Research article

**An indicator of cancer: downregulation of Monoamine Oxidase-A in multiple organs and species**

Leszek A Rybaczyk*1, Meredith J Bashaw2, Dorothy R Pathak3 and Kun Huang*1,4

Address: 1Department of Biomedical Informatics, The Ohio State University, Columbus, OH, USA, 2Department of Psychology, Franklin and Marshall College, Lancaster, PA, USA, 3Departments of Epidemiology and Family Medicine, Michigan State University, East Lansing, MI, USA and 4Comprehensive Cancer Center Biomedical Informatics Shared Resource, The Ohio State University, Columbus, OH, USA

Email: Leszek A Rybaczyk* - rybaczyk.1@osu.edu; Meredith J Bashaw - meredith.bashaw@fandm.edu; Dorothy R Pathak - pathak@msu.edu; Kun Huang* - khuang@bmi.osu.edu

* Corresponding authors

**Abstract**

**Background:** Identifying consistent changes in cellular function that occur in multiple types of cancer could revolutionize the way cancer is treated. Previous work has produced promising results such as the identification of p53. Recently drugs that affect serotonin reuptake were shown to reduce the risk of colon cancer in man. Here, we analyze an ensemble of cancer datasets focusing on genes involved in the serotonergic pathway. Genechip datasets consisting of cancerous tissue from human, mouse, rat, or zebrafish were extracted from the GEO database. We first compared gene expression between cancerous tissues and normal tissues for each type of cancer and then identified changes that were common to a variety of cancer types.

**Results:** Our analysis found that significant down regulation of MAO-A, the enzyme that metabolizes serotonin, occurred in multiple tissues from humans, rodents, and fish. MAO-A expression was decreased in 95.4% of human cancer patients and 94.2% of animal cancer cases compared to the non-cancerous controls.

**Conclusion:** These are the first findings that identify a single reliable change in so many different cancers. Future studies should investigate links between MAO-A suppression and the development of cancer to determine the extent that MAO-A suppression contributes to increased cancer risk.
detecting consistent changes among different types of cancers. One such success is the identification of a mutation in p53, a protein responsible for repairing cellular DNA, which occurs in approximately 50% of all cancers [5]. The discovery of similarities among various cancer tissues is the first step in identifying a common mechanism that contributes to the development of cancer. Once a change is identified, appropriate therapeutic targets can be developed to help physicians identify at-risk individuals and improve patient care. Indeed, novel therapeutic strategies have been developed as a result of the extensive study of p53 [6].

Although best known as a neurotransmitter, only 1% of the tryptophan (Trp) derivative, serotonin (5-HT), is found in the nervous system. The remaining serotonin is found in the periphery and is active in the immune, circulatory, reproductive, musculoskeletal, and gastrointestinal systems [7]. Depending on receptor distributions, serotonin activity can promote or reduce apoptosis [8]. Our previous work explored the activity of serotonin in an array of pathologies, particularly those in which epidemiological data suggests gender differences [7]. We proposed that estrogenic effects on serotoninergic function and receptor distribution could explain gender differences in pathologic incidence, as well as some of the effects of estrogen on breast cancer. Rather than being specific to breast cancer, the role of serotonin and its precursor Trp in cellular physiology suggests that the metabolic pathway of tryptophan and as a result serotonin metabolism may be involved in the promotion or progression of cancers in general. There is some literature supporting this hypothesis [9-17], but further research is needed to understand the exact relationship between Trp or its metabolites and cancer. Recently a mechanism was proposed by which catabolism of Trp by indoleamine 2,3-dioxygenase can be linked to immune evasion in tumor cells [13,18]. Other studies suggest that decreased serum tryptophan levels are predictive of poorer prognosis and quality of life in cancer patients[14]. For serotonin specifically, several studies have shown that the selective serotonin reuptake inhibitors (SSRIs) which prevent the reuptake of serotonin thus increase extracellular serotonin levels, have anti-cancer activity in cancer cell lines [17], decrease incidence of cancer in both animals [19] and humans [9], and can be used as a treatment for lymphoma/leukemia [20].

Using genechip technology to study multiple types of cancer simultaneously we can identify whether multiple cancers have similar gene expression changes [21]. Here, we analyzed an ensemble of cancer genechip datasets focusing on genes involved in tryptophan metabolism, which include serotonergic genes among them. We first compare gene expression between cancerous tissues and normal tissues for each type of cancer and then identify changes that are common to a variety of cancer types.

