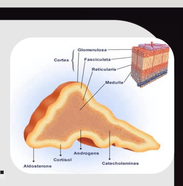
EGFR as potential new molecular target in the medical treatment of Adrenocortical Cancer

Teresa Gagliano[1], Francesco Balboni[1], Carmelina Di Pasquale[1], Erica Gentilin[1,2], Katiuscia Benfini[1], Simona Falletta[1], Marta Bondanelli[1], Paola Franceschetti[1], Carlo Feo[3], Ettore degli Uberti[1,2], Maria Chiara Zatelli[1,2]

[1]Section of Endocrinology, Department of Medical Sciences, University of Ferrara, Via Savonarola 9, 44121 Ferrara, Italy; [2]Laboratorio in rete del Tecnopolo "Tecnologie delle terapie avanzate" (LTTA) of the University of Ferrara, Via Fossato di Mortara 70, 44121 Ferrara, Italy

[3]Department of Morphology, Surgery and Experimental Medicine, University of Ferrara, Via A. Moro, 8, 44124 Ferrara, Italy

BACKGROUND

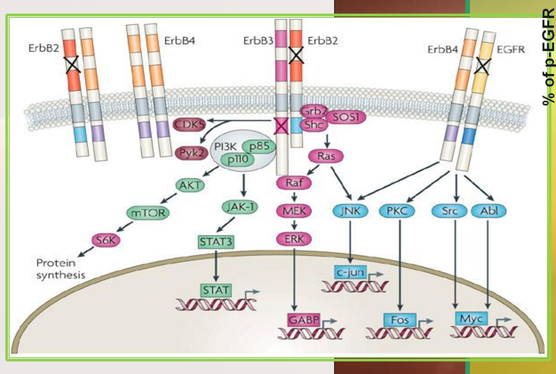


Adrenocortical cancer (ACC) is a rare and aggressive malignancy. Currently the main therapeutic option is surgery, but due to difficult and delayed diagnosis and to the onset of metastases, medical therapy is often tried. ACC treatment is mainly represented by Mitotane alone or in association with chemotherapy, with variable results. Understanding the molecular mechanisms that regulate ACC proliferation could be useful to identify new therapeutic options.

Sunitinib, a multitarget tyrosin-Kinase inhibitor, showed controversial results in phase II trials for advanced refractory ACC. It has been previously demonstrated that Epidermal Growth Factor (EGF) increases HSD3B2 expression in ACC cells.

AIM

The aim of our study is to verify whether EGF pathway could represent a target for Sunitinib in human ACC cells. For this purpose we used 2 human adrenocortical carcinoma cell lines (SW13 and NCI-H295 cells) and human adrenal tumor primary cultures.



METHODS

As an in-vitro model we use 2 ACC cell lines, **NCI-H295** and **SW-13**, and human primary cultures.

We evaluate cell viability **by ATPlite** assay.

Protein expression was evaluated by **western blot** and **Surefire assay.**

By a **Caspase 3/7** assay we determined apotosis.

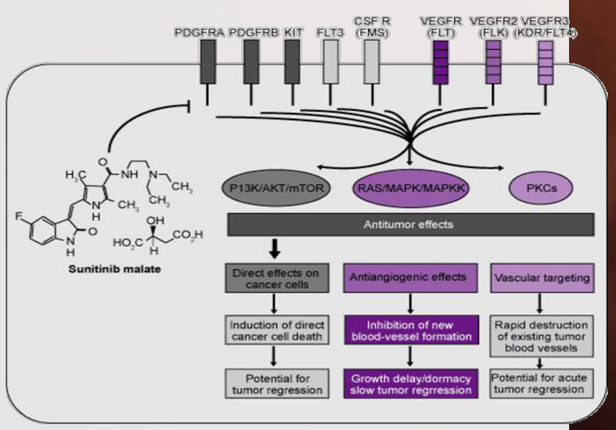
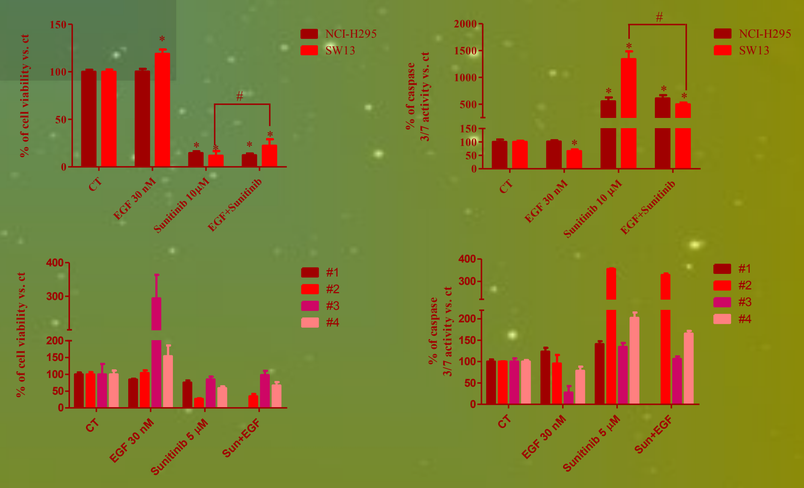


