8.5.10 Rhabdoviruses Rabies and rabies- related ly

8.5.10 Rhabdoviruses: Rabies and rabies- related lyssaviruses 805

8.5.10 Rhabdoviruses 805 diversity. In: Desselberger U, Gray J (eds) Viral gastroenteritis, pp. 317–44. Elsevier Science, Amsterdam. Jones MK, et al. (2014). Enteric bacteria promote human and mouse norovirus infection of B cells. Science 346, 755–9. Kanai Y, et al. (2017). Entirely plasmid-based reverse genetics system for rotaviruses. Proc Natl Acad Sci, 114, 2349–54. Kamada N, et al. (2013). Control of pathogens and pathobionts by the gut microbiota, Nat Immunol 14, 685–90. Karst SM, et al. (2014). Advances in norovirus biology. Cell Host & Microbe, 15, 668–80. Karst SM, Zhu S, Goodfellow IG (2015). The molecular pathology of noroviruses. J Pathol, 235, 206–16. King CK, et al. (2003). Managing acute gastroenteritis among chil- dren: oral rehydration, maintenance, and nutritional therapy. Morb Mort Wkly Rec MMWR, 52, 1–16. Komoto S, et al. (2018). Generation of recombinant rotaviruses ex- pressing fluorescent proteins by using an optimized reverse genetics system. J Virol, 92, pii: e00588-18. Koo HL, et al. (2013). Noroviruses: the most common pediatric viral enteric pathogen at a large university hospital after introduction of rotavirus vaccination. J Pediatric Infect Dis Soc, 2, 57–60. Kroneman A, et al. (2013). Proposal for a unified norovirus nomencla- ture and genotyping. Arch Virol, 158, 2059–68. Kuss SK, et al. (2011). Intestinal microbiota promote enteric virus rep- lication and systemic pathogenesis, Science, 334, 249–52. Leroux-Roels G, et al. (2018). Safety and immunogenicity of different formulations of norovirus vaccine candidate in healthy adults: a randomized, controlled, double-blind clinical trial. J Infect Dis, 217, 597–607. Lundgren O, et al. (2000). Role of the enteric nervous system in the fluid and electrolyte secretion of rotavirus diarrhea. Science, 287, 491–5. Madhi SA, et al. (2010). Effect of human rotavirus vaccine on severe diarrhea in African infants. N Engl J Med, 362, 289–98. Marczinke B, et al. (1994). The human astrovirus RNA-dependent RNA polymerase coding region is expressed by ribosomal frameshifting. J Virol, 68, 5588–95. Matthijnssens J, et al. (2011). Uniformity of rotavirus strain nomen- clature proposed by the Rotavirus Classification Working Group (RCWG). Arch Virol, 156, 1397–413. Mendez E, Arias CF (2013). Astroviruses. In:

Knipe DM, Howley PM, et al. (eds) Fields virology, 6th edition, pp. 609–28. Lippincott Williams & Wilkins, Philadelphia, PA. Mihalov-Kovács E, et al. (2015). Candidate new rotavirus species in sheltered dogs, Hungary. Emerg Infect Dis, 21, 660–3. Nordgren J, et al. (2010). Norovirus gastroenteritis outbreak with a secretor-independent susceptibility pattern, Sweden. Emerg Infect Dis, 6, 81–7. Nordgren J, et al. (2014). Both Lewis and secretor status mediate sus- ceptibility to rotavirus infections in a rotavirus genotype-dependent manner. Clin Infect Dis, 59, 1567–73. Norman, JM, Handley SA, Virgin HW (2014). Kingdom-agnostic metagenomics and the importance of complete characterization of enteric microbial communities. Gastroenterology, 146, 1459–69. Offit PA (1994). Rotaviruses: immunological determinants of protec- tion against infection and disease. Adv Virus Res, 44, 161–202. Patel MM, et al. (2011). Intussusception risk and health benefits of rota- virus vaccination in Mexico and Brazil. N Engl J Med, 364, 2283–92. Payne DC, et al. (2013). Norovirus and medically attended gastro- enteritis in U.S. children. N Engl J Med, 368, 1121–30. Ramani S, et al. (2015). Mucosal and cellular immune responses to Norwalk virus. J Infect Dis, 212, 397–405. Richardson V, et al. (2010). Effect of rotavirus vaccination on death from childhood diarrhea in Mexico. N Engl J Med, 362, 299–305. Robinson CM, Pfeiffer JK (2014). Viruses and the microbiota. Annu Rev Virol, 1, 55–69. Ruvoën-Clouet N, Belliot G, Le Pendu J (2013). Noroviruses and histo- blood groups: the impact of common host genetic polymorphisms on virus transmission and evolution. Rev Med Virol, 23, 355–66. SAGE (2009). Meeting of the Immunization Strategic Advisory Group of Experts, April 2009—conclusions and recommendations. Wkly Epidem Rec, 84, 220–35. Sarker SA, et al. (2013). Anti-rotavirus protein reduces stool output in infants with diarrhoea: a randomized placebo-controlled trial. Gastroenterology, 145, 740–8. Settembre EC, et al. (2011). Atomic model of an infectious rotavirus particle. EMBO J, 30, 408–16. Tate JE, et al. (2016). Global, regional, and national estimates of rota- virus mortality in children <5 years of age, 2000-2013. Clin Infect Dis, 62, S96–105. USAID, UNICEF, WHO (2005). Diarrhaea treatment guidelines including new recommendations for the use of ORS and zinc sup- plementation for clinic-based health care workers. World Health Organization, Geneva. Vesikari T, et al. (2015). European Society for Paediatric Infectious Diseases consensus recommendations for rotavirus vaccination in Europe. Pediatr Infect Dis J, 34, 635–43. Virgin HW (2014). The virome in mammalian physiology and disease. Cell, 157, 142–50. Wold WSM, Ison MG (2013). Adenoviruses. In: Knipe DM, et al. (eds) Fields virology, 6th edition, pp. 1732–67. Lippincott Williams & Wilkins, Philadelphia, PA. Yunus MA, et al. (2010). Development of an optimized RNA-based murine norovirus reverse genetics system. J Virol Methods, 169, 112–18. Zaman K, et al. (2010). Efficacy of pentavalent rotavirus vaccine against severe rotavirus gastroenteritis in infants in developing countries in Asia: a randomised, double-blind, placebo-controlled trial. Lancet, 376, 615–23. 8.5.10 Rhabdoviruses: Rabies and rabies-related lyssaviruses Mary J. Warrell and David A. Warrell ESSENTIALS The Rhabdoviridae are a large family of RNA viruses, two genera of which infect animals: the genus Lyssavirus contains rabies and rabies-related viruses that cause at least 55 000 deaths annually in Asia and Africa. Transmission and epidemiology The risks and problems posed by rabies and other lyssaviruses vary across the world. Viruses can penetrate broken skin and intact mucosae. Humans are usually infected when virus-laden saliva is in- oculated through the skin by the bite of a rabid animal, usually a dog.

