Review

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Pharmacological Chaperoning by Vaptans: Restoring Function of NDI-Causing AVPR2 Mutants *In Vitro*

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Nephrogenic diabetes insipidus (NDI) is a rare disease characterized by the inability of the kidney to respond to vasopressin and concentrate urine. Hereditary NDI is primarily caused by mutations in the arginine vasopressin receptor-2 (*AVPR2*) gene, which enables the response to antidiuretic hormone. Mutations may lead to misfolding of the mutant AVPR2 protein or its retention within the Endoplasmic reticulum of the cell. Therefore, patients cannot respond to antidiuretic hormone and concentrate urine. Consequently, in untreated patients severe dehydration and hypernatremia may occur depending on the severity of the disease. Low-sodium and protein diet, using thiazide diuretics, and amiloride treatment are the conventional and standard treatments for NDI. Besides, many *in vitro* functional analysis studies have been focused on pharmacological chaperones for restoring function of NDI-causing AVPR2 mutants. Vaptans, which are known as vasopressin receptor antagonists and also pharmacological chaperones, are among the current and controversial topics for developing new treatment strategies for NDI. This study aims to review the *in vitro* studies on vaptans which are targeted to restore expression and function of mutant AVPR2s that cause NDI and highlight the importance of their effectiveness according to the type of mutation.

Keywords: AVP; AVPR2; AQP2; NDI; vaptan; pharmacological chaperones

Introduction

Water is the main component of the body and therefore maintaining water homeostasis within the body is very important [1]. It is controlled by the arginine vasopressin (AVP) - Arginine vasopressin receptor-2 (AVPR2) - aquaporin 2 (AQP2) water channel axis [2]. Any loss of function or decreased function of any of the proteins in this axis will cause a disruption of water balance. Hereditary nephrogenic diabetes insipidus (NDI) is one of these diseases primarily caused by defects in the receptor encoded by *AVPR2* gene [3,4]. These kinds of genetic diseases resulting in reduced or complete loss of function due to misfolding of proteins are defined as conformational diseases.

NDI patients suffer from severe dehydration and hypernatremia and life quality of them depends on the degree of the dehydration, hyperosmolality, and consequently severity of the disease. Sufficient water intake and a low-sodium diet is essential for the patients [5]. NDI treatment includes thiazide diuretics, fluids, nonsteroidal anti-inflammatory drugs (NSAIDs) and amiloride [6]. Unfortunately, current treatment options are insufficient to treat the disease and this situation affects the patient's daily life [7]. *In vitro* functional characterization studies of mutant AVPR2s causing NDI may shed light for new treatment strategies [3,4]. Vaptans bind to AVPR2, and they are considered promising treatment options due to their antagonistic effects. Besides their antagonistic treatment properties, they can also act as pharmacological chaperones [8]. Mutant AVPR2 proteins can be misrouted by the cellular quality control system or retained in Endoplasmic reticulum (ER) within the cell, depending on the mutation severity. In this case, the mutant proteins cannot perform their physiological functions and cause several pathologies. Pharmacological chaperones help to rescue these proteins by assisting them in reaching their targets. In this study, we reviewed vaptans, which are considered among the targeted and promising therapeutic strategies at the molecular level for AVPR2 mutations causing NDI.

The AVP, AVPR2 and AQP2 Axis in the Body Water Homeostasis

The primary determining factor of water homeostasis in the human body by increasing water reabsorption through the kidneys is the hormone AVP, which is known as antidiuretic hormone (ADH) [9]. AVP also has other important roles, such as regulation of blood pressure, sodium homeostasis and kidney function [10]. A nine amino acid-peptide hormone AVP, which is encoded by *AVP-neurophysin II* (*AVP-NPII*) gene, synthesized as preprohormone in the magnocellular cells of paraventricular and supraoptic nuclei of the hypothalamus [11,12]. After prepro-AVP-NPII is synthesized, signal peptide is removed and converted to pro-AVP in ER. Pro-AVP is then processed to AVP, its carrier protein NPII and a copeptin during the axonal transport

process [13,14]. Osmotic regulation of AVP hormone stimulation and release are controlled by the response of central and peripheral osmoreceptors to environmental changes such as increased plasma osmolality and decreased blood volume. In response to elevated plasma osmolality, central osmoreceptors increase vasopressin secretion while peripheral osmoreceptors detect the effects of food and fluid intake on osmolality at an early stage [15]. The AVP hormone exerts its effects by interacting with AVPR1a found on various vascular smooth muscle, AVPR2 found on the basolateral membrane of the collecting duct cells of the kidneys, and AVPR1b found on central nervous system cells. These receptors regulate vasoconstriction, renal water reabsorption and central nervous system effect, respectively [16]. The AVPR1a and AVPR1b direct cellular signaling in response to AVP, while the AVPR2 is critically important for the maintenance of water homeostasis [17–19]. On the way of renal water reabsorption, AVPR2 has essential roles. AVPR2 is a member of the G protein-coupled receptor (GPCR) superfamily. Since GPCR superfamily members have important roles in the cellular response after the detection of extracellular signals, they are among the potential targets of drugs which are prescribed in the market [20].

In response to plasma hypertonicity, AVP is secreted and transported to the kidney through the blood stream and binds to AVPR2 in the distal nephron [21,22]. This hormone-receptor complex activates the Gαs proteins to increase the intracellular cyclic AMP (cAMP) levels and activates cAMP-protein kinase A (PKA) signaling cascade which then triggers the AQP2 trafficking in the cell. AQP2 proteins, which are the main water channel proteins in the kidneys located in the subapical storage vesicles of the principal cells in the collecting ducts, are another important protein of the way of water reabsorption. Increased levels of cAMP promote the phosphorylation of Ser256 in the C-terminus of AQP2 via PKA which is critical for the AQP2 trafficking [21,23,24]. This phosphorylation causes the translocation of AQP2-containing storage vesicles to the apical membrane which leads to water reabsorption from urine to the bloodstream. This mechanism is crucial for meeting the organism's water needs by increasing water reabsorption in the renal collecting ducts, thus contributing to urine concentration. A defect occurring at any point in the AVP-AVPR2-AQP2 pathway may lead to water imbalance disorders which are characterized by a risk of hypernatremia [2,25].