Results
By conducting a series of analyses focusing on tryptophan related gene expression data (see Table 1 for a list of genes analyzed) in the GEO database (gene expression omnibus) maintained by NCBI [22], we found that only Monoamine Oxidase A (MAO-A, E.C. 1.4.3.4) showed consistent decreased expression, in cancers among a variety of tissues from humans, rodents, and zebrafish. Specifically, only MAO-A expression was significantly altered in all 13 of the datasets that used non-cancerous patients as controls and half of the paired datasets. Table 2 provides specific p-values and the mean fold change in MAO-A for the datasets analyzed. Although the extent of downregulation varied among patients, cumulatively 95.4% of all of the tissue samples from human cancer patients, and 94.2% of all animal cancer cases showed lower MAO-A expression than the single lowest control sample in their respective dataset. Changes in expression for unpaired data are provided in Figure 1.

Within each dataset, between 67% to 100% of patients had MAO-A expression below the lowest control sample (see Table 2 for individual values). Examining data from individual patients among the paired data revealed a remarkable pattern of downregulation in cancerous tissue among paired samples analyzed. Only a subset of the datasets that compared cancerous tissue to normal tissue from the same patient failed to show significant downregulation. The three datasets that did not contain a significant shift in MAO-A expression after correcting for multiple t-tests were the two papillary thyroid cancer datasets and one gastric cancer dataset. However 69% of thyroid cancer patients and 100% of gastric cancer patients exhibited a decrease in expression compared to non-cancerous tissue from the same patient, this discordance suggests that a less pronounced downregulation of MAO-A occurs in patients with these two types of cancer.

Discussion
To our knowledge there has been no previous report of any genetic mutation or expression change that occurs with such high frequency among patients in a variety of cancers. The regularity of this change suggests that low levels of MAO-A might serve as a biomarker for cancer. In addition, it is possible that the lack of significant differences in thyroid and gastric cancer could be a result of an overall decrease in MAO-A levels in both normal and cancerous tissue. Unfortunately, datasets that compared cancerous thyroid and gastric tissues to those of healthy individuals were not available at the time of this study.
In higher species there are two isoforms of Monoamine Oxidase (MAO): MAO-A and MAO-B. In humans MAO-A preferentially metabolizes serotonin and the dopamine derivatives epinephrine, and norepinephrine; specifically MAO-A is responsible for catalyzing the inactivation of serotonin by removal of a terminal amine group. MAO-B metabolizes phenethylamine, and both MAO-A and MAO-B metabolize dopamine. There is a significant amount of literature regarding the role of dopamine in carcinogenesis [10,23,24], and several studies have indicated that it may have an antiproliferative effect [25]. The dopamine derivatives epinephrine and norepinephrine have also been implicated in carcinogenesis [26,27]. These hormones are released in response to stress and are also metabolized by MOA-A. Both epinephrine and norepinephrine are capable of increasing cell growth in tumor cells [28] and drugs that prevent norepinephrine signaling can reduce cancer risk [29]. In contrast the role of serotonin is less clear, while drugs that increase serotonin seem to have a protective effect in leukemias/lymphoms [20] as well as other cancers [12]. Increased serotonin appears to promote mitosis in vitro [11]. However our findings of a consistent change in MAO-A, but not MAO-B, in conjunction with the literature provide some support for our premise that the critical role of MAO-A in cancer occurs via the serotonergic system rather than the dopaminergic or catecholamine system.

MAO-A is the target of monoamine oxidase inhibitors (MAOIs), a class of antidepressants. There have been several epidemiologic studies that examine the risk between antidepressants and cancer [15,30-32]. While certain studies link the use of MAOIs with an increase risk for cancer [30,32] the literature remains inconclusive about the exact risk [15,30,32,33]. Given the downregulation of MAO-A observed in cancerous tissue here, one might

Table 1: Genes listed in the tryptophan pathway in KEGG.

