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Cell Cycle News & Views

CDK1 links to RARγ in treatment response of cancer cells

Comment on: Hedblom A, et al. Cell Cycle 2013; 12:1251–66;
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Cancer recurrence is associated with treatment failure and is the main cause of death,[1] and thus remains to be a major clinical challenge. Elevated level of the cyclin-dependent kinase 1 (CDK1) correlates with cancer recurrence and treatment resistance in patients with breast cancer or colorectal cancer.[2,3] It is proposed that defects in CDKs may lead to the accumulated genetic defects, which render cells less sensitive to drug-induced growth inhibition and apoptosis.[4] CDK1 is the most essential CDK, as it alone is sufficient to drive cell division cycle in mammalian cells.[4] CDK1 in complex with B-type cyclins regulates the mitotic entry of the cell cycle. Once DNA damages occur, CDK1/cyclin B1 becomes inactivated, and cells are prevented from entering mitosis until the damages are repaired. CDK1 is inactivated by the nuclear vs. cytoplasmic kinases Wee1 and Myt1, which phosphorylate CDK1 on its tyrosin-14 and -15 sites.[5] While it is activated by the antagonist of Wee1 and Myt1, a family of phosphatases of Cdc25A, B and C dephosphorylate CDK1 on the same tyrosin-sites served for Wee1 and Myt1.[6] The proper level and activity of CDK1 is thus kept in balance by these kinases and phosphatases that are involved in the cell cycle progression (**Fig. 1**). However, the role of CDK1 and the regulation of its phosphorylation and subcellular localization upon cellular response to chemotherapeutic drugs are poorly understood.

In a recent issue of *Cell Cycle*, Hedblom et al.[7] showed that altered level of CDK1 is associated with disease recurrence and poor overall survival of patients with acute myeloid leukemia. These novel findings indicate that CDK1 is a critical factor mediating cellular response to ATRA treatment. Only elimination of CDK1, not CDK2, causes a decreased proportion of G_0/G_1 cells and a concomitant increase in mitotic cells, suggesting that cells without proper level of CDK1 had accelerated mitosis. Defects in CDK1 expression level thus confer U-937 leukemic cells to be less sensitive to *all trans retinoic acid* (ATRA)-induced G_0/G_1 cell cycle arrest and differentiation. Hedblom et al.[7] demonstrate that defects in CDK1 expression cause alterations in the expression levels and activities of several proteins, which are the key regulators for cell growth and survival. These include: (1) elimination of CDK1 in U-937 cells leads to a significant reduction in the level of $P27^{kip}$, a key regulator for G_0/G_1 checkpoint; (2) elimination of CDK1 in U-937 cells also results in an increased level of phosphorylated Akt, a key survival factor. These alterations are likely linked to the CDK1-mediated resistance to ATRA treatment.

Hedblom et al. have unravelled several novel mechanisms on how the phosphorylation and subcellular localization of CDK1 is regulated upon cellular response to ATRA. Thus, ATRA receptor RARγ, (not RARα) is required for the regulation of CDK1 expression, phosphorylation and protein stability upon induction of ATRA. RARγ regulates CDK1 level and activity through a direct formation of protein-protein complex in the nucleus of U-937 cells and F9 cells. In the absence of RARγ, ATRA is unable to downregulate CDK1 expression and phosphorylation. This suggests that RARγ is a critical factor to balance the CDK1 level and activity to ensure ATRA to achieve optimal effects on cancer cells. Similar to what is observed in U-937 cells with defects in CDK1 expression; tumor cells with defects in RARγ also display a reduced level of $P27^{kip}$. Thus, RARγ and CDK1 may form a reciprocal regulatory circuit and influence the function and level of $P27^{kip}$ protein (**Fig. 1**).

Hedblom et al.[7] show that ATRA treatment results in a reduced the level of Wee1 kinase and Cdc25A phosphatase in the nucleus, which coincides with the decreased level of nuclear CDK1. This suggests that ATRA inhibits its CDK1 activity in the nucleus by reducing Cdc25A phosphatase, the activator that is responsible for CDK1 activity. It is interesting to note that ATRA treatment also reduces the level of Wee1 kinase, which acts as the inhibitor for CDK1. This suggests that despite that the downregulation of CDK1 is required for ATRA to achieve the optimal effect, a proper level and activity of CDK1 need to be maintained and kept in balance by Wee1 kinase and CDC25A. The novel findings suggest that the regulation of CDK1 is cooperatively mediated by the cell cycle regulators Wee1 kinase and CDC25A and the hormone receptor RARγ in response to ATRA treatment.