Diabetes Insipidus

Diabetes insipidus is a rare disease with a prevalence of approximately 1 in 25,000, occurring equally in both males and females that results in excessive urination (polyuria) and excessive fluid intake (polydipsia) due to imbalances in the body's fluid and electrolyte regulation [26]. This condition arises from issues related to the ADH, either because of ADH deficiency or because of the kidneys' inability to respond to this hormone properly [27]. Patients suffer from excessive urination (more than 50 mL/kg body weight/24 h) and excessive drinking water (more than 3–3.5 L/24 hours in adults or 6.6 mL/kg/hour in children) [5,27,28]. The urine is diluted and low in osmolality, less than 250–300 mOsmol/kg of water, clear and dull [28]. The differential diagnosis of DI is primarily based on distinguishing between primary forms which can be central or renal or secondary form resulting from primary polydipsia [29]. Measuring plasma AVP levels, biological assessment, laboratory diagnostic testing, water deprivation test and desmopressin (dDAVP) challenge, MRI scan and copeptin assay are performed for diagnosis of the disease [5,28,30]. Treatment of the disease depends on the aetiology of the DI type since there are four types of DI: central, nephrogenic, primary polydipsia, and gestational [26,31]. The most common form is central DI (CDI) which can be associated with hereditary form and acquired form. Acquired form of CDI may arise from traumatic reasons or non-traumatic damage of the magnocellular neurons of the hypothalamus. According to Flynn and colleagues, up to 36% of the cases arise from head trauma or transfrontal/transsphenoidal surgery [27]. Inflammation, autoimmune disease, tumors or infiltrative diseases are the other reasons for CDI. In CDI patients, production and secretion of AVP deficiency is observed. Familial or hereditary CDI is a rare disease and typically follows an autosomal dominant inheritance pattern due to mutations in the *AVP* gene. More than 55 mutations reported for familial CDI [27]. On the other hand, NDI occurs as a result of the kidneys' resistance to the AVP hormone which can also be hereditary or acquired. The hereditary form of NDI is caused by mutations in the other two important genes which are the target proteins of AVP; *AVPR2* and *AQP2*. Acquired forms of the NDI are often results from lithium treatment or infiltrative disease [32]. Primary polydipsia is frequently observed in individuals with psychotic disorders such as schizophrenia [33]. This condition can lead to excessive fluid intake during psychotic episodes and may result in complications such as hyponatremia. Gestational DI (GDI), which is another type of the DI, is a temporary condition that occurs during pregnancy especially at the end of the second or early third semester and typically resolves on its own after childbirth. GDI arises due to a deficiency of ADH in the body [26]. Therefore, accurate recognition and management of DI and related conditions are crucial for improving patients' quality of life.

Nephrogenic Diabetes Insipidus

NDI is a rare endocrine disorder that results in reduced water permeability of the kidney's collecting duct principal cells despite the elevated concentrations of AVP [34]. As mentioned before, this form of the disease is associ-

ated with functional defects resulting from mutations in the *AVPR2* and *AQP2* [35]. Under normal conditions, AVP hormone triggers the water reabsorption process by binding the AVPR2 in the kidney. However, this mechanism is impaired in NDI disease which results in preventing the concentration of urine and leads to the production of large volumes of diluted urine [36]. NDI is categorized as hereditary, which typically presents in early childhood and is primarily associated with genetic mutations in the *AVPR2* or *AQP2* genes, or acquired, which develops as a result of various diseases or drug treatments and is more commonly seen in adulthood [30,36].

Acquired NDI is commonly caused by the side effects of long-term lithium therapy, which is used in the treatment of schizophrenia and bipolar spectrum disorders [37–41]. Even if lithium has serious side effects such as NDI, its effectiveness is higher compared to other mood-stabilizing drugs [37]. Hereditary NDI is associated with mutations in the *AVPR2* and *AQP2*, which can exhibit both autosomal recessive and autosomal dominant inheritance traits. While X-linked (*AVPR2* mutations) inheritance accounting for 90% of cases, autosomal recessive or dominant (*AQP2* mutations) inheritance accounting for 10% [23,42]. Mutations in AQP2 gene disrupt its normal function to negatively affect the kidneys' ability to retain water [7,43]. While dominant mutations are functional, they disrupt the tetrameric structure of AQP2, preventing its routing to the apical membrane. In contrast, recessive mutations result in the retention of AQP2 within the ER [19,44]. Approximately 90% of cases of hereditary NDI exhibit an X-linked recessive mode of inheritance due to mutations in the *AVPR2*. *AVPR2* is located on Xq28 and consists of three exons encoding a 371 amino acid-long protein. The receptor has seven transmembrane helices with three extracellular and three cytoplasmic loops [23]. More than 250 mutations have been identified in the *AVPR2*, including missense, nonsense, deletions, and insertions. These *AVPR2* mutations yield different kinds of clinical presentations [45]. The most pathogenic variants in the gene are single nucleotide variants, result in a decreased capacity of the receptor to bind to ligands, the formation of abnormal protein structures, or the accumulation of misfolded proteins in the ER or Golgi apparatus that leading to protein degradation and loss of function [45,46]. For reverting these types of effects, pharmacological chaperone therapies are considered as important for the proper trafficking of the misfolded protein and helping them be functional again. In addition to this, although some mutant proteins may reach to the cell surface, defects in the ligand binding regions prevent the receptor from responding effectively. As a result, a reduction in cAMP signaling occurs, and insufficient transport of AQP2 to the apical plasma membrane limits the kidneys' ability to retain water [7,46]. *AVPR2* mutations classified to five groups [47]. Class I mutations affect the transcription and translation process and therefore, lead to truncated proteins that are degraded. Class II mutations are missense or insertion/deletion mutations which result in accumulation of misfolded proteins in the ER by the cellular quality control system and leading to degradation of the protein as mentioned before. Class III mutations lead to proteins that are expressed in plasma membrane but with reduced affinity to Gαs proteins while class IV mutations have low affinity to AVP. The last type, class V mutations, cause proteins to be misdirected and to travel to different subcellular organelles [7].