| Gene symbol | Gene name | Gene symbol | Gene name |
|-------------|-----------|-------------|-----------|
| AADAT       | aminoadipate aminotransferase | HEMK1      | HemK methyltransferase family member 1 |
| AANAT       | Arylalkylamine N-acetyltransferase | HSD17B10, HADH2 | hydroxysteroid (17-beta-dehydrogenase) |
| ABPI        | Amiloride binding protein I | HSD17B4    | hydroxysteroid |
| ACAT1, ACAT | acetyl-Coenzyme A acetyltransferase 1 | INDO, IDO | indoleamine-pyruvate 2,3 dioxygenase |
| ACAT2       | acetyl-Coenzyme A acetyltransferase 2 | INDOL1    | indoleamine-pyruvate 2,3 dioxygenase-like 1 |
| ACMSD       | Aminocarboxymuconate semialdehyde decarboxylase | INMT      | indolethylamine N-methyltransferase |
| AFMID       | Arylformamidase | KMO        | Kynurenine 3-monooxygenase |
| ALDH1A3     | Aldehyde dehydrogenase 1 family, member A | KYNU      | kynureninase |
| ALDH1B1     | Aldehyde dehydrogenase 1 family, member B | LCMT1     | Leucine carboxyl methyltransferase 1 |
| ALDH2       | Aldehyde dehydrogenase 2 family | LCMT2     | Leucine carboxyl methyltransferase 2 |
| ALDH3A1, ALDH3 | Aldehyde dehydrogenase 3 family | LNX1    | ligand of numb-protein X 1 |
| ALDH3A2     | Aldehyde dehydrogenase 3 family, member A | MAOA | monoamine oxidase A |
| ALDH7A1     | Aldehyde dehydrogenase 7 family, member A | MAOB | monoamine oxidase B |
| ALDH9A1     | Aldehyde dehydrogenase 9 family, member A | METTL2B, METTL2 | methyltransferase like 2B |
| AOC2        | amine oxidase, copper containing 2 | METTL6    | methyltransferase like 6 |
| AOC3        | amine oxidase, copper containing 3 | NFX1      | nuclear transcription factor, X-box binding |
| AOX1        | Aldehyde oxidase 1 | OGDH      | oxoglutarate |
| ASMT        | acetylsertotonin O-methyltransferase | OGDHL | oxoglutarate dehydrogenase-like |
| CARM1       | Coactivator-associated arginine methyltransferase | PRMT2, HRMT1L1 | Protein arginine methyltransferase |
| CAT         | Catalase | PRMT3, HRMT1L3 | Protein arginine methyltransferase |
| CYP1A1, CYP1 | Cytochrome P450, family 1, subfamily A | PRMT5 | Protein arginine methyltransferase 5 |
| CYP1A2      | Cytochrome P450, family 1, subfamily A | PRMT6, HRMT1L6 | Protein arginine methyltransferase 7 |
| CYP1B1, GLC3A | Cytochrome P450, family 1, subfamily B | PRMT7 | Protein arginine methyltransferase |
| DDC         | Dopa decarboxylase | PRMT8, HRMT1L3, HRMT1L4 | Protein arginine methyltransferase |
| ECHS1       | enoyl Coenzyme A hydratase, short chain, 1, enoyl-Coenzyme A | TDO2 | Tryptophan 2,3-dioxygenase |
| EHHADH      | glutaryl-Coenzyme A dehydrogenase | TPH1, TPRH, TPH | Tryptophan hydroxylase 1 |
| GCDH        | glutaryl-Coenzyme A dehydrogenase | TPH2 | Tryptophan hydroxylase 2 |
| HAAO        | 3-hydroxyanthranilate 3,4-dioxygenase | WARS, IFI53 | tryptophanyl-tRNA synthetase |
| HADH, HADHSC | hydroxyacyl-Coenzyme A dehydrogenase | WARS2 | tryptophanyl tRNA synthetase 2, mitochondrial |
| HADHA       | hydroxyacyl-Coenzyme A dehydrogenase | WBCSCR22 | Williams Beuren syndrome chromosome region |

Genes listed in the tryptophan pathway in KEGG. Each dataset was filtered for the tryptophan related genes and all tryptophan related genes included in the dataset were analyzed as described above. The column labeled Gene symbol contains the abbreviated gene symbol. The column labeled gene name contains the full gene name.
Table 2: Descriptive information on datasets extracted from the GEO database used in this study.

### Human Independent Samples (Case-control)

| GSE Number | GEO Description | Species | Control (N) | Cancer Type (N) | Mean Fold Difference in MAO-A | p-value | Percentage of cancer samples < lowest control |
|------------|----------------|---------|-------------|----------------|------------------------------|---------|---------------------------------------------|
| GSE3189    | Cutaneous malignant melanoma | H Normal (7) | Malignant Melanoma (45) | -15.0 | 4.9*10^-21 | 100% |
| GSE1037    | Lung neuroendocrine tumor classification | H Normal Lung (19) | Small Cell Lung Carcinoma (15) | -9.7 | 1.1*10^-10 | 100% |
| GSE2549*   | Human Malignant Pleural Mesothelioma | H Normal Pleura (5) | Malignant pleural mesothelioma (40) | -7.4 | 4.6*10^-10 | 90% |
| GSE61      | Breast tumor characterization | H Normal Breast (10) | Breast-like Tumors (10) | -5.6 | 1.6*10^-4 | 100% |
| GSE2379    | Hypopharyngeal cancer at various stages of progression | H Normal Uvula (3) | Early Stage (4) | -3.4 | 7.7*10^-4 | 100% |
| GSE3744    | Basal-like breast cancer tumors | H Normal Breast (7) | Basal-like Cancer (18) | -5.7 | | 88% |

* Lung tissue and cell lines were excluded. Only primary tumor samples and normal pleura were analyzed.

