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Veterinary Microbiology 114 (2006) 225–239
veterinary microbiology
www.elsevier.com/locate/vetmic

Suitability of canine herpesvirus as a vector for oral bait vaccination of foxes

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Received 22 April 2005; received in revised form 9 December 2005; accepted 13 December 2005

Abstract

Studies were conducted to evaluate the feasibility of using canine herpesvirus (CHV) as a vaccine vector for bait-delivered oral vaccination of wild foxes. To test the viability of CHV in baits, CHV was freeze-dried, incorporated into different baits, stored, and the remaining viral infectivity tested in cell culture after varying periods of time at different storage temperatures. Experimental baits (mouse carcasses) and commercial baits (FOXOFF and PROBAIT) were prepared with either liquid or freeze-dried CHV and tested in two fox trials for their capacity to induce CHV-specific antibodies following oral baiting. Freeze-drying and storage temperatures below 0 °C had a stabilizing effect to virus infectivity. When stored at −20 °C, freeze-dried CHV retained its full infectivity for up to 3 months in PROBAIT baits, the remaining infectivity in FOXOFF baits was 100-fold less. Oral baiting with CHV induced antiviral serum antibodies in all vaccinated foxes (20/20). None of the vaccinated foxes became ill or shed infectious virus into the environment although viral DNA was detected in body secretions as evaluated by PCR. The results indicate that CHV can be freeze-dried and stored over extended periods of time without loosing much of its infectivity. This is the first report of CHV being used for oral bait vaccination of foxes. It appears that CHV is well suited for use as a recombinant vector for wild canids.

Keywords: Fox; Vulpes; Canine herpesvirus; CHV; Canids; Vaccine; Recombinant; Bait; FOXOFF; PROBAIT

1. Introduction

The majority of veterinary antiviral vaccines currently in use for domestic dogs has been developed by the classical laboratory methods of virus attenuation or inactivation. These vaccines have proven to be safe and highly effective in combating many important viral diseases of domestic dogs such as distemper, parvovirosis or rabies (Carmichael, 1999). Rabies vaccines were successfully used also in wild canids, in particular foxes, coyotes and racoons, for which immunization via oral baits is the only practical, large-scale method (Woldehiwet, 2002). However, in addition to the well-known infectious epidemics, wildlife management is likely to face novel challenges in the future, for example, from newly emerging

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doi:10.1016/j.vetmic.2005.12.008

diseases such as severe acute respiratory syndrome among various wildlife species in China, Nippah virus epidemics in pigs in Malaysia or recent zoonotic avian influenza cases in South East Asia (Bengis et al., 2004; Daszak et al., 2001; Peiris et al., 2004; Audsley and Tannock, 2004). Development of vaccines against such emerging infections will be time-consuming and costly, especially when conventional methods such as virus attenuation or inactivation are applied.

Overabundance of certain wildlife species is another challenge for wildlife management. Antifertility vaccination (also known as immunocontraception) is currently being developed in an attempt to address problems associated with overabundant wildlife such as foxes in Australia (Reubel et al., 2005). Here, the overabundance of introduced European red foxes poses not only a major threat to the survival of endangered native fauna, but also considerably impacts on lamb production. Antifertility vaccination is particularly appealing for use in foxes because of its humane, non-lethal approach and the potential to reduce the size of pest animal populations by reducing recruitment rather than increasing mortality (for review see (Ferro, 2002)). Research on antifertility vaccines is fundamentally different from the development of antiviral vaccines. The aim of antiviral vaccination is to confer protective immunity against a viral pathogen whereas the intention of antifertility vaccination is to cause an immune response in the vaccinated individual in a way that critical reproductive processes are interrupted. To elicit such immune responses, the antifertility vaccine needs to contain an antigen with contraceptive properties. This can be achieved by either manufacturing the vaccine using appropriate components derived from native proteins or from transgenic proteins that were harvested and purified from bacterial or fungal sources. A more widely used approach is the genetic manipulation of a live vaccine vector in which the antifertility component of the vaccine is supplied, for example, by a genetically engineered virus (Shellam, 1994). Genetic engineering of suitable viral vectors has proven to be a successful method to overcome some of the shortcomings of conventional vaccine development (Jackson et al., 1998). However, of all vaccines currently used in wildlife on a wide geographical scale only one has been developed using biotechnological genetic engineering. This vaccine is based on a genetically altered vaccinia virus that contains the immunogenic components of rabies virus (Brochier et al., 1991). Unfortunately, the potential of vaccinia virus to indiscriminately infect a wide range of species including humans impedes its general use as a vaccine vector, for example, for applications such as antifertility vaccination of wildlife. It is for this reason that alternative, more species-specific vectors such as canine herpesvirus (CHV) or canine adenovirus (CAdV) are currently being researched.

The use of live viral vectors to deliver vaccine antigens has already been used for various antifertility vaccine prototypes (Jackson et al., 1998; Redwood et al., 2005; Kerr et al., 1999). A number of biological criteria need to be considered that influence the choice of virus as a vaccine vector (Shellam, 1994; Boyle, 1994). Among them are, for example, the species-specificity of the vector, the ability of the virus to disseminate within a population, the potential of pre-existing population immunity to the vector, the intended route of immunization, and, most critically, the suitability of the vector genome to accommodate additional heterologous DNA without disruption of essential viral gene functions. In the case of wildlife oral vaccination, practical aspects such as the virus stability in baits need also be considered.

Based on serological evidence, European red foxes appear to be susceptible to many viral infections commonly described in domestic dogs (Truyen et al., 1998; Garcelon et al., 1992). With regards to the potential of these viruses as vaccine vectors for wild canids, however, very few appear to be suitable candidates. We have focused our attention on CHV as potential vaccine vector for wild foxes for several reasons: In a previous study we have shown that foxes can be experimentally infected with CHV which caused a long-lasting seropositivity in all foxes (Reubel et al., 2001). Orally infected foxes did not shed CHV into the environment and transmission to in contact foxes was not observed. Although viral DNA was detected at the site of latency, the spinal ganglia of infected foxes, no reactivation of virus could be observed even after high dose corticosteroid treatment (Reubel et al., 2004). In conjunction with this, antibody prevalence in domestic dog populations has been reported to reach up to 88% (Gaskell and