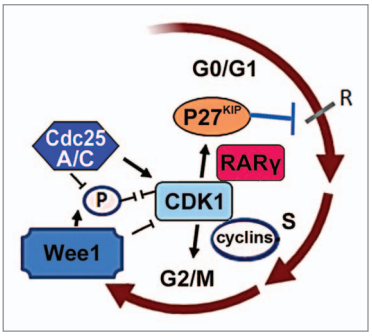

Figure 1. To regulate the proper entry of mitosis, CDK1 is inhibited by Wee1 which phosphorylate CDK1, while it is activated Cdc25A and C, which dephosphorylate CDK1. Hedblom et al. for the first time showed that RARγ and CDK1 form a reciprocal regulatory circuit and influence the function and level of $P27^{kip}$ protein, and control the G_0/G_1 phases of cell cycle.

Retinoid-based therapies are increasingly being utilized to treat various types of cancers. ATRA represents a class of anticancer drug that can induce tumor cells to differentiate and restore their normal function. Because CDK1 is a downstream effect protein of multiple pathways, including Wee1, and Cdc25A and Cdc25C and RARγ, the proper regulation of its expression and activity is essential for sensitizing the cells to respond to treatment. The study by Hedblom et al.[7] suggests that CDK1 and its associated regulators are the ideal targets for cancer therapy, and their novel

findings highlight the therapeutic potential using ATRA for targeting CDK1 and its associated proteins in cancer.

References

1. Kröger N. Curr Opin Oncol 2011; 23:203-8; PMID:21157340; http://dx.doi.org/10.1097/CCO.0b013e328342c6c8
2. Kim SJ, et al. Ann Oncol 2012; 23:891-7; PMID:21821547; http://dx.doi.org/10.1093/annonc/mdr340
3. Zeestraten EC, et al. Br J Cancer 2012; 106:133-40; PMID:22108518; http://dx.doi.org/10.1038/bjc.2011.504
4. Malumbres M, et al. Nat Rev Cancer 2009; 9:153-66; PMID:19238148; http://dx.doi.org/10.1038/nrc2602
5. McGowan CH, et al. EMBO J 1995; 14:2166-75; PMID:7774574
6. Boutros R, et al. Nat Rev Cancer 2007; 7:495-507; PMID:17568790; http://dx.doi.org/10.1038/nrc2169
7. Hedblom A, et al. Cell Cycle 2013; 12:1251-66; PMID:23518499; http://dx.doi.org/10.4161/cc.24313

Progesterone and FOXO1 signaling: Harnessing cellular senescence for the treatment of ovarian cancer

Comment on: Diep CH, et al. Cell Cycle 2013; 12:1433–49; PMID:23574718; http://dx.doi.org/10.4161/cc.24550

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Senescence is an inbuilt cellular response that leads to irreversible cell cycle arrest.[1] It plays a critical role in both aging and tumor suppression. While first observed in culture, cellular senescence happens in vivo. Premature senescence can be triggered by various insults, such as oncogene activation, telomere erosion, irradiation, DNA damage, oxidative stress and toxins, which emphasize the importance of the senescence pathway in arresting growth of prospective cancer cells that have accumulated potentially harmful genetic mutations.[1]

In the May 1, 2013 issue of *Cell Cycle*, Diep and colleagues report that the progesterone receptor (PR) cooperates with the Forkhead transcription factor FOXO1 to trigger cellular senescence in ovarian cancer cells.[2] The authors found that PR not only regulates FOXO1 expression, but also cooperates with this transcription factor to activate genes that encode senescence-associated cell cycle inhibitors, such as $p15^{INK4b}$, $p16^{INK4a}$, $p21^{Cip1}$ and $p27^{Kip1}$ (**Fig. 1**). Importantly, they show that this response is induced upon treatment of cells with a synthetic PR agonist, dependent on the B isoform of PR and attenuated upon FOXO1 knockdown. The inference is that progestins, compounds widely used for a variety of clinical indications, could also be valuable in the management of ovarian cancer, the most lethal of all gynecological malignancies.

