PhD thesis **„ Combination of ellipticine chemotherapy and α5β1 integrin-targeted therapy in human glioblastoma“** by **Mgr. Eva Martínková** are focused on experimental therapy of glioblastoma multiforme *in vitro*. Thema of the thesis is new and may be of grate practical importance in clinical oncology. The aims of the study were: 1/ to describe cytotoxicity of ellipticine in two glioblastoma cell lines (U87 and U373) and investigate its metabolism and activation in those glioblastoma cell lines; 2/ design a combination therapy of integrin α5β1antagonist and ellipticine for glioblastoma. Author fulfils the aims of the study and results of her work has been published in high quality journals.

I have following questions and remarks:

1/ Sentence „Ellipticine has been shown to be a brain-tumor-specific anti-cancer drug..“ is according my meening too optimistic. Ellipticine is not brain tumor specific and it is better say, that it may be effective on brain tumors, because they are neither clinical studies nor animal experiments in brain tumors with this drug.

2/ You maintained that „ellipticine is not used in clinical practice due to its cardiotoxicity“ but as I know the main its toxic effect is nephrotoxicity and I have found the only one short notes about its cardiotoxicity.

3/ It is not necessary to repeat information which are included in Introduction also in Results and discussion e.g. first paragraph of results.

4/ What means hight of collumns in figure 14D left- percentage of AnexinV+ PI- or all AnexinV+ cells? There is recommended to use only description of results of method e.g. AnexinV binding cells and not terms like apoptotic cells.

5/ How did you explain marked decrease of E2F1 mRNA in Fig. 16B?

6/ Do you have any hypothesis about influence of integrins on senescence?

7/ Is there comparison of results obtained from glioblastoma and from colorectal cell lines suitable?

8/ I think that the sentence „ While p53wt-expressing U87MG cell line answered to ellipticine treatment by senescence induction, in U373 (p53mt) ellipticine provoked apoptotic cell death“ is oversimplification. As show figures 14 and 15 in both cell lines ellipticin induced apoptosis and senescence and difference was between ratio of those two responses.

9/ From which type of glioblastoma, primary or secondary, are cell lines U87MG and U373 derived?

There are only few formal mistakes in thesis:

Figures 5 and 6 are similar and it is not necessary to show both; page 50 „increased Bcl/Bax ratio“ insted of Bcl2/Bax; page 58 glioblastoma is everytime grade IV therefore descriprion of lines „human glioblastoma line, WHO grade IV“ is redundat; page 109 unusual cell phenotype after transfection by 5α - figure 31b should be described; page119 ellipticine is not conventional therapy in glioblastoma because as I know it has never been used in this tumor.

Regardless of my remarkas it is the best PhD thesis that I have evaluated to date and I strongly recommend confer a degree PhD.

Psáry, 28.3.2010

prof.MUDr.Tomáš Eckschlager, CSc.

Dept.pediatric hematology/oncology
University Hospital Motol and 2nd Medical school, Charles University