### Paired Samples

| GEO Series Number | Cancer Type | Species | Control (N) | Cancer (N) | Fold Difference in MAO-A | p-value | Percentage |
|-------------------|-------------|---------|-------------|------------|--------------------------|---------|-------------|
| GSE3268           | Human Squamous Cell Carcinoma of the lung | H Control (5) | Cancer (5) | -2.7 | 0.001 | 100% |
| GSE781            | Clear Cell Carcinoma of the human kidney | H Normal (7) | Tumor Carcinoma (7) | -2.2 | 0.002 | 100% |
| GSE2514           | Pulmonary adenocarcinoma | H Adjacent Normal (10) | Tumor (10) | -1.7 | 0.004 | 80% |
| GSE2685**         | Gastric cancer | H Normal (6) | Cancer (6) | -3.6 | 0.031 | 100% |
| GSE3678           | Papillary thyroid cancer | H Normal (7) | Papillary thyroid Cancer (7) | -1.2 | 0.081 | 71% |
| GSE3467           | Papillary thyroid cancer | H Normal (9) | Papillary thyroid Cancer (9) | -1.1 | 0.286 | 67% |

**Paired data was extracted and unpaired samples were excluded.

### Animal Models

| GEO Series Number | Cancer Type | Species | Control (N) | Cancer (N) | Fold Difference in MAO-A | p-value | Percentage |
|-------------------|-------------|---------|-------------|------------|--------------------------|---------|-------------|
| GSE2514           | Urethane-induced lung tumor model of pulmonary adenocarcinoma | M Adjacent Normal (10) | Tumor (29) | -1.7 | 6.6*10^-10 | 86% |
| GSE3519***        | Liver cancer model | Z Normal Liver Tissue (10) | Liver Tumor (10) | -2.2 | 1.7*10^-5 | 100% |
| GSE3348           | LH overexpressing virgin mice (luteinizing hormone overexpression causes spontaneous mammary tumors) | M Wild type Breast tissue (3) | LH-overexpressing Breast tissue (3) | -1.7 | 5.9*10^-5 | 100% |
| GSE2426           | Patched heterozygous model of medulloblastoma | M Granule cell Precursor (4) | Tumor Cells from Heterozygotes (5) | -1.6 | 6.9*10^-5 | 100% |
| GSE422            | Colon cancer | M CS7/B6 wild-type (6) | APC(Min/+)+ mutant (10) | -18.9 | 1.1*10^-4 | 100% |
| GSE1872***        | N-methyl-N-nitrosourea-induced breast cancer model (R) | R Normal (11) | Cancer (9) | -1.7 | 5.9*10^-10 | 100% |
| GSE2528           | Mammary tumorigenesis in MMTV-neu model | M Wild type Normal Breast (3) | Mammary tumors in MMTV-neu Model (7) | -1.8 | 0.001 | 100% |

*** Zebrafish do not have separate MAO-A and MAO-B therefore MAO levels were analyzed.

**** Biological replicates were averaged for the analysis.

Descriptive information on datasets extracted from the GEO database used in this study. Cases are grouped by whether control and cancer tissues came from different human patients (independent samples), the same human patients (paired samples), or animal models. Tissues are from humans (H) mice (Mus musculus, M), rats (Rattus norvegicus, R), or zebrafish (Danio rerio, Z). For each dataset, sample sizes of control and cancer tissues are provided. The fold difference in mean intensities of MAO-A describes the amount of suppression of the expression of that gene in cancerous tissue; for example, a fold difference of -4 indicates 4 times less expression in cancer tissue than in normal tissue. P-values reflect significant fold differences in expression between cancer and control tissues using the appropriate t-test; cancer types are listed in order of decreasing significance of MAO-A expression. All datasets showed a significant change as determined by the sequential Bonferroni-Holm adjustment unless marked §. The percentage column contains the percent of individual cancer tissue samples in each data set that had lower levels of MAO-A expression than the lowest single control sample.
expect that MAO-A-specific inhibitors may increase cancer risk. Indeed, MAO-AIs and other antidepressants increase cell proliferation in animal models [34-37]. If the same is true for humans, then MAO-AI’s may be particularly likely to increase cancer risk.