 Although the greatest threat to man is the persistent cycle of infection in stray dogs, several other terrestrial mammal species are reservoirs of infection. In the Americas, bat viruses are also classic species 1 rabies and insectivorous bats have become the principal vectors of infection to humans in the United States of America. Elsewhere in the world,

there is increasing evidence of widespread rabies-related lyssavirus infection of bats. Unrecognized infection of organ donors has proved fatal to transplant recipients. Clinical features After a highly variable incubation period (usually 20–90 days), pro- dromal symptoms include itching at the site of the healed bite wound. These are followed by symptoms of either furious or para- lytic rabies, reflecting whether infection of the brain or spinal cord predominates. Furious rabies—the diagnostic symptom is hydrophobia, a com- bination of jerky inspiratory muscle spasms, associated with terror, initially provoked by attempts to drink water. Patients may suffer gen- eralized arousal, during which they become wild, hallucinated, fugi- tive, and on rare occasions aggressive. Paralytic rabies—flaccid ascending paralysis develops, starting in the bitten limb. Diagnosis The diagnosis can be made during life using rapid laboratory methods such as immunofluorescence of punch biopsy specimens of skin taken from a hairy area. Polymerase chain reaction tests are used increasingly to detect rabies in saliva and skin biopsy material. However, lack of facilities hampers the confirmation of disease in developing countries where the diagnosis usually relies on recog- nition of hydrophobic spasms and other clinical features of furious rabies. Paralytic disease is rarely identified. Rabies has been misdiag- nosed as cerebral malaria, or even drug abuse. Management and prognosis The few human survivors of rabies encephalomyelitis had received vaccine and, with two exceptions, were left with severe neurological sequelae. Only one unvaccinated patient bitten by a bat in North America has made a good recovery. However, dog rabies virus in- fection remains fatal in man. Patients with furious rabies rarely live more than one week without intensive care but survival can be up to one month with paralytic disease. Infected neurons remain vi- able but dysfunctional, and no treatment has proved effective experimentally. Management—intensive care treatment may be appropriate for patients infected by a bat in the Americas if they present early or are already seropositive. Other patients with rabies should be sedated heavily and given adequate analgesia to relieve their pain and terror. Prevention Highly effective methods for control and prevention of rabies are available. Control of rabies in domestic dogs—99% of human rabies deaths could be prevented by controlling the transmission of dog rabies, but education and resources are lacking. Pre-exposure prophylaxis—a two or three-dose course of rabies vaccine is recommended for travellers and indigenous people in dog rabies endemic areas, but the cost is often prohibitive. Postexposure prophylaxis—at the time of a bite, correct cleaning of the wound and optimum postexposure immunization virtually elim- inate the risk of rabies. Effective prophylaxis demands urgent wound cleaning with copious amounts of soap and water, followed by vac- cine and rabies immunoglobulin. A new 2 visit, four-site intradermal postexposure vaccine regimen could increase the availability of af- fordable treatment. Epidemiology Rabies is a zoonosis of mammals that remains endemic in most parts of the world (Fig. 8.5.10.1). A cycle of infection is maintained in several reservoir species, of which the domestic dog is by far the most important. Many wild mammals including bats are also in- dependent rabies reservoirs (sylvatic infection) with identifiable strains of virus. Any mammalian species is potentially susceptible to rabies and may be a vector (e.g. a cat infected by a dog may then bite and infect a person). However, there is no persistent virus transmis- sion between cats. The animal source of human disease depends on the likelihood of contact with an infected species. Hence domestic dog rabies viruses are the source of 99% of human cases worldwide, mainly in Africa and Asia, but also in parts of South America. Rabies control programmes can reduce the risk of rabies in domestic ani- mals to such an extent that wild animals (e.g. insectivorous bats in the United States of America), become the principal vectors of infec- tion to humans. Rabies in wild mammals is usually spread by bites or by ingestion of infected prey. Rabies and rabies-related viruses The Lyssavirus genus currently includes the classic rabies virus, spe- cies 1, and 15 rabies-related species that are continent-specific in Europe,

Australasia, and Africa, and are, with one exception, zoo- noses of bats (see ‘Rabies-related viruses known to infect humans’). New unclassified lyssaviruses are now emerging. No rabies-related viruses have been found in the Americas and only there do bats have the classic rabies species. All terrestrial rabies reservoir mammal species (dogs and wildlife) carry classic rabies, except for the rare Mokola virus in Africa. Countries currently reported as rabies-free include Iceland, Cyprus, and most other Mediterranean islands, Singapore, Sabah, Sarawak, Antarctica, Oceania (including New Guinea and New Zealand), Hong Kong islands (but not the New Territories), Japan, South Korea, and Caribbean islands with the notable exceptions of Cuba, the Dominican Republic, Grenada, Haiti, and Trinidad and Tobago. The British Isles, together with other Western European countries, Scandinavia, and Australia have no rabies in terrestrial species, but do harbour rabies-related lyssaviruses in bats (Fig. 8.5.10.1). Lyssavirus-seropositive bats have been found in every country where surveillance has been carried out, so unusual con- tact with bats should be considered as a rabies risk anywhere. Inadvertent, usually illegal importation of infected mammals is a global risk. Cyclical epizootics of rabies may result from an uncontrolled in- crease in the population of the key reservoir species, such as the fox epizootic in Europe in the late 20th century. This started in Poland and spread across France, but it has now been eliminated from

 Western Europe. Outbreaks in dogs have followed social unrest and the movement of refugees. Although the fox is one of the species most susceptible to rabies, about 3% of animals survive the infection and become immune. Seropositive bats are not uncommon, and rabies antibody has been found in several other species, exceptionally even in dogs. There is no evidence that animals can become chronically infected or be in- fectious carriers, although an apparently healthy animal may be in- fectious during the prodromal stage of infection. Wild mammal reservoir species Wild mammal reservoir species vary in different areas. North America Reservoir species in the central United States of America and California are striped skunks Mephitis mephitis and, to a lesser ex- tent, spotted skunks Spilogale putorius; in Arizona and Texas grey foxes Urocyon cinereoargenteus and red foxes Vulpes vulpes; and in Alaska arctic foxes Alopex lagopus. However, in the east, rabies is most commonly found in raccoons Procyon lotor that transmit it to skunks and foxes. In North America many insectivorous bats are reservoirs of classic rabies virus, including big brown bats Eptesicus fuscus, Mexican free-tailed bats Tadarida brasiliensis mexicana, little brown bats Myotis lucifugus, and silver-haired bats Lasionycteris noctivagans whose virus is the main cause of human rabies infec- tions in the United States of America (see next) where bat infection has been found in every state. Latin America and the Caribbean Dog rabies persists in some urban areas of South America des- pite control programmes. The three species of true vampire bats Desmodus rotundus, Diaemus youngi, and Diphylla ecaudata (Desmodontinae) occur from sea level to over 3500 m but usu- ally under 1500 m only in Mexico, Central and South America, and some Caribbean islands (Fig. 8.5.10.2). The common vam- pire bat D. rotundus (Fig. 8.5.10.3) is the main reservoir of vampire bat rabies in Trinidad, Mexico, and Central and South America, where humans are occasionally bitten (Fig. 8.5.10.4). Carnivorous bats of the family Megadermatidae, such as the Indian ‘vampire’ Megaderma lyra, have given rise to the myth that vampires occur elsewhere. In Latin America, thousands of cattle are lost each year from vampire bat-transmitted paralytic rabies (derriengue) with locally serious economic consequences. Mongooses Herpestes auropunctatus are reservoirs of sylvatic ra- bies in Central America, Grenada, Puerto Rico, Cuba, Haiti, and the Dominican Republic.

