OMX-0407, a highly potent SIK3 inhibitor, sensitizes tumor cells to cell death and eradicates immune-checkpoint resistant tumors synergistically in combination with PD-1 inhibition

**Introduction**

Using the 10Tage screening platform, salt-inducible kinase 3 (SIK3) was recently identified as a novel epigenetic modulator in cancer therapy. SIK3, a serine/threonine kinase of the AMP-activated protein kinase (AMPK) family, is known for regulating the NF-κB-driven gene landscape through phosphorylation of class IIa histone deacetylases (HDACs), causing the tumor to evade death receptor-mediated killing.

Here we report that OMX-0407, an orally active, single-digit nanomolar inhibitor of SIK3 ablates downstream pro-survival signaling of the SIK3 HDAC4/5 NF-κB axis and potentiates caspase-mediated necroptotic death in murine and human tumor cell lines. OMX-0407 dose-dependent suppression of intratumoral NF-κB activity was shown in vitro and in vivo with an MC38 NF-κB-luc reporter cell line. OMX-0407 monotherapy results in significant tumor growth inhibition (TGI) as well as prolonged survival in the high-immune-infiltrated syngeneic mouse colorectal carcinoma model MC38. Besides its direct inhibitory effects on cancer cells, OMX-0407 reprograms the tumor microenvironment (TME) by strongly decreasing regulatory T cells (Treg) and M2-polarized macrophages in the tumor bed, while not affecting the peripheral T-cell compartment. Using immune checkpoint inhibitor resistant breast (EMT6) and lung (KLN205) cancer models, we demonstrate that OMX-0407 and anti-PD-1 therapy act synergistically by combining the sensitization towards cell death with a reduction in immunosuppressive TME and an increase in cytotoxic T-cell activity. OMX-0407 is an outstanding therapeutic option to overcome TME-induced immune evasion and anti-PD-1 resistance, which will be clinically investigated in the near future.

**Results**

SIK3 inhibits death receptor mediated apoptosis in tumor cells

A) SIK3-KO enhances death receptor mediated tumor cell death

**Conclusion**

OMX-0407 is a strong inhibitor of SIK3 kinase that inhibits the phosphorylation of HDAC4/5 in a dose-dependent manner and thereby abrogates the nuclear activity of pro-tumorigenic transcription factor NF-κB in tumor cells both in vitro and in vivo.

Downregulation of the SIK3-HDAC4/5-NF-κB pathway with OMX-0407 potentiates apoptosis by death receptor ligands, such as TRAIL or TNF, in vitro in tumor cell lines of different origin.

Pharmacokinetics of OMX-0407 in tumor tissue correlates with intra-tumoral abrogation of the HDAC4/5-NF-κB axis.

OMX-0407 shows strong efficacy as monotherapy in the syngeneic tumor model MC38 with a marked repolarization of the tumor microenvironment towards an anti-tumor immune profile.

OMX-0407 acts synergistically with PD-1 blockade in immune checkpoint inhibitor-resistant syngeneic breast and lung cancer models.

The ability of OMX-0407 to repolarize the tumor microenvironment and sensitize tumor cells to death receptor-mediated apoptosis, suggests it as a great potential for combination with high tumor immune model resistance, as monotherapy and in combination with anti-PD-1/PD-L1 immune checkpoint blockade.

OMX-0407 will enter clinical trials in patients with advanced cancer in 2022.

**OMX-0407 inhibits SIK3-triggered phosphorylation of HDAC4/5 and the associated transcriptional activity of NF-κB**

A) OMX-0407 demonstrates dose-dependent depletion of histone deacetylase 4/5.

B) OMX-0407 potentiates TNF-mediated activity of NF-κB pathway.

C) OMX-0407 inhibits transcriptional activity of NF-κB in MC38 tumor cells.

**OMX-0407 reprograms the tumor microenvironment and shows strong anti-tumor efficacy in monotherapy and in combination with PD-1 blockade**

A) Strong anti-tumor efficacy and prolonged survival by OMX-0407 therapy

B) Repolarization toward an anti-tumor immune environment

C) Combination therapy of OMX-0407 and PD-1 blockade acts synergistically in the immune-excluded breast cancer model EMT6 and the immune-excluded lung cancer model

**Statistical information**

Statistical analysis was performed using GraphPad Prism 8.0. For survival curves, Kaplan-Meier tests were used with the log-rank (Mantel-Cox) test for multiple comparisons. Significant differences were set at *p < 0.05, **p < 0.01, ***p < 0.001 compared with DMSO-ctrl.

A) Tumor growth inhibition/remission in the immune-excluded breast cancer model EMT6 and the immune-excluded lung cancer model

A) Strong anti-tumor efficacy and prolonged survival by OMX-0407 therapy

B) Repolarization toward an anti-tumor immune environment

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