# Expression of Serotonin Receptors 5-HT1A, 5-HT1B, 5-HT2B and 5-HT4 in Ovary and in Ovarian Tumours

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**Abstract.** *Background: Recently we have shown that serotonin receptors may be involved in prostate cancer development. In ovarian carcinogenesis, oestrogen may play a role. As oestrogen seems to mediate at least some of its biological effects through serotonin, we decided to evaluate if serotonin receptors are expressed in ovary and in ovarian tumours and if the expression is correlated to ovarian tumour development. Materials and Methods: An immunohistochemical study of the serotonin (5-HT) receptors 5-HTR1A, 5-HTR1B, 5-HTR2B and 5-HTR4 in frozen samples of ovary, and benign, borderline and invasive ovarian tumours was performed. Results: Expression of all four serotonin receptors was strong to intermediate in the ovarian epithelium. In benign and non-invasive cancer cells, strong staining was seen, while in invasive cancer cells, decreased expression was observed. For 5-HTR2B, the decrease was correlated to dissemination of the disease. For none of the serotonin receptors was the expression correlated to survival. In the stromal part, a variable immunoreactivity was observed that was strongest for 5-HTR2B in both ovary and tumours. Staining of blood vessels was observed in ovary and all tumour groups for 5-HTR2B, but only occasionally was a weak expression seen for 5-HTR1A, 5-HTR1B and 5-HTR4. Conclusion: The staining pattern of serotonin receptors in ovary indicates their functional role in ovarian physiology. In ovarian tumours, the expression is in harmony with a tumour suppressor role in ovarian carcinogenesis, which is supported by observations in the literature. Further studies are necessary to resolve the connection between serotonin and ovarian tumour development.*

Among gynaecological neoplasms, ovarian cancer is the leading killer. In spite of advanced cytoreductive surgery and intensive research and clinical trials in chemotherapy the overall 5-year survival has only increased from 37% to 45% during the last three decades (1) and progress in improving survival seems to have stopped. Development of new strategies is urgently required and must be based on knowledge of the molecular biological basis of ovarian tumour development.

The aetiology of ovarian carcinogenesis is still largely unknown, although several molecular mechanisms have been suggested. The potential significance of a variety of growth regulatory factors has been studied by us (2, 3) and several others, as mentioned in recent reviews (4, 5). Hormonal factors are also been claimed to be involved. Thus, both endogenous levels of oestradiol and testosterone (6), as well as hormone replacement therapy (7) have been suspected to increase the risk of epithelial ovarian cancer. Recently an overlooked connection between serotonin (5-hydroxytryptamin, 5-HT) and oestrogen in several aspects was discussed, focusing on new mechanisms for the effects of oestrogen through serotonin (8). We studied four serotonin receptors in prostate cancer and found that they may be involved in tumour progression (9, 10). Besides its hormonal effects, serotonin seems to behave as a growth regulating factor (11), consequently, we decided to perform a similar immunohistochemical study of the expression of serotonin receptors in ovary and epithelial ovarian tumours to reveal if this receptor system is expressed at all and if a possible relationship to tumour development might exist. Finally, we correlated the receptor expression to tumour stage and survival.

## Materials and Methods

*Patient samples.* Fresh surgical specimens were collected prospectively after informed consent during surgery and frozen immediately and kept at –70˚C until analysed. The patient material was anonymized according to the guidelines by the Malmö-Lund Ethical Committee. None of the patients had been subject to treatment prior to surgery. Biopsies were obtained from 5 ovaries removed during surgery for non-malignant gynaecologic disease and showed normal histology. Samples from 16 benign ovarian tumours were obtained. For 5-HT1A samples from 5 serous, 7 mucinous and

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1 benign Brenner tumour were stained. For both 5-HT1B and 5-HT2B the corresponding numbers were 6, 7 and one. Immunohistochemistry for 5-HT4 was performed in 6 serous and in four mucinous samples. In the group of borderline tumours biopsies were obtained from 5 serous and 2 mucinous neoplasms. For the invasive tumour material details of the numbers, stage and histology are shown in Table I and Table II.