The traditional treatment methods for NDI focus on ensuring adequate fluid intake while restricting salt and protein consumption. In this context, medications such as thiazide diuretics and amiloride are frequently used. The goal of these treatment approaches is not to eliminate the cause of the disease but to alleviate existing symptoms; thus, these methods do not offer a solution for the complete cure of the disease. Among the development of new treatment strategies, *in vitro* usage of pharmacological chaperones can facilitate the proper folding of AVPR2 and allow it to escape from the ER [4,48].

Vaptans

History of Vaptans

In 1991, nonpeptide vaptans emerged which antagonise AVPRs. As mentioned above, AVP is a peptide hormone that controls the water and electrolyte homeostasis of the human body through the AVPRs. AVP antagonists are generally used to treat imbalance of this homeostasis such as hyponatremia. At first, peptide compounds were synthesized as potential antagonists based on antidiuretic effects of AVP; however, reduced bioavailability of the peptide structure required the development of longer half-lived nonpeptide compounds. Then the first nonpeptide AVPR1 antagonist, OPC21268 (fuscoside), was identified. OPC21268 is a selective V1 receptor antagonist and counteracts vasopressin-induced vasoconstriction after oral administration in rats [49]. One year later, nonpeptide V2 receptor antagonist OPC31260 (mozavaptan) was discovered and it was the first successful usage of V2 receptor antagonist in humans [50,51]. In the following years, several nonpeptide antagonists were developed and today we know that VPA985 (lixivaptan), OPC41061 (tolvaptan), and SR121463 (satavaptan) are selective AVPR2 antagonists, SR49059 (relcovaptan) and SSR149415 (nelivaptan) are AVPR1a/b antagonists, and YM087 (conivaptan) is a non-selective AVPR1 and AVPR2 antagonist [52–57].

AVP acts on AVPR1a, AVPR1b, and AVPR2. AVPR1a is found in smooth muscle, platelets, liver, adrenal glands, adipocytes, and the heart, uterus, and it leads to vasoconstriction, glycogenolysis, platelet aggregation, and over time vascular smooth muscle proliferation. AVPR1b is located in the central nervous system and the adenohypophysis, stimulating the release of ACTH and prolactin. AVPR2, which are found in the basolateral membrane of

renal collecting ducts, play a crucial role in maintaining fluid homeostasis in the body [57]. When AVP binds to AVPR2, water from the urine is reabsorbed to the bloodstream while electrolyte excretion is unaffected. The entire process through the AVP-AVPR2-AQP2 axis can be disrupted by disorders such as heart failure, liver cirrhosis, euvolemic and hypervolemic hyponatremia, autosomal dominant polycystic kidney disease (ADPKD), and Syndrome of Inappropriate Antidiuretic Hormone Secretion (SIADH). As treatment, vaptans show their antagonistic effects via binding AVPR2 itself. However, while AVP binds to the receptor at the surface, vaptans bind deeper within the transmembrane, thus blocking hormone-receptor interaction [58,59]. They prevent water reabsorption and increase urine volume. Their increased diuresis effect is quantitatively similar to diuretics such as furosemide, but it is qualitatively different because there is not a significant increase in the excretion of urine electrolytes, such as sodium and potassium. Therefore, these antagonists produce aquaresis with a decrease in urine osmolality and an increase in serum [Na]. For this reason, the aquaretic effect is the hallmark of these nonpeptide antagonists and distinguishes them from traditional diuretics [60]. These several antagonists, which are called vaptans, have been developed and tested in humans. In 2004, YM087 was approved by FDA for the treatment of euvolaemic and hypervolaemic hyponatraemia but approval for oral use was withdrawn due to the risk of increased enzyme interactions. Since 2009, OPC41061 has been commercially available in the U.S.A; however, phase III trials for VPA985 were discontinued in 2022 [57,61].

Clinical use of vaptans such as treatment of ADPKD or hyponatremia has revealed important safety considerations. Vaptans can cause aquaretic side effects like thirstiness or dry mouth and polydipsia [60,62]. Hepatotoxicity is also a serious metabolic side effect of vaptans. It was reported that long-term usage of OPC41061 might cause a liver injury due to the significant elevation in liver enzymes [63]. Therefore, OPC41061 treatment is often limited to 30 days for certain conditions, or requires rigorous monthly liver function monitoring. Since vaptans are diuretics, they can cause NDI-like symptoms in healthy people. For the treatment of NDI, they should be used in very low doses. Long-term use of vaptans would require careful monitoring of liver enzymes and general condition of liver metabolism [64].

Vaptans as Pharmacological Chaperones

Beside these antagonistic effects of vaptans, we can call them pharmacological chaperones which are cell-permeable molecules that can rescue proteins from misfolding in cellular quality control systems [65]. The term pharmacological chaperone was introduced to the literature by Morello and colleagues to explain the action of vasopressin antagonists that can promote receptor processing via specific binding to the receptor. pharmacological chaperones are often confused with chemical chaperones because they are both small structures and take part in protein folding. Like osmolytes and some hydrophobic compounds, while chemical chaperones have an effect on protein's surroundings to make them fold, pharmacological chaperones bind specifically to the unfolded or misfolded proteins to lower the folding energy barrier. Therefore, chemical chaperones are non-specific and have an effect on a broad range of proteins, whereas pharmacological chaperones are specific to their targets [66,67]. Pharmacological chaperones can be antagonists, agonists, or allosteric modulators, and their specificity, higher binding affinity, and chaperone activity are their hallmarks [68]. Selectively binding pharmacological chaperones to their targets thermodynamically stabilizes the protein's native conformation. Protein folding intermediates are prone to aggregation like misfolded proteins due to mutation. Pharmacological chaperones assist these misfolded proteins to get their conformation correctly [69,70].

