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G PROTEIN-COUPLED RECEPTORS: ABNORMALITIES IN SIGNAL TRANSMISSION, DISEASE STATES AND PHARMACOTHERAPY

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Abstract: The aim of this review is to present the research results and draw new conclusions about the impact of alterations in the signal transmission through the G protein-coupled receptors (GPCRs) on the formation of diseases and drug therapy. GPCR family is the largest and the most diverse group of membrane receptors. They transmit signals into the cell by interaction with different ligands, which include, *inter alia*, hormones, neurotransmitters, and photons. GPCRs are responsible for the proper conduction of many physiological processes such as vision, intercellular communication, the neuronal transmission, hormonal signaling and are involved in many pathological processes. They are also point on the binding pathway of multiple drugs. They are targets of nearly one third of the drugs at the current pharmaceutical market. The genes encoding GPCRs represent about 4% of the human genome. Mutations that occur in them are associated with a broad spectrum of diseases of diverse etiology. As a mutations result, there is a change in receptor activity (GPCR become inactive, overactive, or constitutively active), in the process of ligand binding and signal transduction. Changes in the GPCRs functioning can cause diseases such as retinitis pigmentosa (rhodopsin mutations), nephrogenic diabetes insipidus (vasopressin receptor mutations), obesity (melanocortin receptor mutations). Many mutational changes in genes encoding GPCR can change drug therapy of already existed diseases: heart failure (adrenergic receptors), asthma (cysteinyl leukotriene receptors). Studies concerning the structure and function of genetically modified GPCRs allow to get know a variety of mechanisms of its action, which in turn can contribute to broaden the knowledge on the etiology and pharmacotherapy of many currently incurable diseases.

Keywords: G protein- coupled receptor, pharmacotherapy

Abbreviations: AC – adenylate cyclase, ADH – autosomal dominant hypocalcemia; AVPR2 – arginine vasopressin receptor 2, β-Arr – β-arrestin, CAM – constitutively active mutants, CaSR – calcium-sensoring receptor, CysLT – cysteinyl leukotrienes, FHH – familial hypocalciuric hypercalcemia, GPCR – G protein-coupled receptor, GRK – G protein-coupled receptor kinase, LT – leukotriene, MC4R – melanocortin-4 receptor, NDI – nephrogenic diabetes insipidus, PLC – phospholipase C, PTH – parathormon, RP – retinitis pigmentosa, SNP – single nucleotide polymorphism

G protein-coupled receptors (GPCRs) represent the largest family of membrane proteins that mediate in the cellular responses process, passing the signal into the cell. They participate in signaling cascades of hormones, neurotransmitters. GPCRs are also important in many physiological processes, play a key role in the vision and the sensing of taste and smell (1). They are also the target for a large group of drugs.

GPCRs have been discovered in 1971 by Martin Rodbell, while scientist studied the formation of cyclic adenosine monophosphate (cAMP) under the influence of glucagon in plasmatic membranes. For the discovery of G-proteins, Martin Rodbell and Alfred Gilman were awarded the Nobel Prize in Physiology or Medicine in 1994. Studies on the GPCR genes sequence revealed the existence of about 800 types of receptors in this family (2). Despite of their large number, the crystal structures are available for less than 20 unique GPCRs of the rhodopsin-like class (3). These findings contributed to the knowledge on the activation mechanism of these receptors. Disorders in GPCR signal transduction are the result of multiple mutations in genes encoding these receptors. Knowledge about the consequences of mutations in the genes encoding the GPCR is particularly important

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due to the fact that, GPCRs are point on the binding pathway for many pharmacologically active substances. Each discovered mutation and its effects, provide important information about the mechanism of the receptor action (4). The purpose of this review was to present research results and new findings concerning the effects of GPCR mutations on the formation of disturbances in cell signaling processes, their importance for the development of diseases and related pharmacotherapy.

Structure and function of receptors coupled with G protein

Receptors located in the cytoplasmatic membrane can be classified based on the number of structures permeating through the membrane on: the ion channels, receptors with tyrosine kinase activity and GPCRs (5). GPCRs have seven hydrophobic transmembrane domains of the α-helix structure, and therefore they have an alternate name – the seven transmembrane receptors (6). GPCRs are known to be very versatile receptors to extracellular signals as diverse as biogenic amines, purines, nucleic acid derivatives, proteins and peptides, odoriferous substances, pheromones, calcium ions, and even photons (6). GPCRs represent a large family of proteins that control many physiological processes and they interact with about 70% drugs used. One of GPCR classifications defined the five family of receptors: rhodopsin-like (family A), secretin-like (family B), glutamate (family C), adhesion receptors and frizzled/taste2 receptors (7). The first family A is the largest of them, also known as a class of rhodopsin like receptors, which comprises nearly 90% of all GPCRs. According to the list created by the International Union of Pharmacology Committee on Receptor Nomenclature and Drug Classification, receptors from family A are encoded by 273 genes including 89, which are so-called orphan receptors, for which ligands are not known. Structure of receptors belonging to this family is similar to the rhodopsin receptor. This is the first known structure of GPCR. The other members of the family A include β1 and β2 adrenergic, opioid, histamine, and dopamine receptors. The second family of GPCRs – secretin-like receptors are encoded by 48 genes. The third family of receptors has a structure similar to metabotropic receptors (encoded by 22 genes). Group called frizzled/taste2 is encoded by 11 genes. They are most closely related to the second family of GPCR (8, 9).

GPCR signal transfer occurs through the activation of heterotrimeric G protein (composed of α, β, and γ subunits) binding guanosine-5'-triphosphate (GTP). This protein, depending on the type of signal, transmits information to the cell through its ability to activate or inhibit a variety of proteins and effector enzymes (10). Subunit α (Gα), depending on the receptor activity (active/inactive), binds guanosine-5'-diphosphate (GDP), or GTP. Ligand interaction with GPCR activates it and causes conformational change of G protein. GTP displaces GDP linked to the α subunit and Gα is disconnected from the Gβγ complex. Active Gα subunit interacts with multiple effectors such as calcium ions, adenylate cyclase (AC), phospholipase C (PLC) and protein kinases (10). Although the β and γ subunits are synthesized separately, they form a biologically inseparable complex Gβγ. Through acetyl and prenyl group it is anchored in the cell membrane, and may regulate the activity of ion channels, PLC, and many other mediators (5).

G-protein type is determined by the type of α subunit. Based on the properties and amino acid similarity in this subunit, four types of G protein can be distinguished: a) Gs protein (stimulating) – activates AC, which is responsible for the conversion of adenosine-5'-triphosphate (ATP) to 3'5'-cyclic adenosine monophosphate (cAMP); b) Gi protein (inhibitory) – inactivates AC by reducing the synthesis of 3'5'-cAMP; c) Gq protein – activate PLC, which hydrolyzes phosphatidylinositol-4,5-bisphosphate (PIP_2) into inositol-1,4,5-tri-4',5'-phosphate (IP_3) and diacylglycerol (DAG); d) $G_{12}/_{13}$ proteins – activate Rho protein (Ras homologous), belonging to a small Ras proteins family (10).