These observations are unexpected for more than one reason. It is generally believed that most cancer cells have disabled the senescence pathway, thus achieving immortality.[1] However, the present study shows that PR- and FOXO1-positive ovarian cancer cells can be tricked into entering senescence in response to progestins. The role of FOXO proteins in cellular senescence is well documented.[3] In keeping with the findings of Diep et al.,[2] it has been shown previously that overexpression or activation of FOXO proteins through inhibition of the upstream phosphatidylinositol-3-kinase (PI3K)/AKT signaling cascade promotes senescence via induction of cell cycle inhibitors, such as $p27^{Kip1}$. Intriguingly, the cellular senescence induced in this way appears to be independent of p53 and $p16^{INK4a}$, molecules important for the maintenance of senescence-associated cell cycle arrest.[3]

The present study not only identifies the PR-FOXO1 axis as a potential therapeutic target in ovarian cancer, but also helps to explain why expression of PR is a prognostic marker for ovarian cancer associated with longer progression-free survival. Similarly, this study provides a mechanistic explanation for why pregnancy, which is associated with high circulating progesterone levels, and the use of progestin-containing oral contraceptives may suppress the growth of premalignant cells in the ovarian cortex, thus protecting against ovarian cancer.[4] There are, however, major

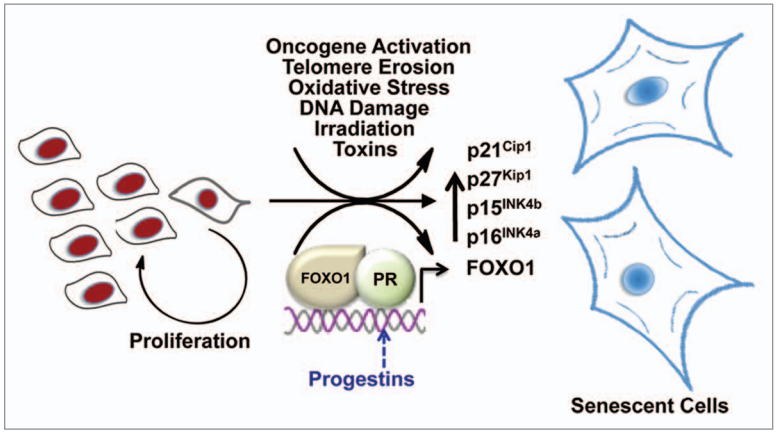

Figure 1. Progestin-dependent activation of the senescence pathway. Cancer cells have increased capacity to proliferate. Cellular senescence can be triggered by various insults. Progesterone promote ovarian cancer cells to enter senescence through activation of the progesterone receptor (PR), which cooperates with FOXO1 to induce expression of senescence-associated cell cycle inhibitors, including FOXO1, p15INK4b, p16INK4a, p21Cip1 and p27Kip1.

obstacles that limit the clinical use of progestins in ovarian cancer. Foremost, ovarian cancer is a heterogeneous disease that consists of etiologically distinct tumors that share an anatomical site. Consequently, progestin sensitivity is likely restricted to certain histological types, such endometrioid and serous cancers.[4] Further, PR as well as FOXO1 are frequently lost in ovarian cancer; the robustness of the senescence response in vivo has not yet been studied, and the contribution of putative non-genomic progestin receptors in modulating cellular responses to hormonal therapies remains poorly understood and controversial.[5] Nevertheless, the observations of Diep and colleagues should help to define molecular markers that identify those tumors likely to be responsive to progestin treatment, alone or combined with a PI3K/AKT inhibitor.

Notably, PR and FOXO1 interactions have also been studied in normal and malignant endometrium.[6,7] In fact, these two transcription factors are also putative determinants of the responsiveness of endometrial cancer cells to chemotherapy and progestin treatment. In the context of reproduction, the induction of FOXO1 and subsequent binding to PR triggers the differentiation of endometrial stromal cells into secretory decidual cells,[7] a process that is indispensable for embryo implantation and the formation of a functional placenta. Few studies have as yet examined senescence in decidual cells, although there is evidence that deregulation of this process can cause preterm labor.[8] Approximately 12.9 million babies are born too soon every year, and more than 1 million die each year as a direct consequence of prematurity. Thus, targeting the FOXO1-PR axis to modulate cellular senescence in the uterus or ovary may unlock hitherto unrecognized therapeutic options of immense clinical value.

