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Relation between Neurotransmitters and NK Cells in Adrenal and Breast Cancer

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Abstract

Purpose: Data on microarray gene expression The Gene Expression Omnibus (GEO) provided information on gene expression. Transcription GEO provided two profiles of human NK cells from breast and adrenal tumors (GSE179509 and GSE143383). Data processing and normalization The Dseq2 tool in the R programming language was used to standardize the raw data from GEO. The following analyses were carried out: fold change and P-value analysis, volcano plot, network analysis, GEPIA, and David pathway analysis. In this paper, using Venny software, we discovered 2 genes that are shared by neurotransmitters and NK cells in breast cancer and adrenal cancer. Between these genes and the pathways, they are a part of, we discovered a network. Pathway analysis revealed that these genes are mostly linked to the neurotransmitter and apoptotic pathways. In breast and adrenal tumors, the genes HRH1 and GABRD were discovered to be connected to NK cells. In response to breast and adrenal tumors, almost all of these genes are effective. It is thus postulated that the diagnosis of breast and adrenal cancer may be affected by the up-or down-regulation of these genes. **Methods:** Microarray gene expression data gene expression data was obtained from the Gene Expression Omnibus (GEO) Transcription 2 profile data of human NK cells from human breast and adrenal cancers were obtained from GEO (GSE179509 and GSE143383). Processing and normalization of data the raw data from GEO were normalized with the Dseq2 package in the R software. Fold change and P value analysis, Volcano plot, network analysis, GEPIA, and David pathway analysis were performed. **Results:** In this article, we found genes common to neurotransmitters with NK cells in adrenal cancer and breast cancer with Venny program, resulting in 2 genes. We identified a network between these genes and pathways they belong to. Pathway analysis showed that these genes are mostly associated with apoptosis and neurotransmitters pathway. **Conclusion:** HRH1 and GABRD genes were found to be associated with NK cells in breast and adrenal cancers. Almost all these genes are effective in response to

breast and adrenal cancers. Therefore, it is hypothesized that downregulation or upregulation of these genes may affect breast and adrenal cancer diagnosis.

Keywords

NK Cells, Breast Cancer, Adrenal Cancer

1. Introduction

Breast tissue can grow into cancer in cases of breast cancer. Breast lumps, altered breast form, dimpling of the skin, milk rejection, fluid emerging from the nipple, an inverted nipple, or a red or scaly patch of skin can all be indicators of breast cancer. Symptoms of distant illness spread include yellow skin, shortness of breath, enlarged lymph nodes, and bone discomfort. Obesity, a lack of exercise, alcoholism, hormone replacement therapy during menopause, ionizing radiation, early age at first menstruation, having children later in life or not at all, being older, having a prior history of breast cancer, and having a family history of breast cancer are risk factors for developing breast cancer [1]. A genetic propensity that is inherited accounts for 5% - 10% of instances and includes, among other things, BRCA mutations. The cells of the inner surface of milk ducts and the lobules that feed these ducts with milk are where breast cancer most frequently manifests itself. Ductal carcinomas are cancers that originate from the ducts, whereas lobular carcinomas are cancers that originate from the lobules. Breast cancer has more than 18 different subtypes. Some form from pre-invasive lesions, including ductal carcinoma *in situ*. A biopsy of the pertinent tissue is used to confirm the breast cancer diagnosis. After a diagnosis is obtained, more tests are performed to see if cancer has progressed outside the breast and to identify the most promising therapies [2]. Breast cancer screening's balance of advantages and disadvantages is debatable. A 2013 Cochrane analysis showed that while most women who test positive for the illness don't really have it, it's unclear if mammographic screening does more damage than good. The U.S. Preventive Services Task Force conducted a study in 2009 and found evidence of benefit in those aged 40 to 70. The group advises women aged 50 to 74 to undergo screening every two years [3]. Those who are at a high risk of getting breast cancer can use the medications tamoxifen or raloxifene to prevent the disease. Another prophylactic treatment is the surgical removal of both breasts in select high-risk women. Cancer patients may have surgery, radiation therapy, chemotherapy, hormone therapy, and targeted therapy, among other therapies. Breast-conserving surgery and mastectomy are two different types of surgery. Breast reconstruction might take place at the time of surgery or subsequently. Treatments for those whose cancer has spread to other bodily areas mostly focus on enhancing comfort and quality of life [4]. The effects of breast cancer vary according to the kind of cancer, the severity of the condition, and the patient's age.