The interaction of ligand with one molecule of GPCR, can activate many G proteins and creates number of second messengers and causes signal amplification. Consequently, one attached molecule can induce a strong physiological response. Appropriate signal amplification depends on the type of G protein involved, the specific characteristics of the receptor and the presence of other proteins, enhancing or extinguishing the signal. A single amino acid substitution in the receptor, can cause a dramatic increase or loss of function, what can cause pathological dysregulation of signal transduction (4). As the result of ligand attachment to the receptor, there is a change in the GPCR conformation and activation of G protein. Gα and Gβγ free subunits transmit the signal through their effectors (E1 and E2, respectively) into the cell and trigger a physiological response. Then, Gα-GTPase hydrolyzes Gα-GTP to Gα-GDP and the phosphoric residue (Pi). This leads to the re-bounding of Gα and Gβγ subunits, GDP connection to Gα and this leads to G protein inactivation (4, 11).

There is a process of desensitization, internalization and renewal of GPCR for the receptor to be again ready to ligand binding. Receptor desensitization protects cell against a constant supply of the signal and prevents uncontrolled stimulation of the receptor. There are two main patterns of desensitization: a) homologous desensitization – associated with an agonist interaction with receptor, specific kinase phosphorylation, and β-arrestin (β-Arr) binding, which inhibits the signal path; b) heterologous desensitization – is independent of agonist and affects less GPCR. In this process, protein kinase A and protein kinase C are involved (4). A key role in the regulation of GPCR desensitization play G protein-coupled receptor-specific kinases (GRK). Receptor upon agonist binding is susceptible to incorporation of kinases, that phosphorylate serine and threonine residues in the C-proximal end of the peptide chain. It should be noted that this process takes place only when the receptor is linked to the ligand. Then, phosphorylated receptor binds to the β-Arr, what blocks G protein-mediated signaling and targets receptors for internalization, and redirects signaling to alternative G protein-independent pathways. The complex receptor-β-Arr is internalized into the cell, in the form of clathrin-coated endosomes (4).

GRK family consists of seven kinases which, on the basis of sequence homology and gene structure, can be divided into three subfamilies: a) GRK1 subfamily – consists of rhodopsin kinase and kinase 7 (GRK7); b) GRK2 subfamily – consists of 2 and 3 kinase β-adrenergic receptor; c) GRK4 subfamily – consists of kinases 4, 5, 6 (12). GRK mutations cause inappropriate desensitization of receptors, resulting in increased activity of GPCRs. For example, kinases from GRK1 subfamily are involved in the pathophysiology of harmful mutations connected with rhodopsin acting, which are related with many diseases associated with hereditary retinal disorders (4).

GPCRs signaling pathway consists of many complex processes, which also involve many regulatory proteins and enzymes. Point mutations of genes encoding GPCRs can cause structural changes in their structure, intracellular interactions, increasing the flexibility of proteins, resulting in a change in the primary activity of receptors (13). Modification of the processes resulting from mutations at each level of signal transduction, may contribute to the pathological changes in a number of communication and may generate molecular phenotypes of numerous diseases, which have a great influence on drug efficacy.

Characteristics of selected GPCR – mutations, pathology and pharmacology

Impaired signaling of various GPCRs is the cause of many congenital and acquired diseases. They are caused by number of mutations, changing the structure and function of receptors (14). These disorders affect GPCR activity, reinforcing the function of receptors (gain-of-function) or loss (loss-of-function). Mutations of the first type are generally acquired, while the second type are mostly inherited. To date, more than 600 mutations detected are deactivating and about 100 mutations activate GPCR, and they are responsible for the formation of more than 30 different diseases. Not every mutation in GPCR initiates the formation of the disease. Mutation can affect pharmacotherapy by changing receptor response under the drug attachment.

The most common types of mutations that occur in the GPCR, are missense and nonsense mutations, small deletions and insertions, that may change the reading frame. GPCR dysfunction due to these different kinds of mutations can be classified depending on which level of receptor maturation they occur. On this basis, four classes of damages have been distinguished (15):

a) the first class – partial or complete deletions and mutations in the gene promoter;
b) the second class – mutations affect mRNA stability, translation, post-translational modifications (nonsense and missense mutations, insertions, deletions, mutations in exons splicing sites) and connected with secretion from the endoplasmic reticulum;
c) the third class – mutations that cause changes in the structure of the receptor and affect the adequate structure formation in the Golgi apparatus. Altered protein is targeted for degradation in the endosome;
d) the fourth class – interfere receptor signaling through changes in the ligand binding domain.

GPCRs are the target for various groups of drugs. Diseases in which drugs action is directed on GPCRs may include cardiovascular, endocrine, may be associated with vision disorders, maintenance of energy homeostasis, coagulation and immune system, and many others (4). This review characterizes some receptors, which impaired function resulting from mutations has led to variety of disease states.

Rhodopsin is a receptor belonging to the family A (rhodopsin like receptors), which is responsible for vision process. It is located in specialized cells of the retina – rod cells. All members of this family are activated by small ligands, such as bio-

genic amines and nucleotides, and rhodopsin is activated by photons. The action of the light is converted into an electrical signal sent to the brain (15). Rhodopsin is the first receptor of GPCRs family, for which high-resolution structure was obtained by the crystallography. Rhodopsin is different from other GPCRs in that it is constantly connected with the inverse agonist – 11-cis-retinal, which maintains the receptor in an inactive state. Photon absorption affects the configuration change of 11-cis-retinal to trans-retinal, and thus the conversion of rhodopsin to its active form – metarhodopsin II. Consequently, transducin (G protein) coupled with rhodopsin is activated by the exchange of GDP to GTP in the α subunit and then initiates a phototransduction cascade (16, 17).

Rhodopsin mutations lead to diseases associated with impaired vision. Twenty percent of them are point mutations that cause improper folding, transport or processing of the receptor (15). Mutations that cause the diseases are often nonsense mutations that lead to a single amino acid substitution in the peptide chain of the receptor. Taking into account the effects of mutations on rhodopsin receptor activity, they can be divided into two groups: a) mutations leading to an increase of receptor activity by creating constitutively active mutants (CAM). These mutant receptors are still capable of activation, even in the absence of exposure to the ligand; b) mutations leading to decreased receptor activity due to changes taking place in the phosphorylation process. The consequence of the most of these mutations is the development of the disease. Most of the known rhodopsin mutations are constitutively active rhodopsin mutants. CAM has modified binding site of an inverse agonist, 11-cis-retinal, and therefore mutated receptor is not inhibited. As a result, there is a dysfunction of rods, resulting in impairment of perception of light in the dark. Constitutively active mutants were first discovered in a severe, progressive disease that is retinitis pigmentosa (RP) (17). RP defines a group of heterogeneous inherited disorders associated with changes and loss of retinal cells. RP is reported to be approximately 1 : 4000 people (18). Mutations in the gene encoding rhodopsin, leading to the formation of CAM, are in positions: Thr4Lys, Asn15Ser, Thr17Met, Pro23His, Pro23Leu, Gln28His, Glu113Gln and Lys296Glu (4). Classic RP begins with the problems in adaptation to the dark vision that during adolescence goes into night blindness (nyctalopia). The next stage of the disease is the progressive loss of peripheral vision in the early years of adulthood. As the disease progresses, there is a total loss of the peripheral vision with the possibility of occurrence of a tunnel vision, and usually to 60 years of age there is a loss of central vision. The symptoms are the result of a progressive retinal dystrophies with reduction of two types of photoreceptors: – rods, which enable vision in black and white in low light intensity; – cone cells, which are responsible for color vision. Degeneration of both types of photoreceptors, occurs in the process of apoptosis. RP is a disease damaging the visual perception, but there are cases in which the disease is associated with other disorders. There are about 30 syndromes co-existing with RP. These include (18):

- Usher syndrome – RP is associated with loss of hearing. There are 3 types of this syndrome. In the first one, hearing loss can be very large and it manifests at birth. Balance difficulties can also occur. In the second type of disease, hearing loss can be moderate or mild and does not increase in time. At last, the third type, hearing loss occurs gradually during adolescence. Usher syndrome is a result of mutations in at least 11 genes;
- Bardet-Biedl syndrome – in this syndrome RP is associated with other disorders such as obesity, hypogonadism, renal failure, or mental retardation. Ten genes have been identified whose mutations are responsible for 70% of RP cases.

