**INTRODUCTION**

Maltose-binding protein (MBP) is part of the maltose/maltodextrin system of *Escherichia coli*. MBP is a relatively small protein (42.5 kDa) approximately 3 x 4 x 6.5 nm in size with surface residues capable of both hydrogen bonding interactions and hydrophobic interactions. Recombinant proteins are often fused to MBP to improve their yield and to increase their solubility. In mice, these fusion proteins showed an enhanced immunogenicity following systemic immunization. More recently, this has been attributed to interaction of MBP with TLR4 on dendritic cells (DCs). TLR4 is also expressed in the enterocytes of the gut. Therefore, we examined if oral administration of MBP-FedF to 4-week-old pigs could be used to induce an immune response against F18+ verotoxigenic *E. coli* in pigs. Also we examined if the oral administration of MBP to pigs is able to induce an immune response. In both experiments chloera toxin was used as oral adjuvant.

**RESULTS**

**Does MBP-FedF induce a protective immune response against F18+ VTEC?**

**Material & Methods**

Two weeks after weaning, six pigs were orally immunized with 1 μg MBP-FedF in PBS, with 56 μg CT as adjuvant. Its negative control two pigs were orally given 1 μl PBS. Five days following the first immunization, the sera from each pig were obtained and the MBP-FedF-specific antibody responses (OD2) were measured. Two weeks after oral immunization, MBP-FedF was administered to each pig by gastric intubation and sera were obtained two weeks following antigen administration. The MBP-specific serum response was measured using a direct sandwich ELISA. Briefly, an optimal dilution of rabbit polyclonal antiserum against MBP was coated on the microtiter plates. After washing, the MBP-FedF was added, followed by the pig sera and goat anti-pig IgM antibody (H+L)-HRP. The plates were washed again and the HRP activity was measured.

MBP binds to pig enterocytes

**Binding of MBP to the intestinal brush border?**

**Material & Methods**

Recombinant MBP (rMBP) positive enterocytes were isolated based on the method described by Ramirez et al. (2004). Approximately 50 000 cells were centrifuged. Subsequently, the cells were resuspended in 1 mL with 50 μg MBP or PBS followed by fixation of the cells with anti-rMBP (sheep: IgG) (1:1000) (MBP specific) at 4°C for 5 to 20 min at room temperature. After each isolation two wells were perfused with 0.05 M Tris-buffered saline.

**Does MBP induce an immune response?**

**Material & Methods**

Two weeks after weaning, six pigs were orally immunized with 1.14 μg MBP in 30 μl PBS; three pigs also received CT as adjuvant. Its negative control two pigs were orally given 1 μl PBS. Five days following the first immunization, the sera from each pig were obtained and the MBP-FedF-specific antibody responses (OD2) were measured. Two weeks after oral immunization, MBP-FedF was administered to each pig by gastric intubation and sera were obtained two weeks following antigen administration. The MBP-specific serum response was measured using a direct sandwich ELISA. Briefly, an optimal dilution of rabbit polyclonal antiserum against MBP was coated on the microtiter plates. After washing, the MBP-FedF was added, followed by the pig sera and goat anti-pig IgM antibody (H+L)-HRP. The plates were washed again and the HRP activity was measured.

**REFERENCES**


**CONCLUSION**

Results showed an enhanced systemic and mucosal immune response against FedF and a significant decrease in the faecal excretion. It is demonstrated that MBP is able to bind to pig enterocytes and after oral uptake can induce a specific intestinal mucosal immune response. Therefore we suggest that the maltose-binding protein has the potential to be a targeting/carrier molecule for mucosal immunisations in pigs.