Local renin-angiotensin system in the reproductive system

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The renin-angiotensin system (RAS) is well known as regulator of electrolytes and blood pressure. Besides this function, there are numerous studies supporting the idea of a local tissue RAS. This system controls the local activity of the different RAS family members, especially of the functional proteins Angiotensin II and Angiotensin (1–7). Those antagonistically acting proteins have been described to be expressed in different organ systems including the human reproductive tract. Therefore, this local RAS has been suspected to be involved in the control and regulation of physiological and pathological conditions in the female reproduction tract. This review of the available literature summarizes the physiological influence of the RAS on the follicular development, ovarian angiogenesis, and placentaland uterine function. In addition, in the second part the role of the RAS concerning ovarian and endometrial cancer becomes elucidated. This section includes possible novel therapeutic strategies via inhibition of RAS-mediated tumor growth and angiogenesis. Looking at a very complex system of agonistic and antagonistic tissue factors, it may be supposed that the RAS in the female reproduction tract will be of rising scientific interest in the upcoming years.

**Keywords:** endometrial cancer, endometrium, ovarian cancer, ovary, renin-angiotensin system, reproductive tract, local

**INTRODUCTION**

The Renin-Angiotensin system (RAS) is of paramount importance for the perpetuation of the circular flow, regulating the electrolyte metabolism thus the blood pressure (1–4). This considerable function of the RAS is mediated by the systemic RAS-pathway. The latter consists of a cascade of peptides, acting as precursors which become transformed by different enzymes into the bioactive end products (5). The main protein of this system is Angiotensinogen, which is synthesized in the liver (6). Following the pathway, Angiotensinogen becomes converted into Angiotensin I, catalyzed by Renin, which is of renal origin. Subsequently, Angiotensin I can be further modulated by the angiotensin-converting enzyme (ACE) I to Angiotensin II or by ACE II to Angiotensin (1–9).

Angiotensin (1–9) then becomes transformed by ACE or neutral endopeptidase (NEP) into Angiotensin (1–7) (7–9). Those two bioactive effector molecules, Angiotensin II and Angiotensin (1–7) act in an antagonistic way by binding to different receptors: angiotensin receptor type 1 (AT1R) and type 2 (AT2R) or Mas-receptor (10). The G-coupled Mas-receptor mediates vasodilatory and anti-proliferative effects and antagonizes actions of the AT1R (11). In recent years, attention has also focused on the evidence of a widespread local tissue RAS (12). Expression of elements of this local RAS has been described in different parts of the human reproductive tract. Apparently, the both antagonistic bioactive proteins of the RAS, in particular Angiotensin II and Angiotensin (1–7) can result from the local tissue RAS (13, 14). This local production of the bioactive peptides is not necessarily dependent on the local expression of all components of the local tissue RAS, since it is also possible to take up components from the circulation, such as renin. Furthermore, besides presence of Angiotensin II and Angiotensin (1–7), expression of AT1R, AT2R, and Mas-receptor human reproductive tissue is needed in order to mediate the local impact of the RAS for physiological and pathological processes, including follicle maturation, fine-tuning of the regulation of reproduction, angiogenesis as well as tumor cell proliferation (15–18). An influencing effect on cancer has been described for different tumor types already during the last two decades (19, 20).

**MATERIALS AND METHODS**

We performed a systematic literature review concerning presence and function of the RAS in the female reproduction tract. This was based on the medical databases Medline, Embase, BIOSIS, and CINHAL. Literature analysis was conducted without a timeframe on all existing publications including 2013. All manuscripts were sighted based on the title and abstract and any duplicate manuscripts occurring in the literature search were excluded. After fulfilling the inclusion criteria (content-related, experimental, and clinical studies, in the case of experimental studies dividing into studies in humans and/or animals) the manuscripts were reviewed and analyzed. Thereby, the data was extracted and content-related articles allocated into two different groups:

- Physiological role of the RAS in the reproductive tract?
- Role of the RAS in gynecologic cancers.
ANGIOTENSIN II IN HUMAN GRANULOSA LUTEIN CELLS