Currently, there are no drugs for selective RP pharmacotherapy. There are many different methods to prevent the progressive loss of vision. These are: – vitamins A and E supplementation – the daily dose of 4.5 mg of retinyl palmitate may delay blindness by up to 10 years; – docosahexaenoic acid supplementation, which belongs to the group of ω-3 acids. In the membranes in which rhodopsin is located, there is significant amount of docosahexaenoic acid; – oxygen therapy – in normal conditions, retinal photoreceptors have high oxygen consumption. It is assumed that the supply of oxygen to the retina, can partially rescue photoreceptors and they are able to carry out the necessary metabolic processes (18).

More methods, which are still in research domain are: – gene therapy; – transplantation of retinal cells; – modification of apoptosis. The last method concerns the use of calcium channel inhibitors such as diltiazem and nilvadipine, having an impact on apoptosis. Analyzing the process of photoreceptor cell death, it was found that it was associated with higher concentration of calcium ions. In studies conducted in mice suffering from RP, it was found that D-cis-diltiazem, by blocking the calcium channels in the photoreceptor cells, causes a reduction in its degeneration in examined mice. Properties and action of nilvadipine prove to

be better than these of diltiazem. Nilvadipine is a hydrophobic drug, making it easier to go to the central nervous system and retina. The result of nilvadipine injection in mice was the reduction of calcium ions concentration in the photoreceptors cells, including individuals that D-cis-diltiazem was not acted on. An additional advantage of nilvadipine is the highest antioxidant potential among calcium channel inhibitors, so it can fight against the products of oxidative stress associated with photoreceptor cell death in RP. To date, the study concerning the effect of nilvadipine on the course of RP was performed only in mice and in a small group of people. It was also noted, that the combination therapy of D-cis-diltiazem, taurine and vitamin E has a beneficial effect on improving the patients vision. Studies concerning the effects of calcium channel inhibitors on apoptosis are still conducted (19). Recently, a new method was described, that will help in the prevention of patients predisposed to developing RP. Treatment consists in placing in the conjunctival sac drops of a mixture of insulin with growth factor, which helps in the recovery of Muller cells. It is a glial type of cells in the retina, that protect photoreceptor cells against excess of glutamate and free radicals. Their protective effect concerns rods mainly, which come from the same progenitor cells as the Muller cells. Research on this method of treatment offers hope to achieve a simple treatment (20).

Another group of disorders associated with rhodopsin are mutations in the GRK encoding genes. An example is the Oguchi disease. It is a rare disease involving disturbances of vision at dark, at night and in situations, where there is a little access to light (scotopic vision). The cause of this disease is a mutation in the gene encoding the GRK1 or arestin, and in result, there is a lack of rhodopsin phosphorylation. In patients, who have this mutation, receptor constantly activated by light, constantly activates transducin. This process occurs until the entire amount of 11-cis-retinal is transformed into 11-trans-retinal. The consequence of this transition is the lack of sensitivity to light, which continues until there is a renewal of 11-cis-retinal. In patients, rhodopsin regeneration takes more than two hours, after which the rods again reach their full sensitivity to light. Rhodopsin kinase is in both, the rods and cones, but in Oguchi disease phosphorylation occurs only in the rods (21). *In vitro* studies have shown that deletion in the 5th exon in the gene encoding GRK1, is a null mutation, which abolishes the enzymatic activity of the kinase. In other research, *in vivo*, there was no phosphorylation of rhodopsin. Based on both studies, we can say that desensitization is one of the key processes determining the proper functioning of rhodopsin receptors (4).

α-Adrenergic receptors (α_1, α_2) and β (β_1, β_2, β_3) in combination with endogenous catecholamines (adrenaline and noradrenaline) regulate the activity of the sympathetic nervous system. They are also the point on the binding pathway of many drugs (agonists or antagonists) for the treatment of diseases such as heart failure, asthma, or obesity (4). Many types of adrenergic receptor polymorphisms, which consists of a single nucleotide substitution in the gene sequence encoding the receptor protein (SNP, single nucleotide polymorphism), are known (22). Polymorphisms caused by mutations in the gene promoter can cause changes in the expression of receptor; mutations in coding region can cause changes in binding with ligand and/or G-protein and interfere signal transduction regulation. For example, β_1 receptor variant (arginine at 389 is converted to glycine) and α_2 receptor variant (asparagine at 251 is converted to lysine). These mutations cause receptor dysfunction in the intracellular signaling process and as a result occurs an increase in the function of second messengers. Depending on the type of polymorphism, a variant can have a major impact on the course of the disease, or only a potential risk of its development. Typically, formation of polymorphic receptor gene is not the cause of the disease. Changed receptor often is associated with some disease and mainly affects receptor response to medicines used in the treatment of a specific disease (4).

Among the β-adrenergic receptors, there are three types of receptors: β_1, β_2 and β_3, which are distributed in many organs. The first (β_1 receptor) is located mainly in the heart, where it influences the pulse rate and myocardial contractility, and in the kidneys, in which it directs the release of renin. Therefore, the β_1-AR stimulation leads to the activation of the renin-angiotensin-aldosterone system (RAA). β_2 Receptors are widely distributed in the lungs, and on the surface of smooth muscle cells of blood vessels. As response to stimulation by endogenous ligands of the sympathetic nervous system, the widening of blood vessels (vasodilation) occurs. Thus β_2-AR plays an important role in blood pressure regulation. β_3 Receptors with β_2 receptors are also found in the heart. All three types of receptors play an important role in the pathophysiology of cardiovascular diseases including hypertension, stable and unstable coronary artery disease (angina pectoris), myocardial infarction, ventricular and

supraventricular arrhythmias and chronic heart failure. β Receptors are also a point of capture of many commonly used drugs as these used in the treatment of bronchial asthma (β_2 agonists), and diseases of the cardiovascular system (β_1 antagonists) (22).

The most common **polymorphic variants of the β_1-adrenergic receptor** are the results of a single amino acid replacement at positions 49 and 389. At position 49 in the N-terminal part of the polypeptide chain of the receptor, serine is substituted by glycine and at position 389 in the proximal part of the C-terminus of the peptide chain, arginine is substituted by glycine. In studies carried on hamsters fibroblasts, there has been shown that substitution of the glycine at position 49 in the receptor resulted in a decrease in the number and density of receptors on the cell surface as a result of its down regulation.