Fig. 8.5.10.1 Global distribution of rabies and rabies-related lyssaviruses which infect humans. Red: Rabies in terrestrial mammal species (Lyssavirus classic rabies) and bat infections by other

lyssavirus species. Yellow: Terrestrial and bat rabies are all the classical species. Green: Bat lyssaviruses, Australian bat lyssavirus European bat lyssaviruses and Irkut, only. White: No lyssaviruses reported. Isla de Margarita Trinidad Fig. 8.5.10.2 Distribution of the three species of true vampire bats (Desmodontinae).

808 section 8 Infectious diseases Africa and Asia Dog rabies predominates but there is sylvatic rabies in Africa in foxes, wolves, jackals, and small carnivores of the families Mustelidae and Viverridae (e.g. the yellow mongoose Cynictis penicillata in South Africa), and in Asia in wolves, jackals, ferret-badgers Melogale moschata in China and Taiwan, and palm civets Paradoxurus hermaphroditus in Indonesia. Europe Foxes, wolves, raccoon dogs Nyctereutes procyonoides, and insect- ivorous bats are infected (see also 'Rabies-related viruses known to infect humans'). Rodents There are reports of rabies virus being isolated from wild rodents in many countries, especially in China. They are unlikely to be a reservoir species. There are very rare Chinese reports of rodent- transmitted human rabies diagnosed clinically. Monkeys Monkey bites are very common in tourists, especially to Asia. Rabies has been reported in nonhuman primates but they are not reser- voirs of infection except for marmosets Siamiri sciuerus in Brazil, which have caused human cases. A variety of other species have been reported to be infected in South America, Africa, and Asia, but transmission to humans has very rarely been clinically diagnosed only in India and Sri Lanka. However, the risk of rabies should al- ways be considered, especially if an animal is behaving abnormally or the bite is severe. Incidence of human rabies The true incidence of human rabies throughout the world is not re- flected in official figures; 59 000 deaths annually have been estimated to occur in Asia and Africa, including about 20 000 in India alone. High mortalities also occur in Bangladesh and Pakistan, and the in- cidence has been rising in China. Surveillance is minimal, especially in Africa. The World Health Organization (WHO) no longer issues country-specific data. In Latin America, the risk of canine rabies persists in Brazil, Bolivia, Dominican Republic, Peru, Guatemala, and Haiti. Vampire bat rabies has been reported recently in Peru, Ecuador, Mexico, and Brazil. In the United States of America there are on average two human deaths annually. Among 24 indigenous infections occurring in 10 years, 21 (88%) were caused by insect- ivorous bats. Europe reported 68 deaths in the last 15 years, mainly from the Russian Federation, Ukraine, and Georgia. Rabies was ap- parently eliminated from the United Kingdom by 1903, but since 2000 there have been five imported cases and one indigenous human European bat lyssavirus infection. Virology The Rhabdoviridae are a family of more than 100 bullet-shaped RNA viruses found in vertebrates, insects, and plants (Fig. 8.5.10.5). Two genera infect animals, Vesiculovirus and Lyssavirus. Vesicular stomatitis virus is a vesiculovirus of cattle and horses, which oc- casionally causes an influenza-like illness in farmers or laboratory workers. The genus Lyssavirus contains rabies and rabies-related viruses. The rabies virion is approximately 180 × 75 nm. Its core is a single spiral strand of negative nonsegmented RNA associated with a nucleoprotein, a phosphoprotein, and an RNA polymerase to form a helical ribonucleoprotein complex. This is enveloped in a matrix protein, host cell-derived lipid, and a coat of protruding glycopro- tein (G) molecules bearing spikes or knobs 10 nm long. The compos- ition of the glycoprotein determines viral virulence. The virus is readily inactivated by ultraviolet light, drying, boiling, most organic lipid solvents including at least 45% ethanol, soap solu- tion, detergents, hypochlorite, and glutaraldehyde solutions. Fig. 8.5.10.3 Desmodus rotundus common vampire bat (Peru). Courtesy of Dr Vargas Meneses, Lima, Peru. Fig. 8.5.10.4 A typical puncture wound inflicted by vampire bat, Madre de Dios, Peru. Courtesy of Dr Vargas Meneses, Lima, Peru.

8.5.10 Rhabdoviruses 809 Genetic sequencing techniques allow the identification of diverse strains of rabies and rabies-related viruses from different geograph- ical areas and vector species. Transmission Virus can penetrate broken skin and intact mucosae. Humans are usually infected when virus-laden saliva is inoculated through the skin by the bite of a rabid dog or other mammal (Fig. 8.5.10.6a, b). Saliva from a rabid animal can infect if the skin is already broken (e.g. by the animal's claws). In North America, contact with bats leading to rabies has passed unnoticed; only 39% of patients reported a bat bite and 34% had no history of exposure to bats. Animals can be infected through the gastrointestinal tract, but there is no evi- dence that this happens in humans. Inhalation of aerosolized virus created by infected nasal secretions of bats may be a method of transmission among cave-dwelling bats. In Texas, two men died of rabies after visiting caves inhabited by millions of Mexican free-tailed bats Tadarida brasiliensis mexicana, some of which were rabid; however, fleeting bat contact is more likely to have caused the infection. Two laboratory workers in the United States of America developed rabies after inhaling aerosol- ized fixed strains of rabies virus during the preparation of vaccines. The accidental use of vaccine in which the virus was not inactivated has led to fixed virus rabies (rage de laboratoire), for example, in Fortaleza, Brazil in 1960. Transmission of rabies between people has been proved in 16 cases of tissue transplantation from donors who had died of un- diagnosed neurological diseases. Six recipients of infected corneal grafts developed retro-orbital headache on the side of the graft 22–39 days after transplantation and died soon afterwards (other infec- tions spread by corneal grafts include Creutzfeldt–Jakob disease and cryptococcosis). In the United States and Germany, eight recipients of kidney, liver, lung, pancreas, or even just a segment of iliac artery, developed rabies encephalitis. Rabies was not suspected in the three donors despite high risk histories of rough travel in India, a bat bite, or raccoon hunting. Recreational drug abuse was detected in two. Rabies developed 18 months after receiving the raccoon rabies in- fected kidney graft, but 3 other recipients of the donor's organs were unaffected. Transplant related deaths also occurred in China and the Middle East. Postexposure prophylaxis following corneal trans- plants from infected donors has been successful. Considering that the saliva, respiratory secretions, and tears of ra- bies patients contain virus, it is surprising that there is no documented case of the disease being spread to intimate relatives and nurses. Transplacental infection has been observed in animals but has only been reported once in humans. Several women with rabies en- cephalitis have given birth to healthy babies. The transmission of rabies from mother to suckling infant via the breast milk has been suspected in at least one human case and is well known in animals. Pathogenesis The mechanism by which the highly neurotropic rabies virus en- ters the nervous system and travels into the brain and out again to Fig. 8.5.10.5 Rhabdoviruses. Virion of rabies virus. (Note the surface projections composed of glycoprotein (G). The marker line is 100 nanometres long) (a) (b)
Fig. 8.5.10.6 Bites inflicted by rabid dogs in (a) Nigeria, and (b) Thailand. These wounds carry a high risk of rabies with a short incubation period. Copyright D. A. Warrell.

