Preclinical studies related to a phase 1/2a TRIAL to investigate the immunologic impact, anti-tumor efficacy and safety of VXM01, an oral T-cell inducing vaccine, in late stage colorectal cancer patients

Investigate immunologic and tumor expression plasmid

VXM01 is an orally applied T-cell inducing immunotherapy based on live attenuated Salmonella typhi vector carrying a eukaryotic expression plasmid coding for vascular endothelial growth factor receptor 2 (VEGFR-2). A recent phase I trial demonstrated safety, immunogenicity and transient anti-angiogenic activity in advanced pancreatic cancer patients. Notably, sustained T-cell responses have been monitored in patients treated monthly with VXM01 after a one-week initiation treatment course. The purpose of the current studies is to gain more insight into the mode of action of VXM01 in preclinical models of colorectal cancer as well as in patients with metastatic colorectal cancer.

**RESULTS**

Quantification of immune cell infiltrates and PD-L1

Figure 4: Immunohistochemistry Staining anti-CD3

A) Empty Vector

B) VXM01 treated animal

CD3 positive cells appear in brown color (see arrow for example)

Eukaryotic expression plasmid

- Encodes desired target antigen cDNA
- Drives strong expression of target antigen in infected cells

Oral vaccine naturally infects cells in the gut

Live attenuated vaccine strain

- Oral administration induces target antigen in infected cells within the Peyer’s patches

VXM01m vaccine

Objective:

Evaluation of the antitumor activity of VXM01m in Balb/c mice bearing subcutaneous CT26 colon tumor cells.

Correlation of tumor volume and immune cell infiltration in tumor tissue

Figure 6: Correlation tumor size and CD3+ cell infiltration in tumor tissue

Fig 6 A and B show the number of CD3+ cells/mm2 tissue section in correlation to tumor volume. One dot represents data from one animal. A) Blue dot = animals treated with empty vector; B) red triangle = animal treated with VXM01m+CV vaccine.

**CONCLUSIONS**

VAXIMM - VXM01 vaccination leads to...

- Significantly reduced tumor size
- Induction of vacccination specific T cells
- 2-fold increase of immune cell infiltrates in the tumor

VXM01 vaccination has significant impact on the immune cell related control of CT26 tumor growth.

**FUTURE PROJECTS:**

1. **PHASE IIb CLINICAL TRIAL**

Objective:

Evaluation of VXM01 treatment in 24 colorectal cancer patients with liver metastases.

- Investigate immunologic and tumor responses.

Endpoints:

- safety and tolerability
- clinical response

Immunological endpoints:

- tumor infiltrating lymphocytes
- tumor vasculature by immunohisto-chemistry on serial liver metastasis biopsies.
- peripheral immune response (ELISpot)

**STUDY DESIGN**

Evaluation of the antitumor activity of VXM01m in Balb/c mice bearing subcutaneous CT26 colon tumor cells.

Correlation of tumor volume and T cells

Figure 7: Correlation tumor size and VEGFR2+ T cells (pentamer analysis splicenocytes)

Fig 7 A and B show (% VEGFR2+T-cells) in correlation to tumor volume. One dot represents data from one mouse. A) Blue dot = animals treated with empty vector; B) red dot = animal treated with VXM01m+CV vaccine.

**RESULTS**

Correlation of tumor volume and immune cell infiltration in tumor tissue

Figure 5: Immune cell infiltrates and PD-L1 mean fold induction vaccinated compared to negative control group.

Data derived from absolute cell count/tissue area (mm2) of immunohistochemistry staining for COB, Foxp3, CD8, PD-1, and PD-L1 single stainings, scanned at x200 magnification on Aperio scanner using Modul Halo Immune Cell Proximity modules.

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Phase IIa clinical trial started.

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