References

1. Collado M, et al. Nat Rev Cancer 2010; 10:51-7; PMID:20029423; http://dx.doi.org/10.1038/nrc2772
2. Diep CH, et al. Cell Cycle 2013; 12:1433-49; PMID:23574718; http://dx.doi.org/10.4161/cc.24550
3. Collado M, et al. J Biol Chem 2000; 275:21960-8; PMID:10791951; http://dx.doi.org/10.1074/jbc.M000759200
4. Modugno F, et al. Endocr Relat Cancer 2012; 19:R255-79; PMID:23045324; http://dx.doi.org/10.1530/ERC-12-0175
5. Gellersen B, et al. Hum Reprod Update 2009; 15:119-38; PMID:18936037; http://dx.doi.org/10.1093/humupd/dmn044
6. Goto T, et al. Oncogene 2008; 27:9-19; PMID:17599040; http://dx.doi.org/10.1038/sj.onc.1210626
7. Labied S, et al. Mol Endocrinol 2006; 20:35-44; PMID:16123151; http://dx.doi.org/10.1210/me.2005-0275
8. Hirota Y, et al. J Clin Invest 2010; 120:803-15; PMID:20124728; http://dx.doi.org/10.1172/JCI40051

Rb and chromatin remodeling in the maintenance of the post-mitotic state of neurons

Comment on: Andrusiak MG, et al. Cell Cycle 2013; 12:1416–23; PMID:23574720; http://dx.doi.org/10.4161/cc.24527

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Cell proliferation and differentiation are two intimately connected fundamental processes. For instance, proliferating neural precursors that undergo terminal neuronal differentiation exit the cell cycle in an irreversible manner, a phenomenon we call mitotic exit. Although we have a good understanding of the gene expression changes that occur during cell cycle arrest, much less is known about the exact molecular mechanisms that enable a permanent shut down of cell cycle gene transcription during mitotic exit. The study by Andrusiak et al.[1] sheds some light on this important question.

Transcription factors of the E2F family play a central role in controlling proliferation: in cells that are growing, E2F proteins turn on the expression of genes enabling cell division, while they contribute to shutting down the transcription of those genes in cells that are exiting the cell cycle. The transcriptional regulatory function of E2F proteins comes in part from their ability to recruit protein complexes that help alter the structure of chromatin. In dividing cells, E2Fs can recruit histone methyltransferases or acetyltransferases to impart chromatin marks that facilitate gene transcription, such as H3K4me3 or H3ac (reviewed in ref. 2). On the other hand, in cells that have stopped to divide, E2Fs interact with one of the pocket proteins, the retinoblastoma protein pRb or its relatives, p107 and p130. By binding to E2Fs, the pocket proteins can passively repress gene expression by blocking E2F transcriptional activation functions. Moreover, they can also actively repress E2F target gene expression by recruiting transcriptional co-repressors that participate in modifying the structure of chromatin (reviewed in ref. 3).

Many of these chromatin remodeling proteins, such as the histone deacetylase HDAC1 or the heterochromatin protein HP1, contain a short stretch of amino acids matching the consensus LXCXE (where X represents any residue), which allows them to interact with the pocket domains of pocket proteins at a site different from the E2F-binding domain. In cultured cells, pRb mutants that lack the LXCXE-interaction domain fail to engage in active transcriptional repression of E2F targets[4] and to properly establish pericentric heterochromatin.[5] Nevertheless, these mutants retain their ability to block E2F-dependent transcriptional activation and to induce cell cycle arrest.

In the brain, Rb is essential to the cell cycle exit and survival of post-mitotic cortical neurons.[6] Andrusiak et al. have now examined whether the role of pRb in enforcing the post-mitotic state depends on its capacity to recruit LXCXE domain-containing chromatin remodeling activities.[1] They made use of inducible loss-of-function alleles of Rb in vivo and in vitro to prevent the emergence of compensatory effects that can occur with constitutive loss-of-function models. The authors show that while the acute deletion of Rb leads to de-repression of E2F-targeted cell cycle genes in cortical neurons, those genes remain silent when the only remaining allele of Rb codes for a protein unable to interact with LXCXE domain-containing proteins (**Fig. 1**). Moreover, they show that the complete elimination of Rb changes the chromatin from a state refractory to transcription (nucleosomes trimethylated on H3K9 and poorly acetylated) to one that is more conducive to gene expression. Finally, the authors demonstrate that E2F proteins are responsible for the change in chromatin structure after