In the United Kingdom and the United States, 5-year survival rates range from 80% to 90%. The percentage of people who survive five years is about lower in poorer nations. With 25% of all occurrences, breast cancer is the most common kind of cancer among women worldwide. In 2018, there were 2 million new cases and 627,000 fatalities consequently. It affects women more frequently than males and is more than 100 times more prevalent in wealthy nations [5]. Adrenal Cancer: Any benign or malignant tumor of the adrenal gland, many of which are famous for their propensity to overproduce endocrine hormones, is referred to as an adrenal tumor or adrenal mass. Malignant adrenal tumors, such as neuroblastoma, adrenocortical carcinoma, and certain adrenal pheochromocytomas, are referred to as adrenal cancer. All adrenocortical adenomas and the majority of adrenal pheochromocytomas are benign tumors that do not penetrate or metastasis into neighboring tissues, but they can have a serious impact on health because they upset the hormone balance [6]. Three distinct layers of endocrine cells make up the adrenal cortex, which is where vital steroid hormones are produced. These include several sex hormones, the mineral corticoid aldosterone, which controls blood pressure and renal function, and glucocorticoids, which are essential for the control of blood sugar, the immune system, as well as the body's reaction to physiological stress. Adrenal cortex tumors, whether benign or malignant, can release steroid hormones with significant clinical repercussions [7]. Adrenocortical adenoma: The benign tumors known as adrenocortical adenomas, which affect the adrenal cortex, are highly prevalent (found in 1% - 10% of people at autopsy). Contrast them with adrenocortical "nodules", which are not actual neoplasms. Adrenocortical adenomas are uncommon in people under the age of 30 and occur equally in both sexes. These neoplasms have two clinical implications. First, since CT scans and magnetic resonance imaging have become more prevalent in a variety of medical contexts, they have been recognized as accidental findings more frequently lately [8]. To rule out the likelihood of early adrenocortical cancer, tests and invasive procedures are required. Second, a small percentage of adrenocortical adenomas (about 15%) are "functional", which means they produce glucocorticoids, mineralocorticoids, and/or sex steroids, leading to endocrine disorders like Cushing's syndrome, Conn's syndrome (hyperaldosteronism), virilization in women, or the feminization of men. Surgical treatment is an option for functional adrenocortical adenomas. Most adrenocortical adenomas are less than 50 grams in weight and less than 2 cm in maximum diameter. However, it is no longer believed that the size and mass of adrenal cortical tumors reliably indicate whether they are benign or malignant. Adrenocortical adenomas are essentially well-circumscribed, solid, homogenous, solitary tumors with a cut surface that is yellow. Bleeding and necrosis are uncommon observations [9]. Adults and is extremely uncommon and severe. Although many adrenocortical adenomas exhibit endocrine failure, certain ACCs may be "functional" in this regard by generating steroid hormones. Most adrenocortical carcinomas are not discovered until they are big due to their placement deep inside the retroperitoneum. They typically spread to the lungs and other organs via

lymphatic and blood channels, as well as major veins like the renal vein and inferior vena cava. Surgery is the most efficient form of therapy, but many people cannot afford it, and the condition has a bad prognosis in general. Other treatments for this condition include hormone therapy, radiation therapy, and chemotherapy [10].

2. Materials and Methods

2.1. Microarray Gene Expression Data

The gene expression data were obtained from the Gene Expression Omnibus (GEO) database. Human NK cells in breast and adrenal cancer tissues were obtained from GEO (GSE179509 and GSE143383).

2.2. Processing and Normalization of Data

The raw data from GEO was normalized with the DESeq2 package in the R software. Normalized transcription profile data consists of 49,373 different genes/49,396 probe sets in human adrenal cancer data and breast cancer data consists of 19,782 different genes/19,782 probe sets in human NK cells. Adrenal data consists of 3 patients with adrenal cancer and 3 control groups and breast data consists of 2 patients with breast cancer and 2 control groups.

2.3. Fold Change and P Value Analysis

Significant genes with fold change greater than |1| (absolute fold change greater than 1) were detected between groups. To group the identified genes, more specifically, genes below 0.05 were selected by calculating the P value, then neurotransmitters in the body and genes common between these two cancer groups and neurotransmitters were selected using the Venny program (Table 1). Analyses were done using GraphPad Prism 9.0.0 (Graphpad Prism 9 Software, San Diego, CA, USA). Genes with a P value less than 0.05 and fold change greater than |1| (absolute fold change greater than 1) were selected.

Genes lFCl greater than 1 and P-value less than 0.05 nin (were performed) Pearson analysis. Network analysis to generate a network based on expression and genetic interactions, the GeneMANIA software was used and genes in similar pathways were identified with Cystoscope.

Table 1. The list of 2 genes which have the most alterations in expression. These genes have fold change greater than |1| (absolute fold change greater than 1) and P value less than 0.05 between all control groups and groups from patients with adrenal and breast cancers. These significant values indicate that the change occurred due to adrenal and breast cancers.

Gene Name	Fold Change	P-value
HRH1	Breast Cancer: −4.767854466 Adrenal Cancer: −4.57780403	Breast Cancer: 1.43E−09 Adrenal Cancer: 2.43E−04
GABRD	Breast Cancer: 1.502207097 Adrenal Cancer: 2.35828359	Breast Cancer: 0.034789099 Adrenal Cancer: 1.46E−04

2.4. Volcano Plot

A scatterplot that displays statistical significance (P value) vs magnitude of change (fold change) is known as a volcano plot. It makes it possible to quickly visually identify genes that have substantial statistical fold changes. These genes could be the ones with the most biological impact. In this plot we wanted to show all genes whose fc and P values are significant. The changes in gene expression in the tissue of breast and adrenal cancer patients were evaluated using whole genome expression data to find their relationship with neurotransmitters. According to the findings, 2 neurotransmitter genes belong to |1| fold change greater than |1| (absolute fold change greater than 1) and a P value less than 0.05. We focused our additional research on genes that influence expression variation among groups. In human breast and adrenal cancer cells from the control groups, one gene was expressed more in the control group and the other gene was expressed more in the cancer groups (Figure 1).

