

# Control of differentiation and programmed cell death: The mammalian ovary as a model system

## Department of Molecular Cell Biology

Tel. 972 8 934 3713      Fax. 972 8 934 4125  
 E-mail: abraham.amsterdam@weizmann.ac.il

Our research is focused on the cellular and the molecular mechanisms, which control programmed cell death in the normal and the neoplastic ovary. We analyzed the effect of pro- and anti-apoptotic stimuli of physiologic and pharmacological nature such as gonadotropin hormones/cAMP, steroid hormones, growth factors and their receptors, cytokines, cytotoxic drugs used in chemotherapy and possible cross talk among the different stimuli. The second major topic of our research is revealing the mechanism exerted by gonadotropins on differentiation of follicular ovarian cells.

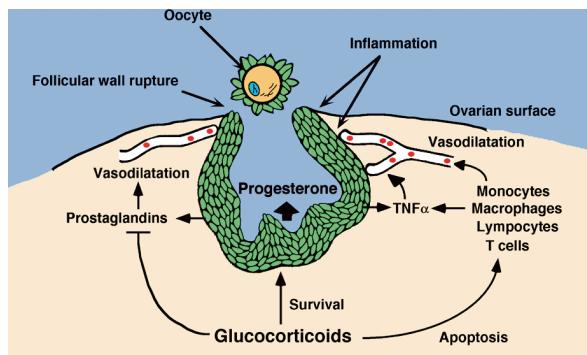
### The anti-inflammatory action of glucocorticoids is mediated by cell type specific regulation of apoptosis.

Glucocorticoids play a major role in attenuation of the inflammatory response. These steroid hormones are able to induce apoptosis in cells of the hematopoietic system such as monocytes, macrophages and T-lymphocytes that are involved in the inflammation reaction. In contrast, it was discovered recently that in glandular cells such as the mammary gland epithelia, hepatocytes, ovarian follicular cells and in fibroblasts glucocorticoids protect against apoptotic signals evoked by cytokines, cAMP, tumor suppressors and death genes. The anti-apoptotic effect of glucocorticoids is exerted by modulation of several survival genes such as Bcl-2, Bcl-xL and NF kappaB, in a cell type-specific manner. Moreover, up regulation or down regulation of the same gene product can occur in a cell type-dependent manner following stimulation by glucocorticoids. This phenomenon is probably due to composite regulatory cross-talk among multiple nuclear coactivators or corepressors, which mediate the transcriptional regulation of the genes, by their interaction with the glucocorticoid receptor. These observations suggest that the anti-inflammatory action of glucocorticoids is exerted by two complementary mechanisms: on the one hand, they induce death of the cells that provoke the inflammation, and on the other hand they protect the resident cells of the inflamed tissue by arresting apoptotic signals (Fig. 1).

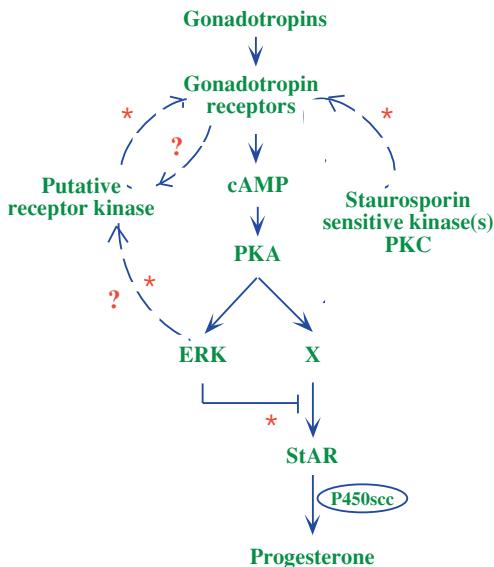
### Mechanisms of gonadotropin desensitization

The gonadotropin hormones, FSH and LH exert a major effect on ovarian and testicular function through interaction with specific seven transmembrane domain glycoprotein receptors.

Desensitization to the hormones, which can occur both *in vivo* and *in vitro*, is essential for prevention of overstimulation of the gonadal cells. The long-term process of desensitization to the gonadotropin hormones is probably mediated, in part, by extensive clustering and internalization of the hormone receptor complex. Short-term desensitization may occur as a result of phosphorylation of serine or threonine residues on the receptor molecules, although a specific receptor kinase has not yet been identified. Recently, we have discovered a novel mechanism of gonadotropin desensitization, which is exerted by down regulation of StAR expression and steroidogenesis mediated by MAPK activation as a result of hormone-receptor interaction, cAMP accumulation and PKA activation. Thus, PKA not only mediates gonadotropin-induced steroidogenesis, it also activates the down-regulation mechanism that can silence steroidogenesis under certain conditions. Moreover, our findings raise the possibility that activation or inhibition of ERK by other pathways could be an important mechanism for diminution or amplification of gonadotropin-stimulated steroidogenesis (Fig. 2). This could contribute to functional luteolysis, a process in which luteinized granulosa cells show reduced sensitivity to LH despite maintenance of LH receptors, or to up-regulation of the steroidogenic machinery during luteinization of granulosa cells. (In collaboration with Dr. Rony Seger)



**Fig. 1** Anti-inflammatory action of glucocorticoids during follicular rupture, suggesting dual regulation (anti-and pro-apoptotic effects) on the cellular components of the ovulatory process.