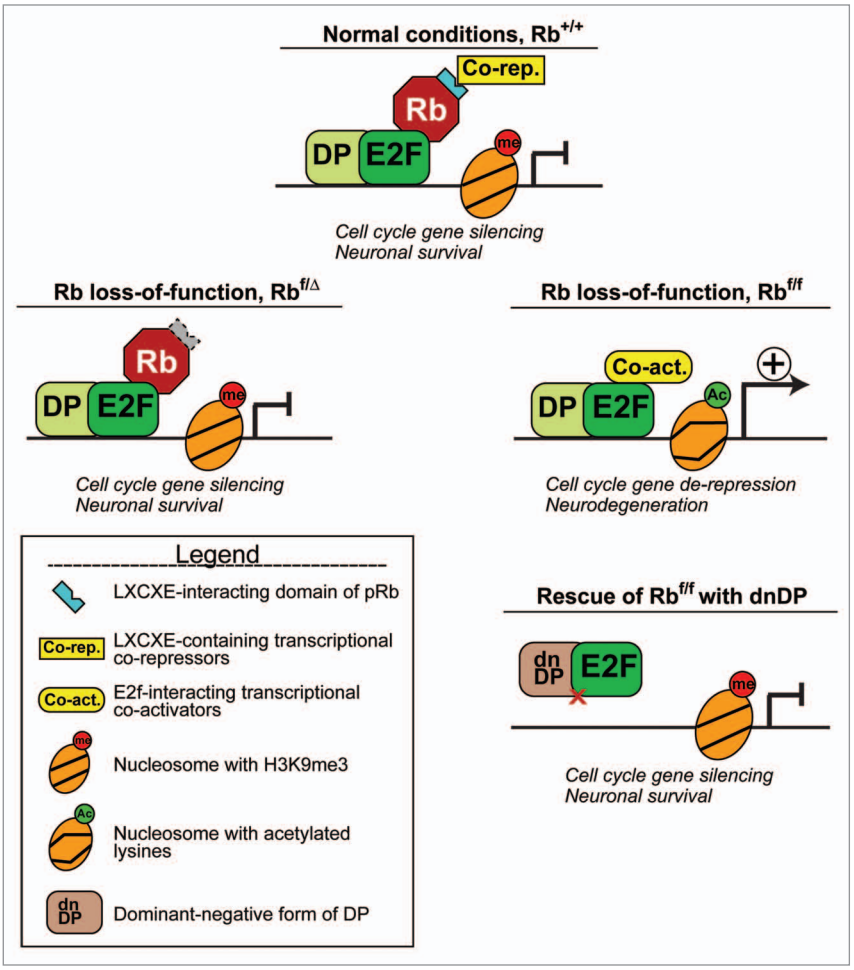

Figure 1. LXCXE-independent chromatin remodeling and cell cycle repression in mitotically arrested cells. Andrusiak et al. reported that in cortical neurons of the adult brain, which have permanently ceased proliferation, the acute loss of pRb leads to chromatin remodeling at cell cycle genes, to cell cycle re-entry and to apoptotic loss of neurons. In contrast, loss of pRb's ability to interact with LXCXE-containing proteins does not lead to changes in chromatin structure or to cell cycle re-entry. Moreover, the loss of pRb phenotype can be reversed if the action of E2F proteins is blocked by the overexpression of a dominant-negative form of DP1 that blocks E2F DNA binding.

complete deletion of Rb: no change in chromatin structure takes place when E2F proteins are prevented from binding to their target genes, and cells remain in their post-mitotic state. This rescue experiment demonstrates that pRb keeps mitotically arrested neurons out of the cell cycle by blocking E2F-mediated transcriptional activation of cell cycle genes, rather than by recruiting LXCXE-containing chromatin remodeling complexes.

These findings inform us on the multiple roles played by pRb in regulating gene silencing. During senescence or after exposure to genotoxic stress, pRb may cooperate with LXCXE-containing proteins to block the expression of target genes and suppress tumorigenesis.[7,8] However, permanent mitotic exit appears to rely on a different function of pRb: that of antagonizing E2F-mediated cell cycle gene activation.