Pharmacological chaperones have been broadly studied for several protein conformational disorders such as Alzheimer's disease, lysosomal storage disease, amyloidoses, NDI, Retinitis pigmentosa, pain perception, familial hypocalciuric hypercalcemia and hyper or hypo-gonadism. Many of them were successful in clinical trials, and a few of them have been approved for use in patients [71–82].

In terms of NDI, *in vitro* therapeutic usage of vaptans is mainly designed for mutant AVPR2s rather than mutant AQP2s. Since the mechanism of vaptans as pharmacological chaperones relies on their high binding affinity for the specific orthosteric pocket of AVPR2, they do not show any affinity to AQP2s [83,84]. Also, these antagonistic small molecules bind with enough affinity to stabilize the misfolded protein and once mutant AVPR2 reaches to the membrane, vaptans can be displaced with natural ligand. Cytosolic AQP2 can be functional in tetramer form then translocate on membrane, and finding a molecule that stabilizes the AQP2 tetramer without permanent binding affinity can be difficult. Therefore, AQP2 based NDI studies mainly focus on by pass strategies that trigger AQP2 membrane insertion via pathways that do not require a functional AVPR2 [83].

Vaptans and Rescue of Mutant AVPR2s

Through the years, vaptans have been widely studied to understand their rescuing effects of mutant AVPR2s since they can somehow restore cell surface expression and function of intracellularly retained mutant receptors. Along these studies, some pharmacological chaperones showed better restoration of functional properties of mutant receptors than others but also the chaperone effect of pharmacological chaperones mostly was found to depend on mutation type. Therefore, in this part, we mentioned *in vitro* studies which demonstrate different rescue potentials of many vaptans together with their common chaperoning patterns and success on implications for functional signaling.

According to the Human Gene Mutation Database, there are 210 known missense/nonsense mutations in the *AVPR2*. While some missense mutations are found on highly conserved residues within mammalian AVPR2 orthologs, the other ones locate on partially conserved (only two possible amino acids) or non-conserved residues of AVPR2 [85]. The structural architecture of AVPR2 comprises extracellular, intracellular, and transmembrane domains; consequently, mutations in conserved residues can severely impair receptor function by altering critical physicochemical properties, such as residue polarity and side-chain size [23]. The cell-permeable antagonists restored cell surface expression and function of mutant receptors through their binding and stabilizing ability to the incompletely folded receptor in the ER. By this way, improperly folded receptor protein could escape the ER quality control system and be processed to the mature form. The rescued receptors were found to be fully functional once they reached the plasma membrane which were capable of inducing cAMP production in response to vasopressin. As a result of many functional analysis studies, we know that vaptans show their rescue effects on mutant AVPR2s at different levels according to the mutation type. In a study, the AVPR2 antagonists SR121463A and VPA985 successfully rescued del62-64, L59P, L83Q, Y128S, S167L, A294P, R322H, and R337X *in vitro* but at different levels [67]. While SR121463A promoted cell surface expression of Y128S and S167L at the same level, cAMP response of S167L was lower than Y128S. Both codon 128 and 167 are conserved residues but the change of residue polarity was dramatic in S167L since serine is a polar amino acid, but leucine is not. Therefore, residue polarity change in a transmembrane domain could affect the function of the receptor even if the mutant receptor could be stabilized and folded properly by a vaptan to translocate on membrane. In another study supporting this result, YM087 was used to rescue Y128S and S167L and the results were seen as nearly same with the SR121463A treatment. YM087 rescued both mutant receptors but again the cAMP accumulation of S167L was lower than Y128S [86]. R137H mutation which was known to promote constitutive B-arrestin-mediated internalization/desensitization of the receptor was rescued from ER quality control by using SR49059. However, treatment with SR49059 could not prevent constitutive internalization of the mutant receptor although its cell surface expression was increased [87]. Like SR49059, YM087 also rescued mistrafficked R137H and restored cell surface expression and function of misfolded receptor but the increase of AVP-stimulated cAMP accumulation was not as high as other mutant receptors that analyzed in that study which could be the result of constitutive internalization of R137H [87]. Robben and colleagues showed that pharmacological chaperones could show broader and stronger rescuing activity rather than chemical chaperones. They used SR121463B and also chemical chaperones such as glycerol, DMSO, and curcumin to understand their effects on nine ER-retained AVPR2 mutants. Only the V206D mutant receptor could be rescued by most chemical chaperones and among nine mutants, both V206D and S167T but specifically S167T mutant receptor was successfully rescued by SR121463B [88]. Another *in vitro* study revealed that the effects of PCs on receptor maturation and rescue potentials were correlated directly with their antagonist affinities. SR49059, OPC31260, OPC41061, and SR121463B were used to analyze nine ER-retained AVPR2 mutants and these four pharmacological chaperones showed different affinities between 0.50 to 75 nM (Ki) [89]. Among nine AVPR2 mutants, eight of them showed different cell surface expression levels and functional rescue except S167L mutant receptor which was unresponsive as mentioned above in similar studies. As a conclusion of this study, functional rescue is a balance between sufficient translocation of a mutant receptor and sufficient displacement by the agonist. It means that functional rescue can be most effective with high-affinity antagonists at clinically relevant concentrations. In addition to this antagonistic effect of pharmacological chaperones, OPC41061 and OPC31260 can have inverse agonistic effects beside their pharmacological chaperone properties. These inverse agonistic effects of them were shown on Y128S and Ser-333del mutants [90]. It was revealed that OPC41061 and OPC31260 exhibited protean agonism in which they acted as pharmacological chaperones for functional rescue of mutants while simultaneously showing their functions as inverse agonists that suppressed constitutive basal activity of the wild type AVPR2. The inverse agonist effect of OPC41061 was also shown in another functional analysis study of S127F mutant receptor [91]. S127F mutant receptor was severely mistrafficked and retained in the ER and also showed only minimal cAMP production in the cell. However, pretreatment with OPC41061 significantly restored cAMP generation of the mutant receptor in response to AVP stimulation. OPC41061 acted as inverse agonist on the wild type receptor through reducing the basal cAMP production but did not show any detectable basal effect on S127F mutant. Many other studies showed that different vaptans showed their pharmacological chaperone effects and rescue potentials on different mutant receptors in varying levels. Some of them fully rescued a kind of mutant receptors while the others partially restored cell surface expression of the mistrafficked mutant receptor [8,76,92–95]. For example, we investigated the effects of YM087 and VPA985 on the restoration of function of T273M mutation and VPA985 was found more effective than YM087 in stabilizing the conformation of the mutant receptor and restoring its function [95].