810 section 8 Infectious diseases many organs is intriguing. The virus may replicate locally in muscle cells or attach directly to nerve endings. It can bind to many types of receptors including the nicotinic acetylcholine receptors at motor endplates, which may concentrate the virus at this postsynaptic site before entry into the presynaptic axon terminal, possibly via the neural cell adhesion molecule. Several other neuronal binding mechanisms may be involved. Once inside peripheral nerves, virus travels in a strictly retrograde direction up the axon. The dynein mo- lecular motor transports the virus within vesicles towards the cell soma, where replication begins. How the virus components move along the dendrite to emerge at the next synapse as complete

virions is unknown. Rabies virus is experimentally inaccessible to serum antibody while concealed in the peripheral nerves. On reaching the central nervous system, the virus replicates mas- sively within neurons and continues to be transmitted cell to cell trans-synaptically. Dramatic symptoms can appear before histo- pathological changes are apparent. Viral virulence is inversely re- lated to neuronal apoptosis. Rabies alters host cell gene expression, but the mechanisms of gross neuronal dysfunction are speculative. Centrifugal spread of virus from the central nervous system, appar- ently in somatic and autonomic efferent nerves, deposits virus in many tissues including skeletal and cardiac muscle, adrenal medulla where infection may be clinically significant, and also in kidney, retina, cornea, pancreas, taste buds, respiratory tract, and the skin in nerve twiglets around hair follicles (see ‘Laboratory diagnosis’). At this stage, productive viral replication occurs, with budding from outer cell membranes in the salivary glands, and then rabies may be transmitted by bites to other mammals. Viraemia has been detected very rarely, only in animals, and is not thought to be involved in pathogenesis or spread. Immunology Immunological response to rabies infection in humans Some patients die without any detectable immune response, because rabies virus evades and suppresses the immune system. Antibody might become detectable in serum seven days or more after the onset of illness and in cerebrospinal fluid a little later. It may rise to high levels in patients whose lives are prolonged by intensive care. A small amount of rabies-specific IgM is sometimes detectable, but is not useful as a means of diagnosis. There is little evidence of a lymphocyte-mediated immune re- sponse to rabies encephalitis. A pleocytosis appears in only 60% of patients, with a mean leucocyte count of 75 × 103/mm. Peripheral blood lymphocyte transformation is minimal if any. Experimentally, in fatal rabies there is inhibition of innate immunity, particularly interferon activity, and the few immune lymphocytes entering the brain undergo apoptosis, whereas survival is associated with in- creased permeability of the blood–brain barrier, neutralizing anti- body in the brain, expression of rabies glycoprotein, and apoptosis of infected neurons. Also in animals, latent infections can be reacti- vated by corticosteroids and stress. This provides a possible explan- ation for occasional reports of long incubation periods Immunological response to rabies vaccination The viral glycoprotein induces neutralizing antibody, which is de- tectable from 2 weeks after the start of primary immunization. In animal studies, the neutralizing antibody titre is the best available measure of protection against death. A titre of 0.5 IU/ml indicates specific seroconversion and is the WHO minimum acceptable level after vaccination. A relatively low response occurs in about 3% of the population, in immunosuppressed patients, and in older people. The nucleoprotein antigens also stimulate antibody that is cross-reactive between lyssaviruses, whereas glycoprotein antibody is more strain-specific. Transient low levels of interferon may be induced after the first dose of rabies vaccine. Although neutralizing antibody is un- doubtedly protective in the early stages after inoculation of virus, it may be deleterious once central nervous system infection is estab- lished. In animals, acceleration of the terminal phase of the enceph- alitis (‘early death phenomenon’) is associated with the presence of low titres of rabies antibody in serum. Rabies in animals All warm-blooded animals can be infected with rabies, but their sus- ceptibility varies. However, only mammals are infected naturally. In dogs, the incubation period ranges from 5 days to 14 months, but is usually between 3 and 12 weeks. The first symptom, as in many humans, is intense irritation at the site of the infection. Despite the popular idea of the ‘mad’ rabid dog, probably only a minority de- velop furious rabies. There is an early and striking change in the dog’s behaviour with dysphagia, ptosis, altered bark, paralysis of the jaw, neck, and hind limbs (Fig. 8.5.10.7), hypersalivation, congested conjunctivae, pruritus, shivering, trembling, snapping at imaginary objects, pica, and extreme restlessness causing the animal to wander miles from home. Dogs with furious rabies attack

inanimate objects, often seriously injuring their mouths in the process. The virus has Fig. 8.5.10.7 Dog with paralytic rabies showing paralysis of the limbs and hypersalivation. Copyright D. A. Warrell.

8.5.10 Rhabdoviruses 811 been found in the saliva 3 days before symptoms appear, and the animal usually dies within the next 7 days. This is the basis for the traditional 10-day observation period for dogs that have bitten humans. Very rare old reports from India, Ethiopia, and Nigeria of persistent or intermittent excretion of virus in the saliva of apparently healthy dogs have not been confirmed by subsequent thorough searches. 'Oulou fato', a clinical variant of canine rabies with reduced virulence, was seen in West Africa 50 years ago. Rabid foxes lose their fear of humans and the majority develop paralytic rabies. An extreme degree of furious rabies is seen in 75% of infected cats. Cattle usually develop paralytic symptoms with dys- phagia, hypersalivation, groaning, trembling, colic, diarrhoea, ten- esmus, and rectal prolapse. Most other domestic ungulates develop paralytic symptoms. Horses often show furious features with sexual excitement. Most wild animals, like foxes, lose their fear of humans and may appear tame. Rabid skunks, raccoons, badgers, martens, and mongooses may become very aggressive. Dysphagia and in- ability to drink is common in rabid animals, but they do not exhibit hydrophobia. Clinical features in humans The incubation period ranges from 4 days to many years, but it is be- tween 20 and 90 days in three-quarters of cases. It tends to be shorter after bites on the face (average 35 days) than after those on the limbs (average 52 days). Prodromal symptoms Often, the first symptom is itching, pain, or paraesthesia at the site of the healed bite wound (Fig. 8.5.10.8). Nonspecific prodromal symptoms include fever, chills, malaise, weakness, tiredness, head- ache, photophobia, myalgia, anxiety, depression, irritability, and symptoms of upper respiratory tract and gastrointestinal infections. Subsequently, symptoms of either furious or paralytic rabies will de- velop, depending on whether the spinal cord or brain are predom- inantly infected. Furious rabies Furious rabies is the more common presentation. Most patients have the diagnostic symptom of hydrophobia, which is a com- bination of inspiratory muscle spasm, associated with terror (Fig. 8.5.10.9a–f). Initially provoked by attempts to drink water, this reflex can be excited by a variety of stimuli including a draught of air ('aerophobia'), water splashed on the skin, irritation of the re- spiratory tract or, ultimately, by the sight, sound, or even mention of water. The inspiratory spasm is violent and jerky. The neck and back are extended, the arms thrown up, and the episode may end with the patient in a position of extreme extension—opisthotonos, having a generalized convulsion complicated by cardiac or a re- spiratory arrest. Patients experience hyperaesthesia and, at times, generalized arousal during which they become wild, hallucinated, fugitive, and sometimes aggressive (Fig. 8.5.10.10). This behaviour alternates with periods of mental lucidity during which patients may become distressingly aware of their predicament. Despite these dramatic symptoms, attributable to brainstem encephalitis, conventional neurological examination may prove to be completely normal, leading to the false assumption of conversion disorder. Reported abnormalities include meningism, cranial nerve lesions (especially III, VI, VII, IX–XII), upper motor neuron lesions, fasciculation, and involuntary movements. Disturbances of the hypothalamus or autonomic nervous system are reflected by hypersalivation (Fig. 8.5.10.11a, b), sweating, lacrimation, hypertension or hypoten- sion, hyperthermia or hypothermia, diabetes insipidus or inappro- priate secretion of antidiuretic hormone, and rarely, priapism with spontaneous orgasms, satyriasis, or nymphomania. Hypersexuality suggests similar aetiology to the Klüver–Bucy syndrome created in rhesus monkeys by bilateral ablation of the hippocampus. Without supportive treatment, about one-third of the patients will die during a hydrophobic spasm

