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The Influence of Recombinant Tyrosinase Expression on Melanin Production in Human Melanoma cells

Ru-Yu Lin and Chih-Chien Lin*
Department of Cosmetic Science, Providence University, Taiwan.

Introduction:

At presents, there are many testing methods for the skin whitening effect evaluation. Most of the studies were used B16-F10 mouse melanoma cells, but less with human cells due to its lower content of melanin that cannot be used as the main cell line for the whitening testing. In order to have better reliability in the results for the development of cosmetic ingredients, we try to express the recombinant human tyrosinase by pcDNA3.1(+)-hTYR plasmid in human melanoma MeWo cells.

Keywords: melanin, human melanoma cells, tyrosinase, cosmetics

Materials & Methods:

Construction of pcDNA3.1(+)-hTYR

The recombinant human tyrosinase gene was cloned as pcDNA3.1(+)-hTYR plasmid (Figure 1), then transfected into human melanoma MeWo cells to test the effects of recombinant human tyrosinase expression on cells.

Measurement of cell viability

Cell viability studies were performed using the MTT assay. The Detroit 551 cells were plated at a density of 1 × 105 cells/well. Add hydrosol for 24h. Next, MTT solution was added to the medium, and the cells were incubated at 37 ℃ for 1h. The MTT-containing medium was removed, and the cells were solubilised in DMSO for 30 mins. The absorbance was measured at 540 nm using an ELISA reader to determine the cell viability.

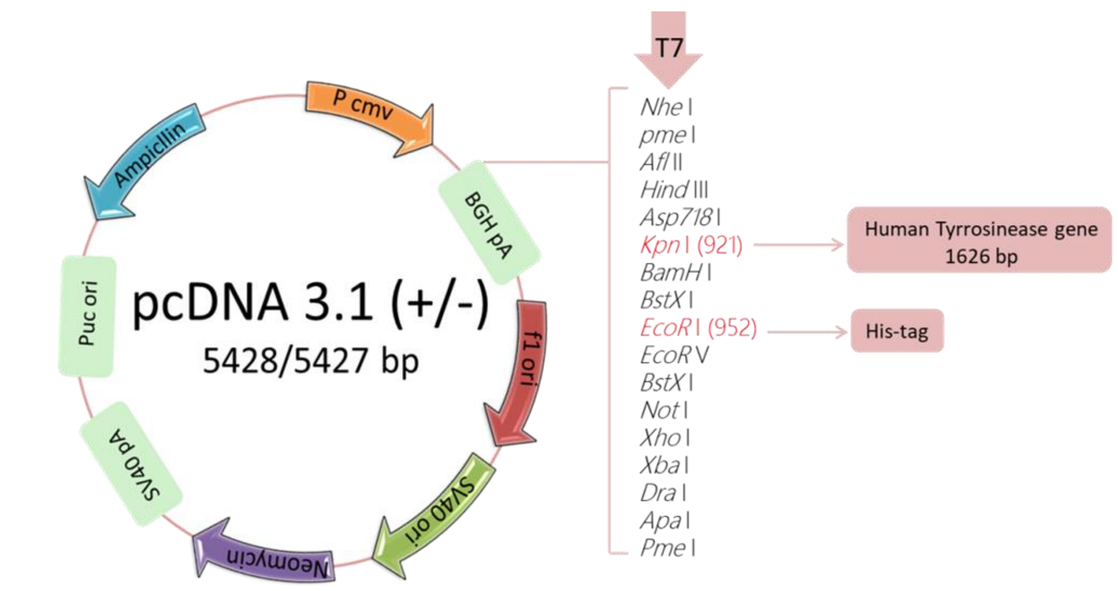

Figure 1. The construction map of pcDNA3.1(+)-hTYR. The pcDNA3.1(+)-hTYR plasmid has a human tyrosinase gene with 1626 bp.

Results & Discussion:

Cellular tyrosinase activity

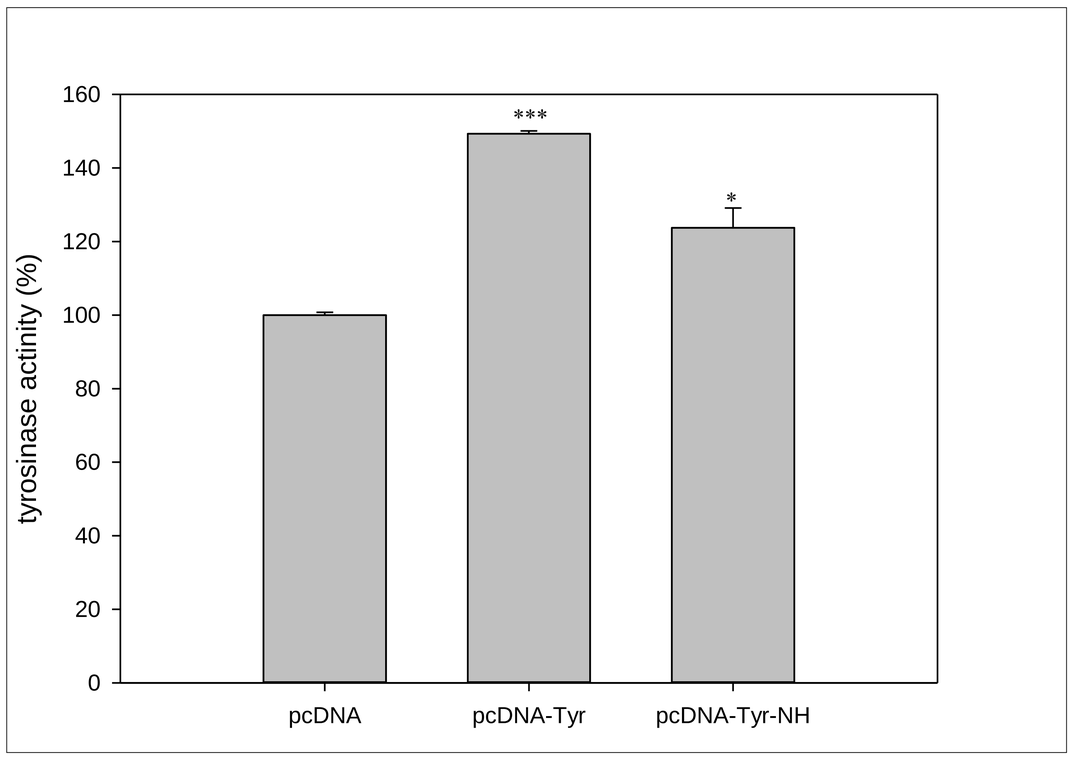

Figure 2. The cellular tyrosinase of plasmid transfected human MeWo cells.

Effects of Rottlerin on the rhTYR expressing cells

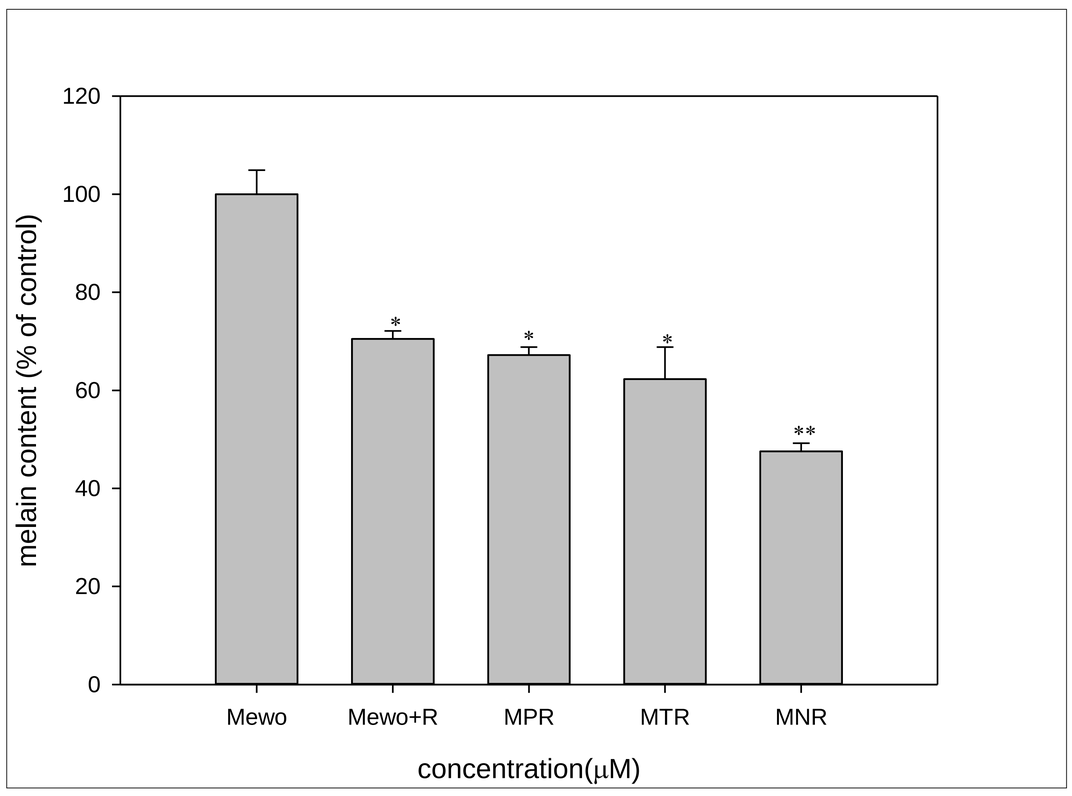

Figure 3. Melanin content of Rottlerin treated rhTYR expressing cells.

Conclusions:

In summary, we suggested that there may have some mechanism to suppress the overproduction of melanin in melanocytes.

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