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AFCo1 provides nasal adjuvant activity against capsular polysaccharides of Neisseria meningitidis serogroup C and Salmonella Typhi

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ABSTRACT

Increasing emphasis is being placed on the mucosal administration of vaccines in order to stimulate mucosal as well as systemic responses. Findings from our group suggest that proteoliposome-derived cochleate (AFCo1) acts as a potent mucosal adjuvant. As an alternative to chemical conjugation, the current study is aimed at determining the benefit of using AFCo1 to improve mucosal and systemic immune responses to capsular polysaccharides from Neisseria meningitidis serogroup C (PsC) and Salmonella Typhi (PsVi). Therefore, intranasal (i.n) immunisations of three doses one week apart with AFCo1 plus PsC or PsVi in adult mice were conducted. High specific anti PsC IgA responses were obtained at site of entry and distant sites such as the vagina. Higher IgG responses after i.n application of PsC or PsVi coadministered with AFCo1 were induced. Our results demonstrate a shift in the isotype pattern elicited in response to PsC and PsVi. Also, the avidities of PsVi antibodies elicited by AFCo1 were higher than those elicited by the polysaccharide alone. B and T cell immunity in spleen against PsVi after a booster intramuscular dose of VaxTyVi were evaluated. In summary, AFCo1 as a nasal adjuvant demonstrated the capability to elicit mucosal, systemic and cellular specific responses against thymus independent antigens

PsVi

oc PsVilgG

INTRODUCTION

MATERIALS AND METHODS

The mucosal surfaces of humans and animals are the major portals of entry and/or sites of diseases caused by microbial pathogens. Mucosal immunizations stimulate both mucosal and systemic immunities, and antigen-specific mucosal immune responses can be expressed at all mucosal sites (2).

Bacterial polysaccharide conjugate vaccines are the gold standard to conferred protection for thyme-independent antigens. Despite efficacy of conjugate vaccines local mucosal immune responses are likely to play an important role in host defense. Adjuvants have been used to improve the immune response to vaccine antigens. Recent progress in the use of mucosal adjuvants to achieve potent immune responses against thyme-independence antigens have been described. AFCo1 had demonstrated be a potent mucosal adjuvant (3,4), a microparticle that showed high stability, antigen delivery capacity and contain multiple MAMPs as immunopotentiators.

The aim of this work was to demonstrate the capacity of AFCo1 to induce systemic, mucosal responses and cellular response against thyme-independent antigens such as: capsular polysaccharides from Neisseria meningitidis serogroup C (PsC) and Salmonella Typhi (PsVi)





Figure 2. Anti PsC systemic and mucosal immune response compared with a conjugated vaccine by i.n route. Anti PsC specific IgA in saliva (A) and IgG (B) in sera, anti PsC IgG subclasses (C), secondary systemic response (IgG, D) and mucosal (E and F) after a booster with PsC alone by nasal route. A p-value <0.05 was considered statistically significant.

Antigens: PsC, PsVi and PL of the meningococcal strain Cu385

(B:4:P1.19,15) was supplied by the manufacturing plant at Finlay Institute, . Cuba (4).

Mice: Female BALB/c and C57BL/6 mice (6-8 weeks old) were housed in the Animal Care Unit during the experiments. Vaccination study: Mice were immunized with AFCo1 mixed with PsC and

PsVi by i.n route in 3 doses, seven days apart. Mice were immunized with a booster dose at 100 days of VaxTyVi vaccine (i.m) or PsC by nasal route. Collection of samples. Saliva were collected 7days after last dose meanwhile

serum samples and vaginal secretion were collected 21 days after the last dose or after booster dose. Antibody and effector T cell analysis: Antibodies specific titers (IgG, IgA and

IgG subclasses) in sera, saliva, and vaginal wash were determined by ELISA. Antibody avidity was measured by ELISA with a caothropic agent (NaSCn). Antibody secreted cells and effector T cells were measure by ELISPOT.

Statistical analysis. Significant differences were determined by a Tukey multiple comparison test using the Graph Pad Prism 4 software (CA, USA).







Figure 4: Antibody secreted cells, antibody afinity, and effector T cell response after a booster dose with VaxTyVi (A) anti PsVi specific IgG kinetics in sera, (B) anti PsVi specific IgM, (C) antiPsVi specific subclasses in sera, (D) anti PsVi specific secreted B cells by ELISPOT. (E) Avidity index of anti PsVi specific IgG antibodies, and (F) effector T cell response anti PsVi specific by ELISPOT. A p-value <0.05 was considered statistically significant.

Summarv

+AFCo1 as a mucosal adjuvant mixed to PsC stimulates mucosal specific responses at site of entry and distant sites like vagina also after a nasal dose of polysaccharide alone •AFCo1 by nasal route generated a systemic specific immune response and mostly IgG against the two polysaccharide antigens (PsC and PsVi) tested

ells

SVi

•AFCo1 as nasal adjuvant influence the isotype distribution to PsC and PsVi changing to a Thymus-dependent behavior

AFCo1 increases anti PsVi antibody afinity in the primary and secondary immune responses

AFCo1 as nasal adjuvant of PsVi is capable to induce B and T cell immunity after a booster dose with a commercial plain polysaccharide vaccine

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