2.5. Plotbox

Box and whisker plots, sometimes called box plots, are a great chart to use when displaying the distribution of data points over a selected metric. These graphs show the ranges of the measured variables. This includes outliers, median, mean, and where most data points fall "in the box." The graph pad prism was used to generate the 9.0 plot box and the difference in expression of genes in cancer and normal cells was shown (Figure 2).

Overall survival and selected genes as a plot box to show the expression of selected genes in normal and cancer groups.

Using a common processing pipeline, it is a newly created interactive web service for examining RNA sequencing expression data of 9736 tumors and 8587 normal samples from the GEPIA, TCGA, and GTEx projects. We examined the

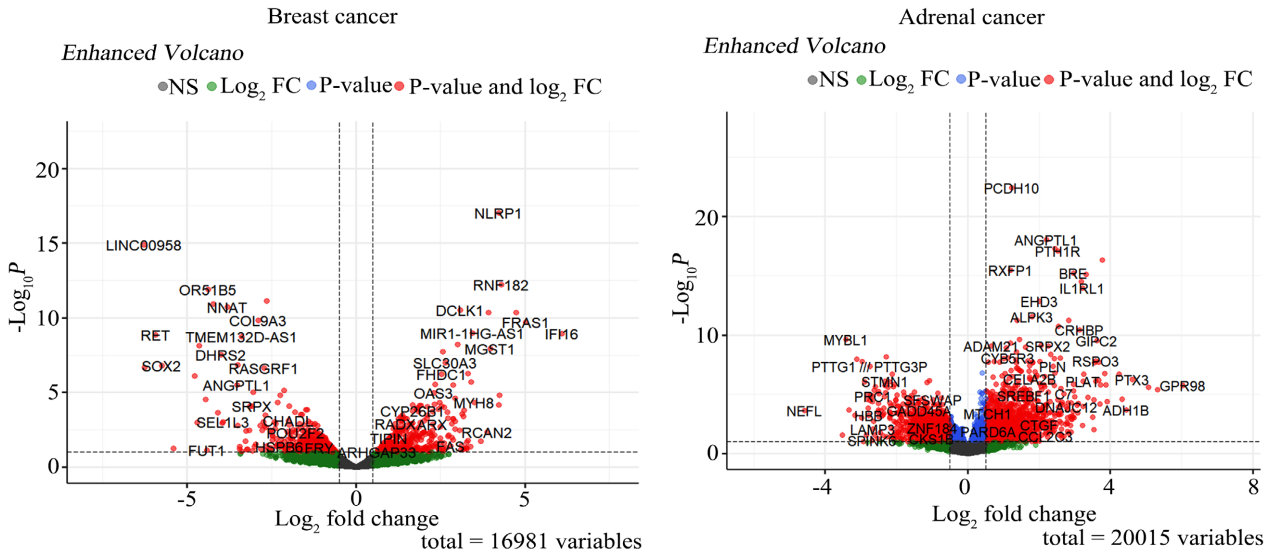

Figure 1. volcano plot shows the log2 of the fold change on the x-axis and minus log10 of the P-value. Genes with P value lesser than 0.05 and fold change greater than |1, 5| (absolute fold change greater than 1.5) are shown.

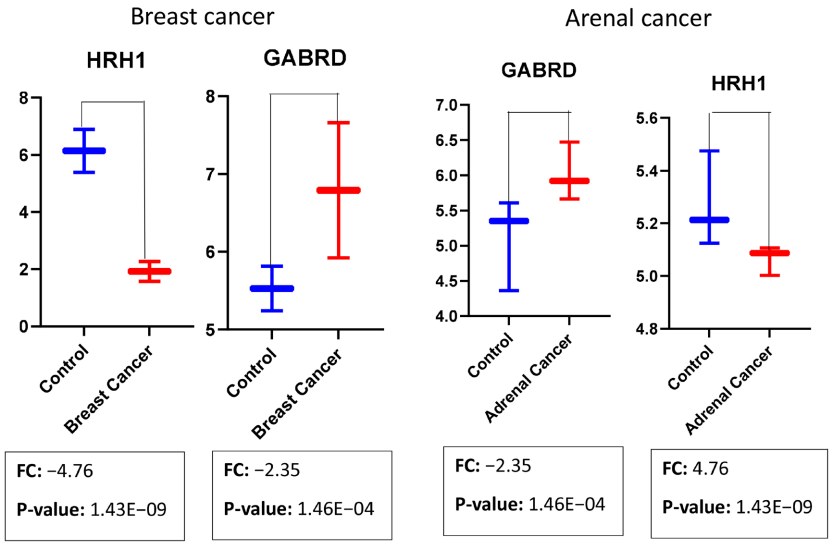

Figure 2. genes that show significant differentially expressed in patients with adrenal and breast cancers NK cells. Statistically significant alterations were detected between control groups adrenal and breast cancer group in NK cells.

expression of genes identified in normal and cancerous tissue using the GEPIA program and showed this expression in a Plotbox, we built an overall survival plot based on these expressions and investigated how low or high expression of these genes affects life. We then compared the plotboxes obtained from the GEPIA program with the plotboxes made with the graphpad prism and showed that the results were fully compatible, which makes our research more robust (**Figure 3**).