SSRIs, another class of antidepressants, have recently been shown to be effective in vitro at inducing apoptosis in biopsy-like Burkitt’s lymphoma cells, and have been shown in vivo to reduce the risk of colon cancer [9,38]. Visual inspection of data from precancerous tissue in MMTV-Neu and Patched heterozygote transgenic mice suggests that there is more severe downregulation in MAO-A expression in cancer than in the pre-cancerous condition. This study adds to the evidence that SSRIs may be a safer alternative than other antidepressants for treatment of depression in patients with a precancerous condition (such as Atypical Ductal Hyperplasia or Barrett’s Esophagus) or cancer [16].
Since genechips measure mRNA levels within a complex sample we were unable to discriminate between actively dividing cells and those at rest. It is possible the downregulation of MAO-A observed occurs in the resting cells. Although the presence of downregulation in many cancers is compelling, this evidence is not adequate to establish causation, and further clinical studies are needed to determine whether MAOIs and specifically MAO-AIs cause an increase in cancer risk.

**Conclusion**

Most current cancer research is focused on tissue-specific genetic mutations. Familial inheritance (e.g., APC in colon cancer), genetic mutation (e.g., p53), and overexpression of growth receptors (e.g., Her2-neu in breast cancer) can each lead to aberrant replication of a cell. Studies of these changes provide tremendous information about tissue-specific effects but are less informative about common changes that occur in multiple tissues. The similarity in the behavior of cancers from different organ systems and species indicates the potential for a universal change among cancers, regardless of the specific tissue or species. This study suggests that downregulation of MAO-A is such a change and could be an important indicator or even a factor in the development and spread of many types of cancers.

Interestingly, MAO-A inhibitors have been identified in tobacco [39]. If decreased expression of MAO-A is demonstrated in clinical trials to be a risk factor for development of cancer, it may be particularly important for individuals with low levels of MAO-A to be advised against smoking. Future studies should investigate links between smoking, MAO-A suppression, and the development of cancer to determine whether MAO-A suppression might be a mechanism by which smoking contributes to increased cancer risk. There are already previously published reports showing that PET scans can detect decreased MAO-A levels in smokers [40]. Our research shows that whole body PET scans might be a non-invasive way to identify MAO-A "cold spots", where localized downregulation indicates increased risk of cancer at that site. This type of a marker would permit more accurate diagnosis, leading to earlier treatment, and improved outcomes. Clinical studies are needed to determine whether changes in MAO-A can be used as a prognostic indicator of cancer risk in patients with a precancerous state.

Although the role of MAO-A in cancer has yet to be fully understood, examination of our results suggests its expression may act as a marker for the development of cancer. The presence of reduced expression of MAO-A in pre-cancerous states implies its levels indicate progression towards cancer, suggesting that MAO-A levels can be used to identify individuals that should receive increased surveillance and testing for the potential onset of cancer.

**Methods**

19 Genechip datasets consisting of cancerous tissue from 10 different organs derived from human, mouse, rat or zebrafish were extracted from the GEO profiles database. Datasets were first identified by using the search terms "cancer" or "metastasis". All datasets in the GEO profiles database as of March 31st 2007 were considered. The genechip data was selected such that both control and cancer samples were contained in the same dataset. Because of the differences in gene expression that are inherent to cell culture and fixation [41,42], only data derived from non-processed primary biopsies was used. By definition cancer implies an invasive phenotype therefore all other tumor types were excluded such as adenomas, and carcinomas in situ. Among datasets that contained multiple types of cancer, the cancer with the closest number of samples to the control was used for analysis.

Table 2 describes the datasets used, which include 12 human cancer datasets, five mouse cancer datasets, one rat cancer dataset, and one dataset from zebrafish. All animal datasets were derived from cancers that were induced using viral, genetic, or by chemical means. No xenografts were included in this analysis. In total, 242 cancerous samples and 139 control samples were used. Prior to analysis, data that was logarithmic was transformed back to its original values, and all "null" values were excluded. Data points that were from more then one patient, also called pooled samples, were also excluded. Previous reports have shown that probe-sets can not be averaged [43] therefore all analyses were preformed on the probe-set with the highest mean expression value. Each genechip dataset was normalized (both intrachip and interchip) before being deposited in the GEO database. We independently validated the normalization in every dataset by inspecting the distribution of expression values. Datasets that were not appropriately normalized were excluded.

