



Rapid development of vaccines against emerging pathogens: The replication-deficient simian adenovirus platform technology



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ABSTRACT

Despite the fact that there had been multiple small outbreaks of Ebola Virus Disease, when a large outbreak occurred in 2014 there were no vaccines or drugs available for use. Clinical development of multiple candidate vaccines was then initiated in parallel with attempts to contain the outbreak but only one vaccine was eventually tested in a phase III trial. In order to be better prepared for future outbreaks of known human pathogens, platform technologies to accelerate vaccine development should be employed, allowing vaccine developers to take advantage of detailed knowledge of the vaccine platform and facilitating rapid progress to clinical trials and eventually to vaccine stockpiles. This review gives an example of one such vaccine platform, replication-deficient simian adenoviruses, and describes progress in human and livestock vaccine development for three outbreak pathogens, Ebola virus, Rift Valley Fever Virus and Middle East Respiratory Syndrome Coronavirus.

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1. The replication-deficient simian adenovirus platform

Replication-deficient adenoviral vectors were originally developed for gene therapy applications, as they can be produced in an efficient manufacturing process, are able to infect cells expressing the coxsackie and adenovirus receptor (CAR) and express the encoded gene of interest within the infected cell. Deleting the E1 gene from the adenoviral genome and supplying it *in trans* from the cell line used to produce the adenoviral vector allows replication-deficient recombinant viruses to be produced, with the novel gene of interest inserted at the E1 locus. However the inherent immunogenicity of adenoviral vectors makes them more suited to use as vaccine vectors, and the ability to drive expression of the vaccine antigen intracellularly after vaccination results in the induction of both T cell and humoral responses to the antigen. Human adenoviruses are highly immunogenic in mice and other species but as many humans have previously been infected by common human adenoviruses such as AdHu5, anti-vector antibodies generated after the initial infection can neutralise a vaccine based on the same adenovirus, thus reducing immunogenicity. In contrast, the level of neutralising antibodies to simian adenoviruses is very low in human populations, and a number of differ-

ent replication-deficient vaccine vectors such as ChAd3, ChAd63 [1] and ChAdOx1 [2] have now been developed from simian adenoviruses.

2. Response to the 2014 Ebola outbreak in West Africa

Between 1976 and 2013 the World Health Organization (WHO) reported 24 localised outbreaks of Ebola virus disease involving a total of 1716 cases, with a fatality rate of between 25% and 90%. No drugs or vaccines were available to treat or prevent this devastating infectious disease. Outbreaks were contained by contact tracing and quarantine [3]. Vaccine development had begun, and by 2014 two different viral vectored vaccines had shown protective efficacy against a high dose Ebola virus challenge in non-human primates (NHP) and were being prepared for clinical development. These were a replication-deficient simian adenovirus vector, ChAd3 [4], and a replication-competent vesicular stomatitis virus (VSV) [5], each expressing the surface glycoprotein of Ebola virus from either the Zaire or Sudan species. Additionally a replication-deficient human adenovirus 5 vaccine was protective in the same NHP challenge model [6] but in a clinical trial, pre-existing immunity to the vaccine vector resulted in variable immune responses to the Ebola glycoprotein [7], with the result that the ChAd3-vectored vaccine, with very low pre-existing immunity to the vaccine vector was the preferred option for further clinical testing.

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The ChAd3-vectored vaccine had already been manufactured in preparation for a clinical trial, and commencement of the first phase I study was accelerated. A phase I trial of a ChAd3-vectored vaccine against hepatitis C had already been completed [8], and the safety and immunogenicity data from that study were used to support rapid regulatory and ethical review of the Ebola vaccine which was then tested in a series of phase I studies [9–11]. By the end of 2014 several trials had produced safety and immunogenicity data and discussions on the location and design of an efficacy study were underway. However the first efficacy trial, of the VSV-vectored vaccine which by that time had also been tested in several phase I trials, did not begin until April 2015 by which time the number of cases had markedly declined. The VSV-vectored vaccine proved to be highly effective [12], but there was no opportunity to test efficacy of the ChAd3-vectored vaccine. A subsequent comparison of total IgG and neutralising antibody responses to each vaccine revealed strikingly similar immunogenicity [10], making it likely that the ChAd3-vectored vaccine would be equally protective, but less reactogenic than the VSV-vectored vaccine. Thus, following an outbreak of a disease which resulted in 28,616 known cases and 11,310 deaths, lasting from the first case in December 2013 to the end of virus transmission in June 2016, and for which two vaccines were well advanced in pre-clinical development and preparation for clinical trials before the outbreak began, it was only possible to test efficacy of one of these vaccines in the declining months of the outbreak.