2.6. Network Analysis

GeneMANIA software was used to create a network based on co-expression and genetic interactions and identified by genes in similar pathways Cytoscape. Unlike two genes expressed from the 2 gene/2 probe sets, were used as input. The datasets were integrated, analyzed, and visualized to determine whether they were functional to identify similar genes related to each other. Related functions for different gene groups in the network. Thus, the network relationship of these genes was: shown. Software scores each gene. Higher scores indicate more genes (**Figure 4**).

2.7. Pathway Enrichment Analysis

The "Database for Annotation, Visualization, and Integrated Discovery" (DAVID) software was utilized to investigate the biological relationship underlying these genes. The pathways linked to our genes have been discovered. Data processing and normalization The Dseq2 tool in the R programming language was used to standardize the raw data from GEO. The following analyses were carried out: fold change and P value analysis, volcano plot, network analysis, GEPIA, and David pathway analysis. In this paper, using Venny software, we discovered 2 genes that are shared by neurotransmitters and NK cells in breast cancer and adrenal cancer. Between these genes and the pathways, they are a part of, we

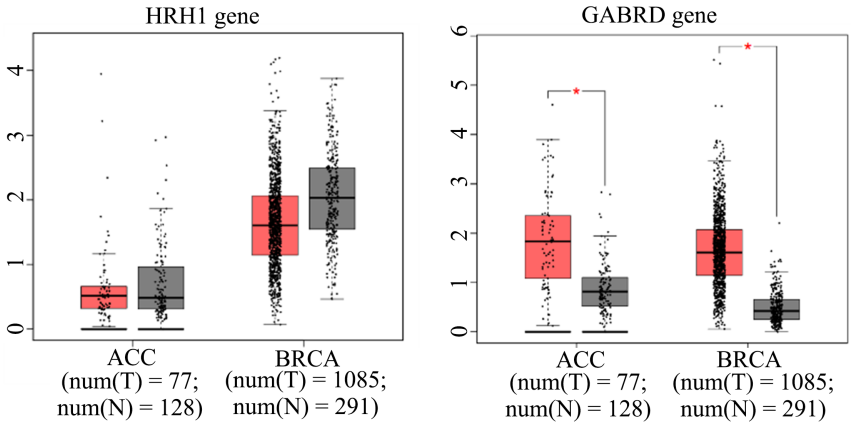

Figure 3. genes that show significant differentially expressed in patients with adrenal and breast cancer. Statistically significant alterations were detected between the control groups cancers group.

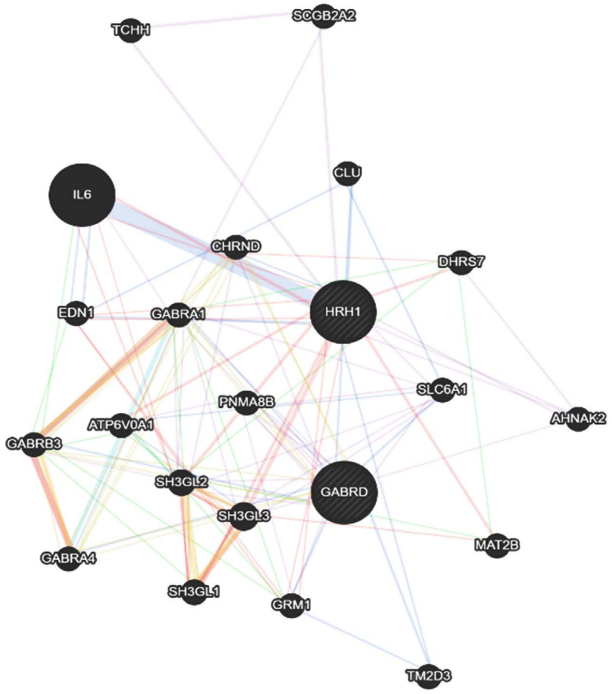

Figure 4. Network analysis of 2 genes that are statistically significant. The figure shows that these 2 genes have a strong network connection (Cytoscape). The linking line between genes illuminates the network of the genes. The thickness of the linking line determines the power of connection of the related genes. This indicates the link formed between these genes has been determined to be stronger by studying more clearly. Additionally, the black nodes indicate the target genes giving by authors. On the other hand, the gray nodes demonstrate the genes which associated genes determined by GeneMANIA application. The size of nodes directly correlated with score.

discovered a network. Pathway analysis revealed that these genes are mostly linked to the neurotransmitter and apoptotic pathways. In breast and adrenal tumors, the genes HRH1 and GABRD were discovered to be connected to NK cells. In response to breast and adrenal tumors, almost all these genes are effec-

tive. It is thus postulated that the diagnosis of breast and adrenal cancer may be affected by the up- or down-regulation of these genes.

