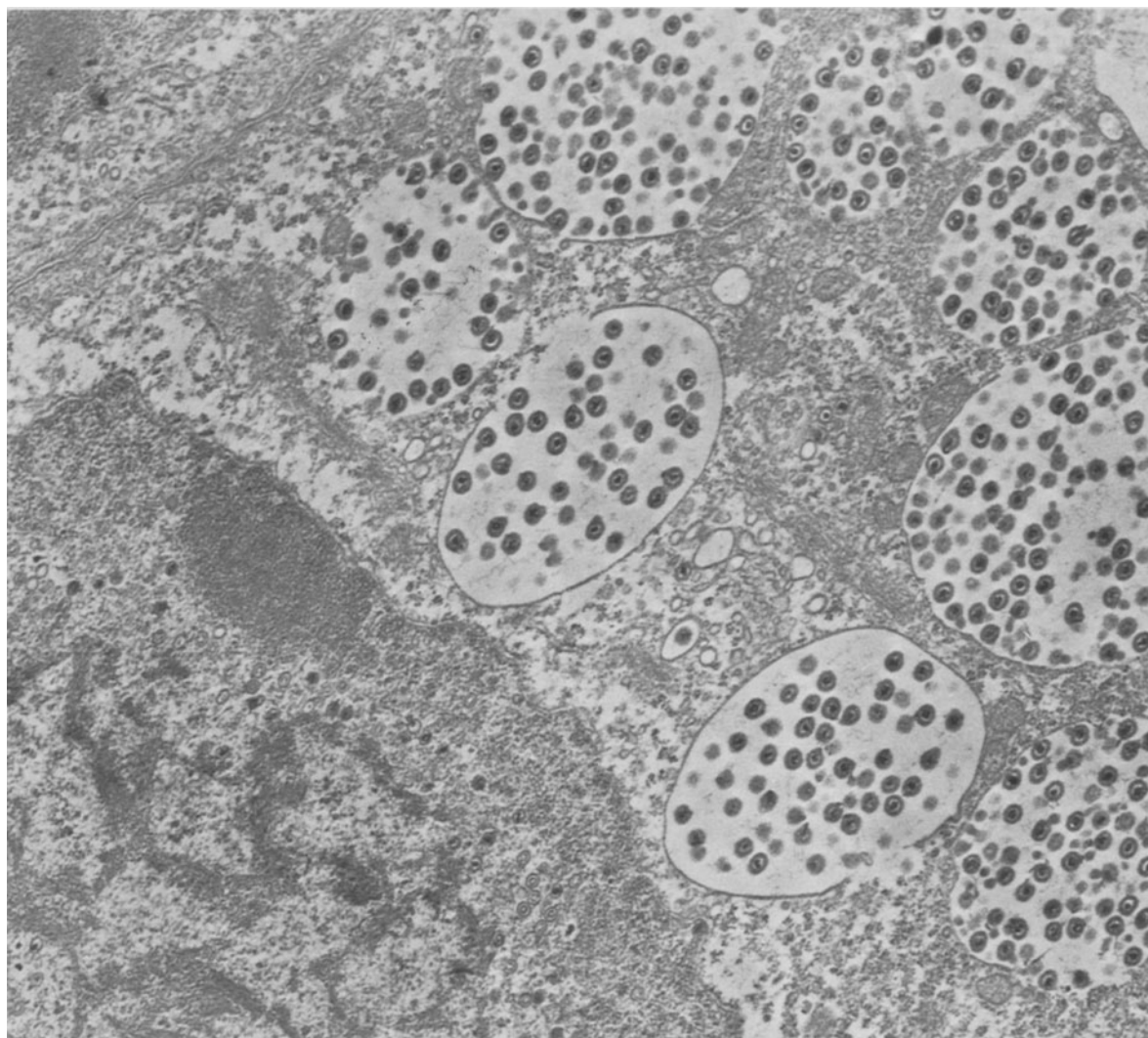
Fig. 174. Salivary gland, mouse, 16 days after infection by cytomegalovirus. Viral particles are in the nucleus and in cytoplasmic vesicles. (Courtesy of Boris Ruebner and *American Journal of Pathology*). Uranyl acetate, TEM × 16300

(Hudson 1979; Lussier 1975; Mannini and Medaris 1961). Within a week of inoculation, young mice develop leukocytosis and viremia with dissemination to multiple organs. Hematocrit, leukocyte and platelet counts decrease, but return to normal by 15 days (Lussier 1975; McCordoc and Smith 1936; Osborn and Shahidi 1973). Intranasal or oral inoculation of low doses into young adult mice results in subclinical pulmonary infection, followed by viremia and dissemination. Virus replicates in monocytes and alveolar macrophages, with septal thickening and edema (Brody and Craighead 1974; Jordan 1978). This probably represents the natural course of infection. Salivary gland infection occurs regularly but virus replication does not take place until later than other organs and infection perists for many months in this location after virus is cleared from other sites. Replication and excretion of salivary virus can occur in the presence of absence of discernible lesions. The affinity of the virus for salivary glands is underscored by the fact that this site is preferentially infected regardless of host age, and natural infections are localized to this site alone (Lussier 1975; Brodsky and Rowe 1958; Mims and Gould 1979; Olding et al. 1976).