Various polymorphisms within the β_1 receptor may affect the body's response to medications. The difference in the individual's response in humans with a mutated form of this protein is particularly important in hypertension, coronary heart disease and heart failure. Therefore, polymorphism Arg389Gly intensifies response to drugs, which are β_1-adrenergic receptor agonists as well as to antagonists for these receptors. It was shown that treatment with dobutamine or adrenaline has better effect on function of the heart of people with Arg389Gly polymorphism, who underwent coronary artery bypass grafting, in relation to persons having a Gly389Arg polymorphism. A similar result was obtained in studies concerning the effect of β_1-AR antagonists on the lowering of blood pressure in healthy subjects and in patients with hypertension and chronic heart failure. It turned out that in patients with a mutation Arg389Gly, a better response to medications was achieved than in patients with Gly389Arg polymorphism. On the basis of this result, it was assumed that persons with Arg389Gly polymorphism in the gene encoding the β_1-AR respond better to treatment with agonists and antagonists of these receptors, in comparison to those with receptor not changed. Therefore, it is suggested that, in order to quicker obtain the favorable therapeutic effects, administration of the higher dose of such drugs to persons without polymorphism Arg389Gly is recommended (22).

In the case of **β_2-AR, polymorphic variants**, positions 16, 27 and 164 of the polypeptide chain of receptor are involved. These mutations have been observed in diseases such as hypertension, asthma, obesity, and certain immune disorders. As in the case of β_1-AR, the presence of polymorphic variants may affect the body's response to a given drug. At the mutation site 16, there is a change from arginine to glycine. Such variant is often associated with elevated level of immunoglobulin E in asthma (4). In addition, those who had Arg16Gly polymorphism, exhibit lower response to β_2-AR agonists in relation to Gly16Arg variant. The receptor with polymorphism at position 16 of the peptide chain, under the influence of long exposure to the agonist is down regulated. Another variant is based on SNP at 27 position, which was converted from glutamine to glycine. As in the case of Gly16Arg, this receptor is also down regulated (14). The third β_2-AR polymorphism results from threonine substitution for isoleucine at 164 position. Despite the fact, that it occurs rarely, it has a significant impact on the function of the receptor. Binding to the G protein and the affinity of β_2-AR to the ligand are reduced. It has also been shown that it causes disturbances in the process of vasodilation, thus contributing to an increase in blood pressure, the frequency of hypertension and other cardiovascular diseases. The study concerning Thr164Ile polymorphism of β_2-AR receptor on 66,770 patients were conducted. In the group of study there were Danish men and women of Caucasian origin. It was demonstrated that the presence of Thr164Ile is associated with increased blood pressure and the other above-mentioned changes in the cardiovascular system in women (23). Thr164Ile polymorphic variant may occur in patients suffering from heart failure and affects response of β_2-AR to agonists. In the context of these results, those who had Thr164Ile polymorphism can be classified into a group of patients for whom there is a need to establish individual therapy treatment of cardiac failure, for example, by increasing the dose of the drug (4). Understanding of the β_2-AR polymorphic variants makes possible to determine their potential impact on the development of the disease and is useful in predicting the response to medications used during treatment.

Heart failure is a disease with a high mortality. The main reasons for its occurrence are: coronary heart disease, hypertension and heart attack. As a result of the above diseases, there is an insufficient blood flow and myocardial ischemia, and necrosis of the tissue. The heart is increasingly attenuated, and the contractions are ineffective. The organism trying to compensate heart disturbances activates systems stimulating heart to work. Mechanisms compensating ineffective myocardial work are: activation of the sympathetic nervous system, stimulation of the RAA system and vasopressin synthesis. As a result of the activation of mechanisms described, there is an increase in cardiac workload,

which leads to the development of heart failure. In the next step, there are changes in the structure of the myocardium. One of them is an abnormal left ventricular hypertrophy, which causes the change of the heart geometry and its dysfunction. In addition, there are risk factors that can facilitate the incidence of heart failure. They can be divided into physiological and genetic. The first group includes age, female gender, and diabetes. The second group includes, among others, changes in the expression of genes encoding the receptors that can influence the activation of signal transduction and transcriptional and translational factors (22). The presence of polymorphic variants of β-adrenergic receptor and their impact on the development of cardiovascular disease are shown in Table 1. In the heart, there are two AR main receptors – β_1 and α_2, and to a lesser amount, β_2 and β_3 receptors. Activation of both types of receptor (β_1 and β_2) causes stimulation of protein Gs and activation of AC, which causes an increase of cAMP concentration. The effect of this signaling cascade is an increase of frequency, strength and speed of contraction of the heart muscle and increase of the relaxation phase of the muscle fibers. β_1-AR is considered to have cardiotoxic properties due to the fact, that its continuous stimulation causes apoptosis of cardiomyocytes. In contrast, β_2 receptor is considered to be cardioprotective.

This receptor may stimulate both Gs and Gi proteins, thereby it can activate two signaling paths. As a result of Gi protein stimulation, kinase Akt is activated, which has anti-apoptotic properties (22, 24). In heart failure, there is a constant stimulation of β_1 receptors by noradrenaline leading to its down

Table 1. Polymorphic variants of β-adrenergic receptors and their influence on the pathogenesis of cardiovascular disease (22, 23).

Receptor	Polymorphism	Signal change	Clinical observations
β_1	Ser49Gly	Gly49 – increase of the susceptibility of receptor on down regulation	Gly49 – reduced risk of deepening failure, heart transplantation or death
	Gly389Arg	Arg389 – binds stronger Gs protein – receptor more sensitive to stimulation with agonist and antagonist	Homozygotes Arg389 – better treatment effects of β-blockers in the form of an increase in left ventricular ejection fraction than Gly389, an increase the risk of arrhythmia. Homozygotes **Gly389** with heart failure – lower oxygen consumption during exercise
β_2	Thr164Ile	Ile164 – reduced affinity for catecholamines, reduced binding of Gs protein and weaker activation of adenylate cyclase Thr164 – increased sensitivity to stimulation of the receptor with agonist	Ile164 – there is an increase in blood pressure, the risk of coronary heart disease
	Arg16Gly	Gly16 – increased susceptibility to agonist-induced receptor down regulation	No apparent effect on the pathophysiology of diseases of the cardiovascular system
	Gln27Glu	Glu27 – increased resistance to agonist-induced receptor down regulation	

regulation. In extreme cases, it may lead to the disappearance of up to 50% of β_1 receptors. Using during long period of β_1-receptor antagonist can restore the original state of these receptors. As a result of continuous stimulation of β_2 receptors, there is not its down regulation but there is an increase of the inhibitory Gi protein concentration. In consequence, there is a weakening of the response to continuous stimulation of the sympathetic nervous system (22).

