**Introduction**

*Staphylococcus aureus* pneumonia is associated with high mortality, irrespective of antibiotic susceptibility. Up to six cytolytic toxins are produced by *S. aureus*: alpha-hemolysin (Hla) and the five bi-component leukocidins (HlgAB, HlgCB, LukED, LukSF, and LukGH/LukAB) that are important for pathogenesis. We previously described two human IgG1 monoclonal antibodies (mAbs), ASN-1 and ASN-2, that in combination (as ASN100) neutralize these six toxins. Unlike mice, rabbits are sensitive to all of these toxins and therefore, the prophylactic efficacy and key pharmacokinetic (PK) parameters of ASN100 were determined in this species.

**Methods**

Male New Zealand White rabbits were passively immunized with ASN100 (0.08 to 20 mg/kg of ASN-1 and ASN-2 administered in a 1:1 ratio), 24 hours prior to intratracheal challenge with USA300 CA-MRSA, USA100 HA-MRSA, or two MSSA strains. Survival was monitored for 7 days post-challenge. Alternatively, animals were sacrificed at 12 hours post-challenge and gross necropsy, lung histopathology, and microbiological analyses were performed. In the PK studies, serum and bronchoalveolar lavage fluid (BALF) samples were serially collected from uninfected animals immunized with 40 mg/kg of ASN100. Human mAb and rabbit IgG concentrations were determined by ELISA using anti-idiotope (ASN-1, ASN-2) or human as well as rabbit IgG-specific detection antibodies.

**Dose-dependent prophylactic efficacy of ASN100 against different *S. aureus* strains in a rabbit model of pneumonia**

Bacterial CFU in the lungs and histopathological analysis of stained lung tissues were evaluated from rabbits treated with different protective doses of ASN100 (1.25-20 mg/kg) and sacrificed 12 hours post-challenge with a USA3000 CA-MRSA strain.

**ASN100 reduces lung pathology at early phase of pneumonia**

The three highest doses of ASN100 (1.25-20 mg/kg) were evaluated in a blinded manner for their effect on lung pathology of infected animals sacrificed 12 hours post-challenge with a USA3000 CA-MRSA strain.

**Pharmacokinetics of ASN100 in naïve rabbits**

Serum and BALF antibody concentrations were determined from serially collected samples of rabbits immunized with 40 mg/kg of ASN100.

**Conclusions**

- ASN100 elicited high protective efficacy against all *S. aureus* strains tested in lethal rabbit models of pneumonia irrespective of the antibiotic susceptibility and toxin expression profiles (both pvl- and pvl+) of the challenge strains.
- Significant protection was observed at an ASN100 dose of as low as 1.25 mg/kg, however, the highest doses achieved 100% survival.
- Reduction of bacterial counts, lung edema rate as well as macroscopic and histopathological lung scores of infected animals were the most pronounced at the 20 mg/kg ASN100 dose.
- PK profile of ASN100 was as expected for human mAbs in rabbits. ASN100-concentrations in BALF confirmed efficient penetration to lung epithelial lining fluid, reaching 50% of the serum levels by 24 hours, and peak levels by 48 hours post-dosing.

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**Disclosure and Contact**

Disclosure: All authors are employees and shareholders in the companies performing this work.

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