The most outstanding data in the literature has been published concerning the regulatory character of the RAS on vascular function and angiogenesis in the ovary. The invoking effects on the

Vessels are thereby first of all adapted from the Angiotensin II-AT1R-pathway (41–44). To be contrary to this, the restitution of the luteal vasculature is mediated by the AT2R-pathway (45). Anyway, Angiotensin II obviously influences the microvascular endothelial function in the corpus luteum (42). Hayashi et al. demonstrated that microvascular endothelial cells (MVE) in the corpus luteum express ACE and are capable to convert Angiotensin I into Angiotensin II. The Angiotensin II production thereby increases significantly under stimulation with estradiol in combination with vascular endothelial growth factor (VEGF) (41). MVE furthermore possess AT1R and AT2R (41, 42). Interestingly, the expression of those two receptors differ throughout the cycle: AT1R remains constant but AT2R-expression is lowest during the mid luteal phase and highest during the late luteal phase (41, 46). The regulation of angiogenic processes is urgently needed to ensure the constant flow of growth, maturation, and demise of the corpus luteum. It has been shown by our group, human granulosa lutein cells collected during in vitro fertilization (IVF) are expressing several components of the RAS (47). In addition, we demonstrated that exogenous Angiotensin II stimulation increased VEGF synthesis via AT1R signaling in vitro (47). This data may implicate the regulatory effect of the RAS on angiogenesis in the corpus luteum. In agreement with its meaning concerning control of systemic blood pressure, the individual family members of the local RAS also regulate perfusion and vascular tone in the ovary (36, 44).

PLACENTA

The human placenta is one of the most interesting tissues in the reproductive tract, because the utero-placental unit provides a transposition of nutritive substances and oxygen between mother and fetus. It has been assumed by many authors that the RAS influences the placental function (48–52), since all different components of the RAS are expressed in human placenta cell lines (53) as well as in placental tissue (54, 55). However, functional data of the placental RAS is very rare. Obviously, the different RAS-proteins are expressed differentially in the various areas of this organ: angiotensinogen, Renin, Angiotensin I, Angiotensin II, ACE, AT1R, and AT2R were localized to maternal decidua (56, 57) and Angiotensin II and ACE were additionally found in pericytes of endometrial spiral arteries. However, Angiotensinogen and renin also have been detected in fetal capillaries (58). The AT1R, which is predominantly expressed in the placenta, was found in cytotrophoblast and syncytiotrophoblast cells as well as in fetal capillaries, while little is known concerning localization of the AT2R (59, 60). The antagonistic proteins to Angiotensin II, namely Angiotensin I (1–7) and ACE2 were found to be expressed in syncytiotrophoblast, cytgotrophoblast, and vascular smooth muscle cells of primary and secondary villi (58). The above mentioned members of the RAS family can be detected from 6 weeks of gestation until birth. Obviously, there are some variations in the course of pregnancy: it has been shown that mRNA of ACE is increasing during gestation but decreases near term. In addition, AT1R mRNA and AT1R protein levels are rising throughout the entire pregnancy, reaching highest levels at the end (61). Since a direct connection between Angiotensin II and AT1R has been observed in the placenta, it has been supposed that this fact indicates a
regulating effect of Angiotensin II on the AT1R expression (62). From a more clinical point of view, there are several references that the placental RAS is involved in trophoblast invasion and angiogenesis (63, 64), being a possible cause of defect for the development of conditions with disordered utero-placental perfusion, namely preeclampsia (see below).

**FALLOPIAN TUBE**

Data concerning the RAS and the oviduct is rare. Any clinical relevant findings have not been published. However, Angiotensin II has been localized in blood vessel endothelium and in stromal cells. Both binding and Angiotensin II type-2 receptor mRNA were detected at high levels, but no differences in receptor concentration could be detected in fallopian tubes ipsilateral or contralateral to the corpus luteum (65).

**UTERUS**

Outline above, the data of uterine RAS are of descriptive nature and mostly limited to the endometrium. Studies investigating the functional relevance of the RAS in the uterus are rare.