References

1. Andrusiak MG, et al. Cell Cycle 2013; 12:1416-23; PMID:23574720; http://dx.doi.org/10.4161/cc.24527
2. Blais A, et al. Curr Opin Cell Biol 2007; 19:658-62; PMID:18023996; http://dx.doi.org/10.1016/j.ceb.2007.10.003
3. Dick FA, et al. Nat Rev Mol Cell Biol 2013; 14:297-306; PMID:23594950; http://dx.doi.org/10.1038/nrm3567
4. Chan HM, et al. Oncogene 2001; 20:6152-63; PMID:11593423; http://dx.doi.org/10.1038/sj.onc.1204793
5. Isaac CE, et al. Mol Cell Biol 2006; 26:3659-71; PMID:16612004; http://dx.doi.org/10.1128/MCB.26.9.3659-3671.2006
6. Andrusiak MG, et al. J Neurosci 2012; 32:14809-14; PMID:23077065; http://dx.doi.org/10.1523/JNEUROSCI.1912-12.2012
7. Bourgo RJ, et al. Mol Cell 2011; 43:663-72; PMID:21855804; http://dx.doi.org/10.1016/j.molcel.2011.06.029
8. Narita M, et al. Cell 2003; 113:703-16; PMID:12809602; http://dx.doi.org/10.1016/S0092-8674(03)00401-X

Progestins: Pro-senescence therapy for ovarian cancer?

Comment on: Diep CH, et al. Cell Cycle 2013; 12:1433–49; PMID:23574718; http://dx.doi.org/10.4161/cc.24550

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Increasing epidemiological evidence suggests a role of steroid hormones in the development and progression of certain molecular subtypes of ovarian cancer. In general, the data suggest that estrogens are associated with increased risk of ovarian cancer, while progesterone and progestins may have a protective role (for recent review on this topic, see ref. 1). Despite these epidemiological associations, there remains a surprisingly limited understanding of the underlying biology and mechanisms of steroid receptor action in early and late stages of ovarian tumorigenesis. However, given that hormonal therapies represent the best of "targeted therapy in personalized medicine," with limited toxicities, there is renewed interest in the static and maintenance roles for hormonal therapy in ovarian

cancer recurrence. In addition, there might be a role for endocrine therapy in treatment of early-stage ovarian cancer, when expression of nuclear receptors is high.

In the May 1, 2013 issue of *Cell Cycle*, Carol Lange and colleagues published an exciting study, which provides mechanistic insight into how progesterone receptor (PR) mediates the protective effects of progestins in ovarian cancer.[2] The authors used a number of established ovarian cancer cell lines of different histological subtypes to show that progestins activate nuclear PR, leading to elevated FOXO1 expression and ultimately cellular senescence. Senescence is at least in part due to the induction of the cell cycle inhibitor p21. It is important to point out that in addition to the elegant mechanistic studies, the authors provide very valuable and extensive data on PR protein expression in the different molecular subtypes of human ovarian cancer, showing low PR expression in clear cell and mucinous tumors (3 and 18%, respectively) and moderate to high expression in serous (35%), low-grade serous (64%) and endometriod tumors (67%) (**Fig. 1**).

As with other scientific studies, this manuscript provides clues to mechanism of action of progestins but at the same time raises many new questions. For example, what is the cause for the biphasic dose response to progestins? Does this imply a critical role for the other progesterone-binding proteins? And can this explain prior discrepant results showing both progesterone-induced apoptosis and proliferation? Importantly, given the increasing realization of diverse roles of the two PR isoforms, PR-A and PR-B, what can we expect to see from loss (or overexpression) of the PR-A isoform? And, finally, what is the role of PR action in the tumor microenvironment?

This study has important translational implications, in that PR agonists (and antagonists) are clinically available. However, to date, small clinical trial cohorts have demonstrated modest activity in ovarian cancer.[3] Many of these trials have limitations, and it might be time to revisit the potential use of progestins and anti-progestins in ovarian cancer therapy. Several steps can be taken. First and foremost is the need to use biomarkers to predict active PR signaling in the tumor (and possibly the stroma). In addition, clinical trials mostly occurred in chemotherapy-resistant, late-stage disease; however, data at hand suggest that there is progressive loss of PR during tumor progression. This might imply a place for progestin in early-stage tumorigenesis, perhaps from precursors like endometriosis, which predispose to endometrioid, clear cell and low-grade serous ovarian tumors (**Fig. 1**). In support of this rationale is the fact that PR expression is highest in low-grade serous and endometrioid (although not clear cell) tumors,[2,4] further suggesting the early roles of PR-mediated pathogenesis and the potential for PR-targeted therapy or prevention, at least in some histological subtypes. And finally, the idea of using synthetic lethal approaches, i.e., combining progestin with drugs showing increased efficacy against senescent cells would be warranted. One approach that comes to mind is the use of immune therapies. Senescence of epithelial tumor cells leads to the release of inflammatory proteins and mediators of extracellular matrix (also called the senescence- messaging secretome or SMS),[5,6] which can act as activators of the innate immune response, leading to tumor cell clearance. In addition, active immunization with senescent cell-derived, tumor antigen-based vaccines may act in synergy to the potentially immunogenic in vivo inducers of senescence (like progestins, for example), providing additional benefits in both therapeutic as well as preventive approaches, although the efficacy of such approaches remains to be determined.