Melanocortin-4 receptor

More and more people around the world suffer from the problem of obesity. The causes of this civilization disease are lifestyle changes, diet and genetic factors. Significant influence on the development of obesity has a mutation in the gene encoding the melanocortin-4 receptor (MC4R). MC4R participates in regulations relevant to the proper functioning of the body's processes, it is responsible for controlling hunger and satiety. Receptor belongs to the family A, rhodopsin-like GPCRs. It is small, has only 332 amino acids. The receptor may be associated with three types of G proteins: Gs, Gi/o or Gq. Its stimulation results in a further activation of signal transmission by the relay of the second messengers (cAMP), which affects the increase of intracellular calcium ion concentration. In classical signal transduction through the receptor, protein Gs is stimulated (25). MC4R is located in the paraventricular nucleus in the hypothalamus, where it is a part of the melanocortin signaling of appetite and energy balance. MC4R modulation depending on the attached ligand can have opposite effects. After binding with α-melanocortin, receptor stimulates protein Gs, which activates AC, and then executes a further signal transduction cascade. The effect of α-melanocortin interaction with MC4R is mute the center of hunger and stimulation of satiety center. After a meal, a person endowed with genetically modified MC4R do not feel satiety (26). On the contrary, when inverse agonist – agouti-related protein is connected to the receptor, satiety disappears and hunger center is triggered. *In vitro*, MC4R connected with the agouti-related protein shows a continuous inhibition on the hunger center (27). MC4R mutations are the most common cause of monogenic obesity, usually autosomal dominant. It is estimated that such mutations occur with a frequency of 1–6% in children and adults with severe cases of obesity. On the basis of more than 50 types of mutations described so far, three main pathomechanisms of receptor acting were distinguished (26): a) impaired activation of the receptor after ligand interaction; b) decreased expression of receptor on the cell membrane; c) reduced constitutive activity of the receptor. The first group of disorders is determined by mutations in the gene encoding MC4R, changing receptor agonist response. After the interaction of the receptor with α-melanocortin, there is no answer or it is limited and this could happened because of the reduced receptor affinity for agonists or weak signal transduction. Disorders classified in the second group are caused by mutations, that cause the reduction of expression of the receptor on the cell surface. It was demonstrated that 80% of children with severe obesity have showed a partial or complete intracellular retention of the MC4 receptor. In the last group of receptor disorders, there are mutations that result in a loss of constitutive activity of the receptor. It has been shown that the continuous sending satiety signal provided by the constitutive activity of the receptor is required to maintain the energy balance of the body. Mutations that cause this kind of disruption in the functioning of the receptor are the most common defects observed in obese individuals.

Obesity is mainly based on the excessive accumulation of body fat, which substantially affects the conditions of life. Severity of obesity can be measured by the scale of body mass index (BMI). BMI is body mass divided by height in meters to the second power. This index helps to identify individuals who have overweight (BMI = 25) and obesity (BMI = 30) (28). Carried studies have shown that obesity results from environmental and genetics factors. The first of them is disturbed homeostasis between the energy supplied with food and energy spent on physical activity. As a result, people have a positive energy balance, resulting in the deposition of energy reserves in the body fat. Genetic factors constitute 50–90% of the causes of obesity (29). There are 3 types of inherited obesity): a) multigenic obesity – is the most common, but so far it is the least known; b) obesity, which is a part of syndrome – is rare, obesity is one of the phenotypic characteristics of the disease, for example, in Prader and Willi syndrome it is accompanied with hyperphagia and behavioral disorders; c) monogenic obesity – it is rare, but is important in understanding the mechanism of appetite regulation (30). Mutations in the gene encoding MC4R are the most common cause of monogenic obesity. Frequency of monogenic obesity, determined by mutations in the gene MC4 was analyzed within 500 people with severe obesity which started in childhood, and specific phenotype of disease was set up. It has been shown that mutations in genes encoding MC4R occurs in 5.8% of the studied population. Higher growth and higher bone

density was demonstrated in persons with mutations in the MC4 receptor than in obese persons without this mutation (25).

Due to the fact that obesity is a global problem and is classified as a chronic disease, there are many studies on its pharmacotherapy. One of the therapeutic options are drugs binding to the MC4 receptor. Currently, a number of clinical studies are conducted on substances that can be used as medicaments for the treatment of obesity. MC4 receptor agonists must fulfill several requirements. Due to the fact that the receptors are in the brain, such drugs should have a good penetration of the blood-brain barrier. In addition, they must be strictly selective for the MC4 receptor, because the group of melanocortin receptors show a high degree of homology to each other (26, 27). Research is also conducted on drugs that can be used in the treatment of disorders associated with mutations in genes encoding a second class of receptors. Due to the retention of the receptor in the cell, potential drug would have a mechanism of action such as pharmacoperons (25). Universal and often used method in the treatment of obesity caused by excessive eating or genetic factors is increased physical activity. Regular exercise can cause increased energy consumption, cause lose weight and prevent obesity.

Mutations in the arginine vasopressin receptor 2

Vasopressin (antidiuretic hormone) is a hormone released from the pituitary in hypovolemia or hypernatremia. Its function is to control the water reabsorption in the kidney collecting duct cells. Vasopressin interacts with arginine vasopressin receptor 2 (AVPR2), which is located in the membrane of cells of the distal convoluted tubule and

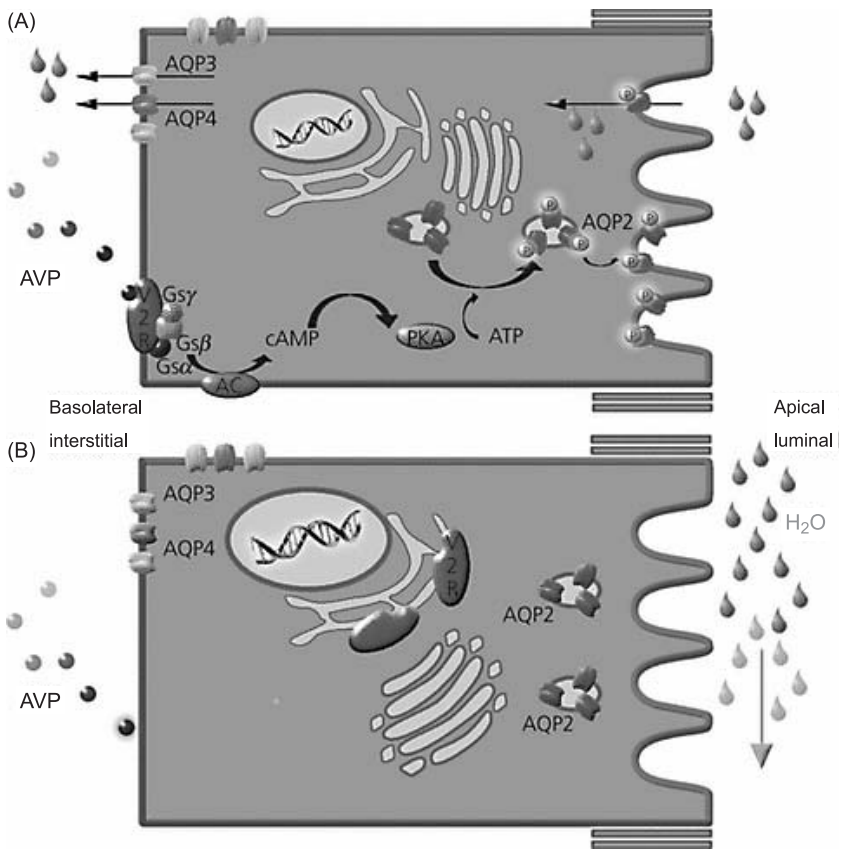

Figure 1. The mechanism of water transport in the collecting duct principal cells in a healthy individual (**A**) and a patient with NDI (**B**). AVP – arginine vasopressin; V2R – vasopressin receptor 2; AC – adenylate cyclase; cAMP – cyclic adenosine monophosphate; PKA –protein kinase A; ATP – adenosine-5'-triphosphate; AQP2 – aquaporin 2; AQP3 – aquaporin 3; AQP4 – aquaporin 4 (33). The description in the text