Being an indispensable part of the reproductive tract, the endometrium underlies a cyclic change of growth and degradation. Basically, all elements of the local tissue RAS are expressed in the endometrium (66), however this expression diversifies during the cycle (46, 67): angiotensin II underlies cyclic variances within the endometrium and is increased during the proliferative phase, and decreased during the secretory phase (68). Angiotensin-(1–7) and its receptor MAS is also present throughout the menstrual cycle but increases in the glandular endometrium in the mid and late secretory phase. Although AT1R and AT2R are expressed in the endometrium, expression of AT2R is more frequent and varying (68) and it is down-regulated during pregnancy (69). The AT2R-expression is thereby most prominent in the myometrium (up to 90%) as compared to AT1R-expression (up to 10%). Unfortunately, data concerning functional effects of the RAS in the uterus is rare. Since endometrium is controlled by female sex hormones, it has been supposed that the RAS might also be influenced by those hormones. This assumption is supported by the finding, that the local expression and production of renin is increased after stimulation with progesterone (70).

**PATHOPHYSIOLOGICAL ROLE OF THE RAS IN THE REPRODUCTIVE TRACT**

During the past few years, the primary small number of publications concerning pathophysiologic aspects of the RAS has been markedly increased. This affects aspects of reproduction, in essence preeclampsia, as well as of the role of the RAS controlling gynecological cancers. Thereby, the most resilient data is available regarding the regulatory aspects on tumor cell proliferation, vascular function, and angiogenesis (71).

**RAS AND REPRODUCTION**

In patients with polycystic ovary syndrome, the intra-follicular renin, which is needed for synthesis of the bioactive peptides of the RAS, affects maturation and oocyte quality (43). Follicles with high levels of renin indicating a high local RAS activity, were associated with better oocyte quality and showed higher VEGF concentrations during IVF procedures (43). Furthermore, a strong activation of the local ovarian RAS by beta-hCG has been observed during IVF treatment. This process was also associated with an increased VEGF concentration. Consequently, it has been assumed, that the activation of the ovarian RAS and consecutive high levels of VEGF might act synergistically during pathogenesis of ovarian hyperstimulation syndrome (OHSS).

Although the role of the RAS concerning invasion of trophoblast and placentation is poorly investigated, there is evidence that dysfunction of this system may cause hypertension and preeclampsia (56, 72, 73, 74): patients with preeclampsia present with increased expression of Angiotensin II and AT1R in maternal decidua cells and in the placenta itself (74, 75). In pre-eclamptic pregnancies Angiotensin II and AT1R was been observed to be increased, whereas levels of Angiotensin I, Angiotensin (1–7), ACE, and ACE2 were normal as compared to healthy pregnancies (48).

Current data describe a relevant clinical link between RAS and preeclampsia: women with a male fetus who developed gestational hypertension had increased Angiotensin (1–7) levels at 15 weeks gestation compared with women with normal pregnancies, suggesting that these women were on an early trajectory for the development of hypertension. Therefore, the authors proposed measurement of Ang-(1–7) during early pregnancy in order to predict new-onset hypertension (76).

In addition Valdes et al. also reported elevated Angiotensin (1–7) concentrations in spontaneously aborted and ectopic early pregnancy placentas, which lead the authors to hypothesize that the ACE2–Angiotensin (1–7) axis plays a functional role in placental development (77). Further recent data underlines the connection between RAS and preeclampsia, since there is an association with a polymorphism of Angiotensinogen in Chinese women. This finding might cause disordered vasculogenesis contributing to the development of preeclampsia (78).