Among other vaptans, the rescue potential of OPC41061 has come forward on a large scale through the years. Many studies showed its great rescue potential of mutant AVPR2s. In a latest study, therapeutic potential of OPC41061 for AVPR2 mutations through a massive,

protein-wide functional screen was addressed [96]. For finding a universal pharmacological chaperone, massively parallel measurements via SUNi mutagenesis and FACS-based multiplexed assay were used to quantify the expression of thousands of AVPR2 variants simultaneously. They found that 87% of poorly expressed AVPR2 variants that cause NDI were rescued at least moderately expressed levels. For the computationally predicted pathogenic AVPR2 variants, 86.5% of them were rescued by OPC41061. OPC41061 was sufficient to overcome the small destabilization caused by most missense mutations, regardless of their location on the AVPR2 structure. This study suggested that general pharmacological chaperones could offer an effective and widespread therapeutic strategy for many rare diseases caused by loss of protein abundance.

While the success of vaptans *in vitro*, *in vivo* and clinical studies have also been conducted for using them as a treatment of misfolded AVPR2s. However, not every successful *in vitro* result showed its treatment potential in clinical studies. Phase III clinical studies of SR121463, SR49059, and VPA985 were discontinued [57]. Therefore, finding a molecule that both has a rescue potential with restoring the cell surface expression of the mutant receptors and shows minimal side effects at clinical studies has an importance for NDI treatment strategies. Providing a foundation for the development of AVPR2-targeted therapies, researchers have been tried to understand what are the structural properties of antagonist recognition by the AVPR2. For this purpose, OPC41061 and YM087, which are approved for the treatment of hyponatremia, were analyzed [97]. AVP binds to the large binding pocket of AVPR2 and this orthosteric binding pocket is suitable for small molecule ligands like vaptans. They showed that OPC41061 was deeply inserted into the binding pocket while YM087 showed a shallower binding pose and extended toward the first extracellular loop of AVPR2. Structural analysis reveals that antagonist binding reverses the seventh transmembrane domain distortion caused by AVP, thereby restoring helical continuity at the bottom of the binding pocket. Consequently, the necessary conformational change for G protein coupling was prevented. This conformational dependent antagonism can suggest a distinct inactivation mechanism for AVPR2s. Through this understanding, more effective therapeutic agents can be developed in the near future.

Conclusions

There have been many studies conducted about *in vitro* usage of vaptans as pharmacological chaperones for restoring the function of mutant AVPR2s since developing new treatment strategies for rare diseases like NDI has a great importance on human health. Approximately 25 years ago, vaptans were started to be used *in vitro* as pharmacological chaperones and then some of them, especially most successful ones at rescuing mutant receptors from mistrafficking in the cell, were involved in clinical studies. Through these clinical studies, some of them were elected at the critical phases of clinical studies because of their side effects. Long-term use of vaptans can cause hepatotoxicity; therefore, if it is necessary to use them as a long-term, liver metabolism and related enzymes of the patients should be carefully monitored. Finding molecules that have both antagonistic effect for the treatment NDI and also can be usable as pharmacological chaperones for rescuing most of the mutant AVPR2s at clinically safe doses would have a great impact on treatment strategies.

Availability of Data and Materials

Not applicable.

Author Contributions

ESO and BE conceptualized the review. ESO, BE and EMA conducted the literature search and drafted the manuscript. All authors contributed to important editorial changes in the manuscript. All authors reviewed and approved the final version of the manuscript. All authors have participated sufficiently in the work to take public responsibility for appropriate portions of the content and agreed to be accountable for all aspects of the work in ensuring that questions related to its accuracy or integrity.

Ethics Approval and Consent to Participate

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Conflict of Interest

The authors declare no conflict of interest.

References

[1] Balla A, Hunyady L. Nephrogenic Diabetes Insipidus. Experientia Supplementum (2012). 2019; 111: 317–339. https://doi.org/10.1007/978-3-030-25905-1_15.

[2] D'Acierno M, Fenton RA, Hoorn EJ. The biology of water homeostasis. Nephrology, Dialysis, Transplantation: Official Publication of the European Dialysis and Transplant Association - European Renal Association. 2025; 40: 632–640. https://doi.org/10.1093/ndt/gfae235.

[3] Makita N, Manaka K, Sato J, Iiri T. V2 vasopressin receptor

mutations. Vitamins and Hormones. 2020; 113: 79–99. https://doi.org/10.1016/bs.vh.2019.08.012.

[4] Erdélyi LS, Hunyady L, Balla A. V2 vasopressin receptor mutations: future personalized therapy based on individual molecular biology. Frontiers in Endocrinology. 2023; 14: 1173601. https://doi.org/10.3389/fendo.2023.1173601.