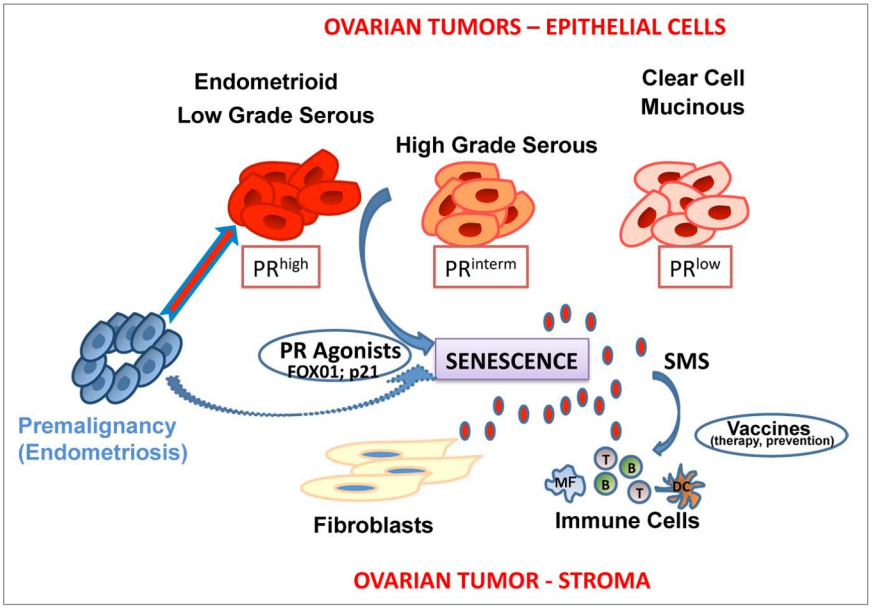

Figure 1. Schematic representation of PR action as target in different subtypes of ovarian cancer. SMS, senescence messaging secretome.

In summary, a renewed effort to understand steroid hormone action in ovarian cancer (in both the epithelium and stroma) may lead to novel approaches for targeted therapies with limited toxicities and therapeutic benefit.

References

1. Modugno F, et al. Endocr Relat Cancer 2012; 19:R255-79; PMID:23045324; http://dx.doi.org/10.1530/ERC-12-0175
2. Diep CH, et al. Cell Cycle 2013; 12:1433-49; PMID:23574718; http://dx.doi.org/10.4161/cc.24550
3. Zheng H, et al. Int J Gynecol Cancer 2007; 17:325-38; PMID:17362310; http://dx.doi.org/10.1111/j.1525-1438.2006.00749.x
4. Langdon SP, et al. Clin Cancer Res 1998; 4:2245-51; PMID:9748145
5. Kuilman T, et al. Nat Rev Cancer 2009; 9:81-94; PMID:19132009; http://dx.doi.org/10.1038/nrc2560
6. Hannani D, et al. Cancer J 2011; 17:351-8; PMID:21952286; http://dx.doi.org/10.1097/PPO.0b013e3182325d4d

Escaping death to quiescence: Avoiding mitotic catastrophe after DNA damage

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PMID:23574719; http://dx.doi.org/10.4161/cc.24528

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When a mammalian cell exits G_1 and enters the S-phase, it becomes committed to the completion of the entire cell cycle until the next G_1 phase, where a quiescent status can be reached. However, when DNA damage is induced, mammalian cells may activate checkpoints that arrest the cell cycle during specific phases, such as G_1, intra-S, and G_2. The mechanisms dictating the exit from cycle arrest have major implications on cell fate and thus affect the outcome of DNA damage-based cancer therapies.