during the first few days. The rest lapse into coma and generalized flaccid paralysis, and rarely survive for more than a week without intensive care. Paralytic or dumb rabies This is the clinical pattern recognized in less than one-fifth of human cases except in the case of bat-transmitted rabies, especially vampire bat infection, which is usually paralytic. Patients may become liter- ally dumb (‘rage muette’) because their laryngeal muscles are para- lysed, but symptoms are quieter (‘rage tranquille’) than in furious rabies. The largest reported outbreak was in Trinidad between 1929 and 1936 when there were 53 human cases, initially misattributed to poliomyelitis or botulism; others have been described from Mexico, Guyana, Brazil, Peru, Ecuador, Bolivia, and Argentina. The paralytic form of rabies was also seen in patients with postvaccinal rabies, in the two patients who inhaled fixed virus, and is said to be more likely to develop in patients who have received antirabies vaccine. After the same prodromal symptoms, especially fever, headache, and local par- aesthesias, flaccid paralysis develops, usually in the bitten limb, and ascends symmetrically or asymmetrically with pain and fasciculation in the affected muscles and mild sensory disturbances. Paraplegia and sphincter involvement then develop, and finally fatal paralysis of deglutitive and respiratory muscles (Fig. 8.5.10.12). Hydrophobia is unusual, but may be represented by a few pharyngeal spasms in the terminal phase of the illness. Even without intensive care, patients with paralytic rabies have survived for up to 30 days. Other manifestations and complications Respiratory system Asphyxiation and respiratory arrest can complicate the hydrophobic spasms or generalized convulsions of furious Fig. 8.5.10.8 This man developed intense itching in the right leg, provoking scratching and excoriation, 8 weeks after being bitten in that limb by a rabid dog. He died with furious rabies a few days later. Copyright David A. Warrell.

812 section 8 Infectious diseases rabies and the bulbar and respiratory paralysis of dumb rabies. Bronchopneumonia is a predictable complication if life is pro- longed by intensive care, but a primary rabies pneumonitis may occur. Various abnormal patterns of respiration have been de- scribed, including cluster and apneustic breathing. There are some similarities to palatal myoclonus. Pneumothorax may complicate inspiratory spasms. Cardiovascular system A variety of dangerous cardiac arrhythmias have been reported, including supraventricular tachycardias, sinus bradycardia, atrioventricular block, and sinus arrest, together with T wave and ST segment changes (Fig. 8.5.10.13). Hypotension, pulmonary oedema, and congestive cardiac failure are attributable to myocarditis. Nervous system Raised intracranial pressure resulting from cerebral oedema or in- ternal hydrocephalus has been reported in a few cases, but spinal fluid opening pressure is usually normal and papilloedema is rarely seen. There is clinical and electrophysiological evidence of diffuse axonal neuropathy, consistent with histological appearances of de- generation of peripheral nerve ganglia and axons. (b) (a) (c) (d) (e) (f) Fig. 8.5.10.9 Hydrophobic spasms (a–e) in a 14-year-old Nigerian boy with furious rabies. Note the violent contraction of inspiratory muscles (sternomastoids and diaphragm) depressing the xiphisternum. (f) In a Thai man who had just asked to drink water. Copyright D. A. Warrell.

8.5.10 Rhabdoviruses 813 Gastrointestinal system ‘Stress’ ulcers and the Mallory–Weiss syndrome are possible explan- ations for the haematemesis often reported in rabies. Clinical and differential diagnosis Rabies should be suspected in any patient who develops neuro- logical symptoms after being bitten by a mammal in a rabies en- demic area. However, some patients fail to remember that they have been bitten and others may be infected while they are asleep possibly by contact with lip mucosae (North American insectiv- orous bats) or near-painless bites by vampire bats in parts of Latin America. Furious rabies Pathognomonic inspiratory spasms with associated emotional

re- sponse are provoked by asking the patient to swallow accumulated saliva or by directing a draught of air on to the face. • Psychiatric conditions: Rabies encephalitis has been misdiag- nosed as a variety of psychiatric conditions, including conversion disorder and behavioural disturbances attributed to recreational drugs. Conversely, patients with a morbid fear of rabies (rabies phobia, lyssaphobia, pseudohydrophobia) may simulate the more melodramatic features of the disease but hydrophobia is unlikely to be mimicked accurately, the incubation period after the bite (hours or a few days) is usually much too short for rabies enceph- alitis, and the prognosis is, of course, excellent. • Otolaryngological conditions: Pharyngeal and upper airway symptoms of hydrophobia may be misinterpreted as pharyngitis or laryngitis so that the patient is referred to an otolaryngologist. • Tetanus: This can also follow an animal bite and is similar to ra- bies in some respects, especially the pharyngeal form of cephalic tetanus (‘hydrophobic tetanus’). It is distinguished by its shorter incubation period (usually less than 15 days in severe tetanus),
Fig. 8.5.10.10 Episode of intense arousal in a Nigerian patient with furious rabies. Copyright D. A. Warrell. (a) (b) Fig. 8.5.10.11 Autonomic overactivity in rabies encephalomyelitis. (a) Sweating and hypersalivation. (b) Salivation and lacrimation. Copyright D. A. Warrell. Fig. 8.5.10.12 Paralytic rabies. Copyright D. A. Warrell.

 the presence of trismus, the persistence of muscle rigidity between spasms, the absence of meningoencephalitis (cerebrospinal fluid is universally normal), and the better prognosis. • Other encephalopathies/encephalitides: The typical encephalitic progression from severe headache to continuous coma is un- usual in furious rabies. Hydrophobia with intermittent excita- tion and lucid intervals of full consciousness does not occur in other encephalitides. Among children with suspected cerebral malaria in Malawi, some were proved by biopsy to have died of rabies. Toxic encephalopathies: Delirium tremens, some drugs (pheno- thiazines, amphetamines, modafinil, cocaine, and other recre- ational drugs), and plant poisonings (e.g. thorn apple, Datura stramonium) can cause excitable and aggressive behaviour that might be confused with rabies. Paralytic rabies Other causes of ascending (Landry-type) paralysis may enter the differential diagnosis. • Postvaccinal encephalomyelitis (see next): This usually develops within 2 weeks of the first dose of the now rarely used nervous tissue rabies vaccines. • Poliomyelitis: Objective sensory disturbances are absent, and fever rarely persists after paralysis has developed. • Acute inflammatory polyneuropathy (Guillain–Barré syn- drome): Cerebrospinal fluid examination will help to distinguish this condition. • Cercopithecine herpesvirus (B virus) encephalomyelitis: Bites and other types of contact with Asian macaque monkeys (genus Macaca), especially rhesus (M. mulatta) and cynomolgus (M. fas- cicularis) transmit this dangerous infection. The incubation period (3–4 days) is usually shorter than in rabies and symptoms develop within 1 month of contact. Vesicles may be found in the monkey’s mouth and at the site of the bite, and the diagnosis can be confirmed virologically. Pathology The brain, spinal cord, and peripheral nerves show ganglion cell de- generation, perineural, and perivascular mononuclear cell infiltra- tion, neuronophagia, and glial nodules. Inflammatory changes are most marked in the midbrain and medulla in furious rabies and in the spinal cord in paralytic rabies. Negri bodies (Fig. 8.5.10.14), eosinophilic intracytoplasmic in- clusions, function as viral factories containing rabies RNAs and translated proteins. They can be demonstrated by haematoxylin and eosin stains in histological sections of grey matter in up to 75% of human cases, especially in hippocampal pyramidal cells and cere- bellar Purkinje cells. In view of the appalling prognosis of rabies encephalitis, neuro nolysis is often surprisingly mild and patchy, and death can occur without any inflammatory response. Vascular lesions such as throm- bosis and haemorrhage have also been described. The