As our interest was in tryptophan-related genes, we examined all human cancer datasets, mouse cancer datasets, and zebrafish tryptophan pathway genes (~60 genes depending on the species and gene chip) listed in Kyoto Encyclopedia of genes and genomes (KEGG) using BRB-array tools [44] developed by the National Cancer Institute's Biometrics Research Branch (NCI BRB). Table 1 lists all the tryptophan genes in KEGG and Figure 2 is a graphical representation of the tryptophan genes included in this analysis, and the number of datasets in which these genes were differentially expressed. First, differences in the expression levels of the genes were examined within each of the individual cancer datasets. We performed either related samples or independent samples t-tests (as appropriate, see Table 2) on
Figure 2
A pathway representation of all the tryptophan related genes analyzed. The figure was created by Ingenuity Pathway Studio, by importing the frequency of differential expression along with the gene name. The genes were then mapped and the frequency of differential expression was overlaid on top of the respective gene. The numbers below each gene represent the number of datasets in which a t-test of the gene’s expression level resulted in $p \geq 0.05$. The intensity of the coloration is provided to show respectively the number of datasets in which differential expression occurred. Gene names are capitalized and small molecules are lower case.
the change in expression of the genes between control and cancer samples for each dataset. Since we focused on the behavior of individual genes across multiple types of cancer rather than groups of genes in individual datasets we only compensated for the multiple t-tests over the composite number of datasets. The probability that we would observe the same false positive from among approximately 60 genes, in at least 16 out of the 19 separate datasets, is at most of the order of 7.8 × 10^{-17}, an unlikely outcome. Therefore finding one specific gene (MAO-A) which is consistently down-regulated in 16 out of 19 datasets is an indication that the observed down-regulation of MAO-A in cancer tissues cannot be attributed to chance alone. Nonetheless, we did adjust for multiple t-tests among the 19 datasets by using the Bonferroni-Holm adjustment [45,46]. Specifically, we sorted the p-values for t-tests of expression of each gene from smallest to largest and compared the i-th p-value to the original alpha level (0.05) divided by the number of data sets+1-i. Thus we compared the smallest (first) p-value with 0.05/18 = 0.0028 and the largest (last) p-value with 0.05/1 = 0.05. For each dataset a list of tryptophan related genes whose expression level was significantly changed using these criteria was generated. Then the frequency of each gene appearing in all the lists was counted and the genes were sorted by the frequency from high to low. Only MAO-A was deregulated in the majority of the datasets analyzed.

**Abbreviations**

MAO-A, Monoamine Oxidase A; qPCR, real time Polymerase Chain Reaction; 5-HT, 5-hydroxytryptamine or Serotonin; MAO-B Monoamine Oxidase B; GEO, National Center for Biotechnology Information Gene Expression Omnibus; KEGG, Kyoto Encyclopedia of genes and genomes; MMTV, Mouse Mammary Tumor Virus; MAO, Monoamine Oxidase; MAOI, Monoamine Oxidase Inhibitor; PET, Positron Emission Tomography, Trp, Tryptophan

**Authors' contributions**

LR developed the original hypothesis for this work, conducted the analysis, participated in the development of the methodology, and contributed to the writing of the final version. MB participated in the development of the methodology, the writing of the manuscript, and validation of the data. DP participated in the validation of the statistics, and the writing of the manuscript. KH participated in the analysis, development of the methodology, validation of the data, and the writing of the manuscript. All authors read and approved the final manuscript.

**Acknowledgements**

This work was partially supported by start-up funds provided by The Department of Biomedical Informatics at The Ohio State University. We thank Kelly Rybaczyk for her support during the research leading up to and the writing of this paper. In addition, we thank Fred Owens for helpful comments on an earlier draft of this manuscript. For their insights in the editing of this paper we thank Dr. Don Holtschu, Kristin Circle, and Dr. Jared Butcher. We are grateful to Lee Cooper for his help with generating the graphics in the manuscript. We also acknowledge Professor P.K. Pathak for his review of our statistical methods.