**OVARIAN CANCER**

The published data apropos RAS and invasive epithelial ovarian cancer provides a role concerning proliferation and dissemination of cancer cells and tumor-angiogenesis. Ovarian cancer cells express Angiotensin II and AT1R (79) but there is still missing evidence of the other components of the local RAS in ovarian cancer. It has been shown that levels of AT1R are higher in borderline lesions and in invasive epithelial ovarian cancer as compared with normal ovaries (80). Being in line with this finding, ovarian cancer patients presenting with high levels of AT1R, have a worse prognosis in comparison with tumors lacking the AT1R expression. Obviously, the Angiotensin II → AT1R pathway is able to influence ovarian cancer cell proliferation (80). Since it has been shown that levels of VEGF as well as rates of angiogenesis are increased due to Angiotensin II, the link between the RAS and Angiogenesis has been established in epithelial ovarian cancer cells (80). At least in vivo, this connection even acts quantitatively: stimulation of the ovarian cancer cell line Skov 3 with Angiotensin II caused increased VEGF expression (80) and high levels of AT1R are associated with significantly increased VEGF production and micro-vessel density (MVD) (79). All those findings might indicate a therapeutical approach. Therefore, inhibition of AT1R has been performed in mice with peritoneal carcinomatosis, leading to a significant decrease of ascites and peritoneal
tumor cell dissemination. In patients, current data at least indicate that agonistic auto antibodies against AT1R may be associated with advanced progression of early ovarian cancer (81). These findings implicate that AT1-AA might be selected as a detectable biomarker and potential therapeutic target in diagnosis and treatment of EOC patients. Summing up, it appears that two substantial mechanisms, increased tumor cell proliferation and angiogenesis are mediated by the RAS. Therefore, targeting the Angiotensin II → AT1R pathway could provide a future treatment strategy for invasive epithelial ovarian cancer.

ENDOMETRIAL CANCER

Similarly to ovarian cancer, there are proofs for a possible influence of the local RAS concerning endometrial cancer. It has been published that in endometrial cancer, prognosis, tumor cell proliferation, and angiogenesis are affected by the RAS (82). According to the situation in the ovary, again increased local levels of Angiotensin II are associated with poorer prognosis in endometrial cancer patients (82, 83). This finding might be due to the fact, that those higher levels of Angiotensin II were found in patients with an advanced tumor stage (82). In this study, 81.9% were positive for Ang II and 59.6% positive for the AT1R. However, it seems as not only progression of disease but also an increased risk for developing endometrial cancer might be mediated by the RAS: a polymorphism of ACE has been described to be associated earlier in the fact that low Angiotensin II activity is associated with less aggressive behaviors in endometrial cancer patients (82, 83). This finding might be due to the fact, that the published data indicates a potential involvement of the local RAS in affecting physiological angiogenesis in a dose-dependent way (83). Recently, it has even speculated that Angiotensin II modulates the VEGF type-2 receptor KDR via AT1R (85). Inversely regarded, the connection of the RAS with angiogenesis is supported by the fact that low Angiotensin II activity is associated with less VEGF and a decreased MVD. This relation was basis for functional experiments: the treatment with the AT1R-blocker Losartan has a anti-proliferative effect in endometrial cancer tissue in vitro (86). In summary, high activity of the local RAS in endometrial cancer is associated with higher incidences, earlier onset, and increased rates of angiogenesis. The roles of Ang-(1–7) and the AT2R as well as clinical randomized study data is completely lacking and need to be further investigated.

CONCLUSION

This review summarizes the available literature concerning the local tissue RAS in the reproductive tract with regards to physiological and pathological clinical situations. The majority of the published studies remain on a non-functional descriptive level, but nevertheless, a role of the local tissue RAS as regulator in the human reproductive tract can be supposed. Obviously, the RAS affects oocyte maturation and quality, endometrial lining as well as hormone production and may therefore be considered as important system for regulation of physiologic pathways. Furthermore, the published data indicates a potential involvement of the local RAS in affecting physiological angiogenesis in the reproductive tract. Currently, pathologic conditions are better investigated than physiology. The Angiotensin II → AT1R pathway promotes tumor growth and angiogenesis in malignancies, arising new treatment strategies by inhibition of the AT1R. Data concerning stimulation of the antagonistic pathways such as the AT2R or Angiotensin (1–7) pathway as treatment modality for ovarian- or endometrial cancer is lacking. Due to current data, it is clear that most conclusions made are speculative since only a negligible number of functional studies have been conducted and clinical randomized data is missing completely. However, regarding a very complex and variable system of agonistic and antagonistic tissue factors, it may be hypothesized that the RAS in the female reproduction tract will be of increasing interest in the near future.

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