Functional enrichment of genes and pathway connections: DAVID software was used to do a pathway analysis of biological processes to discover the link between these 2 genes and cellular activities and pathways, as well as to better grasp their new significance. During adrenal and breast cancer, As a result of the analysis, the GABA-A receptor activity of the GABA-A pathway of these 2 genes, ion channel activity, extracellular ligand-gated ion channel activity, chloride channel activity, protein binding, neurotransmitter receptor activity, donor-gated ion channel activity involved in the regulation of postsynaptic membrane potential, Neuroactive ligand-receptor synapse, GABA-receptor synapse, GABA-receptor-histaminergic synapse interaction, abscissa, gene dependency. activity, G-protein coupled serotonin receptor activity, and neurotransmitter receptor activity showed that.

3. Results

Breast tissue can grow into cancer, and symptoms of distant illness spread include yellow skin, shortness of breath, enlarged lymph nodes, and bone discomfort. Risk factors include obesity, a lack of exercise, alcoholism, hormone replacement therapy, ionizing radiation, early age at first menstruation, having children later in life, and having a family history of breast cancer. Breast cancer is inherited and most often manifests in the inner surface of milk ducts and lobules. It has 18 subtypes, and a biopsy is used to confirm the diagnosis. Breast cancer screening's benefits and disadvantages are debated, and the U.S. Preventive Services Task Force recommends screening every two years. High-risk women can use tamoxifen or raloxifene to prevent breast cancer and may have surgery, radiation therapy, chemotherapy, hormone therapy, and targeted therapy. Breast cancer is the most common kind of cancer among women worldwide, with a 5-year survival rate of 80% to 90% in wealthy nations. Adrenal cancer is a malignant tumor of the adrenal gland that can have a serious impact on health by disrupting the hormone balance. Adrenal cortex tumors can release steroid hormones, which can have significant clinical repercussions. Adrenocortical adenomas are highly prevalent and occur equally in both sexes and have been recognized as accidental findings by CT scans and magnetic resonance imaging. Tests and invasive procedures are needed to rule out early adrenocortical cancer, and functional adrenocortical adenomas can lead to endocrine disorders. Adrenocortical adenomas are well-circumscribed, solid, homogenous, solitary tumors with yellow cut surfaces. Adrenocortical carcinoma (ACC) is a rare and severe condition that affects both children and adults. Surgery is the most effective form of therapy, but it has a bad prognosis. Other treatments include hormone therapy, radiation therapy, and chemotherapy (Table 2).

The family of G protein-coupled receptors like rhodopsin includes the H1 receptor, which is a histamine receptor. The biogenic amine histamine triggers activation of this receptor. It is expressed in smooth muscle, vascular endothelial

Table 2. Pathways related to the genes are linked. It is seen that important pathways in cancer progression and autoimmune disease are related to our genes. Most of these genes are linked to Ca^{2+} signaling pathway, Neuroactive ligand-receptor interaction and Inflammatory mediator regulation signaling pathway from the database for annotation, visualization, and integrated discovery (DAVID).

GABRD	Gamma-aminobutyric acid type A receptor subunit delta
GOTERM_BP_DIRECT	chloride transport, signal transduction, chemical synaptic transmission, ion transmembrane transport, regulation of membrane potential, neurological system process, excitatory postsynaptic potential, chloride transmembrane transport
GOTERM_CC_DIRECT	plasma membrane, integral component of plasma membrane, integral component of membrane, axon, dendrite, chloride channel complex, neuron projection, neuronal cell body, synapse, postsynaptic membrane, GABA-ergic synapse, integral component of postsynaptic membrane, GABA-A receptor complex
GOTERM_MF_DIRECT	GABA-A receptor activity, ion channel activity, extracellular ligand-gated ion channel activity, chloride channel activity, protein binding, neurotransmitter receptor activity, transmitter-gated ion channel activity involved in regulation of postsynaptic membrane potential
INTERPRO	Gamma-aminobutyric acid A receptor, Neurotransmitter-gated ion-channel transmembrane domain, Neurotransmitter-gated ion-channel, Neurotransmitter-gated ion-channel ligand-binding, Gamma-aminobutyric-acid A receptor delta subunit, Neurotransmitter-gated ion-channel, conserved site
KEGG_PATHWAY	Neuroactive ligand-receptor interaction, Retrograde endocannabinoid signaling, GABAergic synapse, Morphine addiction, Nicotine addiction
OMIM_DISEASE	Epilepsy, idiopathic generalized, 10, Epilepsy, juvenile myoclonic, susceptibility to, Generalized epilepsy with febrile seizures plus, type 5, susceptibility to
UP_KW_BIOLOGICAL_PROCESS	Ion transport, Transport
UP_KW_CELLULAR_COMPONENT	Membrane, Postsynaptic cell membrane, Synapse, Cell membrane
UP_KW_DISEASE	Disease variant, Epilepsy
UP_KW_DOMAIN	Signal, Transmembrane, Transmembrane helix
UP_KW_LIGAND	Chloride
UP_KW_MOLECULAR_FUNCTION	Ion channel, Receptor, Chloride channel
UP_KW_PTM	Glycoprotein, Phosphoprotein, Disulfide bond
HRH1	**histamine receptor H1**
GOTERM_BP_DIRECT	inflammatory response, G-protein coupled receptor signaling pathway, G-protein coupled receptor signaling pathway, coupled to cyclic nucleotide second messenger, phospholipase C-activating G-protein coupled receptor signaling pathway, chemical synaptic transmission, memory, visual learning, positive regulation of inositol trisphosphate biosynthetic process, regulation of vascular permeability, positive regulation of vasoconstriction, inositol phosphate-mediated signaling, regulation of synaptic plasticity, eosinophil chemotaxis, cellular response to histamine, G-protein coupled serotonin receptor signaling pathway
GOTERM_CC_DIRECT	cytosol, plasma membrane, integral component of plasma membrane, dendrite, synapse
GOTERM_MF_DIRECT	G-protein coupled receptor activity, histamine receptor activity, G-protein coupled serotonin receptor activity, neurotransmitter receptor activity