collecting ducts. This receptor belongs to the family B of GPCR, for which characteristic is the detection of large particles and a long N-terminal domain. The interaction of vasopressin with receptor results in the activation of specific proteins that are responsible for the transport of water into the cells, aquaporin 2. They are tetrameric proteins that form in the cell membrane of the renal tubule channels with diameter corresponding to water molecule (31). In the inactive state, aquaporin 2 is arranged in vesicles inside the cell. After connection to the vasopressin receptor, aquaporin 2 moves from the alveoli to the cell membrane, where they form a transverse channels, increasing membrane permeability to water (31). Currently, 221 mutations are known in the gene encoding AVPR2, that cause nephrogenic diabetes insipidus (NDI), and 21 mutations that do not initiate this disease (23). All mutations can be divided into 15 types within the 4 classes, taking into account the impact of specific disorders on the several steps of receptor maturation process. The most common type of mutations that occur in AVPR2 and the most important in terms of initiating NDI are missense mutations. These belong to the second class of disorders and constitute up to 48% of all mutations occurring in AVPR2 (31). Transcellular water transport is running properly in the body thanks to AVPR2. By the vasopressin connection with AVPR2 in the basal-lateral membrane, Gα subunit is disconnected from the trimeric G protein, then it comes to activation of AC and increase of cAMP levels. Second messenger activates protein kinase A, which stimulates aquaporin type 2 to move to the basement membrane of the follicle cells. In this way, it is possible to move the water through the membrane, from the tubular lumen to the main cell, and transcellular transport, then with aquaporin 3 and 4, the flow to the interstitial nephron. After obtaining a suitable concentration of urine, vasopressin detaches from receptor, and aquaporins 2 are endocytosed or are excreted with the urine (32). The most common disorder arising from mutation of a receptor AVPR2 is NDI. In the people with NDI, there is disruption of the receptor maturation. AVPR2 is permanently attached to the endoplasmic reticulum and cannot be connected to vasopressin in the basal-lateral membrane of the principal cell tubular nephron. In consequence, no water resorption leads to polyuria (33). The process is illustrated in Figure 1. The proper conduction of the mechanism of water transition from collecting duct to the interstitial space is responsible for maintaining the body's water balance and blood pressure regulation. Patients with NDI are unable to concentrate urine and they produce its large quantities. Depending on the initial cause, there are two the most occurring types of diabetes insipidus (32): – central diabetes insipidus – is associated with synthesis or secretion of antidiuretic hormone from the pituitary; NDI – is caused by insensitivity to the renal tubular vaso-

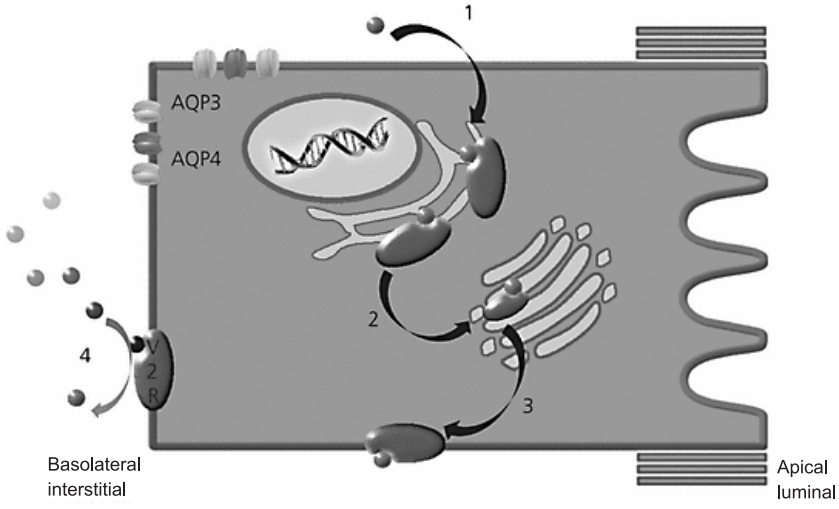

Figure 2. The mechanism of action of the antagonist (pharmacoperons) on the formation of AVPR2. (1) Antagonist passage through the cell membrane into the cytoplasm, an interaction with the mutant receptor in the endoplasmic reticulum (ER) and stabilization of the structure. (2) Escape of receptor from the ER. (3) Aging in the Golgi and location in a cell membrane. (4) High levels of vasopressin displaces antagonist from receptor and activates it

pressin. In these patients, the urine cannot been concentrated despite normal hormone level in the blood. NDI may be inherited or acquired. In almost 90% of cases is inherited and passed on as a recessive disorder linked to the X chromosome. Mutations in the gene encoding AVPR2 usually cause the loss of receptor function. In such cases, the water resorption is disturbed in renal collecting tubule, due to the interference signal of AVPR2 dependent on it expression, and their transport to the main cell membrane (34). The remaining 10% are mostly autosomal recessive mutations or less dominant. These are mutations in the gene encoding aquaporin 2, which result in the molecules that are not displaced from the cell membrane vesicles to the collecting tubule. There are also pathological conditions, which promote NDI formation, which include hypokalemia, hypercalcemia, low-protein diet and lithium therapy (31). Characteristic symptoms occurring in patients with NDI are, *inter alia*, polydipsia, polyuria (from 15 to 20 liters of diluted urine per day) and nocturnal urination (nocturia) (32). In newborns with NDI may occur disorders in eating, difficulties in putting on weight and symptoms of dehydration, which include dryness and loss of elasticity of the skin and dark circles under the eyes. In the case of giving up treatment, among young people there is a disturbance in the development and reduction of growth. Probably, this is due to the problems of nutrition, excessive water intake or recurrent episodes of dehydration. In addition, electrolyte abnormalities such as hypernatremia or hiperchloremia and constant dehydration can cause permanent brain damage, mental retardation and problems with growth and development (33).

The patients with NDI, which undergo therapy, can live to adulthood. In children, quick diagnose and beginning of the treatment can prevent the expansion of mental retardation. The treatment is based primarily on the administration to patient a large amounts of water to prevent dehydration. This is a hindrance in daily life, because the constant fluid intake and an inability to concentrate urine is associated with the need for frequent urination. In addition, elderly patients may have difficulty in the sensation of thirst or in maintaining urine. The use of low-sodium diet, thiazide diuretics and indomethacin are the other methods by which partial reduction of urine amount is possible (32). The expectancy for improving the treatment of NDI, there is a therapy with new class of drugs called pharmacological chaperones or pharmacoperons. In the endoplasmic reticulum (ER), which is the site of synthesis, formation and transport of proteins, there are chaperone proteins. Their role is to join and stabilize newly formed proteins, preventing their reaction with other peptides and assistance for transport to the other places in the cell. The pharmacoperons allow for proper folding and the inherent spatial structure of the protein and receptor transport from the ER to the plasma membrane. It is important that, when attached to new proteins, chaperones do not affect their functions. Mutated protein, which failed to pass the checks in the ER, cannot leave the endoplasmic reticulum and is degraded. The pharmacoperons are small lipophilic compounds, which penetrate through the membrane into the cell and bind to the structurally modified, as a result of mutations, proteins. These proteins have impaired spatial structure, they accumulate in the endoplasmic reticulum and are degraded, what can cause the disease. The pharmacoperons allow proper folding and achieving proper spatial structure of proteins and receptor transport to the ER membrane cell. This mechanism is presented in Figure 2. Yhe pharmacoperons interact selectively with mutated receptor and allow for escape of proteins from ER according to proper process of protein synthesis. The structure of pharmacoperons affect its efficiency, which determines the selectivity of the target protein, severity of the damage and the location of the mutation in the protein (for example, the mutation should not occur in the part of the gene responsible for encoding the part of protein interacting with ligand) (35). Examples of pharmacoperons for AVPR2 are its antagonists SR121463, SR49059, OPC41061 and OPC31260. So far, there was only one clinical trial in which five patients were treated with an antagonist SR49059 (Relcovaptan). As a result of the experiment, decreased levels of daily urine were observed. Vaptans class of drugs (for example Tolvaptan) are used in the treatment of hypernatremia. Their use in the treatment of NDI is still in the research phase (33).