Macrophages are important in early virus clearance and restriction (or replication) (Brautigam et al. 1979). Neutralizing antibody appears early after infection and provides a protective effect against challenge, but it does not seem to be involved in recovery (Hudson 1979). Cell-mediated

immunity is important in host recovery (Ho 1980; Hudson 1979; Lussier 1975) and lymphocytic infiltration heralds termination of infection (Henson and Strano 1972). Immunity can result in recovery, but more often results in two types of persistent infections. In chronic infections, the virus is localized to salivary glands, where it replicates and is orally excreted for up to a year after infection (Brodsky and Rowe 1958). More often, latent infections develop, in which replicating virus cannot be detected (Olding et al. 1976). Salivary glands, macrophages, bone marrow-derived (B) lymphocytes, and reproductive tissues of latently infected mice harbor cytomegalovirus but not brain, thymus, liver, or kidney (Brautigam et al. 1979; Cheung et al. 1980; Olding et al. 1976). Latent infection can be reactivated in vivo and in vitro by immunosuppression and allogeneic reactions (Gardner et al. 1974; Hudson 1979; Lussier 1975; Montplaisir 1979).

Cytomegalovirus has been shown to have a number of immunosuppressive effects on infected mice due to alterations in T-cell, B-cell, and macrophage functions and interferon response (Hudson 1979). A synergistic effect on mouse mortality occurs in combined cytomegalovirus and *Pseudomonas aeruginosa* infections (Hamilton and Overall 1978). Chronically and latently infected mice can develop immune complex glomerular lesions and antinuclear antibodies (Olding et al. 1976).

Etiology. Cytomegaloviruses are herpesviruses which are species specific in vivo. Several strains have been isolated from laboratory and wild mice, and vary in virulence. Different cytomegaloviruses have been described in mice, rats, hamsters, guinea pigs, nonhuman primates, moles, pigs, dogs, feral rodents, sheep, horses, and man (Lussier 1975).

Frequency. Based on observation of salivary gland inclusions or isolation of virus, infection is not common among laboratory mice and is rare among hamsters and rats. With appropriate techniques, infection is found to be very common among wild mice (Lussier 1975; Mannini and Medearis 1961). Recently, molecular hybridization techniques have revealed latent cytomegaloviral DNA in specific pathogen-free mice, in which virus was undetectable by other means. It is likely many laboratory mice may be latently infected, but presence of viral DNA does not necessarily mean that the virus can be reactivated or will interfere with experimental procedures (Cheung et al. 1980).

Comparison with Other Species

Cytomegaloviruses frequently infect salivary glands in many species of animals and lesions are often present as incidental findings. Human infection mimics the murine disease in many ways, including its propensity for generalized infection following immunosuppression and allogeneic reactions. As in mice, fatal cytomegalovirus infections in man are characterized by pneumonia and disseminated cytomegalic inclusion disease (Hudson 1979; Lussier 1975; Mims and Gould 1979; Montplaisir 1979).

References

Bentvelzen P, Hilgers J (1980) Murine mammary tumor virus. In: Klein G (ed) Viral oncology. Raven, New York, pp 311-355

Brautigam AR, Dutko FJ, Olding LB, Oldstone MBA (1979) Pathogenesis of murine cytomegalovirus infection: the macrophage as a permissive cell for cytomegalovirus infection, replication and latency. J Gen Virol 44: 349-359

Brodsky I, Rowe WP (1958) Chronic subclinical infection with mouse salivary gland virus. Proc Soc Exp Biol Med 99: 654-655

Brody AR, Craighead JE (1974) Pathogenesis of pulmonary cytomegalovirus infection in immunosuppressed mice. J Infect Dis 129: 677-689

Chantler JK, Misra V, Hudson JB (1979) Vertical transmission of murine cytomegalovirus. J Gen Virol 42: 621-625

Cheung K-S, Huang E-S, Lang DJ (1980) Murine cytomegalovirus: detection of latent infection by nucleic acid hybridization technique. Infect Immun 27: 851-854

Cross SS, Parker JC, Rowe WP, Robbins ML (1979) Biology of mouse thymic virus, a herpesvirus of mice, and the antigenic relationship to mouse cytomegalovirus. Infect Immun 26: 1186-1195

Gardner MB, Officer JE, Parker J, Estes JD, Rongey RW (1974) Induction of disseminated virulent cytomegalovirus infection by immunosuppression of naturally chronically infected wild mice. Infect Immun 10: 966-969

Hamilton JR, Overall JC Jr (1978) Synergistic infection with murine cytomegalovirus and *Pseudomonas aeruginosa* in mice. J Infect Dis 137: 775-782