*Antibodies and immunohistochemistry*. Immunohistochemistry on frozen tissue sections was performed as described elsewhere (9). Antibodies used were anti-5-HTR1A (diluted 1:300), anti-5-HTR1B (diluted 1:400), anti-5-HTR2B (diluted 1:400) and anti-5-HTR4 (diluted 1:500), all obtained from Santa Cruz, CA, USA. The final estimation of stainings was performed by one of the authors (RH) by microscopy at two different occasions without knowledge of the results and with almost complete coincidence. The estimation was semiquantitative, being strong if more than two thirds of the cells were immunoreactive, intermediate if one third to two thirds stained positively and weak if less than one third of the cells were positively stained.

*Statistical methods*. Differences in antigen expression between the groups were estimated with the two-tailed Fisher exact probability test (12). The survival times were assessed from the day of operation. Cumulative survival curves were constructed according to the Kaplan-Meier method (13) and differences in survival were estimated with the log-rank test (14).

## Results

Results are shown in detail in Table III and are briefly summarized here. Examples of the stainings are shown in Figure 1.

*5-HT1A*. Principally, the same staining pattern was observed in ovaries and benign and borderline tumors with a strong staining in the epithelial fraction and only weak or no expression in the stromal part. In invasive tumour cells, however, the expression of 5-HT1A was strongly decreased. In 22 samples a strong staining was observed, but in 10 less than one-third of the tumour cells were positively stained and two samples were unreactive. The reduced staining did not reach significance compared to that of benign ($p$=0.29) or the borderline ($p$=0.08) groups. Furthermore, no reduced staining was noted in more advanced tumours compared to stage 1 tumours ($p$=1.0). In stroma a weak staining was seen in 4 samples while the remaining 30 were completely negative. No blood vessel seemed to harbour 5-HT1A.

*5-HT1B*. In epithelial cells as well in benign and non-invasive tumor cells the pattern was similar to that of 5-HT1A. In stroma no reaction was detected. In invasive tumour cells we observed a reduced or no staining of 5-HT1B in 15 and 10 samples respectively; while in 8 it was strong. The difference was significant against the benign ($p$=0.02) but not the borderline ($p$=0.13) group. Compared to stage 1 tumours a reduced expression was observed in more advanced tumours ($p$=0.14). In contrast to the other tissue groups a weak staining of the stroma was seen in 12 of the 34 malignant samples. No blood vessels turned positive.

*5-HT2B*. The strong expression of 5-HT2B corresponded well with that of 5-H1B in epithelial cells and benign and

Table I. *Staging of the invasive ovarian tumours in all staining groups. In a few cases information of the staging was missing.*

| Stage | I | II | III | IV | Missing |
|---|---|---|---|---|---|
| 5-HT1A | 11 | 3 | 14 | 2 | 4 |
| 5-HT1B | 10 | 3 | 13 | 2 | 5 |
| 5-HT2B | 11 | 3 | 13 | 2 | 4 |
| 5-HT4 | 5 | 2 | 5 | 1 | 3 |

Table II. *Histology of the invasive ovarian tumours used in the stainings. Serous (ser), mucinous (muc), endometrioid (end), clear cell (cle), mixed (mix) and poorly differentiated (poo).*

| Histology | Ser | Muc | End | Cle | Mix | Poo |
|---|---|---|---|---|---|---|
| 5-HT1A | 11 | 4 | 3 | 7 | 7 | 2 |
| 5-HT1B | 11 | 5 | 7 | 2 | 6 | 2 |
| 5-HT2B | 11 | 5 | 7 | 2 | 6 | 2 |
| 5-HT4 | 5 | 1 | 4 | 1 | 4 | 1 |