Continued

INTERPRO	G protein-coupled receptor, rhodopsin-like, Histamine H1 receptor, GPCR, rhodopsin-like, 7TM
KEGG_PATHWAY	Calcium signaling pathway, Neuroactive ligand-receptor interaction, Inflammatory mediator regulation of TRP channels
SMART	SM01381
UP_KW_CELLULAR_COMPONENT	Membrane, Cell membrane
UP_KW_DOMAIN	Transmembrane, Transmembrane helix
UP_KW_MOLECULAR_FUNCTION	G-protein coupled receptor, Receptor, Transducer
UP_KW_PTM	Glycoprotein, Phosphoprotein, Disulfide bond

cells, the heart, and the central nervous system. The enzyme phospholipase C and the inositol triphosphate (IP3) signaling pathway are both activated by the intracellular G protein (Gq) to which the H1 receptor is linked. Antihistamines that block this receptor are prescribed to treat allergies. The crystal structure of the receptor was determined (shown right/below) and used to find new histamine H1 receptor ligands in structure-based virtual screening experiments [11]. The expression of NF-B, the transcription factor that regulates inflammatory processes, is promoted by the constitutive activity of the H1 receptor as well as by agonists that bind to the receptor [12]. Histamine-activated histamine H1 receptor (H1R) signaling regulates the expression of many genes primarily through protein kinase C (PKC)/extracellular signal-regulated kinases (ERK) signaling. Other signaling pathways have also been shown to be involved, including NF-B, Wnt, RUNX-2 and Rho-A signaling pathways. In addition, cAMP production through activation of the H1R signal has been reported. The H1R gene itself is also upregulated through activation of H1R signaling by histamine. The results suggest that molecular signaling and transcriptional regulation of the H1R gene differ between neuronal and non-neuronal cells [13].

Humans have a gene called GABRD that produces a protein called gamma-aminobutyric acid receptor subunit delta. The delta (δ) subunit in the mammalian brain co-assembles to generate distinct GABAA receptor subtypes, resulting in the subunit-containing GABAA receptors (-GABAA receptors) [14]. One of the hetero-pentameric -GABAA receptors' subunits, the delta () (extra and incorrectly inserted sign) subunit, is a decisive subunit for the unique cellular localization of these GABAA receptors. In the mammalian brain, GABA is the main inhibitory neurotransmitter that interacts with a variety of GABAA receptors, which are ligand-gated chloride channels. It is put together from a variety of subunits, including assembly from a family of 19 subunits [15]. The delta (δ) component is encoded by the GABRD gene. Tonic inhibition, which is slower than classical inhibition (phasic or synaptic inhibition), is specifically mediated by the -subunit, which is often expressed in GABAA receptors linked to extra synaptic activity. The gamma subunit, which is present in the majority of

