WETENSCHAPPELIJK INSTITUUT VOLKSGEZONDHEID
INSTITUT SCIENTIFIQUE DE SANTÉ PUBLIQUE

Novel GMO-based vaccines against tuberculosis: state-of-the art and biosafety considerations

Amaya Leunda, Ph.D.

ABSA 57th Annual Biological Safety Conference

October 3-8, 2014

Rue Juliette Wytsmanstraat 14 | 1050 Brussels | Belgium
T +32 2 642 51 11 | F +32 2 642 50 01 | email: info@wiv-isp.be | www.wiv-isp.be

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Vaccines **2014**, *2*, 463-499; doi:10.3390/vaccines2020463

OPEN ACCESS

vaccines

ISSN 2076-393X

www.mdpi.com/journal/vaccines

Review

Amaya Leunda [1,*], Aline Baldo [1], Martine Goossens [1], Kris Huygen [2], Philippe Herman [1] and Marta Romano [2,*]

[1] Biosafety and Biotechnology Unit, Scientific Institute of Public Health, 14 Juliette Wytsman Street, Brussels 1050, Belgium;

[2] Immunology Unit, Scientific Institute of Public Health, 642 Engeland Street, Brussels 1180, Belgium

Novel GMO-based vaccines against tuberculosis

- Introduction
- Risk assessment of activities involving GMO-based vaccines against Tuberculosis (TB): general regulatory considerations in Europe

- Biosafety considerations of clinical studies with GMO-based vaccines
  - ✓ BCG replacement with genetically modified mycobacteria
  - ✓ TB vaccine candidates based on recombinant viral vectors as “booster” sub-unit vaccines

Novel GMO-based vaccines against tuberculosis: Introduction

Mycobacterium tuberculosis (*Mtb*) and mycobacteria of *Mtb* complex:

- tuberculosis, a severe human disease
- Easy spread into the community (airborne)
- Therapeutic exists (except XDR-TB)
- RG 3 microorganism

Novel GMO-based vaccines against tuberculosis: Introduction

=> Need of novel antibiotics

=> New treatment schemes

=> Effective vaccines

BCG vaccination

- protects children against TB meningitis and disseminated TB
- Low efficacy against pulmonary TB

Poor understanding of the immunity

Novel GMO-based vaccines against tuberculosis: Introduction

This presentation focuses on **novel GMO-based vaccines** (or recombinant vaccines) currently in clinical trials and

Protection of the general population and the environment against an exposure to the recombinant vaccines during clinical trials

Not the protection of the vaccinee

Novel GMO-based vaccines against tuberculosis: Regulation

Directive 2009/41/EC: on the ***contained use*** of Genetically Modified Microorganisms (GMM), for protection of the general population and the environment

Directive 2001/18/EC: on the ***deliberate release*** in the environment and placing on the market of Genetically Modified Organisms (GMO), for the protection of the general population and the environment

Directive 2000/54/EC

Novel GMO-based vaccines against tuberculosis: Regulation

Directives 2009/41/EC and 2001/18/EC:

- **risk assessment** of the recombinant vaccine
- **environmental risk assessment** (ERA) in case of release of the vaccine candidate and contact with the general population and the environment

Novel GMO-based vaccines against tuberculosis: Regulation

⇒ potential of the GMO to cause **adverse effects** on persons, animals, plants and other microorganisms exposed to it and

=> **probability** that these adverse effects will occur

Novel GMO-based vaccines against tuberculosis: Regulation

Risk assessment of the recombinant vaccine takes into account:

- genetic stability and possible interaction with other organisms
- intrinsic characteristic of the strain used and of the transgene
- biodistributon and level of dissemination
- possibility of recombination
- risk classification
- pathways of exposure through which it may interact with humans and the environment

Genetically modified mycobacteria: VPM1002

Modified BCG

- expressing listeriolysin **LLO** a toxin from *Listeria monocytogenes (*formation of pores in the phagosome)
- deleted in *ureC* to obtain optimal pH

=> facilitates translocation and subsequent MHC1 loading of mycobacterial antigens

=> might activate cell apoptosis

Genetically modified mycobacteria: VPM1002: risk assessment

In Phase II clinical trial (new-born infants in South Africa)

BCG is largely used in vaccination against TB and is of RG1 (2)