Table III. *Expression in different tissue compartments of 5-HTR1A, 5-HTR1B, 5-HTR2B and 5-HTR4 estimated as strong/weak/zero as defined in the Materials and Methods.*

| 5-HT receptor | 5-HT1A | 5-HT1B | 5-HT2B | 5-HT4 |
|---|---|---|---|---|
| Ovary | | | | |
| Epithelium | 2/1/0 | 1/1/0 | 1/1/0 | 5/0/0 |
| Stroma | 0/2/1 | 0/0/3 | 0/3/0 | 0/0/5 |
| Blood vessels | 0/1/2 | 0/0/3 | 2/0/1 | 0/0/5 |
| Benign ovarian tumour | | | | |
| Tumour cells | 11/2/0 | 9/5/0 | 8/6/0 | 6/0/0 |
| Stroma | 0/3/10 | 0/0/14 | 5/8/1 | 0/0/6 |
| Blood vessels | 0/0/13 | 0/0/14 | 1/12/1 | 0/0/6 |
| Borderline ovarian tumour | | | | |
| Tumour cells | 7/0/0 | 3/2/0 | 4/0/1 | 3/0/0 |
| Stroma | 0/0/7 | 0/0/5 | 1/2/2 | 0/0/3 |
| Blood vessels | 0/0/7 | 0/0/5 | 1/2/2 | 0/0/3 |
| Malignant ovarian tumour | | | | |
| Tumour cells | 22/10/2 | 8/15/10 | 8/11/14 | 10/5/1 |
| Stroma | 0/4/30 | 2/12/19 | 12/17/4 | 0/1/15 |
| Blood vessels | 0/0/34 | 0/0/33 | 3/20/10 | 0/0/16 |