brainstem, limbic system, and hypothalamus appear to be most severely affected and, in paralytic disease, the spinal cord and medulla. Outside the nervous system, there is focal degeneration of salivary and lacrimal glands, pancreas, adrenal medulla, and lymph nodes. An interstitial myocarditis with round cell infiltration is found in about 25% of cases. Laboratory diagnosis If a mammal suspected of being rabid has bitten, scratched, or other- wise may have infected a person, it should be killed, and its brain examined without delay. The best way to detect rabies antigen in Fig. 8.5.10.13 Electrocardiogram in a Nigerian patient with furious rabies showing sinus tachycardia, atrial and ventricular premature beats, and a wandering atrial pacemaker. Copyright D. A. Warrell. Fig. 8.5.10.14 Street virus in human cerebellar Purkinje cells as seen with the light microscope. Several Negri bodies can be seen (one is arrowed). Magnification × 615. Courtesy of Armed Forces Institute of Pathology 73–12 330.

8.5.10 Rhabdoviruses 815 acetone-fixed brain impression smears is by the direct immuno-fluorescent antibody (IFA) test. Alternatively, if no fluorescent microscope is available, rapid enzyme immunodiagnosis can be used. Virus isolation takes about 4 days in cell culture. In humans, rabies can be confirmed early in the illness by dem- onstration of viral antigen by the direct IFA test in frozen sections of full-thickness skin biopsies taken from a hairy area, usually the nape of the neck. Specific diagnostic staining is seen in nerve twiglets around the base of hair follicles (Fig. 8.5.10.15). This rapid method is positive in 60–100% of cases, and no false-positive results have been reported. Antigen can also be found in brain biopsies, but tests on corneal impression smears are very unreliable. Reverse transcription polymerase chain reaction (RT-PCR) is now used to detect rabies in saliva, skin biopsy material, and occasionally cerebrospinal fluid. During the first week of illness, virus may be detected in saliva, brain, cerebrospinal fluid, and very rarely urine. Rabies antibodies are not usually detectable in serum or cerebrospinal fluid before the eighth day of illness in unvaccinated patients. The IFT antibody test can cross-react with other viruses, so low levels alone are not diagnostic. Serum antibody may leak into the cerebrospinal fluid in patients with postvaccinal encephalomyelitis, but a very high titre suggests a diag- nosis of rabies. A specific IgM test has not proved useful diagnostically. Prognosis There is no specific antirabies therapeutic agent. Rabies was formerly regarded as a universally fatal disease, but there are reports of 10 cases of recovery or prolonged survival following intensive care. The diagnoses were made serologically except in three cases where virus was identified by PCR. Nine patients who had received some vaccine before the onset of symptoms, survived months or years with inten- sive care. However, seven had profound neurological impairment but two recovered. A boy in Ohio USA infected by an insectivorous bat in 1970, had delayed treatment with duck embryo vaccine and completely recovered after intensive care therapy. Also, a Turkish man who had one dose of vaccine 4 days after a dog bite, was said to have recovered completely from rabies encephalitis, according to a brief report. Neutralizing antibody was present on the second day in hospital and he recovered spontaneously without intensive care. The first unvaccinated patient to recover from rabies has re- turned to near normal life following intensive care and antiviral therapy. She was bitten by a bat in Wisconsin in 2004, had no ra- bies prophylaxis, and developed typical encephalitis without hydro- phobia. Rabies neutralizing antibody was detected on the sixth day of illness. Treatment comprised coma induction and antiviral drugs. She made a slow recovery over 5 years and has returned to normal life, although with minor neurological deficits. The antiviral treatments have not proved effective against rabies experimentally; however, she developed antibody at an early stage of the disease. Her treatment possibly maintained her vital functions until her immune response eliminated the virus, probably with loss or mal- function of

infected neurons. In animal experiments, American bat rabies virus infection differs from that of canine virus in that it is slower to evolve and progress, virus replication is not restricted to neurons, and histopathological changes are milder. This suggests that the virus may be less pathogenic and may also explain the re- covery of the boy in Ohio bitten by a bat. It is likely that he too had rabies antibody present at an early stage of illness. Treatment No treatment has yet proved effective in animal models. Antiserum, antiviral agents, interferon-α, corticosteroid, and other immuno- suppressants have proved useless. Human rabies of canine origin remains 100% fatal in unvaccinated patients. The 'Milwaukee' treatment protocol used in Wisconsin has since been used unsuc- cessfully in 30 other patients with rabies encephalitis who were infected by bats or dogs. There is no evidence that it is superior to supportive intensive care. Until a new treatment is proved effective experimentally, palliation of the patient's symptoms and immunization of contacts is recommended. Patients must be sedated heavily and given adequate analgesia to relieve their pain and terror. Intensive treatment may be appropriate for patients infected by an American bat, who present early, and are already seroposi- tive. Intensive care is inappropriate for canine virus infection, especially in developing countries, and the cost is prohibitive. If intensive care is undertaken, the aim is to prevent complications such as cardiac arrhythmias, cardiac and respiratory failure, raised intracranial pressure, convulsions, fluid and electrolyte disturb- ances, including diabetes insipidus and inappropriate secretion of antidiuretic hormone, and hyperpyrexia. A future treatment could be intrathecal live attenuated rabies virus to induce intracerebral neutralizing antibody to eliminate infection. Fig. 8.5.10.15 Diagnosis of human rabies during life. Vertical section through a hair follicle and shaft showing fluorescence of nerve cells around the follicle indicating the presence of rabies antigen. Magnification × 250. Copyright M. J. Warrell.