Ebola virus is only one of many ‘emerging pathogens’ that may cause an outbreak in humans in the future. The WHO has announced a priority list (see Table 1) for which measures (diagnostics, therapeutics, vaccines) should be developed in advance of any further outbreaks. Vaccine development should encompass pre-clinical testing, preferably determining vaccine efficacy in a suitable animal model, and phase I clinical trials can be completed to assess safety and immunogenicity, where possible comparing immune responses to known correlates of protection in animal models. Vaccine stockpiles could then be produced, with plans for an efficacy trial put in place in readiness to test the vaccine for efficacy as soon as an outbreak occurs. However vaccine development is a slow process, typically taking 10–15 years to achieve vaccine licensure, at considerable cost. For emerging pathogen vaccines, employing platform technologies to develop vaccines against multiple diseases could save considerable amounts of time and money in the efforts to protect the world against novel pathogens. Replication-deficient simian adenovirus-vectored vaccines are

already in development for all of the pathogens on the WHO priority list as well as others, and progress on two of them is summarised below.

3. Rift Valley Fever (RVF)

RVF is caused by a mosquito-borne *bunyavirus* with a wide host range including sheep, goats, cattle, camels and humans [13]. Virus transmission between susceptible hosts occurs via numerous mosquito species but humans can also acquire infection from handling contaminated animal fluids or tissue [14]. Following its first isolation in 1930 in Kenya [15], RVF virus has spread throughout much of Africa and the Arabian Peninsula, causing recurrent outbreaks of human and livestock illness [14]. Disease in livestock is characterised by high rates (>90%) of neonatal mortality and abortion in pregnant animals [13]. In humans RVF presents as an acute self-limiting febrile illness but severe manifestations can also occur, including haemorrhage and encephalitis, with high (>30%) case fatality rates [16,17]. However, RVF is probably under-reported and its true burden remains unknown. For instance, miscarriage in pregnant women has only recently been identified as a possible outcome of RVF infection [18], underscoring the need for better surveillance in areas that are endemic for RVF transmission.

No licensed RVF vaccines are currently available for human use, but live or inactivated whole RVF virus vaccines are available for livestock [19]. Despite their widespread use in Africa and the Arabian Peninsula, these livestock vaccines have major safety concerns, including residual virulence [19]. Safe, efficacious alternatives are urgently needed, and technically, their co-development for livestock and human use should be straightforward for two main reasons: (1) the viral envelope glycoproteins (Gn and Gc) targeted by protective neutralising antibody are highly conserved and, (2) vaccination readily elicits neutralising antibody levels within the range acquired in convalescence and associated with long-lived immunity.

Single-dose immunisation with ChAdOx1 RVF, a replication-deficient simian adenovirus vaccine encoding RVF viral glycoproteins, has demonstrated 100% efficacy against viral challenge in livestock, including sheep, goats and cattle, outperforming the most widely used licensed product in Africa [20]. ChAdOx1 RVF will now undergo testing in large field trials in livestock (sheep, goats, cattle, camels) in East Africa in 2017, including pregnant animals. In parallel, ChAdOx1 RVF will be evaluated in human phase I clinical trials in the UK and East Africa, which could provide for the

Table 1
Priority pathogens for vaccine development. The descriptions here are intended to give an overview of the diseases with outbreak potential and the information contained is not exhaustive. More detailed, and updated information may be obtained from the WHO website <http://www.who.int/mediacentre/factsheets/en/>.

Disease	Characteristics of disease	Insect vector	Main geographic location	Livestock affected
Ebola virus disease	Haemorrhagic fever	None	Central and West Africa	None, reservoir in wild animals
Marburg	Haemorrhagic fever	None	Angola, Democratic Republic of the Congo, Kenya, and Uganda	None, reservoir in wild animals
Lassa fever	Haemorrhagic fever	None	Benin, Ghana, Guinea, Liberia, Mali, Sierra Leone, and Nigeria	None, reservoir in rodents
Crimean Congo haemorrhagic fever	Haemorrhagic fever	Ticks	Africa, the Balkans, the Middle East and Asia, in countries south of the 50th parallel north	Cattle, sheep, goats, ostriches
MERS	Chronic respiratory syndrome	None	Middle East	Camels infected but illness not apparent
SARS	Acute respiratory syndrome	None	China	None, reservoir in wild animals
Nipah	Acute respiratory syndrome, encephalitis	None	Malaysia, Bangladesh, India	Pigs, reservoir in bats
Rift Valley fever	Haemorrhagic fever	<i>Aedes</i> and <i>Culex</i> mosquitoes	Africa, Middle East	Sheep, cattle, goats, camels
Zika	Generally mild, but causes severe birth defects	<i>Aedes</i> mosquitoes	South America	None

first time a vaccine that can be deployed against the same pathogen in multiple diverse species. This One Health approach to vaccine development is well suited for emerging pathogens as it may allow rapid approval of vaccines for emergency use in humans based on levels of the protective response (in this case, RVF virus neutralising antibody) in livestock plus safety data in humans.