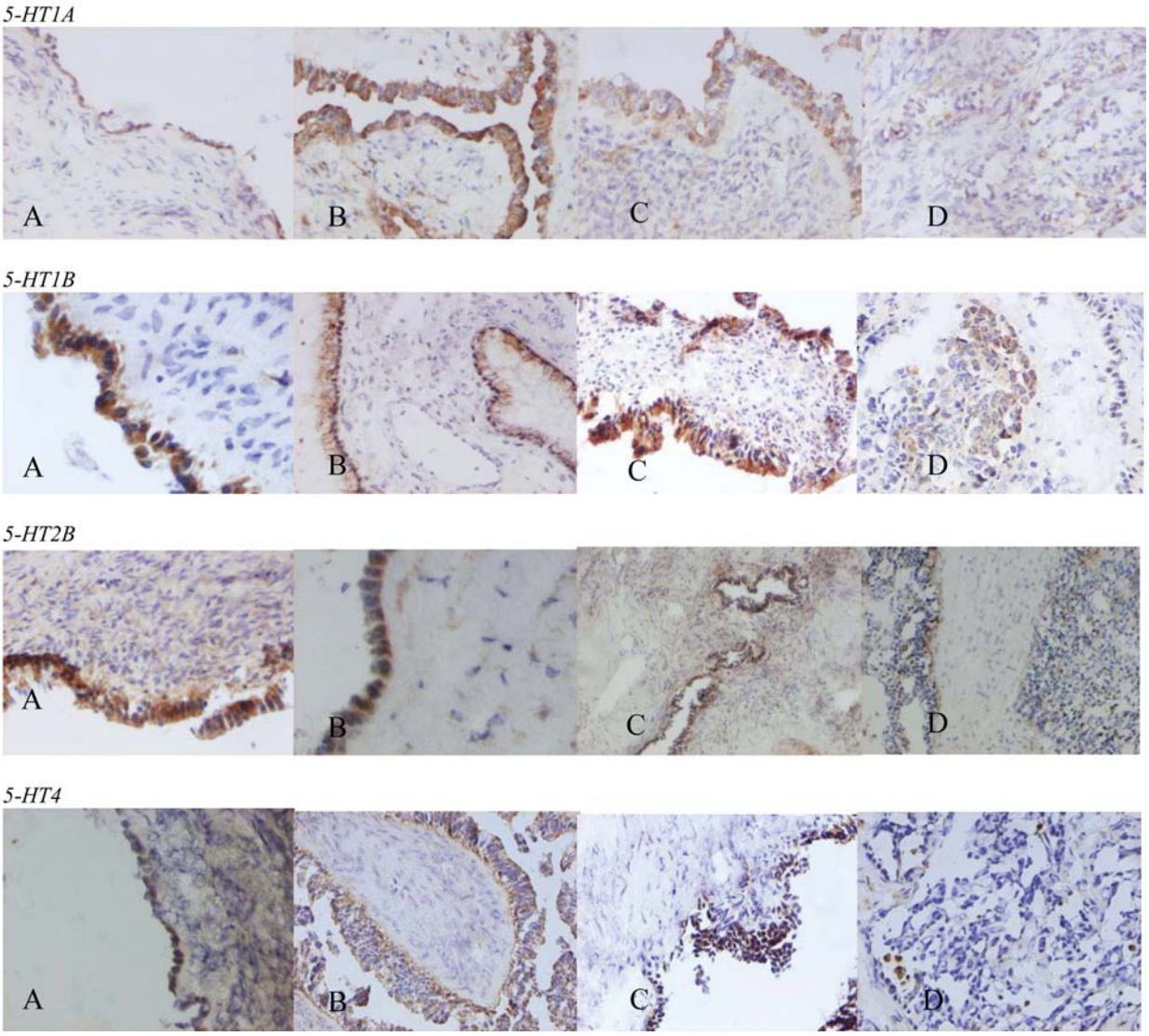


Figure 1. *Expression of 5-HT1A in ovary (A), benign seropapillary ovarian tumour (B), borderline serous ovarian tumour (C), and malignant seropapillary ovarian tumour (D). Magnification, ×200 for all figures. Expression of 5-HT1B in ovary (A), benign mucinous ovarian tumour (B), borderline mucinous ovarian tumour (C) and malignant mixed endometrioid and serous ovarian tumour (D). Magnification, (A) ×400, (B) to (D) ×200. Expression of 5-HT2B in ovary (A), benign mucinous ovarian tumour (B), borderline serous ovarian tumour (C) and malignant seropapillary ovarian tumour (D). Magnification (A) ×200; (B) ×400; (C) and (D) ×100; Expression of 5-HT4 in ovary (A), benign serous ovarian tumour (B), borderline ovarian tumour (C) and malignant mixed endometrioid and serous ovarian tumour (D). Magnification, ×200 for all, except (B) with magnification ×100.*

non-invasive tumor cells. In contrast to 5-H1B, the stroma stained positive in most samples and interestingly, in blood vessels the antigen could be detected in the main part of the samples. The staining pattern of 5-HT2B in the invasive tumour cells was almost equal to 5-HT1B with significant reduction against the benign ($p$=0.04) and the borderline ($p$=0.03) groups. Decreased staining strongly correlated to dissemination of the disease ($p$=0.009). Similar to ovarian and tumour samples blood vessels in the malignant tumours were expressed in a varying degree.

*5-HT4*. The staining pattern in all tissue groups was principally similar to that observed for 5-H1A and 5-H1B. The observed decrease in disseminated disease compared to stage 1 did not reach significance ($p$=0.08), probably because of smaller number of samples.

*Survival analysis*. For all cancer patients a survival analysis was calculated. For none of the receptors the expression was correlated to survival. For 5-HTR2B the Kaplan-Meier plot indicated a decreased survival correlated to reduced staining, however without obtaining significance by the log-rank test ($p$=0.11).

## Discussion

In this study, we have shown that serotonin receptors are expressed in normal ovary and in ovarian tumours, which indicates a role in ovarian physiology and pathology. Although serotonin is best known as a neurotransmitter, only 1% is actually found in the nervous system. The remainder is found in the periphery and is ascribed a number of physiological functions. It may therefore be involved in both the aetiology and pathogenesis of a growing number of pathological conditions (8). A few studies have indicated a role of the serotonin system in ovarian physiology. Two decades ago presynaptic 5-HT receptors on adrenergic nerves in bovine ovarian follicle were characterized (15). In addition, recently, the 5-HT transporter was described in oocytes and the rate limiting step in the production of serotonin, tryptophan hydroxylase, was found in cumulus cells (16). This finding, together with our observed localization of the serotonin receptors in different compartments of the ovary including blood vessels makes serotonin a candidate in regulation of ovulation with the monthly repetitive process of cell proliferation and wound healing. Such a regulatory role is further supported by the recent description of a local 5-hydroxytryptaminergic system in peripheral arteries (17). Moreover, 5-HT was to be found to be almost as effective as noradrenalin in the contraction of ovarian vein preparations (18) and has been discussed in relation to systemic hypertension (19), as well as pulmonary arterial hypertension (20-22) and pulmonary vascular remodelling (23). Taken together with our observed strong expression of 5-HT2B in ovarian blood vessels, vascular regulation by serotonin is probable and deserves further studies.

