## ABSTRACT (English)

**Objective:** Ovarian cancer can be considered as an endocrine-related cancer. The pleiotropic effects of estrogen are mediated by estrogen receptor ERα and ERβ. However, expression rates of ERs are limited of prognostic values. A subfamily of orphan receptors, estrogen receptor-related receptors (ERRs), has been demonstrated to modulate transcription of some estrogen responsive genes via variants estrogen responsive elements (EREs). This study was performed to determine whether human estrogen receptor-related receptor -α, -β, -γ and their isoformal transcripts (hERRα-1 and hERRβ-2) might be associated with ovarian cancers. The association between the expression of ERRs and clinicopatholigical parameters were also analyzed.

**Materials and Methods:** To analyze the subcellular location of ERRs, a Green Fluorescent Protein (GFP)-reporter plasmid of hERRα was constructed and transfected into the cell lines SKOV-3 and OVCAR-3. To distinguish the cellular cytoplasm and nucleus, DAPI staining was performed. A Double-color confocal scan microscope was used to observe the reporter signal. Western-Blot analysis was performed to detect the HA-tag-hERRγ fusion protein in the transfected cell lines. Two-step LightCycler real-time RT-PCR was used to quantitatively analyze the expression of hERRα, hERRβ, hERRγ ,hERα and hERβ mRNA in ovarian cancer cell lines SKOV-3, OVCAR-3 OAW-42, MDAH-2774, ES-2 as well as 33 primary ovarian cancer samples and 12 normal ovarian tissue samples. The immunochemistry and immunohistology were also used to detect the ERRs protein expression. The serum CA-125 was detected in all samples by ELISA assay. To discuss the potential role of ERRs as biomarkers in ovarian cancer, the expressions of ERRs family were analyzed with multi-clinicopathological parameters and overall survival analysis. Progression-free survival time and overall survival time of patients with different expression of ERRs were analyzed by the Kaplan-Meier method.

**Results:** hERRα-GFP fusion protein showed hERRα expressed chiefly in the cell nucleus. In ovarian cancers, 19 cases (57.6%) were hERRα mRNA and protein positive-expression, 3 cases (9.1%) were hERRβ mRNA and protein positive-expression and 16 cases (48.5%) were hERRγ mRNA and protein positive-expression; in normal ovaries 2 cases (16.7%), 0 cases (0%) and 4 cases (33.3%), respectively. Compared with the normal ovaries, higher expression rate of hERRα and hERRγ were detected in patients with ovarian cancer (p=0.002 and p=0.045). A high co-expression of hERα and hERRα was found in the ovarian cancer. High expression of ERRα seems to be

associated with a lower expression of hERα. Compared with normal ovaries, the ratio of hERα/hERRα seems to be decreased in the ovarian cancer. Combination of the hERRα expression and serum CA-125 level analysis showed CA-125 level in hERRα positive-group (1954.79 U/ml) was significantly higher than in the hERRα negative-group (448.56 U/ml) (p=0.012); However, the difference of serum CA125 levels between hERRβ positive - and negative- group (p=0.795) and hERRγ positive- and negative-group (p=0.515) are not significant. In the multivariate analysis, expression of hERRα was an independent prognostic factor for poor survival (RI, 3.02: 95% CI: 1.27 to 6.06). High expression of hERRα is associated with increasing serum CA-125, advanced FIGO stage, poor differentiation (grade) and decreased overall survival time. High expression of hERRγ is associated with early FIGO stage (p=0,040) and longer progression-free survival time (p=0,020). Moreover, increasing expression of hERRγ tends to be correlated with decreased serum CA-125 and low ascites volume. Survival analysis showed that the hERRα-positive group had a reduced overall survival time (p=0.015) whereas the hERRγ-positive group showed a longer progression-free survival time (p=0.020).

**Conclusion**: Based on the results showed in my dissertation, I concluded follows: ERRs are highly expressed in the cell nucleus of ovarian cancer samles. Both hERRα and hERRγ seem to play an important role in the ovarian cancer. High expression of hERRα is strongly associated with more advanced clinical stages of ovarian cancer and more aggressive malignant biological behavior. Moreover, hERRα and hERRγ are potential endocrinal therapy targets of ovarian cancer.

## Key words

**Orphan receptor; estrogen receptor-related receptor; ovarian cancer, tumor marker, prognosis**