The transgene is inserted in bacterial chromosome making horizontal gene transfer highly improbable

LLO activity is limited to phagosome membranes

Genetically modified mycobacteria: VPM1002: risk assessment

BCG and mycobacteria in general are showed to be genetically stable with poor replicative characteristics

No serious adverse effects were reported in animal models and in volunteers in phase I clinical trial

Persistence in macrophages showed to be lower than BCG VPM1002 is rapidly eliminated

Genetically modified mycobacteria: VPM1002: risk assessment

VPM1002 has been classified **in RG 1**

Biodistribution and ERA:

In phase I trial, surveillance of VPM1002 shedding: analysis of blood, saliva, urine and stool by PCR => no VPM1002 detection

No case of transmission of the vaccine to other persons reported

Genetically modified mycobacteria: VPM1002: risk assessment

⇒ Probability of dissemination into the environment considered very low

⇒ Limited environmental impact if released into the environment

Genetically modified mycobacteria: MTBVAC

Attenuation of a *Mtb* strain from human origin by 2 deletions:

- the transcription factor *phoP* that contributes to *Mtb* virulence
- the gene *fadD26* required in pathogen protection against host defence and for *Mtb* multiplication in mouse lungs

=> 2 unlinked non-reverting mutations in *Mtb* (Geneva Consensus)

In Phase I trial (healthy humans)

Genetically modified mycobacteria MTBVAC: risk assessment

Mtb is classified in **RG3**, a microorganism that may cause a severe disease and able to propagate easily to the community

Poor replicative characteristics and high genetic stability of mycobacteria in general

However, exchange of genetic material of recombinant *Mtb* with environmental mycobacteria and consequences of these exchange should be explored (complementation)

Genetically modified mycobacteria MTBVAC: risk assessment

Until now, lack of evidence of gene reversion complementation or horizontal gene transfer

In animal models, MTBVAC is showed to be more attenuated than BCG

MTBVAC is classified in **RG1 for animals**

In humans, more data are needed

Genetically modified mycobacteria MTBVAC: risk assessment

Biodistribution and ERA:

Surveillance of shedding by analysis of urine and stool (no data on blood or saliva) to detect MTBVAC => reported negative in mice

In guinea pigs, presence was detected at the injection site after vaccination

No data on bio-distribution and shedding **in human**

Genetically modified mycobacteria MTBVAC: risk assessment

⇒ Probability of dissemination into the environment considered very low

⇒ If released into the environment, the environmental impact is expected to be low and no adverse effects of MTBVAC are anticipated in a person

VPM1002 and MTBVAC: Risk management measures

Containment level 1 (Directive 2009/41/EC)

For personnel, primary hazards are:

- Inhalation of recombinant vaccines (airborne)
- Exposure to contaminated droplets or aerosols of mucous membranes or broken skin
- Accidental projection
- Inadvertent parental inoculation
- Unintentional contamination via close contact with contaminated material.

VPM1002 and MTBVAC: Risk management measures

These bio-incidents may be the origin of unintentional dissemination in the environment

- Appropriate personal protection equipment (PPE): lab coat, gloves, goggles, mask
- Use of Biosafety Cabinet for open phase manipulations with recombinant vaccines
- Avoid use of sharped objects as far as possible
- Never re-cap nor remove needles from syringes

VPM1002 and MTBVAC: Risk management measures

- Appropriate procedures for decontamination of material and surface
- Contaminated waste and PPE inactivated before final disposal

Vaccinated volunteer at home should protect injection site, manage waste and avoid closed contact with old and young people or persons who are immuno-compromised

Recombinant viral vectors as booster sub-unit vaccines

Boosters to be administrated in a vaccine regimen involving BCG vaccination at birth followed by a boost vaccination

AdHu5Ag85A and AERAS-402:

recombinant replication deficient Adenovirus serotype 5 (Ad5) and 35 (Ad35) respectively, expressing mycobacterial antigens

MVA-85A and MVA-85A-IMX313:

recombinant of the Modified Vaccinia Ankara (MVA) strain expressing a mycobacterial antigen

Recombinant viral vectors as booster sub-unit vaccines: risk assessment

Wild type Adenovirus is **RG2** for humans

Ad5 and Ad35 vectors:

- are made replication deficient
- remain essentially episomal in transduced cells

=> **Ad5, Ad35 are of RG1**

Recombinant viral vectors as booster sub-unit vaccines: risk assessment

MVA:

- is a highly attenuated vaccinia strain
- Unable to propagate in most mammalian cells
- Has a fully cytoplasmic cycle of propagation
- Has poor replicative characteristics

=> **MVA is of RG1**

Recombinant viral vectors as booster sub-unit vaccines: risk assessment

AdHu5Ag85A and MVA-85A express Ag85A, a major component of the *Mtb* cell wall, immunodominant

AERAS-402 expresses Ag85A, 85B and TB10.4

The function of TB10.4 is unknown, is target for immune response

MVA-85A-IMX313 expresses Ag85A and IMX313 used to potentiate immune effect

Recombinant viral vectors as booster sub-unit vaccines: risk assessment

Transgenes in boosters:

- used to induce and amplify cellular response against *Mtb*
- No adverse effects in healthy humans observed in trial
- No known toxic, allergic effects when expressed in human
- do not change safety profile

Recombinant viral vectors as booster sub-unit vaccines: risk assessment

- Possibility of recombination of AERAS-402 or AdHu5Ag85A during co-infection with wild-type adenovirus and risk of replication competent adenovirus (RCA) => not observed

- Insertion of the gene of interest may alter the safety profile of the recipient viral strains => not observed

- Results from first clinical trials show no adverse effects

AERAS-402 and AdHu5Ag85A are classified in **RG1**

Recombinant viral vectors as booster sub-unit vaccines: risk assessment

- Possibility of recombination of MVA vectors during co-infection of the same cell with homologous non-human orthopox virus (OPV) is very low, except in animals.

MVA-85A used in cattle => possible recombination

- Results from first clinical trials with MVA-85A show no adverse events

MVA-85A is classified into **RG1**

MVA-85A-IMX313 more data are needed

Recombinant viral vectors as booster sub-unit vaccines: risk assessment

Biodistribution and ERA:

Intramuscular administration of recombinant adenovirus leads to systemic biodistribution and shedding via almost all excreta

If recombinant adenoviral vectors are released and in case of RCA:

- immune system would rapidly eliminate RCA
- no harmful effects of the expressed proteins
- in immunosuppressed persons infection could lead to adverse effects

Recombinant viral vectors as booster sub-unit vaccines: risk assessment

Adenovirus are species specific and Ad5 and Ad35 not pathogenic to animals

The consequences of release in the environment are not known

Concerning MVA-85A, no data on dispersion and shedding are available

MVA-85A-IMX313 phase I trial is still ongoing and no data available

Recombinant viral vectors as booster sub-unit vaccines: Risk management measures

Containment Level 1 (Directive 2009/41/EC)

During production, all batches should be tested for the presence of replication competent virus

For personnel, primary hazards consist in:

- exposure to droplets or aerosols of mucous membranes or broken skin
- Accidental projection into the eye or other mucous membranes

Recombinant viral vectors as booster sub-unit vaccines: Risk management measures

For personnel, primary hazards consist in:

- inadvertent parental inoculation
- unintentional contamination via close contact with contaminated material.

These bioincidents may lead to unintentional dissemination in the environment

Recombinant viral vectors as booster sub-unit vaccines: Risk management measures

- Adequate PPE: lab coat, gloves, goggles, mask
- Use of a Biosafety cabinet
- Work with needles and other sharp objects strictly limited
- Never re-cap nor remove needles from syringes
- Appropriate disinfectant for surface decontamination (spill)
- Waste and PPE inactivated using an appropriate method before disposal

Recombinant viral vectors as booster sub-unit vaccines: Risk management measures

Vaccinated volunteer at home should protect injection site and follow procedure for waste management

When vaccinated with Ad, volunteer should avoid closed contact with old and young people or persons who are immuno-compromised

Novel GMO-based vaccines against tuberculosis: conclusion

Other recombinant vaccines currently in research and based on BCG or *Mtb*

Other schemes of vaccination combining recombinant vaccines are currently in trials

Beside safety and efficacy, biosafety should be considered to protect general population and the environment

- Identification of potential risks of the GMO-based vaccine
- Probability of occurrence

=> Risk management measures

Thank you for your attention