The pathways in ovarian carcinogenesis are not fully understood. A direct transformation from ovarian epithelium to malignant tumour has been described but is rare. To what extent the malignant transformation passes the stages of benign and borderline tumours is unknown. In benign ovarian tumours, we observed a staining pattern of 5-HT receptors principally as in the ovary. Almost nothing is known about proliferation or other biological parameters *in vivo* of the benign tumours. During earlier studies of tumour markers in ovarian tumour development, we never found proliferation of the benign tumour cells as estimated by the proliferation marker Ki 67 (3).

Strong expression of 5-HT2B was detected in the benign tumour stroma, while the blood vessels showed variable immunoreactivity. In these compartments, only weak and sporadic staining for the remaining receptors was seen. As in the ovaries, their functional significance remains unknown.

In the borderline tumours, non-invasive tumour cells revealed, in principal, the same staining pattern of serotonin receptors as their benign counterparts. At least a fraction of borderline tumours will progress to invasive cancer and should therefore undergo aggressive surgical treatment. Correspondingly, we earlier found indication of proliferative activity by positive staining of Ki 67 in non-invasive malignant tumour cells (3).

In invasive malignant ovarian neoplasms, we observed a decrease in expression in the tumour cells of all serotonin receptors. Furthermore, except for 5-HTR1A the reduction was more pronounced in advanced tumours compared to localized neoplasms. This difference from non-invasive cancer cells may indicate a tumour suppressor role during the invasive step of ovarian carcinogenesis and studies of the metabolism of serotonin indirectly support such a protective role. Thus, in colon carcinoma, serotonin did not influence tumour growth, but did strongly reduce cell invasion (24). As a precursor for serotonin, lower levels of tryptophan are expected to result in lower levels of serotonin. In a study of Schroecksnadel and co-workers, measurements of tryptophan and its degradation products in patients suffering from gynaecological cancer revealed an increased degradation of tryptophan by the enzyme indolamine-(2,3)-dioxygenase (IDO) (25). Furthermore, reduced levels of tryptophan in blood were observed in patients suffering from progressive disease (26). Others have found increased synthesis of IDO to be a marker of poor prognosis in patients suffering from the serous type of epithelial ovarian cancer (27-29). In addition, recent data in a nude mouse xenograft model further substantiate that IDO is positively involved in ovarian cancer progression (29). It is thought that catabolising tryptophan to N-formyl-kynurenine IDO may starve T cells from this important amino acid, creating inappropriate immune responses (30) and recently a trial was performed to reverse the IDO-mediated arrest of T-cell proliferation in serous ovarian cancer (31). A lower level of serotonin has also been suggested to be carcinogenic through reduced apoptosis by a complex interaction with oestrogen (8).

In contrast, in other organ systems, increased levels of serotonin have been positively related to cancer development. Thus, monoamine oxidase-A (MAO-A), an enzyme that down regulates serotonin by metabolization was reduced in several types of non-gynaecological cancer tissues compared to corresponding normal tissues (32). Whether this is true for ovarian cancer has to await future studies. However, the effect may occur through other mechanisms as besides serotonin, MAO-A metabolizes dopamine and its derivates epinephrine and norepinephrine, all of which have been suspected to play a role in cell proliferation and carcinogenesis (reviewed in

32). Because of the supposed increase in cancer risk coupled to down regulation of MAO-A, it has been recommended to use other antidepressive medicine with different modes of action rather than MAO inhibitors. Selective serotonin re-uptake inhibitor (SSRI) is such a pharmacological agent for treatment of depression, and its use in populations has been studied to reveal a possible connection to cancer development. For breast (33) and ovarian cancer (34), no association with SSRI use was observed. For colon cancer, a high daily dose in fact reduced the risk (35). In addition, the intake of SSRI seemed to be associated with a reduced risk of lung cancer (35). SSRI was reported to inhibit the growth of transformed cells and to have a pro-apoptotic effect (37). These data lends further support for a tumour suppressor role of serotonin, at least in some tissues.