**References**

1. Cowling VH, Cole MD: Mechanism of transcriptional activation by the Myc oncoproteins. Semin Cancer Biol 2006, 16(4):242-252.
2. Pacal M, Brenner R: Insights from animal models on the origins and progression of retinoblastoma. Curr Mol Med 2006, 6(7):759-781.
3. Contessa JN, Hampton J, Lammering G, Mikkelsen RB, Dent P, Valerie K, Schmidt-Ullrich RK: Ionizing radiation activates Erb-B receptor dependent Akt and p70 S6 kinase signaling in carcinoma cells. Oncogene 2002, 21(25):4032-4041.
4. Codegoni AM, Niccoliti MI, Buraggi G, Valoti G, Giavazzi R, D’Incalci M, Landoni F, Mancos A, Broggini M: Molecular characterisation of a panel of human ovarian carcinoma xenografts. Eur J Cancer 1998, 34(9):1342-1349.
5. Uramoto H, Sugio K, Oyama T, Nakata S, Ono K, Nozoe T, Yasumoto K: Expression of the p53 family in lung cancer. Anticancer Res 2006, 26(3A):1785-1790.
6. Gabrilovich DI: INGN 201 (Adexrin(R)): adenoviral p53 gene therapy for cancer. Expert Opin Biol Ther 2006, 6(8):823-832.
7. Rybaczyk LA, Bashaw MJ, Pathak DR, Moody SM, Gilders RM, Holzschu DL: An overlooked connection: serotonergic mediation of estrogen-related physiology and pathology. BMC Womens Health 2005, 5:12.
8. Adayev T, Ray I, Sondhi R, Sobocki T, Banerjee P: The G protein-coupled 5-HT1A receptor causes suppression of caspase-3 through MAPK and protein kinase Calpha. Biochem Biophys Acta 2003, 1640(1):85-96.
9. Xu W, Tamin H, Shapiro S, Stang MR, Collet JP: Use of antidepressants and risk of colorectal cancer: a nested case-control study. Lancet Oncol 2006, 7(4):301-308.
10. Sakakura C, Takemura M, Hagiwara A, Shimomura K, Miyagawa K, Nakashima S, Yoshikawa T, Takagi T, Kin S, Nakase Y, Fujyama J, Hayasaki Y, Okazaki Y, Yamagishita S, Nakashima S, Yoshikawa T, Takagi T, Kin S, Nakase Y, Fujyama J, Hayasaki Y, Okazaki Y, Yamagishita S: Serotonin and serotonin antagonists and risk of colorectal cancer: a nested case-control study. Lancet Oncol 2006, 7(4):301-308.
11. Groth L, Kottke M, Hesse E, Klingebiel T, Griesinger C, Knapp W, Förster A: Diminished quality of life in patients with cancer correlates with tryptophan degradation. J Cancer Res Clin Oncol 2007, 133(7):477-485.
12. Harlow BL, Cramer DW, Baron JA, Titus-Ernstoff L, Greenberg ER: Tissue specific expression of dopa decarboxylase in peritoneal dissemination of gastric cancer and its potential as a novel marker for the detection of peritoneal micrometastases with real-time RT-PCR. Br J Cancer 2004, 90(3):665-671.
13. Siddiqui Ej, Shabbir MA, Mikhailidis DP, Muratza FH, Thompson CS: The effect of serotonin and serotonin antagonists on bladder cancer cell proliferation. BJU Int 2006, 97(3):634-639.
14. Abdul M, Anezins PE, Logothetis CJ, Hoover NM: Growth inhibition of human prostatic carcinoma cell lines by serotonin antagonists. Anticancer Res 1994, 14(3A):1215-1220.
15. Zamanakou M, Germenis AE, Karanikas V: Tumor immune escape mediated by indoleamine 2,3-dioxygenase. Immunol Lett 2007, 111(2):59-75.
16. Schroeksnadel K, Fieg M, Prass K, Winkler C, Denz HA, Fuchs D: Diminished quality of life in patients with cancer correlates with tryptophan degradation. J Cancer Res Clin Oncol 2007, 133(7):477-485.
17. Harlow BL, Cramer DW, Baron JA, Titus-Ernstoff L, Greenberg ER: Psychotrophic medication use and risk of epithelial ovarian cancer. Cancer Epidemiol Biomarkers Prev 1998, 7(8):697-702.
18. Tan K, Rodriguez LA, Hernandez-Diaz S: Use of antidepressants and risk of lung cancer. Cancer Causes Control 2007, 18(10):1055-1064.
19. Rossetto M, Frasselli M, Tessei A, Zoli W, Conti M: Cytotoxicity of different selective serotonin reuptake inhibitors (SSRIs) against cancer cells. J Exp Ther Oncol 2006, 6(1):23-29.
20. Tankiewicz A, Dzieniamczyk D, Buczko P, Szarmach IJ, Grabowska SZ, Pawlak D: Tryptophan and its metabolites in patients with oral squamous cell carcinoma: preliminary study. Adv Med Sci 2006, 51 Suppl 1:221-224.
21. Martarelli D, Martarelli B, Pediconi D, Nabissi MI, Perfumi M, Pompei P: Hypericum perforatum methanolic extract inhibits growth of human prostatic carcinoma cell line orthotopically implanted in nude mice. Cancer Lett 2004, 210(1):27-33.
22. Schuster C, Fernbach N, Rix U, Superti-Furga G, Holy M, Freiressmuth M, Szcze HH, Sexl V: Selective serotonin reuptake inhibitors--a
new modality for the treatment of lymphoma/leukaemia? Biochem Pharmacol 2007, 74(9):1424-1435.
21. Rhodes DR, Yu J, Shanker K, Deshpande A, Varambally R, Ghosh D, Barrette T, Pandey A, Chinnaiyan AM: Large-scale meta-analysis of cancer microarray data identifies common transcriptional profiles of neoplastic transformation and progression. Proceedings of the National Academy of Sciences of the United States of America 2004, 101(25):9309-9314.

