Antibody delivery to the lungs against Ricine intoxication by inhalation: Drug and device development

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AeroRiMac project: context

Castor bean also called Ricin is a toxin classified as a priority of biological agents by the French Defense Ministry, and belongs to the biological agents of B category according to the "Centers for Disease Control and Prevention" (CDC). Ricin risk is mainly associated to aerosol diffusion. Vaccines showed limited efficacy to induce neutralizing antibody in a pulmonary ricin intoxication (Pincus S.H. *et al.*, 2011), while the local delivery of recombinant neutralizing antibodies raised against the A subunit of ricin (Poli M.A. *et al.*, 1996 ; Guo J. *et al.*, 2006), such as the 43RCA antibody developed by IRBA (Pelat T. *et al.*, 2009) led to animal survival up to 6 hours after intoxication, thereby representing a promising therapeutic approach. Recently, the CEPR (INSERM U1100/EA6305) in Tours, specialized in the aerosol delivery of drugs, has demonstrated the feasibility and the clinical interest of delivering antibodies through the airways as an aerosol to treat lung affections (Maillet A. *et al.*, 2008 ; Maillet A. *et al*., 2011).

Ricin: Biological toxin with high potential of bioterrorism

Castor Beans (*Ricinus communis*)

- **[Ricin]** per beans 1 to 10 %
  (ingestion of 3 castor beans = death)
- **Lethal dose in Human through inhalation:** 1 to 10 µg/kg
  (10x lower than ingested ricin)

- **Vaccines: Limited** efficacy in case of ricin inhalation
- **Therapeutic approach proposed:**
  Aerosoltherapy with neutralizing monoclonal antibody (IgG 43RCA, IRBA)
  (Local delivery of 43RCA protected mice against pulmonary intoxication to ricin)
- **Inhibition of 5 DL50 ricin:** 4.5 mg of 43RCA in lung

Drug & Device Development

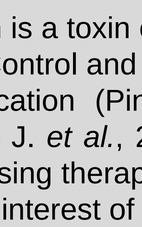
DRUG DEVELOPMENT

0 Initial State

43 RCA Fomulation:
- Frozen Anti-ricin Mab
- PBS 1X ~ *15 mg/mL*

Nebulizer:
- *VMD 4 µm*
- *Alveolar deposition: 10 %*

1 Formulation

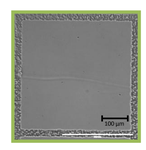


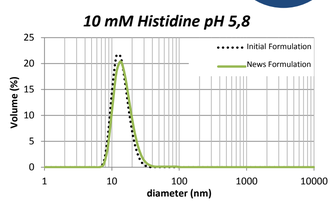
- Lyophilization
- Nebulization

Requirements
- Storage at room temperature
- "Biocompatible" solution:
  - pH, Iso-osmolarity
  - Pulmonary tolerance
- Low amounts of aggregates to avoid immunogenicicty and loss of activity
- Maximum affinity

Results/Technical improvements:

Histidine 10 mM pH 5,8





- **Affinity formulation = affinity initial state**
- **Nebulization: Add Tween**

2 Reconstitution

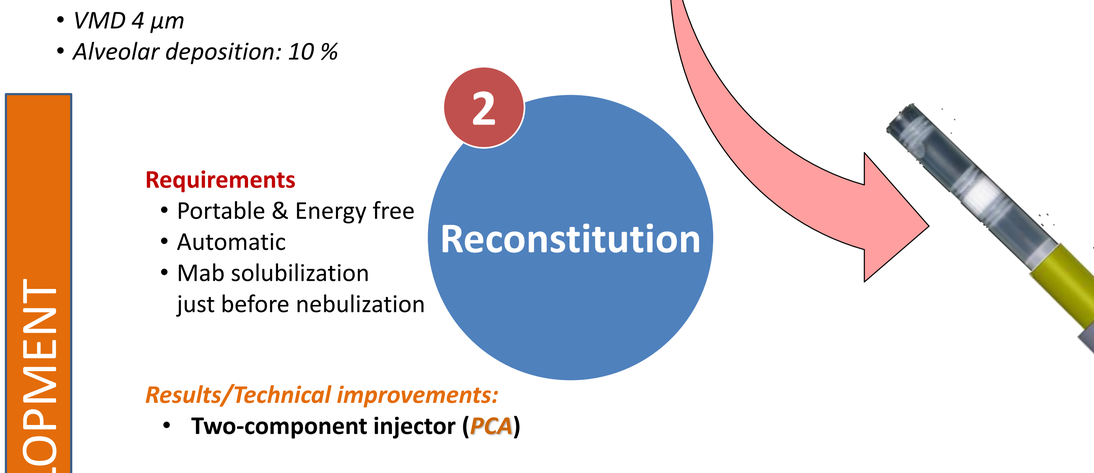
Requirements
- Portable & Energy free
- Automatic
- Mab solubilization just before nebulization