However, substantial evidence exists for a tissue specific positive role of serotonin, in tumour development. Thus, in prostate cancer, we observed an increased expression of 5-HT1A, 5-HT1B, 5-HT2B and 5-HT4 receptors compared to normal prostate (9, 10). Furthermore, we found that serotonin antagonists inhibited growth of PC cell lines, indicating a tumour promoting role for serotonin in the prostate (10). In a study of bladder cancer cells, antagonists to 5HT1A and 5HT1B both inhibited bladder cancer cell growth (38). In liver regeneration, the expression of 5-HT2A and 5-HT2B receptors in the liver parenchyma increased after hepatectomy (39). That the serotonin system played a role in liver regeneration was supported by the observation that serotonin agonist in thrombocytopenic mice restored liver regeneration. Thus, solid data exist for both stimulatory and inhibitory roles of serotonin in cell proliferation and carcinogenesis.

Ovarian steroids may be an important factor in ovarian carcinogenesis as receptor studies suggest an increased ER $\alpha$: ER $\beta$ ratio implicating a selective growth advantage for ER $\alpha$ positive cells which by reducing 5HT1A receptors (40) may increase the risk of malignancy through reduced apoptosis (reviewed by 41). Furthermore, observations of regeneration of liver, as well as in hepatocellular carcinoma in rats, indicated that activated 5-HT1A receptor inhibited hepatocyte DNA synthesis and that 5-HT1A receptor function was of significance in the control of cell proliferation (42). This is in agreement with our observation that reduced 5-HT1A receptor expression in invasive cancer compared to borderline and benign tumours might be a permissive step.

As discussed above, another perspective of serotonin in carcinogenesis is the vasoconstrictor effect. This is another possible contribution to the growth of ovarian tumours by local vasodilatation in parallel to decreased expression of 5-HT2B in blood vessels with resultant increased blood flow necessary for tumour cell proliferation.

In conclusion, serotonin receptors are widely expressed in ovary and in ovarian tumours. The decreased expression in invasive neoplasms may indicate a tumour suppressor role in ovarian carcinogenesis. In fact, several observations in the literature support such a role. On the other hand, several reports indicate a positive role of serotonin in the carcinogenesis of some non-gynaecological tumours. A possible explanation may be that the serotonin receptors may react in an organ specific manner or that the biological response to serotonin is determined by the combined effect of several receptors.

## Conflict of Interest

The Authors declare that they have no conflict of interest.

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## References

1 Jemal A, Siegel R, Ward E, Murray T, Xu J, Smigal C and Thun MJ: Cancer statistics, 2006. CA Cancer J Clin *56*: 106-130, 2006.

2 Henriksen R, Funa K, Wilander E, Bäckström T, Ridderheim M and Oberg K: Expression and prognostic significance of platelet-derived growth factor and its receptors in epithelial ovarian neoplasms. Cancer Res *53*: 4550-4554, 1993.

3 Henriksen R, Strang P, Backstrom T, Wilander E, Tribukait B and Oberg K: Ki-67 immunostaining and DNA flow cytometry as prognostic factors in epithelial ovarian cancers. Anticancer Res *14*: 603-608, 1994.

4 Kurman RJ and Shih IM: Pathogenesis of ovarian cancer: lessons from morphology and molecular biology and their clinical implications. Int J Gynecol Pathol *27*: 151-160, 2008.

5 Yap TA, Carden CP and Kaye SB: Beyond chemotherapy: targeted therapies in ovarian cancer. Nat Rev Cancer 9: 167-181, 2009.

6 Eliassen AH and Hankinson SE: Endogenous hormone levels and risk of breast, endometrial and ovarian cancers: prospective studies. Adv Exp Med Biol *630*: 148-165, 2008.

7 Zheng H, Kavanagh JJ, Hu W, Liao Q and Fu S: Hormonal therapy in ovarian cancer. Int J Gynecol Cancer *17*: 325-338, 2007.

