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Medizinische Fakultät Mannheim

Dissertations-Kurzfassung

Enzastaurin inhibits invasion and metastasis in non-small cell lung cancer via inverse regulation of essential genes relevant for tumour progression

Autor: Alexandra Körner

Institut / Klinik: Abteilung für Experimentelle Chirurgie und Molekulare Onkologie Solider Tumore

Doktormutter: Prof. Dr. H. Allgayer

Worldwide, lung cancer is the most lethal form of cancer for men and women. Especially in NSCLC, occurring with an incidence rate of 75 % of all lung cancers, early resistance to conventional chemotherapy and radiotherapy and its fast progression makes treatment difficult. There is still a great need for novel compounds for the treatment of NSCLC which show less resistance to therapy, acceptable side-effects and better overall survival rates. Enzastaurin is a serine/threonine kinase inhibitor blocking specifically PKC β and PI3K/Akt signalling pathways, known to be activated in NSCLC and correlating with tumour progression. In early clinical trials, Enzastaurin has been well tolerated with no significant Grade 3 or 4 toxicities. Although Enzastaurin is considered promising for lung cancer therapy, very little is known about the effect of Enzastaurin on the differential regulation of specific genes associated with invasion and metastasis, and as to what molecular patterns might indicate or predict response or resistance to Enzastaurin therapy in NSCLC.

The present study focuses on the ability of Enzastaurin to inhibit migration, invasion, and *in vivo* metastasis in NSCLC, and to explore first mechanisms contributing to Enzastaurin-induced inhibition of migration, invasion, and metastasis-related processes in NSCLC. The ability of Enzastaurin to inhibit migration, invasion, and metastasis and to target molecules involved in tumour progression was investigated in NSCLC by performing wound-healing, matrigel, and *in vivo* chorionallantoic membrane (CAM) assays as well as by real-time validated microarray. In this study, we show that Enzastaurin significantly reduces migration and invasion in NSCLC. Furthermore, we for the first time elucidate that Enzastaurin inhibits *in vivo* metastasis to lungs and liver in NSCLC. Genes promoting cancer progression (u-PAR, Axl, HIF 1α, VEGF C) and tumour suppression (VHL, RASSF1, FHIT) in NSCLC were significantly down- or upregulated upon Enzastaurin-treatment in H460, A549, and H1299 cells, respectively. Moreover, gene expression arrays suggest that Enzastaurin negatively regulates a panel of genes which have been implicated as important deregulated genes in NSCLC in previous publications. By luciferase, gelshift, and chromatin immunoprecipitation analysis, respectively, we show for the first time that Enzastaurin transcriptionally controls u-PAR expression by promoter inhibition through Sp1, Sp3, and c-Jun (AP-1). Furthermore, we show that Enzastaurin transcriptionally controls Axl expression via suppressing the binding of Sp family transcription factors to the Axl promoter. siRNA knockdown of u-PAR and Axl, respectively, re-sensitises relatively resistant NSCLC cells to Enzastaurin and further reduces the basal invasive potential of these cells, suggesting u-PAR and Axl as potentially clinically relevant markers for Enzastaurin resistance. Taken together, the regulation of genes, normally up- or downregulated in NSCLC and playing an important role in the pathogenesis and metastasis of NSCLC, may reflect a promising potential of this compound for the treatment of NSCLC and for survival benefits of patients.