ZKBS
Zentrale Kommission
für die Biologische Sicherheit

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Opinion of the ZKBS on the risk assessment of the recombinant adenovirus-based vaccine virus Ad26.ZEBOV according to § 5 paragraph 1 GenTSV

General

Ad26.ZEBOV is a replication-defective human adenovirus of serotype 26 (Ad26) that contains the gene for the glycoprotein (GP) of Zaire ebolavirus (ZEBOV). Ad26 belongs to the species *Human mastadenovirus D* and was first isolated in 1956 from an anal swab of a nine-month-old boy [1] . In the few studies published to date, gastrointestinal symptoms and conjunctivitis have been observed in infections with viruses of the species [2] . Ad26 is assigned to **risk group 2** as a donor and recipient organism for genetic engineering work.

Compared to the wild-type virus, the E1 gene region of Ad26.ZEBOV is completely deleted and the E3 gene region is partially deleted. The deletion of the E1 gene region causes the replication defect of the vaccine virus. At the genomic location of the E1 gene, the coding nucleic acid portion of the complete ZEBOV-GP was inserted. The expression of the protein is under the control of the cytomegalovirus promoter. In addition, the ORF6/7 within the E4 gene region was replaced with the homologous nucleic acid segment of human adenovirus 5 (Ad5). The recombinant cell line PER.C6 is used to produce Ad26.ZEBOV particles. This is a human embryonic retinal cell line transformed by introducing a plasmid containing the E1 gene of Ad5. Expression of the two gene products, E1A and E1B, complements the replication defect of the genome of Ad26.ZEBOV, resulting in the release of replication-defective adenoviral particles from the cells. Restoration of the replication capacity of the adenovirus by recombination is not expected due to the lack of homologies to the adenoviral genomic segment present in the cell line [3] .

Ad26.ZEBOV was licensed in the EU on 1 July 2020 as a vaccine against Zaire ebolavirus for use in adults and children aged one year and older. However, as ZEBOV is not endemic in Europe, the actual use of the vaccine within the EU is generally limited to research laboratory staff and medical personnel. For the safety of the vaccine, the European Medicines Agency (EMA) has summarized eleven phase I, II and III clinical trials [4] . Of these, the results of four Phase I and one Phase II study have since been published in medical journals. [5-9] . The analysis of data from 1901 healthy adults and 649 children and adolescents aged 1 to 17 years, who were each initially vaccinated intramuscularly (i. m.) with 5 x 10^{10} vector particles of Ad26.ZEBOV and after at least 28 days i. m. with 10^{8} infection units of a second Ebola vaccine candidate based on the Modified Vaccinia Virus Ankara (MVA) (also licensed as MVA-BN-Filo on 1 July 2020), showed a good overall safety profile. Local adverse events occurred in 51 % of adults and 27 % of children and adolescents. Of these, the most common local adverse event was pain at the injection site (48 % and 24 %, respectively). Systemic side effects

occurred in 67 % of adults and 37 % of children and adolescents. The most common systemic side effects[1] were fatigue (46 % and 15 %, respectively), headache (45 % and 24 %, respectively), muscle pain (37 % and 8 %, respectively), and chills (24 % and 10 %, respectively). Side effects were predominantly mild to moderate in adults as well as in children and adolescents and lasted only a few days. Severe systemic adverse reactions occurred in 4 % of adults and 1 % of children and adolescents. The proportion of different serious adverse events roughly reflected the relative frequencies of systemic adverse events overall. There was no difference in unexpected serious adverse events after vaccination with Ad26.ZEBOV compared with the placebo group. However, in one subject, an association was seen between vaccination with Ad26.ZEBOV and small fiber neuropathy. Even with the inclusion of additional subjects who were infected with human immunodeficiency virus or who deviated from the standard protocol in the order of vaccines or the interval between vaccinations, there were no significant differences in the frequency and severity of adverse events. When vaccinated twice with Ad26.ZEBOV 14 days apart, the frequency of adverse events increased [4] .

The vector backbone used for Ad26.ZEBOV is also used for several other vaccine viruses. These include the vaccine viruses Ad26.COVS1 (ref. 45242.0180, July 2020), Ad26.Mos2S.Env, Ad26.Mos1.Env, Ad26.Mos1.Gag-Pol, Ad26.Mos2.Gag-Pol and Ad26.RSV.preF (all ref. 45242.0188, April 2021), which have already been assigned **to risk group 1** by the ZKBS. In addition, rAd26-S-CoV2, another Ad26-based vaccine virus, was assigned to **risk group 1** by the ZKBS (ref. 45242.0193, June 2021). All these vaccine viruses proved to be well tolerated in clinical studies.

Recommendation

According to § 5 paragraph 1 GenTSV in conjunction with the criteria in Appendix 1 GenTSV, the recombinant adenovirus Ad26.ZEBOV is assigned to **risk group 1** as a genetically modified organism.

Justification

The recombinant adenovirus Ad26.ZEBOV is a replication-defective virus carrying a gene from ZEBOV without its own hazard potential. The vaccine virus is licensed in the EU for adults and children over the age of one year. In clinical studies, the vaccine virus was well tolerated and mostly caused only mild or moderate side effects.

[1] The information on specific systemic adverse reactions in children and adolescents refers to children aged 4 years and older according to the definitions in the studies.

Literature

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