816 section 8 Infectious diseases Control of rabies in animals The elimination of dog rabies would reduce the human mortality by over 99% and drastically reduce the need for human vaccin- ation. Rabies control has been achieved most effectively where the principal reservoir is the domestic dog, as in 19th-century United Kingdom, Malaysia, and Japan, and since then in other areas including Western Europe, Taiwan, North America, and parts of urban Latin America. In countries where rabies is enzootic The control strategy depends on the local pattern of rabies oc- currence in wild and domestic animals. Education and publicity about rabies is always needed. Domestic animals can be protected by regular vaccination. Owned dogs can be muzzled or kept off the streets. People should be discouraged from keeping wild car- nivores such as skunks, raccoons, coatis, and mongooses as pets. Unnecessary contact with mammals should be avoided (e.g. stroking stray dogs or apparently friendly wild animals, exploring bat-infested caves). Culling reservoir species has proved an un- popular and ineffective method of long-term control. Impressive reduction of urban rabies in stray dogs has proved possible in India by vaccination, population control, and reducing available food and shelter by removing refuse. Effective oral vaccination of dogs is not yet practicable. Control of sylvatic rabies has been achieved by vaccination of key wild animal reservoir populations with live oral vaccines dis- tributed in bait. Repeated campaigns distributing attenuated rabies vaccine have eliminated fox rabies in Western Europe, and vaccinia- recombinant vaccine expressing rabies glycoprotein has been used in North American coyotes, foxes, and raccoons. New vaccines are being developed for other species. Vaccination of bats is unlikely to be feasible. Vampire bat rabies is controlled by destroying roosts and poisoning the bats with anticoagulants. In countries where rabies is not endemic The inadvertent importation of a mammal incubating rabies is a universal risk. The movement of potential vectors, especially do- mestic dogs and cats, wild carnivores, and bats, should be strictly

controlled. Serological evidence of successful vaccination should be provided for imported mammals, or they should be vaccinated on arrival and quarantined. Prevention of human infection Pre-exposure prophylaxis Pre-exposure vaccination is the most effective form of rabies pre- vention. No rabies deaths have been reported in anyone who had had pre-exposure vaccine and then postexposure booster doses. It is recommended for people who handle imported animals, workers in zoos and rabies laboratories, and those who are resi- dent in or intend to travel to dog rabies-endemic areas, especially children. Others particularly at risk in certain areas include veter- inarians, dog catchers, farm workers, cave explorers, naturalists, and animal collectors. In dog rabies-endemic areas, pre-exposure prophylaxis is advisable but is rarely used. Travellers should be educated to seek immediate local medical help if they are bitten, scratched, or licked by mammals. However, recommendations vary in different areas and local advice may be unreliable. Tissue culture vaccine and especially rabies immune globulin may not be readily available. Primary pre-exposure vaccine course A course of three doses of tissue culture rabies vaccine (see next) is given intramuscularly (IM) into the deltoid, or the anterolateral thigh in children, on days 0, 7, and 21–28. The 2018 WHO recommenda- tions include a 2 dose IM regimen, on days 0 and 7. An effective economical alternative is intradermal (ID) injections of 0.1 ml, 3 doses at the same intervals, but the latest recommendations give a 2-site, one week ID alternative regimen on days 0 and 7. If the ID injection is too deep to produce a papule, withdraw the needle and repeat the procedure. Rabies vaccines do not contain preservatives. Strict aseptic precautions are mandatory to avoid contamination. One vaccine ampoule can be shared but must be used within a day or discarded. If chloroquine is being taken for malaria prophylaxis (unlikely today), or in other cases of suspected immunosuppression, the intramuscular route must be used. Many travellers cannot afford three doses of an expensive vaccine, so the economical intradermal route is ideal for family, student, or other groups who can be vaccin- ated on the same day. Pre-exposure booster doses A booster dose 1–2 years after the primary course enhances and prolongs the presence of antibody. Although the titre falls more rapidly after intradermal than intramuscular inoculation, the re- sponse to a booster dose is still prompt. Confirmation of serocon- version is recommended only if immunosuppression is suspected. Further booster doses for those at higher risk may be given ID or IM at intervals of 2–10 years. Boosters are not necessary if the ra- bies neutralizing antibody level is at least 0.5 IU/ml. Laboratory staff at high risk should have more frequent serology tests. Travellers who will have rapid access to vaccine if exposed need not have further immunization, but if medical resources will be unreliable, a booster vaccination should be given before departure if 5 years have elapsed since the previous dose. A personal record of immunization must be kept, and urgent treatment is essential after possible exposure. Lyophilized rabies vaccine is relatively stable even at tropical ambient temperatures. It is sensible to take a dose on expeditions to remote rabies endemic areas. An extra emergency injection can then be given immediately after a risky encounter with an animal. If more than one person is exposed, the ampoule can be shared by giving multiple ID doses to each, using the whole dose (see Postexposure prophylaxis', next). This does not replace the normal postexposure treatment, which must still be given as soon as possible. Postexposure prophylaxis Despite intensive care, rabies encephalomyelitis of canine origin re- mains 100% fatal in unvaccinated patients. At the time of the bite, however, correct cleaning of the wound (see next) and optimum postexposure immunization reduce the risk of rabies to nearly zero compared to about 35–57% for untreated bites by proven rabid ani- mals. The risk varies with the biting species and the site and severity of the bites. It is highest following bites to the head by proven rabid

8.5.10 Rhabdoviruses 817 wolves, which carries a case fatality exceeding 80% in unvaccinated people. The decision to give postexposure treatment depends on an assessment of the risk of infection. Intact skin is a barrier against the virus. Ask about the precise geographical location of the exposure; its severity, whether it was a bite or lick on broken skin; the site of the lesion; and the nature, appearance, behaviour, and fate of the biting animal, and whether it had been recently vaccinated against rabies (Box 8.5.10.1). The animal's brain must be tested for rabies if possible. If there is any doubt, the patient should be given full postexposure prophylaxis, even if the bite is several months old. If the exposure was more than a year previously no rabies immuno- globulin is needed. The aim of prophylaxis is to neutralize inoculated virus before it can enter the nervous system. Wound cleaning and active and passive immunization must be implemented as soon as possible. Wound cleaning This is effective in killing virus in superficial wounds, but is often neglected. First aid includes vigorous cleaning of the wound with soap or detergent and water under a running tap for at least 5 min. Foreign material should be removed and a viricidal agent such as povidone iodine, or 40 to 70% alcohol, should be applied liber- ally. Quaternary ammonium compounds such as benzalkonium chloride are inactivated by soap and so are not recommended. Hospital treatment of wounds involves thorough exploration, debridement, and irrigation of deep lesions, if necessary under local or general anaesthetic. Suturing should be avoided or de- layed, and the wound left without occlusive dressings. Attention should be given to tetanus prophylaxis and the large range of viral, bacterial, and fungal pathogens particularly associated with mammal bites. These include Cercopithecine herpesvirus (B virus) from Asian macaques (Chapter 8.5.2); Pasteurella multo- cida (Chapter 8.6.18), Francisella tularensis (Chapter 8.6.19), Streptobacillus moniliformis, and Spirillum minus (Chapter 8.6.13) from rodents; and Pasteurella multocida, Capnocytophaga cani- morsus, and Bartonella henselae (Chapter 8.6.42) from dogs and/ or cats. Most of the bacteria are sensitive to amoxicillin/clavulanic acid, cefoxitin, or tetracycline. Active immunization Rabies vaccines Three highly immunogenic tissue culture vaccines that meet the WHO recommended standards are purified chick embryo cell (PCEC) vaccine, purified Vero cell rabies vaccine (PVRV), and human diploid cell vaccine. Several tissue culture vaccines are produced, mainly for national use, in China, India, Japan, and Russia. Obsolete nervous tissue rabies vaccines are not sanctioned by the WHO, but suckling mouse brain (Fuenzalida) vaccine is still used in a few countries in South America and in Algeria. Daily subcuta- neous doses for 7–14 days, followed by booster doses, are usually given over the abdominal wall. Neurological reactions including postvaccinal encephalomyelitis could still occur. Postexposure tissue culture vaccine regimens The IM Essen regimen has become 4 × 1-ml (PVRV 0.5 ml) doses injected into the deltoid (or anterolateral thigh in children) on days 0, 3, 7 and 14–28. There is no change to the alternative 2-1-1 IM regimen, of two full doses injected into the deltoids on day 0, and one dose on days 7 and 21. The intramuscular regimens are unaffordable in many countries. However, economical multisite ID methods are available, requiring less vaccine than the IM regimens. Each of the intradermal injection sites drains to a different group of lymph nodes, intended to stimu- late more lymphoid tissue to produce antibody. Aseptic precautions are required as for pre-exposure ID treatment. The simplified four-site ID regimen replaces the eight-site ID regimen. It consists of a whole ampoule of vaccine divided be- tween four intradermal injections over the deltoid and the thigh or suprascapular areas. The volume per site is about 0.1 ml for PVRV and the equivalent dose for vaccines containing 1 ml per ampoule is 0.2 ml. On day 7, two intradermal injections of 0.1/0.2 ml in the deltoid areas are followed by a single intradermal dose on day 28. Since giving half the dose, 0.1 ml of PCECV (1 ml/ampoule) was found to be immunogenic in trial conditions, there is a wide safety margin in case of inexperience with ID injection technique or for immunosupressed