Results/Technical improvements:
- **Two-component injector (*PCA*)**

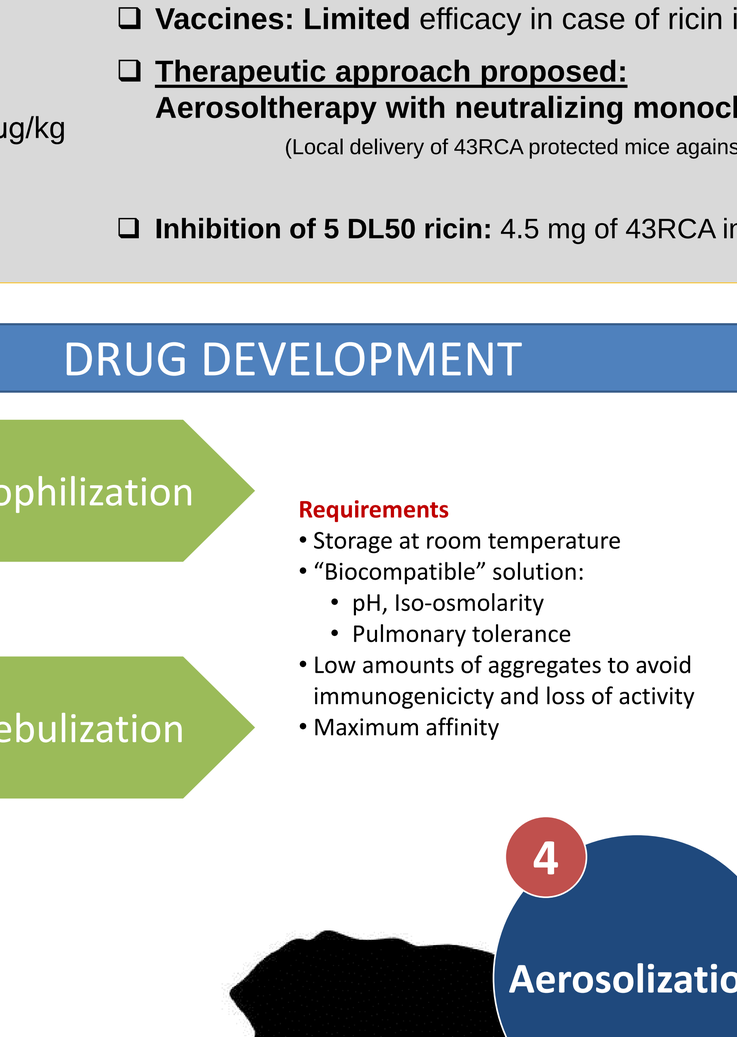
3 Nebulizer

Requirements
- Small size
- Standalone energy device
- Use: Medical Staff
- Terms of Use: *Temp.: 5/40 °C RH: 0 /100 %*

Results/Technical improvements:
- **Membrane OnQ™ Low VMD (*Aerogen*) 2 µm**
- **Storage chamber (*DTF*): Inhalable fraction 80 %**

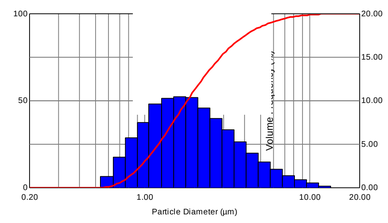


4 Aerosolization



Requirements
- Alveolar target: 2 µm
- Time < 20 minutes

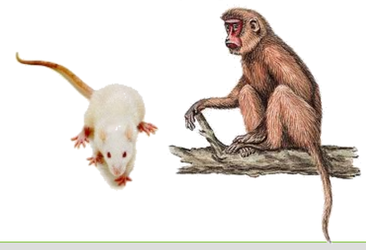
Results/Technical improvements:
- **VMD 2 µm**
- **Alveolar deposition: 20 %**
- ***Works in progress***



5 *In-vivo* studies (rat, non-human primates)

Requirement
- Pharmacokinetics
- Toxicity
- Quantification of lung deposition

Results/Technical improvements:
- ***Works in progress***



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