FIG.1

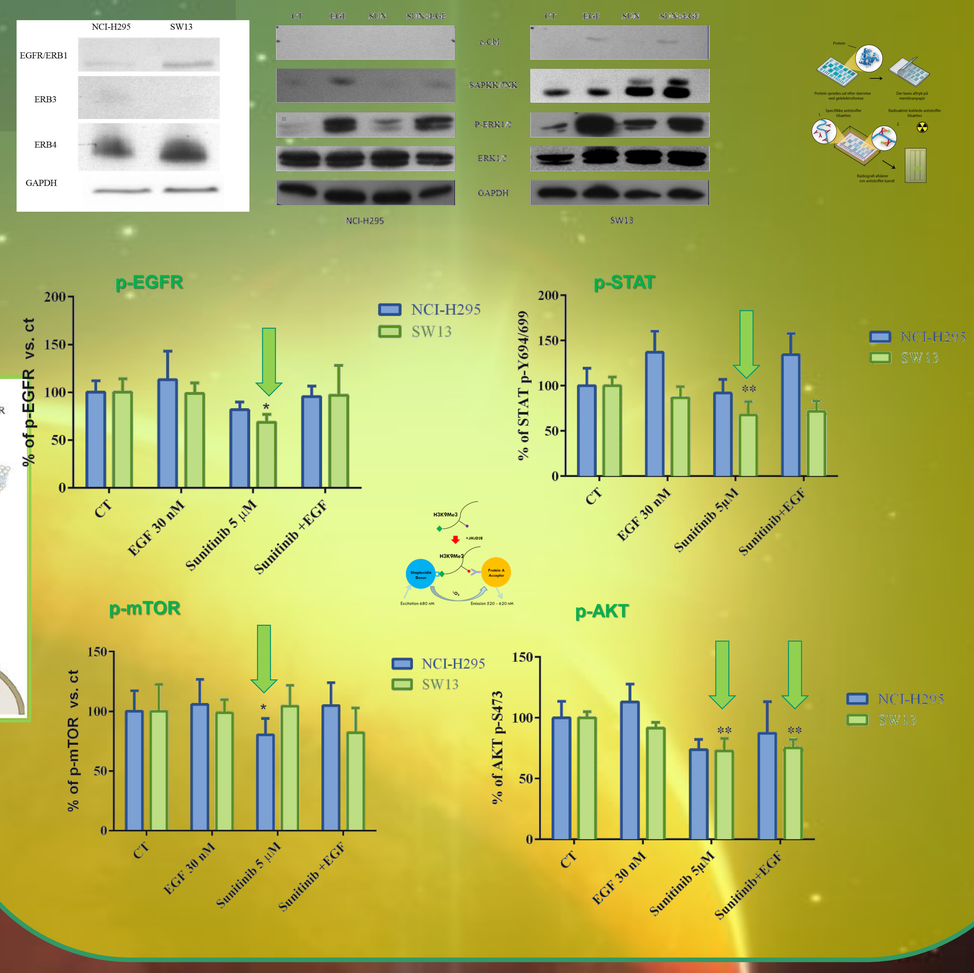
Cell Viability and Apoptosis assay



We found that EGF increased proliferation and reduced apoptosis not only in SW13 but also in adrenal tumour primary culture, while it did not affect NCI-H295 cells. Sunitinib reduced cell viability in both cell lines, being counteracted by EGF in SW13 cells and in the majority of primary cultures analyzed.

FIG.2

Western blot analysis & alpha screen assay



We investigated in both cell lines the expression of EGFR family members, which are more expressed in SW13 cells as compared to NCI-H295 cells. Moreover, we investigated the intracellular signal transduction pathway of EGF in ACC cells. Our results show that in SW13 cells Sunitinib inhibited EGFR phosphorylation on tyrosine 1068, and counteracted EGF-induced phosphorylation of ERK1/2. In SW13 Sunitinib increased the expression of SAPK/JNK leading to caspase 3/7 activation. In NCI-H295 Sunitinib did not reduce EGFR phosphorylation, but inhibits PI3K/mTOR/AKT pathway.

