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**Comparative characterization of yeast and human 3-ketosteroid reductase, two moonlighting proteins that share a job**

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Yeast and human 3-ketosteroid reductase (ERG27 and human HSD17B7, respectively) catalyze the reduction of 3-keto-intermediates of sterol biosynthesis into the corresponding beta-hydroxy compounds[1]. Besides this shared function, each of the two proteins possess an additional ability that up to now has not been reported to be shared with the other protein: yeast ERG27 behaves as a chaperonine-like protein towards oxidosqualene cyclase, an upstream enzyme of ergosterol biosynthesis; HSD17B7 is highly effective in transforming estrone into estradiol. In previous experiments with recombinant yeast strains, the inability of mammalian 3-ketosteroid reductase to protect yeast oxidosqualene cyclase was unambiguously established[2].

In order to distinguish the intrinsic properties of the proteins from those depending on cellular/tissue environment, yeast ERG27 and human HSD17B7were expressed in *E. coli*,and a series of parallel experiments with cell homogenates were designed: (i) proteins would be assayed for their 3-ketoreductase activity or their estrone reductase activity through separate incubation of cell homogenates with radioactive 4-methyl zymosterone or radioactive estrone, respectively; (ii) the inhibitory effect of molecule designed as inhibitors of estrogenic activity of human HSD17B7 would be assayed; (iii) the ability of yeast ERG27 to restore oxidosqualene cyclase functionality in ERG27-deficient cells would be tested by assaying the oxidosqualene cyclase activity of homogenates of cells combined with bacterial homogenates containing yeast 3-ketoreductase. Part of the designed experiments of the project is presented here.

Both proteins proved to be active in transforming radioactive 4-methyl zymosterone into the corresponding hydroxyl derivative, whereas the estrogenic activity was displayed only by the human enzyme. This expected result poses an interesting question about the evolution of the protein: indeed, one wonders when and thanks to the alteration of which structural part this protein became a moonlighting protein and acquired the estrone reduction ability as its second job.

Inhibitors of the estrogenic activity of the human HSD17B7 resulted scarcely active against the 3-ketosteroid reductase activity of the same enzyme, pointing out the requirement of a different designing strategy to synthesize inhibitors of the latter enzymatic activity of the protein.

Experiments on chaperonine-like properties of yeast 3-ketoreductase are in progress.

References

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