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Development of a humanized monoclonal antibody against Neisseria gonorrhoeae

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Background

Monoclonal antibody (mAb) 2C7 recognizes a gonococcal lipooligosaccharide (LOS) epitope expressed by > 95% of clinical isolates. Mab 2C7 is bactericidal and hastens gonococcal vaginal clearance in mice. Activity of mAb 2C7 in vivo is dependent on killing by the membrane attack complex of complement. We aim to develop a humanized version of mAb 2C7 with Fc engineered to enhance complement activation and plasma half-life as an immunotherapeutic against AMR Neisseria gonorrhoeae.

Aim/Methods

Humanization of mAb 2C7 was performed by CDR grafting. Humanized mAb 2C7 (HuMab 2C7) variants expressed in ExpiCHO cells were tested for binding to gonococcal LOS and intact bacteria. Fc variants to improve function included: i) human IgG3 (hIgG3; hIgG3 is the most potent complement-activating IgG subclass) and ii) human IgG1 Fc with triple (Q311R/M428E/N434W: called ‘REW’) mutations to enhance complement activation and plasma half-life. We tested the bactericidal activity of HuMab 2C7 variants in vitro, efficacy in a vaginal colonization model of gonorrhea using transgenic (Tg) mice expressing human complement inhibitors (FH/C4BP mice; gonococci bind only human, but not mouse complement inhibitors to escape killing by complement) and half-life using Tg mice that expressed human FcRn (Tg32 mice).

Results

Of 20 purified HuMabs (5 VH x 4 VL combinations), one (VH5/VL2) bound favorably to gonococci. Four back-mutations (one in VH5 and three in VL2) introduced simultaneously further improved binding and bactericidal activity, and this variant was selected for Fc optimization. HuMab 2C7 with IgG3 Fc (IgG1 hinge) showed the most potent bactericidal activity (~2-3-fold reduction in IC50 (concentration of mAb that yielded 50% killing) compared with a human IgG1 Fc with complement-enhancing mutation), and effectively cleared gonococci in FH/C4BP Tg mice following a single intravenous dose of 3.3 µg. HuMab 2C7 with IgG1-REW Fc showed superior bactericidal activity in vitro and almost doubled the half-life (14 d vs 8 d) in Tg32 mice compared to HuMab 2C7 with wild-type Fc.

Conclusions

HuMab 2C7s with Fc engineered to enhance complement activation and half-life may represent promising ‘passive vaccines’ or adjunctive treatments against the rapidly evolving threat of multidrug-resistant gonorrhea.