Anti-cancer effects of artesunate in a panel of chemoresistant neuroblastoma cell lines
Michaelis, M.; Kleinschmidt, M.C.; Barth, S.; Breitling, R.; Mayer, B.; Deubzer, H.; Witt, O.; Doerr, H.W.; Cinatl, J.; Cinatl Jr., J.

IMPORTANT NOTE: You are advised to consult the publisher's version (publisher's PDF) if you wish to cite from it. Please check the document version below.

Document Version
Publisher's PDF, also known as Version of record

Publication date:
2009

Link to publication in University of Groningen/UMCG research database

Citation for published version (APA):

Copyright
Other than for strictly personal use, it is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), unless the work is under an open content license (like Creative Commons).

Take-down policy
If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

Downloaded from the University of Groningen/UMCG research database (Pure): http://www.rug.nl/research/portal. For technical reasons the number of authors shown on this cover page is limited to 10 maximum.
We have previously demonstrated that non-small cell lung cancer cell, H460, had strong drug resistance to TGFβ and can grow and metastasize successfully in animal model. Despite the fact TGFβ can suppress the growth of gastric carcinoma and hepatoma cells, it also promotes the epithelial mesenchymal transition (EMT) and/or metastasis of liver and breast cancers. We therefore were interested to know would TGFβ also promote the EMT and/or tumor invasion. According to our early observation, PPARγ was already known to play a critical role in the early development of TGFβ resistance of H460. Our current results showed that TGFβ-induced cell scattering of H460 first appeared at day 3 after the TGFβ treatment followed by a morphological shift (from round to fibroblast or spindle-like shape) at day 7 and 14. The results clearly demonstrated a TGFβ-induced EMT in H460. Seven days after TGFβ treatment, the migration and invasion of H460 were significantly increased in accompany with the induced expression of PPARγ and cell survival. The up-stream regulators (p38, ERK, p38α, and COX-2) of PPARγ were also activated (phosphorylated) by TGFβ at early time points (1−6 h). To further confirm the role of PPARγ in TGFβ-induced EMT and cell invasion in H460, we added PPARγ inhibitor (GW9662) into TGFβ-treated H460 and found that not only survival of H460 was decreased, TGFβ-induced EMT and cell invasion were also interrupted. The results suggested that PPARγ was critical in the EMT induction of H460 from TGFβ-mediated growth inhibition and also promoted TGFβ-induced EMT and cell invasion in H460. In overall, TGFβ-induced EMT and cell invasion in H460 have been confirmed and proved to be PPARγ dependent. Results from the study not only provided information about the drug resistance and metastasis of H460 in response to TGFβ treatment but also implied the therapeutic value of PPARγ inhibitor (GW9662) in the treatment of NSCLC.

Conclusions: It is suggested that Compound3 should be considered for further exploration and development and that induction of monoastral formation may not be critical as a predictive biomarker. Taken together, a novel mitotic kinesin Eg5 inhibitor Compound3 may have other mechanisms of action for its growth inhibitory effect on cancer cells and further investigation on alternative biomarkers is necessary to develop Eg5 inhibitors as an anticancer drug.

Background: Artemisinin derivatives are well-tolerated anti-malaria drugs that also exert anti-cancer activity. Here, we investigated artemisinin and its derivatives dihydroartemisinin and artemesin in a panel of chemoresistant neuroblastoma cell lines.

Materials and Methods: Cell viability was determined by MTT assay or by determination of ATPase activity. Apoptosis was examined by staining for activated caspase-3 and detection of cells with low DNA content (sub-G1) by flow cytometry. Bioinformatic analysis of gene microarray data was used to identify genes relevant for neuroblastoma cell response to artemisinin.

Results: Only dihydroartemisinin and artemesin affected neuroblastoma cell viability with artemesin being more active. Of 16 cell lines and two primary cultures, only UKF-NB-3’CDDP showed low sensitivity to artemesin. Artesunate induced apoptosis and reactive oxygen species in neuroblastoma cells. L-Buthionine-S-R-sulfoximine, an inhibitor of GCL (glutamate-cysteine ligase), mesothelial killed in part UKF-NB-3’CDDP cells to artemesin. This finding together with bioinformatic analysis of gene expression signatures showed that this pathway is involved in artemesin resistance.

Conclusions: These data indicate that neuroblastoma represents a artemesin-sensitive cancer entity including chemoresistant cells. Characteristic gene expression signatures based on a previous analysis of artemesin resistance in the NCI60 cell line panel clearly separated UKF-NB-3’CDDP from the other cell lines.