patients. Without sharing ampoules, a maximum of three doses are needed. The two-site ID regimen was designed for use with PVRV. A dose of 0.1 ml for PVRV, or 0.2 ml for vaccines formulated in vials con- taining 1 ml, is given ID at two sites in the deltoid area on days 0, 3, 7 and 28. An ID dose of 0.1 ml per site is usually used with PCEC 1 ml vaccine but higher-potency vaccines are demanded by most coun- tries using this lower dose. Omitting the day 28 dose is now permitted by the WHO. The 2-site ID regimen becomes 3 visits in one week, and vaccine vials can be shared on each occasion. The 4-site ID regimen becomes 2 visits: 4-site ID using a whole vial on day 0 and 2-site using half a vial on day 7. For possibly im- munosuppressed patients, a day 28 single site ID booster dose is still advisable. For all other vaccines, the manufacturer's instructions should be followed. Box 8.5.10.1 Specific postexposure prophylaxis for use in a rabies endemic areaa following contact with a domestic or wild rabies vector species, whether or not the animal is available for observation or diagnostic tests Minor exposure (including minor scratches, or abrasions without bleeding) • Start vaccine immediately • Stop treatment if animal remains healthy for 10 days • Stop treatment if animal's brain proves negative for rabies by appro- priate laboratory tests Major exposure (including licks of broken skin or mucosa, minor bites on arms, trunk or legs, or major severe bites, i.e. multiple or on face, head, fingers, or neck) • Immediate rabies immune globulin and vaccine • Stop treatment if domestic cat or dog remains healthy for 10 days • Stop treatment if animal's brain proves negative for rabies by appro- priate laboratory tests a This scheme is a simplification of the recommendations of the World Health Organization Expert Consultation on Rabies (2018).

 Postexposure vaccine boosting regimen for people who have already received vaccination If a complete pre-exposure or postexposure course of a potent tissue culture vaccine has been given in the past, or if the neu- tralizing antibody level has been over 0.5 IU/ml, rabies immune globulin is not required and only two doses of tissue culture vac- cine are given IM on days 0 and 3. Alternatively, a the one day booster regimen is four 0.1 ml ID injections in deltoid and thigh areas. Vaccine should not be wasted, so if immediate sharing is not possible, a whole vial of vaccine is divided between four intra- dermal sites. Side effects of tissue culture vaccines Mild and transient local redness, itching (especially after ID injec- tion), or pain at the site of injection are not uncommon. Influenza-like symptoms and rashes are infrequent. Type I immediate hypersensi- tivity occurs rarely during primary courses. No fatal reactions have been reported. Very rarely neurological symptoms including poly- neuritis or Guillain–Barré syndrome, have been reported in patients receiving tissue culture vaccines but no more frequently than for other commonly used virus vaccines. Passive immunization: Rabies immune globulin Rabies immune globulin (RIG) has proved valuable in providing protection before neutralizing antibody has been actively gener- ated, presumably by neutralizing rabies virus during the first week after initial vaccination. It is recommended as part of primary postexposure treatment, but it is vital following severe bites (on the head, neck, hands, and multiple or deep bites) (see Box 8.5.10.1). The dose of human RIG is 20 IU/kg body weight and for equine RIG is 40 IU/kg. Reactions to equine and human RIG have been observed in 1.8% and 0.09% of recipients, respectively, and serum sickness in 0.72% and 0.007%, respectively. These are not predicted by previous ID hypersensitivity testing and so this must not be used. Adrenaline (epinephrine) should always be available in case of reactions. All the RIG is infiltrated into and around the bite wound if ana- tomically possible, but any remaining is injected intramuscularly preferably into the thigh, not the buttock, at a site distant from the vaccine. RIG should be given at the start of vaccination. If it is given hours or days before the first dose, the active immune response will be impaired but it can be given up to 7 days after the first vaccine dose. RIG is prohibitively expensive and is not avail- able or affordable for 99% of

people in developing countries for whom postexposure treatment is indicated. If supplies of RIG are limited or if it is unaffordable, wound infiltration alone may be given. Failures of postexposure prophylaxis Deaths from rabies have occurred despite prophylaxis. Failures are attributable to delay in starting vaccination, incomplete vaccine course, use of a substandard vaccine or omission of RIG. Failure to wash the wound or infiltrate RIG around it, injection of vaccine into the buttock, or impaired immune responsiveness of the pa- tient may also contribute. Vaccine protection against rabies-related lyssaviruses may be less efficient than against the classic rabies spe- cies (see next), but no case of vaccine failure has been attributed to this phenomenon. Rabies-related virus infections of humans The genus Lyssavirus contains 12 species: type 1, classic rabies and 15 rabies-related species, but only 6 of these in three phylogroups are known to have infected man (Table 8.5.10.1). Rabies-related viruses occur in Africa, Europe, Asia and Australia. They are not found in the Americas. With the exception of Mokola, all are viruses of bats. Phylogroup I contains classic rabies and five rabies-related bat species which are known to cause rabies-like encephalitis in humans. In phylogroup II, Mokola virus of shrews causes a milder disease in man. European bat lyssaviruses have occasionally been detected in terrestrial species, but diagnostic tests are available only in specialized laboratories, infection is rarely suspected, and the routine tests for classic type 1 rabies virus may be weakly positive or negative. The true prevalence of lyssaviruses is unknown. Only 13 human cases of rabies-related virus infections have been reported, and disease is likely to remain unrecognized and misdiagnosed. African lyssaviruses • Mokola virus (species 3 phylogroup II) has been isolated from shrews (Crocidura spp.) and rodents, as well as cats and dogs Table 8.5.10.1 Lyssaviruses known to infect humans Species phylogroup I Virus Distribution Reservoir mammal species Human deaths 1 Rabies Almost worldwide Terrestrial mammal species and bats in
the Americas Unknown 60 000? 4 Duvenhage virus Africa Insectivorous bat 3 5 European bat lyssavirus type 1 Northern and Eastern Europe Insectivorous bat 2 6 European bat lyssavirus type 2 Western Europe and Scandinavia Insectivorous bat 2 7 Australian bat lyssavirus Australia Fruit bats and insectivorous bats 3 10 Irkut virus Eurasia Insectivorous bat 1 Phylogroup II 3 Mokola virus Africa Shrew, rodent 2?

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