[5] Mutter CM, Smith T, Menze O, Zakharia M, Nguyen H. Diabetes Insipidus: Pathogenesis, Diagnosis, and Clinical Management. Cureus. 2021; 13: e13523. https://doi.org/10.7759/cureus.13523.

[6] Vaz de Castro PAS, Bitencourt L, de Oliveira Campos JL, Fischer BL, Soares de Brito SBC, Soares BS, *et al*. Nephrogenic diabetes insipidus: a comprehensive overview. Journal of Pediatric Endocrinology & Metabolism: JPEM. 2022; 35: 421–434. https://doi.org/10.1515/jpem-2021-0566.

[7] Milano S, Carmosino M, Gerbino A, Svelto M, Procino G. Hereditary Nephrogenic Diabetes Insipidus: Pathophysiology and Possible Treatment. An Update. International Journal of Molecular Sciences. 2017; 18: 2385. https://doi.org/10.3390/ijms18112385.

[8] Mouillac B, Mendre C. Pharmacological Chaperones as Potential Therapeutic Strategies for Misfolded Mutant Vasopressin Receptors. Handbook of Experimental Pharmacology. 2018; 245: 63–83. https://doi.org/10.1007/164_2017_50.

[9] Nigro N, Grossmann M, Chiang C, Inder WJ. Polyuria-polydipsia syndrome: a diagnostic challenge. Internal Medicine Journal. 2018; 48: 244–253. https://doi.org/10.1111/imj.13627.

[10] Cuzzo B, Padala SA, Lappin SL. Physiology, Vasopressin. StatPearls Publishing: Treasure Island (FL). 2023.

[11] Caldwell HK, Lee HJ, Macbeth AH, Young WS, 3rd. Vasopressin: behavioral roles of an "original" neuropeptide. Progress in Neurobiology. 2008; 84: 1–24. https://doi.org/10.1016/j.pneurobio.2007.10.007.

[12] Deniz F, Acar C, Saglar E, Erdem B, Karaduman T, Yonem A, *et al*. Identification of a Novel Deletion in AVP-NPII Gene in a Patient with Central Diabetes Insipidus. Annals of Clinical and Laboratory Science. 2015; 45: 588–592.

[13] Turkkahraman D, Saglar E, Karaduman T, Mergen H. AVP-NPII gene mutations and clinical characteristics of the patients with autosomal dominant familial central diabetes insipidus. Pituitary. 2015; 18: 898–904. https://doi.org/10.1007/s11102-015-0668-z.

[14] Rotondo F, Butz H, Syro LV, Yousef GM, Di Ieva A, Restrepo LM, *et al*. Arginine vasopressin (AVP): a review of its historical perspectives, current research and multifunctional role in the hypothalamo-hypophysial system. Pituitary. 2016; 19: 345–355. https://doi.org/10.1007/s11102-015-0703-0.

[15] Vincent JL, Su F. Physiology and pathophysiology of the vasopressinergic system. Best Practice & Research. Clinical Anaesthesiology. 2008; 22: 243–252. https://doi.org/10.1016/j.bpa.2008.03.004.

[16] Treschan TA, Peters J. The vasopressin system: physiology and clinical strategies. Anesthesiology. 2006; 105: 599–612; quiz 639–640. https://doi.org/10.1097/00000542-200609000-00026.

[17] Boone M, Deen PMT. Physiology and pathophysiology of the vasopressin-regulated renal water reabsorption. Pflugers Archiv: European Journal of Physiology. 2008; 456: 1005–1024. https://doi.org/10.1007/s00424-008-0498-1.

[18] Bankir L, Bichet DG, Morgenthaler NG. Vasopressin: physiology, assessment and osmosensation. Journal of Internal Medicine. 2017; 282: 284–297. https://doi.org/10.1111/joim.12645.

[19] Hagströmer CJ, Hyld Steffen J, Kreida S, Al-Jubair T, Frick A, Gourdon P, *et al*. Structural and functional analysis of aquaporin-2 mutants involved in nephrogenic diabetes insipidus. Scientific Reports. 2023; 13: 14674. https://doi.org/10.1038/s41598-023-41616-1.

[20] Venkatakrishnan AJ, Deupi X, Lebon G, Tate CG, Schertler GF, Babu MM. Molecular signatures of G-protein-coupled receptors. Nature. 2013; 494: 185–194. https://doi.org/10.1038/nature11896.

[21] Chou CL, Limbutara K, Kao AR, Clark JZ, Nein EH, Raghuram V, *et al*. Collecting duct water permeability inhibition by EGF is associated with decreased cAMP, PKA activity, and AQP2 phosphorylation at Ser^{269}. American Journal of Physiology. Renal Physiology. 2024; 326: F545–F559. https://doi.org/10.1152/ajprenal.00197.2023.

[22] Stockand JD. Vasopressin regulation of renal sodium excretion. Kidney International. 2010; 78: 849–856. https://doi.org/10.1038/ki.2010.276.

[23] Erdem B, Schulz A, Saglar E, Deniz F, Schöneberg T, Mergen H. Functional characterization of AVPR2 mutants found in Turkish patients with nephrogenic diabetes insipidus. Endocrine Connections. 2018; 7: 56–64. https://doi.org/10.1530/EC-17-0236.

[24] Saglar Ozer E, Moeller HB, Karaduman T, Fenton RA, Mergen H. Molecular characterization of an aquaporin-2 mutation causing a severe form of nephrogenic diabetes insipidus. Cellular and Molecular Life Sciences: CMLS. 2020; 77: 953–962. https://doi.org/10.1007/s00018-019-03219-w.

[25] Valenti G, Tamma G. The vasopressin-aquaporin-2 pathway syndromes. Handbook of Clinical Neurology. 2021; 181: 249–259. https://doi.org/10.1016/B978-0-12-820683-6.00018-X.