Henson D, Strano AJ (1972) Mouse cytomegalovirus. Necrosis of infected and morphologically normal submaxillary gland acinar cells during termination of chronic infection. Am J Pathol 68: 183-202

Ho M (1980) Role of specific cytotoxic lymphocytes in cellular immunity against murine cytomegalovirus. Infect Immun 27: 767-776

Hudson JB (1979) The murine cytomegalovirus as a model for the study of viral pathogenesis and persistent infections. Arch Virol 62: 1-29

Jordan MC (1978) Interstitial pneumonia and subclinical infection after intranasal inoculation of murine cytomegalovirus. Infect Immun 21: 275-280

Lussier G (1975) Murine cytomegalovirus (MCMV). Adv Vet Sci Comp Med 10: 223–247

Lyon HW, Christian JJ, Miller CW (1959) Cytomegalic inclusion disease of lacrimal glands in male laboratory rats. Proc Soc Exp Biol Med 101: 164–166

Mannini A, Medearis DN Jr (1961) Mouse salivary gland virus infections. Am J Hyg 73: 329–343

McCordock HA, Smith MG (1936) The visceral lesions produced in mice by the salivary gland virus of mice. J Exp Med 63: 303–310

Medearis DN Jr (1964) Mouse cytomegalovirus infection. 3. Attempts to produce intrauterine infections. Am J Hyg 80: 113–120

Mims CA, Gould J (1979) Infection of salivary glands, kidneys, adrenals, ovaries and epithelia by murine cytomegalovirus. J Med Microbiol 12: 113–122

Montplaisir S (1979) Latency and activation of cytomegalovirus in man and in mice. Can J Microbiol 25: 261–266

Olding LB, Kingsbury DT, Oldstone MBA (1976) Pathogenesis of cytomegalovirus infection. Distribution of viral products, immune complexes and autoimmunity during latent murine infection. J Gen Virol 33: 267–280

Osborn JE, Shahidi NT (1973) Thrombocytopenia in murine cytomegalovirus infection. J Lab Clin Med 81: 53–63

Priscott PK, Tyrrell DAJ (1982) The isolation and partial characterisation of a cytomegalovirus from the brown rat, Rattus norvegicus. Arch Virol 73: 145–160

Ruebner BH, Miyai K, Slusser RJ, Wedemeyer P, Medearis DN Jr (1964) Mouse cytomegalovirus infection. An electron microscopic study of hepatic parenchymal cells. Am J Pathol 44: 799–821

Ruebner BH, Hirano T, Slusser R, Osborn J, Medearis DN Jr (1966) Cytomegalovirus infection. Viral ultrastructure with particular reference to the relationship of lysosomes to cytoplasmic inclusions. Am J Pathol 48: 971–989

Schwartz JN, Daniels CA, Klintworth GK (1975) Lymphoid cell necrosis, thymic atrophy, and growth retardation in newborn mice inoculated with murine cytomegalovirus. Am J Pathol 79: 509–522

Sialodacryoadenitis (SDA) Infection, Rat

Robert O. Jacoby

Synonyms. Rat coronavirus infection.

Gross Appearance

Lesions develop during the 1st week after infection and occur primarily in the submaxillary and parotid salivary glands, which are located on the anteroventral and anterolateral aspects of the neck, respectively (Fig. 175). The glands are unilaterally or bilaterally enlarged ans pale yellow to white in contrast to their normal tan color (Fig. 176). Periglandular connective tissue is often edematous and, together with glandular enlargement, may cause grossly visible cervical swelling in the living rat. The parotid salivary gland of the rat is lobulated and sometimes difficult to delineate from surrounding adipose tissue when inflammatory edema is severe. The cervical lymph nodes may also be enlarged and edematous. They are frequently congested, especially early in infection, and occasionally flecked with red spots. Gross lesions of the oropharynx and tubular portions of the gastrointestinal tract have not been reported.

Microscopic Features

The lesions of SDA are characterized by necrosis and inflammation during acute stages of disease and by squamous metaplasia as tissues are repaired (Jonas et al. 1969; Jacoby et al. 1975). Serous and miced salivary glands in the oropharynx and neck can be affected, but mucous salivary glands are resistant. Earliest lesions are detected in the submaxillary salivary glands, where necrosis of ductular epithelium can begin by 4 days postinfection (Fig. 177). Immunohistochemical stains reveal viral antigen in the cytoplasm of infected epithelial cells. Infection and necrosis of adjacent acinar parenchyma begins shortly thereafter and spreads quickly to involve large areas of the parenchyma (Fig. 178). Necrosis is accompanied by inflammatory edema which engulfs periglandular connective tissues. The inflammatory cells consist of small and large mononuclear cells and polymorphonuclear leukocytes. Necrosis and inflammation in the parotid salivary glands develop in a similar pattern and lesions are sometimes found in salivary tissues of the oropharynx. The sublingual salivary glands, which are mucin-secreting glands in the rat and which lie at the ante-