**Fig. 2** Schematic representation of the signaling pathways controlling gonadotropin-induced steroidogenesis. The stars indicate possible involvement of phosphorylation that may lead to desensitization to gonadotropin stimulation. (Modified from Seger et al., 2001)

### Selected Publications

Amsterdam, A., Gold, R.S., Hosokawa, K., Yoshida, Y., Sasson, R., Jung, Y., Kotsuji, F. (1999) Crosstalk among multiple signaling pathways controlling ovarian cell death. *Trends Endocrinol. Metab.* 10, 255-262.

Yoshida, Y., Hosokawa, K., Dantes, A., Tajima, K., Kotsuji, F. and Amsterdam, A. (2000) Theophylline and cisplatin synergize in down regulation of BCL-2 induction of apoptosis in human granulosa cells transformed by a mutated p53 (p53 val135) and Ha-ras oncogene. *Int. J. Oncol.* 17, 227-235.

Almog, B., Gold, R., Tajima, K., Dantes, A., Salim, K., Rubinstein, M., Barkan, D., Homburg, R., Lessing, J.B., Nevo, N., Gertler, A. and Amsterdam, A. (2001) Leptin attenuates follicular apoptosis and accelerates the onset of puberty in immature rats. *Mol. Cell. Endocrinol.* 183, 179-191.

Amsterdam, A., Hanoch, T., Dantes, A., Tajimaa, K., Strauss III, J.F., Seger, R. (2001) Mechanisms of gonadotropin desensitization. *Mol. Cell. Endocrinol.* (in press).

Amsterdam, A., Kannan, K., Givol, D., Yoshida, Y., Tajima, K. and Dantes, A. (2001) Apoptosis of granulosa cells and female infertility in achondroplastic mice expressing mutant fibroblast growth factor receptor 3 (G374R). *Mol. Endocrinol.* 15, 1610-1623.

Amsterdam, A., Sasson, R. (2001) The anti-inflammatory action of glucocorticoids is mediated by cell type specific regulation of apoptosis. *Mol. Cell. Endocrinol.* (in press).

Ginath, S., Menczer, J., Friedmann, Y., Aingorn, H., Aviv, A., Tajima, K., Dantes, A., Glezerman, M., Vlodavsky, I. and Amsterdam, A. (2001) Expression of heparanase, Mdm2, and erbB2 in ovarian cancer. *Int. J. Oncol.* 18, 1133-1144.

Sasson, R., Tajima, K. and Amsterdam, A. (2001) Glucocorticoids protect against apoptosis induced by serum deprivation, cyclic adenosine 3',5'-monophosphate and p53 activation in immortalized human granulosa cells: involvement of Bcl-2. *Endocrinology* 142, 802-811.

Seger, R., Hanoch, T., Rosenberg, R., Dantes, A., Merz, W.E., Strauss, J.F., 3rd and Amsterdam, A. (2001) The ERK signaling cascade inhibits gonadotropin-stimulated steroidogenesis. *J. Biol. Chem.* 276, 13957-13964.

Tajima, K., Babich, S., Yoshida, Y., Dantes, A., Strauss, J.F., 3rd and Amsterdam, A. (2001) The proteasome inhibitor MG132 promotes accumulation of the steroidogenic acute regulatory protein (StAR) and steroidogenesis. *FEBS Lett.* 490, 59-64.

Yoshida, Y., Hosokawa, K., Dantes, A., Kotsuji, F., Kleinman, H.K. and Amsterdam, A. (2001) Role of laminin in ovarian cancer tumor growth and metastasis via regulation of Mdm2 and Bcl-2 expression. *Int. J. Oncol.* 18, 913-921.

Amsterdam, A., and Sasson, R. (2001) The anti-inflammatory action of glucocorticoids is mediated by cell type specific regulation of apoptosis. *Mol. Cell. Endocrinol.* (in press)

Tajima, K., K. Hosokawa, Y. Yoshida, A. Dantes, R. Sasson, F. Kotsuji and A. Amsterdam (2001) Establishment of follicle-stimulating hormone responsive cell lines by transfection of preovulatory human granulosa cells with mutated p53 (p53val135) and Ha-ras genes. *Mol. Hum. Reprod.* (in press).

### Acknowledgements

The work was supported by grants from Israel Academy of Science, The Levine Center for applied research and the Yad Avraham center for cancer Research at WIS.

AA is the incumbent of the Joyce and Ben B. Eisenberg Professorial Chair of Molecular Endocrinology and Cancer Research.