Calcium-sensoring receptor (CaSR)

This receptor belongs to the C family G-protein coupled receptors. Extracellular domain binding calcium ions and other cations is characteristic for this family. CaSR is located on the surface of parathyroid cells in hormone-secreting glands, which are responsible for maintaining levels of calcium in the extracellular fluid and blood serum. A large number of receptors are present on the surface of kidney, they are less in the bone and intestine. CaSR is very sensitive to calcium ions, and therefore can effectively regulate the level of its concentration in the extracellular fluid and blood. If the

concentration of calcium ions is less than the physiological level, receptor stimulates parathyroid cells to secrete parathyroid hormone (PTH). This hormone enhances calcium ions level, by affecting the bone structure, the glomerular reabsorption from the initial and calcitriol synthesis in the small intestine (36).

Rare mutations in the CaSR encoding gene, contribute to the formation of disorders manifested by hypocalcemia and hypercalcemia (14). Mutations causing loss of CaSR function occur in familial hypocalciuric hypercalcemia (FHH) and in neonatal severe hyperparathyroidism. The incidence of both diseases is < 1/10000. Mutations causing increase of CaSR function occur in autosomal dominant hypocalcemia (ADH). In FHH, there is a loss of function of the receptor as a result of mutations and their effect is a reduced CaSR sensitivity to calcium ions. In most cases, in families suffering from FHH, the level of calcium ions is gently increased, but this does not require an increase of PTH level above physiological. Patients with FHH, in contrast to patients with primary hyperparathyroidism, have not severe symptoms of hypercalcemia such as the formation of kidney stones and disorders of the skeletal system. In most patients, there are no symptoms, and if they are, they have a gentle nature. These include dizziness, anxiety, feeling faint, tartar, muscle pain and weak memory. In the FHH treatment calcimimetics are used, which are modulators of CaSR. The action of this class of drugs is to increase the sensitivity of the receptor to calcium ions, which in turn leads to increased signal transduction. Research concerning the effects of cinacalcet (drug used to treat people who have drug-resistant hypercalcemia caused by mutations in the CaSR gene) were conducted. Among four people with FHH three of them were cognates. The drug was administered at a dose of 30 mg to 60 mg once a day for three months. The result of the study was an amelioration of well-being of patients and improvement in calcium homeostasis, which was maintained up to three years without increasing the dose. In three related patients, complete disappearance of the symptoms of hypercalcemia has been shown. None of the patients experienced adverse reactions. During cinacalcet therapy, bone mineral density was not improved (37). Mutations in the CaSR encoding gene, which increase the receptor activity, have been identified among patients with ADH. Factor, initiating the ADH creation, is constitutively active mutation in the gene encoding CaSR, which reduces the EC_{50}. This value represents the concentration needed to cause 50% of the maximum response to the agonist. The result of activating mutation in the calcium receptor is its increased sensitivity to calcium ions. Therefore, the receptor does not respond properly to the reduced concentration of calcium ions in serum and does not stimulate secretion of parathyroid hormone (17). Patients with the inherited form of hypocalcemia in most cases have no symptoms of the disease. Children during fever may have seizures and be sensitive. Patients usually have mild or moderate form of hypocalcemia with abnormal serum PTH level and a partial or complete hypercalciuria with increased urinary calcium excretion, despite its low concentration in serum blood. Supplementation with calcium and vitamin D is one of the ADH treatment. Is followed until the patient reaches a suitable serum calcium level and gets rid of the symptoms of the disease. People with ADH who have an increased secretion of calcium, can have an impaired kidney function and the formation of kidney stones. In this case, it is often required to monitor urinary excretion and administration of the drug from thiazide diuretics group, which can reduce the concentration of calcium in the urine (36).

Cysteinyl leukotrienes receptor

Leukotrienes (LT) define a group of biologically active molecules with lipid structure. One of the subgroups are cysteinyl leukotrienes (CysLT), which include leukotrienes C4 (LTC4), D4 (LTD4) and E4 (LTE4). They are important mediators of inflammation and allergic reactions in the course of diseases such as asthma and allergic rhinitis. CysLT interact with two types of receptors: CysLT1 and CysLT2. The cysteinyl leukotrienes are synthesized in the 5-lipoxygenase pathway from arachidonic acid, which is a component of the phospholipids of cell membranes. The source of leukotrienes are mainly mast cells, eosinophils, basophils, and macrophages. They exert their effects through reactions with specific membrane receptors belonging to the comprehensive GPCR family. They cause bronchoconstriction and vasodilation. As a result of increased vascular permeability and exudation of macromolecules, swelling of the tissue is also formed. Mutations in genes encoding leukotriene receptors play an important role in the pathophysiology of asthma and inflammation and warrants the creation of atopic asthma. Atopic asthma is a chronic, inflammatory disease, that is characterized by bronchial hypersensitivity to allergens. After an exposure occurs bronchospasm and reduction of air flow,which results in difficulty in breathing (38, 39). Polymorphisms of CysLT1 and CysLT2 receptors are not the main cause of the disease, but in most