In one scenario, cells can recover from temporary arrest after DNA damage is repaired. This results in the resumption of cell cycle progression, although a fraction of the surviving cells may contain an altered genome due to mutagenic DNA repair. In another scenario, cells escape cycle arrest with residual DNA damage. This can lead to delayed cell death or genomic instability.[1] Following release from G_2 arrest, such events have the potential to induce cell death through mitotic catastrophe. Lastly, permanently arrested cells have the option to enter a quiescent G_1/G_0 status known as senescence. It is not difficult to see how long-term G_1 arrest permits entrance into senescence, but it is unclear how G_2-arrested cell can enter this state.

In a recent article,[2] Ye et al. reported that the induction of long-term G_2 arrest resulted in the complete omission of M-phase and facilitated the entrance into a tetraploid quiescent G_1 phase. The investigators used a cell line that was previously shown to be able to undergo long-term G_2 arrest after a high dose of ionizing radiation.[3] They found that after 10 Gy of γ-irradiation, these cells underwent a long-term G_2 arrest, which was never followed by mitosis. Critical genes involved in the G_2-M transition were downregulated in the G_2-arrested cells. Furthermore, these tetraploid quiescent cells displayed a senescent phenotype. This led to the conclusion that these G_2-arrested cells directly bypassed mitosis and entered into a G_1/G_0 quiescent status. This is the first report that suggests G_2 arrest induced by DNA damage can result in G_2 slippage that bypasses mitosis, although it has been previously reported that long-term arrest at metaphase causes the abortion of mitosis (mitotic slippage) and subsequent entrance into G_1.[4]

Unlike G_1 phase, G_2 phase is only a transient phase of the cell cycle. It is conceivable that if the damaged cells adapt to the long-term G_2 arrest and prematurely enter mitosis with DNA damage, mitotic catastrophe is likely the outcome. Therefore, G_2 slippage might be a mechanism for the cells to escape reproductive mitotic death to enter a quiescent status in the event of severe DNA damage at G_2 or late S phase. The study by Ye et al. raises several questions relevant to radiation cancer therapy.

First, DNA damage induced senescence is considered to be a form of accelerated cellular senescence (ACS).[5] The ACS cells retain some metabolic activities, but a small portion of the ACS cells may reenter the cell cycle.[5] The senescent tetraploid G_1 cells observed by Ye et al.[2] were induced by a high dose of radiation and may be considered a form of ACS. It remains to be determined whether a small fraction of the tetraploid ACS cells formed after G_2 slippage reentered the cell cycle, as this has significant implications for cancer therapy.

Second, a major mechanism of radiation induced proliferative cell death is through mitotic catastrophe. It was noted that the long-term G_2 arrest and subsequent senescence by G_2 slippage is more predominant at a high dose of irradiation than at a modest dose.[2,3] This raises the interesting question of whether G_2 slippage is a predominant outcome after high dose irradiation. The answer to this question would be relevant to the emerging concept of high dose stereotactic ablative radiotherapy.[6,7] High doses could efficiently induce senescence that would quickly control tumor mass, but it would also enable cells to avoid cell death via mitotic catastrophe. Thus, ablative radiotherapy would be beneficial for the initial and local control of tumor mass, but it may also leave the possibility of tumor recurrence, because a small fraction of the therapy induced ACS cells may re-enter the cell cycle.[5]

References

1. Shen Z. J Mol Cell Biol 2011; 3:1-3; PMID:21278445; http://dx.doi.org/10.1093/jmcb/mjq057
2. Ye C, et al. Cell Cycle 2013; 12:1424-32; PMID:23574719; http://dx.doi.org/10.4161/cc.24528
3. He J, et al. Cell Cycle 2011; 10:1468-76; PMID:21455017; http://dx.doi.org/10.4161/cc.10.9.15510
4. Brito DA, et al. Curr Biol 2006; 16:1194-200; PMID:16782009; http://dx.doi.org/10.1016/j.cub.2006.04.043
5. Wu PC, et al. Exp Oncol 2012; 34:298-305; PMID:23070015
6. Dahele M, et al. Cancer Res Treat 2011; 43:75-82; PMID:21811422; http://dx.doi.org/10.4143/crt.2011.43.2.75
7. Tree AC, et al. Lancet Oncol 2013; 14:e28-37; PMID:23276369; http://dx.doi.org/10.1016/S1470-2045(12)70510-7