GABAA receptors, enables the receptor to bind benzodiazepines. The term "benzodiazepine-insensitive" GABAA receptors are often used to describe receptors that include -subunits (also known as -GABAA receptors). These results are not entirely supported by the literature, but they do demonstrate an extraordinarily high sensitivity to ethanol compared to the benzodiazepine-sensitive receptors, which do not respond to ethanol [16]. The ventral tegmental area (VTA) pathway in the brain's hippocampus is known to relate to the -subunit-containing receptors, which suggests that they may have an impact on learning, memory, and reward [17]. GABAA receptors were first cloned using the traditional approach, which involved creating synthetic DNA probes to search brain cDNA libraries using the peptide sequences acquired from purified (bovine brain) receptors. Finally, using this method, the majority of the gene family's isoforms, including subunits 1 through 6, 1 through 3, and 1, were identified [18]. It has been demonstrated that GABA inhibits immunological responses and suppresses T-cell proliferation by binding to active GABA receptors. Human CD4+ and CD8+ T lymphocytes' ability to produce IFN- was reduced by the positive allosteric modulator of GABA known as diazepam [19]. A new study has discovered that GABA is secreted by B cells. By reducing cytotoxic T cell and macrophage invasion and activity, it has been discovered that GABA produced by B cells inhibits antitumor responses. Tumor-infiltrating CD8+ T and NK cells with increased cytotoxic and inflammatory marker expression were more common in patients with B-cell depletion. Additionally, treatment with a wild-type GABAA-R-specific antagonist markedly boosted the production of inflammatory markers in tumor-associated macrophages and the cytotoxic activity of CD8+ T lymphocytes that infiltrate tumors. These result that pharmacological suppression of GABAA-Rs on T-cells or adoptive transfer of GABAA-R-deficient tumor-reactive T cells or APCs may improve the effectiveness of cancer therapies [20]. Additionally, it has been shown that activating GABARDs alters the metabolic and signaling systems of certain tumor cells, preventing, or encouraging migration and replication. According to the kind of tumor, the unique impact of GABARD activation on tumor cell proliferation and migration has recently been examined. Epidemiological research has linked benzodiazepine usage to a higher risk of developing several malignancies. When interpreting these results, care should be taken because some benzodiazepines, including diazepam, have a strong affinity for the widely expressed mitochondrial translocator protein (TSPO), formerly known as the "peripheral benzodiazepine-binding receptor" and a protein modulator [21]. Recent research has revealed that certain tumor cells produce and emit GABA. This tumor-expressed GABA promotes GSK-3 inactivation, which in turn promotes improved -catenin signaling and tumor development. It also suppresses tumors infiltrating CD8+ T and NK cells. These findings demonstrate that in several forms of solid tumors, the GABA/GABRD system can assist tumor cells in dodging immune surveillance. As a result, specifically targeting GAD67/GAD65 or GABRD may be useful as a treatment to slow the growth of certain kinds of

cancers [22].

4. Conclusions

Histamine activates the intracellular calcium pathway by binding to the HRH1 receptor on NK cells, and because of the activation of this pathway, the NK cell also granulates and causes the apoptosis of cancer cells. In this paper, we identified 2 neurotransmitter genes on NK cells that are fully associated with AML. We showed that HRH1 is a type of Gq receptor and functions in the cell by activating DAG and IP3. As a result of histamine binding to HRH1 on NK cells, the NK cell degranulates and secretes perforin and granzyme, which kill the cancer cell. Based on this invention, we speculate that HRH1 antagonists will prevent NK cells from degranulating. The use of HRH1 antagonist drugs for different allergies not only suppresses allergies but also prevents apoptosis of NK cells and may be effective in cancer progression. Histamine, as an immune response activator, stimulates all immune cells, even NK cells. At the same time, it increases the permeability of the vessels, which causes inflammation in the cancer area and finally destroys the tumor cells. Based on this, the use of antihistamine reduces the activation of NK and other immune cells, which in turn reduces the immune response to the tumor. We found that it can reduce the immune response to the tumor, reduce the possibility of cancer, and get rid of cancer. One of them is histamine, which binds HRH1 in NK cells, activates NK cells, and initiates a great fight with cancer cells.

GABA prevents the degranulation and apoptosis of these cells over GABRD on NK cells, thus allowing tumor cell proliferation and metastasis. The other gene we found, GABRD, is a Cl channel, and when GABA binds to this channel, this channel opens, and Cl goes into the cell. The fact that this event occurs in NK cells affects the intracellular signals of these cells and reduces the immune response of the cell a lot, but there are also GABARs in some tumors. And in this respect, this cancer has a positive effect on cancer by reducing the metastasis and proliferation of cells. This Gene reduces migration and motility in all cells, it performs this function by connecting to a CL-channel called GABRD, as a result, it decreases the speed of NK cells. NK cells are suppressed, and the response of NK cells to tumor cells is decreased. But from another point of view, it was found that the probability of GABA binding to tumor cells decreases as well. It is not known to be found in tumor cells, which means that GABA neurotransmitter binds to GABRD in NK cells, suppressing NK suppression and the resultant tumor NK cell response.

Conflicts of Interest

The author declares no conflicts of interest regarding the publication of this paper.

References

[1] Gøtzsche, P.C. and Jørgensen, K.J. (2013) Screening for Breast Cancer with Mam-

mography. *Cochrane Database of Systematic Reviews*, **2013**, CD001877. https://doi.org/10.1002/14651858.CD001877.pub5

[2] Siu, A.L. (2016) Screening for Breast Cancer: U.S. Preventive Services Task Force Recommendation Statement. *Annals of Internal Medicine*, **164**, 279-296. https://doi.org/10.7326/M15-2886

[3] Nelson, H.D., Tyne, K., Naik, A., Bougatsos, C., Chan, B., Nygren, P. and Humphrey, L. (2009) Screening for Breast Cancer: Systematic Evidence Review Update for the US Preventive Services Task Force. Agency for Healthcare Research and Quality (US), Rockville, Report No. 10-05142-EF-1.