[26] Christ-Crain M, Bichet DG, Fenske WK, Goldman MB, Rittig S, Verbalis JG, *et al*. Diabetes insipidus. Nature Reviews. Disease Primers. 2019; 5: 54. https://doi.org/10.1038/s41572-019-0103-2.

[27] Flynn K, Hatfield J, Brown K, Vietor N, Hoang T. Central and nephrogenic diabetes insipidus: updates on diagnosis and management. Frontiers in Endocrinology. 2025; 15: 1479764. https://doi.org/10.3389/fendo.2024.1479764.

[28] Leroy C, Karrouz W, Douillard C, Do Cao C, Cortet C, Wémeau JL, *et al*. Diabetes insipidus. Annales D'endocrinologie. 2013; 74: 496–507. https://doi.org/10.1016/j.ando.2013.10.002.

[29] Christ-Crain M, Winzeler B, Refardt J. Diagnosis and management of diabetes insipidus for the internist: an update. Journal of Internal Medicine. 2021; 290: 73–87. https://doi.org/10.1111/joim.13261.

[30] Moeller HB, Rittig S, Fenton RA. Nephrogenic diabetes insipidus: essential insights into the molecular background and potential therapies for treatment. Endocrine Reviews. 2013; 34: 278–301. https://doi.org/10.1210/er.2012-1044.

[31] Arima H, Azuma Y, Morishita Y, Hagiwara D. Central diabetes insipidus. Nagoya Journal of Medical Science. 2016; 78: 349–358. https://doi.org/10.18999/nagjms.78.4.349.

[32] Chasseloup F, Tabarin A, Chanson P. Diabetes insipidus: Vasopressin deficiency…. Annales D'endocrinologie. 2024; 85: 294–299. https://doi.org/10.1016/j.ando.2023.11.006.

[33] Christ-Crain M, Gaisl O. Diabetes insipidus. Presse Medicale (Paris, France: 1983). 2021; 50: 104093. https://doi.org/10.1016/j.lpm.2021.104093.

[34] Bockenhauer D, Bichet DG. Nephrogenic diabetes insipidus. Current Opinion in Pediatrics. 2017; 29: 199–205. https://doi.org/10.1097/MOP.0000000000000473.

[35] Angelousi A, Alexandraki KI, Mytareli C, Grossman AB, Kaltsas G. New developments and concepts in the diagnosis and management of diabetes insipidus (AVP-deficiency and resistance). Journal of Neuroendocrinology. 2023; 35: e13233. https://doi.org/10.1111/jne.13233.

[36] Kavanagh C, Uy NS. Nephrogenic Diabetes Insipidus. Pediatric Clinics of North America. 2019; 66: 227–234. https://doi.org/10.1016/j.pcl.2018.09.006.

[37] Kaiser M, Edemir B. Lithium Chloride and GSK3 Inhibi-

tion Reduce Aquaporin-2 Expression in Primary Cultured Inner Medullary Collecting Duct Cells Due to Independent Mechanisms. Cells. 2020; 9: 1060. https://doi.org/10.3390/cells9041060.

[38] Jobbagy S, Vitturi DA, Salvatore SR, Pires MF, Rowart P, Emlet DR, *et al*. Nrf2 activation protects against lithium-induced nephrogenic diabetes insipidus. JCI Insight. 2020; 5: e128578. https://doi.org/10.1172/jci.insight.128578.

[39] Inoue M, Nakai K, Mitsuiki K. Triamterene in lithium-induced nephrogenic diabetes insipidus: a case report. CEN Case Reports. 2021; 10: 64–68. https://doi.org/10.1007/s13730-020-00517-2.

[40] Liu M, Deng M, Luo Q, Sun P, Liang A, Li X, *et al*. Metabolic reprogramming of renal epithelial cells contributes to lithium-induced nephrogenic diabetes insipidus. Biochimica et Biophysica Acta. Molecular Basis of Disease. 2023; 1869: 166765. https://doi.org/10.1016/j.bbadis.2023.166765.

[41] Jinnouchi T, Yoshimoto M, Ogino K, Oji T, Hayashi M. Lithium-induced Nephrogenic Diabetes Insipidus with Efficacy of Desmopressin in Combination with Thiazide Diuretics and Non-steroidal Anti-inflammatory Drugs: A Case Report with a Review of the Literature. Internal Medicine (Tokyo, Japan). 2024; 63: 1399–1404. https://doi.org/10.2169/internalmedicine.2437-23.

[42] Bichet DG, Bockenhauer D. Genetic forms of nephrogenic diabetes insipidus (NDI): Vasopressin receptor defect (X-linked) and aquaporin defect (autosomal recessive and dominant). Best Practice & Research. Clinical Endocrinology & Metabolism. 2016; 30: 263–276. https://doi.org/10.1016/j.beem.2016.02.010.

[43] Hureaux M, Vargas-Poussou R. Genetic basis of nephrogenic diabetes insipidus. Molecular and Cellular Endocrinology. 2023; 560: 111825. https://doi.org/10.1016/j.mce.2022.111825.

[44] Gao C, Higgins PJ, Zhang W. AQP2: Mutations Associated with Congenital Nephrogenic Diabetes Insipidus and Regulation by Post-Translational Modifications and Protein-Protein Interactions. Cells. 2020; 9: 2172. https://doi.org/10.3390/cells9102172.

[45] Manoel D, Mohammed I, Hussain K, Saraiva LR. Functional characterization and cAMP-mediated rescue of a novel truncating AVPR2 mutation causing nephrogenic diabetes insipidus. American Journal of Physiology. Endocrinology and Metabolism. 2025; 329: E764–E773. https://doi.org/10.1152/ajpendo.00325.2025.

[46] Strych L, Černá M, Hejnalová M, Zavoral T, Komrsková P, Tejcová J, *et al*. Targeted long-read sequencing identified a causal structural variant in X-linked nephrogenic diabetes insipidus. BMC Medical Genomics. 2024; 17: 29. https://doi.org/10.1186/s12920-024-01801-1.