8 Rybaczyk LA, Bashaw MJ, Pathak DR, Moody SM, Gilders RM and Holzschu DL: An overlooked connection: Serotonergic mediation of estrogen-related physiology and pathology. BMC Womens Health *5*: 12, 2005.

9 Dizeyi N, Bjartell A, Nilsson E, Hansson J, Gadaleanu V, Cross N and Abrahamsson PA: Expression of serotonin receptors and role of serotonin in human prostate cancer tissue and cell lines. Prostate *59*: 328-336, 2004.

10 Dizeyi N, Bjartell A, Hedlund P, Taskén KA, Gadaleanu V and Abrahamsson PA: Expression of serotonin receptors 2B and 4 in human prostate cancer tissue and effects of their antagonists on prostate cancer cell lines. Eur Urol *47*: 895-900, 2005.

11 Fanburg BL and Lee SL: A new role for an old molecule: serotonin as a mitogen. Am J Physiol *272*: 795-806, 1997.

12 Armitage BG: Statistical Methods in Medical Research. Second edition. Blackwell Scientific Publications, Oxford, UK, 1987.
13 Kaplan EL and Meier P: Nonparametric estimation from incomplete observations. J Am Stat Assoc *53*: 457-481, 1958.
14 Mantel N: Evaluation of survival data and two new rank order statistics arising in its consideration. Cancer Chemother Rep *50*: 163-170, 1966.
15 Kannisto P, Owman C, Schmidt G and Sjöberg NO: Characterization of presynaptic 5-HT receptors on adrenergic nerves supplying the bovine ovarian follicle. Br J Pharmacol *92*: 487-497, 1987.
16 Dubé F and Amireault P: Local serotonergic signaling in mammalian follicles, oocytes and early embryos. Life Sci *81*: 1627-1637, 2007.
17 Ni W, Geddes TJ, Priestley JR, Szasz T, Kuhn DM and Watts SW: The existence of a local 5-hydroxytryptaminergic system in peripheral arteries. Br J Pharmacol *154*: 663-674, 2008.
18 Stones RW, Beard RW and Burnstock G: Pharmacology of the human ovarian vein: responses to putative neurotransmitters and endothelin-1. Br J Obstet Gynaecol *101*: 701-706, 1994.
19 Watts SW: The beginning of a fantastic, unanswered question: is 5-HT involved in systemic hypertension? Am J Physiol Heart Circ Physiol *295*: H915-H916, 2008.
20 Martin K, Klinger JR and Rounds SI: Pulmonary arterial hypertension: new insights and new hope. Respirology *11*: 6-17, 2006.
21 Kaumann AJ and Levy FO: 5-Hydroxytryptamine receptors in the human cardiovascular system. Pharmacol Ther *111*: 674-706, 2006.
22 MacLean MR and Dempsie Y: Serotonin and pulmonary hypertension--from bench to bedside? Curr Opin Pharmacol *9*: 281-286, 2009.
23 Esteve JM, Launay JM, Kellermann O and Maroteaux L: Functions of serotonin in hypoxic pulmonary vascular remodeling. Cell Biochem Biophys *47*: 33-44 2007.
24 Huhnt W and Lübbe AS: Growth, microvessel density and tumor cell invasion of human colon adenocarcinoma under repeated treatment with hyperthermia and serotonin. J Cancer Res Clin Oncol *121*: 423-438, 1995.
25 Schroecksnadel K, Winkler C, Fuith LC and Fuchs D: Tryptophan degradation in patients with gynecological cancer correlates with immune activation. Cancer Lett *223*: 323-329, 2005.
26 Schroecksnadel K, Fiegl M, Prassl K, Winkler C, Denz HA and Fuchs D: Diminished quality of life in patients with cancer correlates with tryptophan degradation. J Cancer Res Clin Oncol *133*: 477-485, 2007.