22. NCBI Gene Expression Omnibus profiles [http://www.ncbi.nlm.nih.gov/sites/entrez?db=geo]
23. Pimenta Fj, Horta MC, Vidigal PV, De Souza BR, De Marco L, Romano-Silva MA, Gomez RS: Decreased expression of DARPP-32 in oral premalignant and malignant lesions. Anticancer Res 2007, 27(4B):2339-2343.
24. Bodei L, Hofland LJ, Ferone D, Mooy CM, Kros JM, Paridaens DA, Baarsma SG, Ferdeghini M, Van Hagen MP, Krenning EP, Kweekoom DJ: In vivo and in vitro detection of dopamine d2 receptor in basal melanomas. Cancer Biother Radiopharm 2003, 18(6):895-902.
25. Senogles SE: D2 dopamine receptor-mediated antiproliferation in a small cell lung cancer cell line, NCI-H69. Anticancer Drugs 2007, 18(7):801-807.
26. Lloyd P: The role of cyclic AMP and oxygen intermediates in the inhibition of cellular immunity in cancer. Cancer Immun Immunother 1996, 43(1):1-9.
27. Entschladen F, Drell TL, Lang K, Joseph J, Zaanker KS: Neurotransmitters and chemokines regulate tumor cell migration: potential for a new pharmacological approach to inhibit invasion and metastasis development. Current pharmaceutical design 2005, 11(3):403-411.
28. Sood AK, Bhaty K, Kamat AA, Landen CN, Han L, Thaker PH, Li Y, Gershenson DM, Lutgendorf S, Cole SW: Stress hormone-mediated invasion of ovarian cancer cells. Clin Cancer Res 2006, 12(2):369-375.
29. Algazi M, Plu-Bureau G, Flahault A, Dondon MG, Le MG: [Could treatments with beta-blockers be associated with a reduction in cancer risk?]. Rev Epidemique Sante Publique 2004, 52(1):23-65.
30. Wallace RB, Sherman BM, Bean JA: A case-control study of breast cancer and psychotropic drug use. Oncology 1982, 39(5):279-283.
31. Chudoba J, Buiss DS, Boudreau DM, Roos MA, Lumley T, Weiss NS: Breast cancer recurrence risk in relation to antidepressant use after diagnosis. Breast Cancer Res Treat 2007.
32. Cotterchio M, Kreiger N, Darlington G, Steingart A: Antidepressant medication use and breast cancer risk. Am J Epidemiol 2000, 151(10):951-957.
33. Sternbach H: Are antidepressants carcinogenic? A review of preclinical and clinical studies. J Clin Psychiatry 2003, 64(4):1153-1162.
34. Li YF, Zhang YZ, Liu YQ, Wang HL, Yuan L, Luo ZP: Moclobemide up-regulates proliferation of hippocampal progenitor cells in chronically stressed mice. Acta Pharmacol Sin 2004, 25(1):1408-1412.
35. Brandes Lj, Arron Rj, Bogdanovic RP, Tong J, Zaborniak CL, Hogg GR, Warrington RC, Fang W, LaBella FS: Stimulation of malignant growth in rodents by antidepressant drugs at clinically relevant doses. Cancer Res 1992, 52(13):3796-3800.
36. Sato G, Chimoto T, Aoki T, Hosokawa S, Sumigama S, Tsukidate K, Sagami F: Toxicological response of rats to a novel monoamine oxidase type-A inhibitor, (5R)-3-[2-((1S)-3-cyano-1-oxiranylmethyl)benzoxazol-6-yl]-5-methoxymethyl-2-oxazolidinone (E2011), orally administered for 13 weeks. J Toxicol Sci 1999, 24(3):165-175.
37. Abdous M, Albert PK, Drobesky E, Filip GJ, Kouassi E: 5-HT1A-mediated promotion of mitogen-activated T and B cell survival and proliferation is associated with increased translocation of NF-kappaB to the nucleus. 2004, 18(1):24-234.
38. Serafeim A, Holder MJ, Grafton G, Chamba A, Drayson MT, Luong QT, Bunce CM, Gregory CD, Barnes NM, Gordon J: Selective serotonin reuptake inhibitors directly signal for apoptosis in biopsy-like Burkitt lymphoma cells. Blood 2003, 101(8):3212-3219.
39. Khaliil AA, Steyn S, Castagnoli N Jr: Isolation and characterization of a monoamine oxidase inhibitor from tobacco leaves. Chem Res Toxicol 2000, 13(1):31-35.