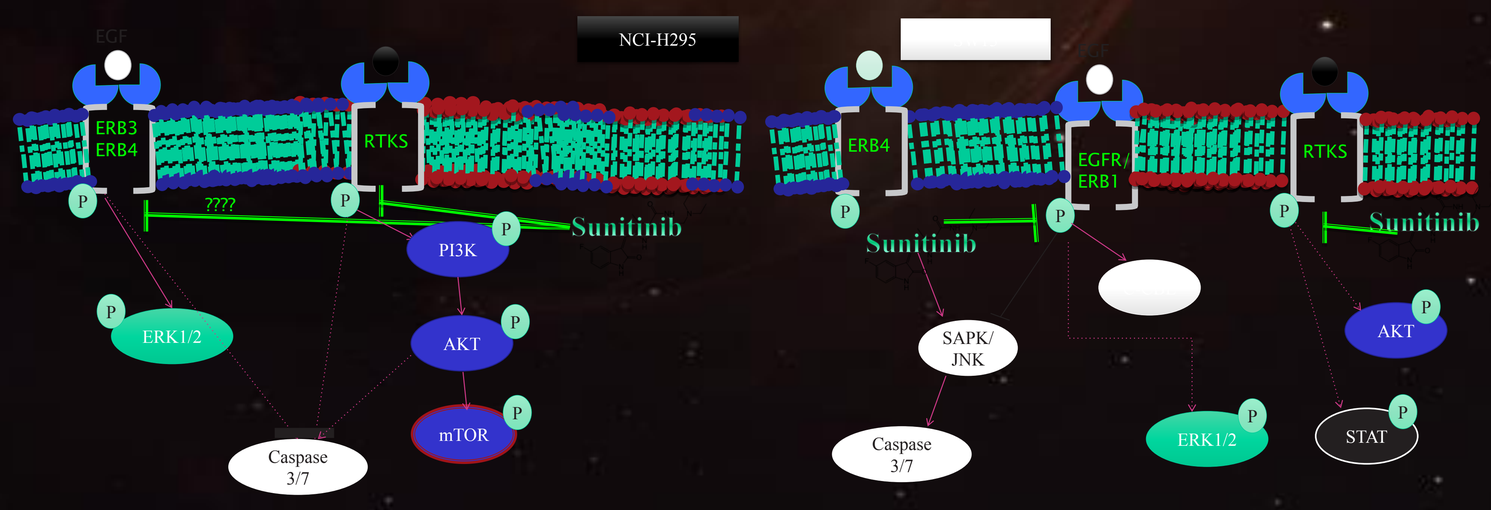
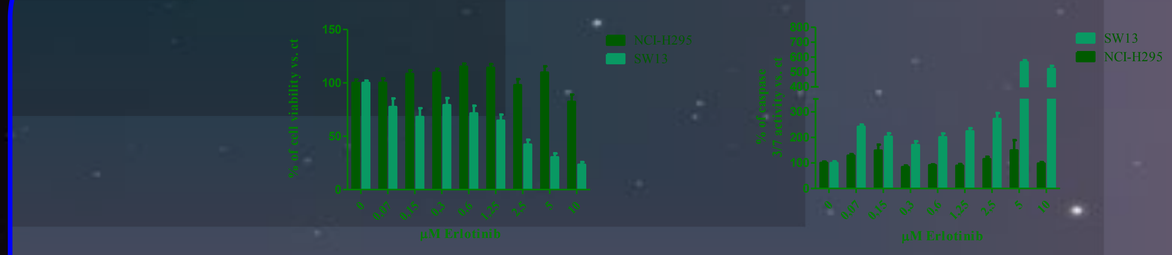


FIG.3

Erlotinib



To verify the involvement of EGFR in regulating ACC cell viability we tested Erlotinib, a selective EGFR inhibitor, in the 2 cell lines. We found that Erlotinib was capable of dose dependently reducing cell viability and activating caspase 3/7 in SW13 cell line, having no effects on NCI-H295 cells.

Conclusion: **EGF may be important in regulating EGFR expressing ACC cell proliferation.**
In conclusion our data suggest that EGF pathway could represent a new molecular target in drug design for treatment of ACC that display enhanced EGFR expression

17 ECE
5-GP-29
Endocrine tumours
Teresa Gagliano
DOI: 10.3252/pso.eu.17ece.2015
Poster presented at: ECE2015 16-20 May 2015 DUBLIN, IRELAND 17th EUROPEAN CONGRESS OF ENDOCRINOLOGY