27 Okamoto A, Nikaido T, Ochiai K, Takakura S, Saito M, Aoki Y, Ishii N, Yanaihara N, Yamada K, Takikawa O, Kawaguchi R, Isonishi S, Tanaka T and Urashima M: Indoleamine 2,3-dioxygenase serves as a marker of poor prognosis in gene expression profiles of serous ovarian cancer cells. Clin Cancer Res *11*: 6030-6039, 2005.
28 Takao M, Okamoto A, Nikaido T, Urashima M, Takakura S, Saito M, Saito M, Okamoto S, Takikawa O, Sasaki H, Yasuda M, Ochiai K and Tanaka T: Increased synthesis of indoleamine-2,3-dioxygenase protein is positively associated with impaired survival in patients with serous-type, but not with other types of, ovarian cancer. Oncol Rep *17*: 1333-1339, 2007.
29 Inaba T, Ino K, Kajiyama H, Yamamoto E, Shibata K, Nawa A, Nagasaka T, Akimoto H, Takikawa O and Kikkawa F: Role of the immunosuppressive enzyme indoleamine 2,3-dioxygenase in the progression of ovarian carcinoma. Gynecol Oncol *115*: 185-192, 2009.
30 Zamanakou M, Germenis AE and Karanikas V: Tumor immune escape mediated by indoleamine 2,3-dioxygenase. Immunol Lett *111*: 69-75, 2007.
31 Qian F, Villella J, Wallace PK, Mhawech-Fauceglia P, Tario JD Jr, Andrews C, Matsuzaki J, Valmori D, Ayyoub M, Frederick PJ, Beck,A, Liao J, Cheney R, Moysich K, Lele S, Shrikant P, Old L and Oduns IK: Efficacy of levo-1-methyl tryptophan and dextro-1-methyl tryptophan in reversing indoleamine-2,3-dioxygenase-mediated arrest of T-cell proliferation in human epithelial ovarian cancer. Cancer Res *69*: 5498-5504, 2009.
32 Rybaczyk LA, Bashaw MJ, Pathak DR and Huang K: An indicator of cancer: downregulation of monoamine oxidase-A in multiple organs and species. BMC Genomics 9: 134, 2008.
33 Coogan PF, Palmer JR, Strom BL and Rosenberg L: Use of selective serotonin reuptake inhibitors and the risk of breast cancer. Am J Epidemiol *162*: 835-838, 2005.
34 Moorman PG, Berchuck A, Calingaert B, Halabi S and Schildkraut JM: Antidepressant medication use [corrected] and risk of ovarian cancer. Obstet Gynecol *105*: 725-730, 2005. Erratum in: Obstet Gynecol. 105: 1495, 2005.
35 Xu W, Tamim H, Shapiro S, Stang MR and Collet JP: Use of antidepressants and risk of colorectal cancer: a nested case-control study. Lancet Oncol *7*: 301-308, 2006.
36 Toh S, Rodríguez LA and Hernández-Díaz S: Use of antidepressants and risk of lung cancer. Cancer Causes Control *18*: 1055-1064, 2007.
37 Serafeim A, Holder MJ, Grafton G, Chamba A, Drayson MT, Luong QT, Bunce CM, Gregory CD, Barnes NM and Gordon J: Selective serotonin reuptake inhibitors directly signal for apoptosis in biopsy-like Burkitt lymphoma cells. Blood *101*: 3212-3219, 2003.
38 Siddiqui EJ, Shabbir MA, Mikhailidis DP, Mumtaz FH and Thompson CS: The effect of serotonin and serotonin antagonists on bladder cancer cell proliferation. BJU Int *97*: 634-639, 2006.
39 Lesurtel M, Graf R, Aleil B, Walther DJ, Tian Y, Jochum W, Gachet C, Bader M and Clavien PA: Platelet-derived serotonin mediates liver regeneration. Science *312*: 104-107, 2006.
40 Lu NZ and Bethea CL: Ovarian steroid regulation of 5-HT1A receptor binding and G protein activation in female monkeys. Neuropsychopharmacology *27*: 12-24, 2002.
41 Leung PC and Choi JH: Endocrine signaling in ovarian surface epithelium and cancer. Hum Reprod Update *13*: 143-162, 2007.
42 Sulaiman P, Joseph B, Kaimal SB and Paulose CS: Decreased hepatic 5-HT1A receptors during liver regeneration and neoplasia in rats. Neurochem Res *33*: 444-449, 2008.

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