4. Middle East Respiratory Syndrome (MERS)

MERS is caused by a coronavirus that was first identified in 2012 [21]. Since then there have been >1850 laboratory-confirmed cases of infection and >650 deaths, with the majority of cases in Saudi Arabia where there have been several outbreaks within hospitals. The original viral reservoir may have been in bats, but the majority of camels in the Middle East are now seropositive for MERS [22,23]. Having crossed the species barrier into camels, the virus is now infectious for humans as the DPP4 cell surface receptor that the virus uses for cell entry is highly conserved between camels and humans [24]. In humans, the cases that have been identified suffer from a severe lower respiratory tract infection [25]. However individuals who are exposed to camels have a significantly higher rate of seropositivity to MERS than the general population and it is likely that mild infections in otherwise healthy individuals are the source of infection for some other human cases [26,27].

The major surface antigen of MERS-CoV is the Spike (S protein). ChAdOx1 expressing MERS S induces the production of anti-S antibodies, including neutralising antibodies, after a single intramuscular immunisation in mice (Alharbi, manuscript in preparation) and studies in camels are now planned. A replication-deficient poxvirus, Modified vaccinia virus Ankara (MVA) expressing MERS S was able to reduce virus shedding after MERS challenge of camels in a pilot study [28]. Both ChAdOx1 and MVA-vectored vaccines are now being prepared for clinical trials.

The most effective vaccination strategy to prevent further morbidity and mortality resulting from human MERS-CoV infection may be difficult to define. Severe or fatal infections occur in older adults who are to some extent immunocompromised [29]. This population may not make strong immune responses following vaccination, and it would be necessary to vaccinate large numbers to prevent further cases. Vaccinating camels to eliminate the infection in the Middle East would prevent human exposure, but will likely require government intervention since the disease in camels is mild and does not appear to result in economic losses to the camel owners. Vaccination of imported camels would need to be maintained, and in the short-term vaccination of those occupationally exposed to camels, or healthcare workers, may be beneficial to prevent further human to human spread. The burden of disease in African countries where MERS seropositive camels have been found is not yet understood. This presents a complex series of public health problems, which will be difficult to solve. The ability to employ a well-characterised platform technology for both camel and human vaccine development would result in some reduction of the complexity.

5. Summary and future perspectives

The use of platform technologies to produce vaccines against multiple diseases has the potential to greatly reduce the time and cost required to develop novel vaccines to the point at which safety and immunogenicity has been established, master seed stocks of the vaccines have been produced and stockpiles have been prepared for use should an outbreak occur. By selecting a small number of platform technologies to use it will be possible to plan all aspects of vaccine manufacturing and clinical develop-

ment with advance knowledge of the expected thermostability, reactogenicity and immunogenicity of vaccines against multiple different diseases. Prior knowledge of the safety of a given vaccine platform technology in adults, children, infants, older adults and HIV positive individuals will aid in the design and conduct of vaccine trials to be conducted rapidly when an outbreak occurs. In some cases, parallel development of a veterinary and human vaccine could result in deployment of the veterinary vaccine to control the animal reservoir of disease and prevent human exposure at the same time as preparing a stockpile for use in humans should an outbreak occur. Additionally, when the next previously unknown pathogen emerges, the options for rapid vaccine development, the steps to be taken, the facilities to be used and the time needed to prepare doses for human use will be far better understood.

The pathway to regulatory approval for vaccines against emerging pathogens is not yet clearly defined. For vaccines against pathogens which are already widespread, such as Chikungunya or Zika it will be possible to follow the usual route through clinical trials, demonstrating the safety, immunogenicity and efficacy of the novel vaccine in the population for which it is needed, including the full age range. For vaccines against pathogens that typically cause small outbreaks, such as Rift Valley Fever virus, it may be necessary to seek regulatory approval based on safety and immunogenicity in humans, plus efficacy in a relevant animal species, but the requirements for this approach have not yet been agreed with regulatory authorities. Once again, the use of a platform technology will be advantageous as vaccine safety data from the clinical use of all vaccines using the same platform will be relevant.

Since the 2014 Ebola outbreak, the need for rapid development of vaccines against known emerging pathogens and the ability to respond swiftly to newly identified pathogens has been highlighted, and a new organisation, the Coalition for Epidemic Preparedness Innovations (www.cepi.net) has been formed to address these needs. At the time of writing, preproposals for the development of vaccines against MERS, Nipah and Lassa are under review by CEPI.

6. Conflict of interest

SCG is a co-founder of, consultant to and shareholder in Vacitech plc which is developing a vectored MERS vaccine.

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