[4] Boyd, N.F., Guo, H., Martin, L.J., Sun, L., Stone, J., Fishell, E., Jong, R.A., *et al.* (2007) Mammographic Density and the Risk and Detection of Breast Cancer. *The New England Journal of Medicine*, **356**, 227-236. https://doi.org/10.1056/NEJMoa062790

[5] Kleer, C.G., van Golen, K.L. and Merajver, S.D. (2000) Molecular Biology of Breast Cancer Metastasis. Inflammatory Breast Cancer: Clinical Syndrome and Molecular Determinants. *Breast Cancer Research*, **2**, 423-429. https://doi.org/10.1186/bcr89

[6] Cingam, S.R., Mukkamalla, S.K.R. and Karanchi, H. (2023) Adrenal Metastasis. StatPearls Publishing, Treasure Island.

[7] Lenders, J.W., Eisenhofer, G., Mannelli, M. and Pacak, K. (2005) Phaeochromocytoma. *The Lancet*, **366**, 665-675. https://doi.org/10.1016/S0140-6736(05)67139-5

[8] Arnold, D.T., Reed, J.B. and Burt, K. (2003) Evaluation and Management of the Incidental Adrenal Mass. *Proceedings* (*Baylor University. Medical Center*), **16**, 7-12. https://doi.org/10.1080/08998280.2003.11927882

[9] Willatt, J.M. and Francis, I.R. (2010) Radiologic Evaluation of Incidentally Discovered Adrenal Masses. *American Family Physician*, **81**, 1361-1366.

[10] Arezzo, A., Bullano, A., Cochetti, G., Cirocchi, R., Randolph, J., Mearini, E., *et al.* (2018) Transperitoneal versus Retroperitoneal Laparoscopic Adrenalectomy for Adrenal Tumours in Adults. *Cochrane Database of Systematic Reviews*, **12**, CD011668. https://doi.org/10.1002/14651858.CD011668.pub2

[11] Mitsuchashi, M. and Payan, D.G. (1989) Molecular and Cellular Analysis of Histamine H1 Receptors on Cultured Smooth Muscle Cells. *Journal of Cellular Biochemistry*, **40**, 183-192. https://doi.org/10.1002/jcb.240400207

[12] Braman, S.S. (1987) Histamine Receptors in the Lung. *New England and Regional Allergy Proceedings*, **8**, 116-120. https://doi.org/10.2500/108854187778994446

[13] Mizuguchi, H., Kitamura, Y., Takeda, N. and Fukui, H. (2022) Molecular Signaling and Transcriptional Regulation of Histamine H1 Receptor Gene. *Current Topics in Behavioral Neurosciences*, **59**, 91-110. https://doi.org/10.1007/7854_2021_256

[14] Arslan, A. (2021) Extrasynaptic δ-Subunit Containing GABAA Receptors. *Journal of Integrative Neuroscience*, **20**, 173-184. https://doi.org/10.31083/j.jin.2021.01.284

[15] Korpi, E.R., Gründer, G. and Lüddens, H. (2002) Drug Interactions at GABA(A) Receptors. *Progress in Neurobiology*, **67**, 113-159. https://doi.org/10.1016/S0301-0082(02)00013-8

[16] Goetz, T., Arslan, A., Wisden, W. and Wulff, P. (2007) GABA(A) Receptors: Structure and Function in the Basal Ganglia. *Progress in Brain Research*, **160**, 21-41. https://doi.org/10.1016/S0079-6123(06)60003-4

[17] Vashchinkina, E., Panhelainen, A., Aitta-Aho, T., Korpi, E.R. (2014) GABAA Receptor Drugs and Neuronal Plasticity in Reward and Aversion: Focus on the Ventral Tegmental Area. *Frontiers in Pharmacology*, **5**, Article No. 256.

https://doi.org/10.3389/fphar.2014.00256

[18] Seeburg, P.H., Wisden, W., Verdoorn, T.A., Pritchett, D.B., Werner, P., Herb, A., *et al*. (1990) The GABAA Receptor Family: Molecular and Functional Diversity. *Cold Spring Harbor Symposia on Quantitative Biology*, **55**, 29-40. https://doi.org/10.1101/SQB.1990.055.01.006

[19] Bhandage, A.K. and Barragan, A. (2021) GABAergic Signaling by Cells of the Immune System: More the Rule than the Exception. *Cellular and Molecular Life Sciences*, **78**, 5667-5679. https://doi.org/10.1007/s00018-021-03881-z

[20] Laurie, D.J., Seeburg, P.H. and Wisden, W. (1992) The Distribution of 13 GABAA Receptor Subunit mRNAs in the Rat Brain. II. Olfactory Bulb and Cerebellum. *Journal of Neuroscience*, **12**, 1063-1076. https://doi.org/10.1523/JNEUROSCI.12-03-01063.1992

[21] Christie, S.B., Li, R.W., Miralles, C.P., Yang, B. and De Blas, A.L. (2006) Clustered and Non-Clustered GABAA Receptors in Cultured Hippocampal Neurons. *Molecular and Cellular Neuroscience*, **31**, 1-14. https://doi.org/10.1016/j.mcn.2005.08.014

[22] Tian, J. and Kaufman, D.L. (2023) The GABA and GABA-Receptor System in Inflammation, Anti-Tumor Immune Responses, and COVID-19. *Biomedicines*, **11**, Article No. 254. https://doi.org/10.3390/biomedicines11020254