cases it accompanied. Their presence also affects the course of the mechanism of action of drugs, thereby constituting an important reference point for the further search for more effective pharmacologically active substances (38, 40). CysLT1 receptors are the point of capture of "classical" antagonists (montelukast, zafirlukast, pranlukast, pobilukast and MK571), while in the case of CysLT2 receptors, signal transduction is not inhibited by mentioned antagonists. The only common antagonist of both groups is BAYu9773. CysLT1 and CysLT2 are homologous only in 38% to each other. CysLT1 occurs widely in leukocytes, spinal cord, less in the lungs, pancreas, small intestine, and in a small extent in other organs. In asthma, CysLT1 receptors are widely distributed in most inflammatory cells, and their number is significantly increased. CysLT2, due to the fact that there is not known its selective antagonist, it was not as well studied as CysLT1. CysLT2 widely occurs in the heart, in the pulmonary veins and in various parts of the brain. To a lesser extent, it is located in the spinal cord, kidneys and other organs (39). Signal transduction after leukotriene interaction with receptor is mediated by G protein. Both CysLT1 and CysLT2 are linked to protein Gq. Different amounts of CysLT released from mast cells and macrophages, and the transmitted signal strength affect the severity of asthma. In the treatment of atopic asthma blocking leukotrienes were used primarily (41). After administration of selective antagonists of leukotriene receptor such as montelukast, pranlukast, in most of patients CysLT1 receptor inhibition has occurred. The rest of patients who did not exhibited any pharmacological effects following administration of drugs, could have mutational changes that reduced receptor functions. CysLT2 receptor is also an important target in the treatment of asthma. In many cases, it forms, together with CysLT1 receptor, a highly active dimers with unique properties. Patients unresponsive to CysLT1 blockers, could be treated with drugs directed at CysLT2. Currently, only BAYu9773 operates antagonistically to both types of receptors, in that partially as an agonist on CysLT2. Research on CysLT1 and CysLT2 receptor variants allowed to determine the way in which they influence the response after joining agonist and their association with asthma. CysLT2 receptor has at least four polymorphic variants. Met201Wal variant causes partial inactivation of receptor, and Arg292Gly/Arg315Lys variant increases its activity compared to the wild-type of receptor. Probably, the part of the ligand binding site was changed, what causes an increased activity of agonist connected with Arg292Gly/Arg315Lys receptor variant and reduced agonist potential for Met201Wal. Paradoxically, only Met201Wal variant was observed in people with atopy or asthma. Therefore, it is uncertain whether the reduction in the sensitivity of the receptor to the ligand (as compared to the wild type) has a protective effect in lung diseases. These considerations are the subject of ongoing studies to determine risk factors for atopic asthma (4).

Neurotransmitter receptors

Antipsychotics bind to multiple receptors belonging to the family of GPCR. These are dopaminergic, serotonergic, muscarinic and opioids receptors. Dopamine is a major neurotransmitter in the central nervous system. It participates in neuroregulation, is responsible for motor activity, secretion of hormones and emotional states. There are five types of dopaminergic receptors, which are divided in terms of structure and pharmacological properties on the two groups: D1, which includes the D1 and D5 receptors and D2 receptors including the D2, D3 and D4 ones. Drugs for Parkinson's disease are dopaminergic receptors agonists, and in schizophrenia they are their antagonists. In contrast to the first group, the second one has a lot of polymorphic variations, changing effect of antipsychotic medications. An example is bromocriptine, which antagonist acting to D4 receptor is two times weaker than to the D2 receptor.

The opposite situation occurs in the case of clozapine, the activity of which is twofold higher to D4 than when connected to D2 or D3 receptors. Intensive studies on dopamine receptors have shown that a group of mutations in genes encoding D2 receptors is associated with mental disorders. A large number of medicinal substances has the point where the drug first associates in the serotonergic system. These include drugs acting as antidepressants and antipsychotically. However, there is often a resistance to some drugs, for example to clozapine, in the case of Cys22Ser mutation in the gene encoding 5-hydroxytryptamine receptor 2C (5-HT_{2C}). Altered response to a drug also occurs when there is a mutation Cys23Ser in $5HT_{2C}$ and Gly22Ser in $5HT_{1A}$. Pharmacogenetics research on serotonin receptors may contribute to the improvement of pharmacotherapy of disease associated with this system.

Opioid receptors are also in the interests of researchers, because their stimulation is associated with the development of addiction. An example is the receptor having several polymorphic variants, such as Asn40Asp, Asn152Asp, His260Arg,

His265Arg and Ser268Pro, that may change the effects of opioids. Receptors with Asn40Asp and Asn152Asp have four times greater affinity for the β-endorphins as compared to the wild type receptor. When the Ser268Pro mutation occurs, receptor desensitization process is reduced, which in turn makes that they are often in the active state. This may contribute to increased predisposition to addiction of persons with the Ser268Pro mutation (4, 28).

SUMMARY

GPCRs are the largest family of membrane receptors binding to many, different ligands, which include, *inter alia*, hormones, neurotransmitters, and photons. GPCRs are responsible for the proper conduction of a number of processes such as vision, intercellular communication, neural transmission and hormonal signaling. A characteristic feature of this family is their binding with heterotrimeric G protein, which upon activation changes its conformation. Depending on the type of protein G attached to GPCR, there is a cascade of different secondary messengers transmitting a signal into the cell. Through desensitization, inactivation and internalization, there is a controlled process of extinction of the transmitted signal (11). GPCRs control many physiological processes, and are also involved in many pathological processes. They also interact with a large group of drugs *inter alia* in the treatment of heart failure, asthma, or of renal diabetes insipidus (23, 31, 41, 43). Mutations, which occur at different levels of receptor maturation, initiate changes in receptor activity (inactive, overactive, or constitutively active GPCR), signaling processes and expression in the cell membrane. They lead to the diseases of different etiologies (4). Rhodopsin is one of the first GPCR for which a structure and mechanism of activation has been known. This receptor is responsible for the process of seeing. Some of mutations in genes encoding rhodopsin causes: formation of CAM, decrease of receptor activity or disturbance in the correct folding, transport and processing of the receptor protein. Retinitis pigmentosa (RP) is a complex disease characterized by progressive blindness resulting from the apoptosis of rod cells. One of the reasons of RP occurrence are mutations giving rise to a CAM. Constantly, research searching for an effective method of pharmacotherapy RP are conducted (42). Genetic disorders that cause loss of rhodopsin function result *inter alia* in Oguchi disease (4, 16). Receptor sensitive to calcium ions – CaSR – regulates its level in blood serum by stimulating parathyroid to the secretion of parathyroid hormone or inhibition of it secretion. FHH is a genetic disease, which arised as the result of a mutation that reduces the sensitivity of the CaSR to calcium. Studies on the use of calcimimetics in the treatment of FHH are conducted. Mutations that increase receptor activity led to a CAM and are the cause of autosomal dominant hypocalcemia (17). The most commonly occurring mutational changes within adrenergic receptors are SNP mutations leading to changes in the protein, and more specifically in its expression, interaction with a ligand, binding to a G protein and the regulation of signal transduction. Polymorphic variants can initiate disease or be a potential risk to their development. Examples of such diseases are hypertension, asthma and cardiac failure. In addition, as a result of the SNP, a change in the actions of many drugs may occur (4). Mutations in the gene coding for AVPR2, that reduce receptor activity, are the cause of NDI. It is characterized by abnormal process of collecting and concentrate urine in the kidney tubules. Currently, studies are being conducted on the use of specific compounds known as pharmacoperones in NDI therapy (33).

Melanocortin 4 receptor (MC4R) controls hunger and satiety center. Mutations in the gene encoding MC4R cause monogenic obesity in children and adults. In the majority, these are missense mutations, which in different ways influence the activity of the receptor (26).

Cysteinyl leukotriene receptors, by responding to inflammatory mediators, play an important role in allergic reactions. Mutations in the genes encoding the CysLT1 receptor and the CysLT2 cause polymorphs in these receptors, which affect the treatment of atopic asthma (4).

Dopamine, serotonin and opioid receptors are also important in pharmacotherapy. Mutations in the genes encoding these receptors cause many changes in the course and treatment of mental illness (4). Studies on the structure and function of genetically modified GPCRs allow to know a variety of mechanisms of action, which in turn can contribute to better knowledge on the etiology and pharmacotherapy of many currently incurable diseases.

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