[47] Robben JH, Knoers NVAM, Deen PMT. Characterization of vasopressin V2 receptor mutants in nephrogenic diabetes insipidus in a polarized cell model. American Journal of Physiology. Renal Physiology. 2005; 289: F265–F272. https://doi.org/10.1152/ajprenal.00404.2004.

[48] Ulloa-Aguirre A, Zariñán T, Gutiérrez-Sagal R, Tao YX. Targeting trafficking as a therapeutic avenue for misfolded GPCRs leading to endocrine diseases. Frontiers in Endocrinology. 2022; 13: 934685. https://doi.org/10.3389/fendo.2022.934685.

[49] Yamamura Y, Ogawa H, Chihara T, Kondo K, Onogawa T, Nakamura S, *et al*. OPC-21268, an orally effective, nonpeptide vasopressin V1 receptor antagonist. Science (New York, N.Y.). 1991; 252: 572–574. https://doi.org/10.1126/science.1850553.

[50] Yamamura Y, Ogawa H, Yamashita H, Chihara T, Miyamoto H, Nakamura S, *et al*. Characterization of a novel aquaretic agent, OPC-31260, as an orally effective, nonpeptide vasopressin V2 receptor antagonist. British Journal of Pharmacology. 1992; 105: 787–791. https://doi.org/10.1111/j.1476-5381.1992.tb09058.x.

[51] Ohnishi A, Orita Y, Okahara R, Fujihara H, Inoue T, Yamamura Y, *et al*. Potent aquaretic agent. A novel nonpeptide selective vasopressin 2 antagonist (OPC-31260) in men. The Journal of Clinical Investigation. 1993; 92: 2653–2659. https://doi.org/10.1172/JCI116881.

[52] Serradeil-Le Gal C, Wagnon J, Garcia C, Lacour C, Guiraudou P, Christophe B, *et al*. Biochemical and pharmacological properties of SR 49059, a new, potent, nonpeptide antagonist of rat and human vasopressin V1a receptors. The Journal of Clinical Investigation. 1993; 92: 224–231. https://doi.org/10.1172/JCI116554.

[53] Freidinger RM, Pettibone DJ. Small molecule ligands for oxytocin and vasopressin receptors. Medicinal Research Reviews. 1997; 17: 1–16. https://doi.org/10.1002/(sici)1098-1128(199701)17:1<1::aid-med1>3.0.co;2-5.

[54] Tahara A, Tomura Y, Wada KI, Kusayama T, Tsukada J, Takanashi M, *et al*. Pharmacological profile of YM087, a novel potent nonpeptide vasopressin V1A and V2 receptor antagonist, in vitro and in vivo. The Journal of Pharmacology and Experimental Therapeutics. 1997; 282: 301–308.

[55] Yamamura Y, Nakamura S, Itoh S, Hirano T, Onogawa T, Yamashita T, *et al*. OPC-41061, a highly potent human vasopressin V2-receptor antagonist: pharmacological profile and aquaretic effect by single and multiple oral dosing in rats. The Journal of Pharmacology and Experimental Therapeutics. 1998; 287: 860–867.

[56] Verbalis JG. AVP receptor antagonists as aquaretics: review and assessment of clinical data. Cleveland Clinic Journal of Medicine. 2006; 73: S24–S33. https://doi.org/10.3949/ccjm.73.suppl_3.s24.

[57] Hajnal K, Ágota-Noémi B. Chemical and pharmacological characterization of vasopressin antagonists. Bulletin of Medical Sciences. 2022; 95: 206–223. https://doi.org/10.2478/orvtudert-2022-0015.

[58] Miyazaki T, Fujiki H, Yamamura Y, Nakamura S, Mori T. Tolvaptan, an orally active vasopressin V(2)-receptor antagonist - pharmacology and clinical trials. Cardiovascular Drug Reviews. 2007; 25: 1–13. https://doi.org/10.1111/j.1527-3466.2007.00001.x.

[59] Lava SAG, Zollinger C, Chehade H, Schaffner D, Sekarski N, Di Bernardo S. Diuretics in pediatrics. European Journal of Pediatrics. 2023; 182: 2077–2088. https://doi.org/10.1007/s00431-022-04768-2.

[60] Peri A. Clinical review: the use of vaptans in clinical endocrinology. The Journal of Clinical Endocrinology and Metabolism. 2013; 98: 1321–1332. https://doi.org/10.1210/jc.2012-4082.

[61] Titko T, Perekhoda L, Drapak I, Tsapko Y. Modern trends in diuretics development. European Journal of Medicinal Chemistry. 2020; 208: 112855. https://doi.org/10.1016/j.ejmech.2020.112855.

[62] Cao P, Wang Q, Wang Y, Qiao Q, Yan L. Safety assessment of tolvaptan: real-world adverse event analysis using the FAERS database. Frontiers in Pharmacology. 2025; 15: 1509310. https://doi.org/10.3389/fphar.2024.1509310.

[63] Bellos I. Safety Profile of Tolvaptan in the Treatment of Autosomal Dominant Polycystic Kidney Disease. Therapeutics and Clinical Risk Management. 2021; 17: 649–656. https://doi.org/10.2147/TCRM.S286952.

[64] Hammond S, Meng X, Barber J, Mosedale M, Chadwick A, Watkins PB, *et al*. Tolvaptan safety in autosomal-dominant polycystic kidney disease; a focus on idiosyncratic drug-induced liver injury liabilities. Toxicological Sciences: an Official Journal of the Society of Toxicology. 2025; 203: 11–27. https://doi.org/10.1093/toxsci/kfae142.

[65] Tao YX, Conn PM. Chaperoning G protein-coupled receptors: from cell biology to therapeutics. Endocrine Reviews. 2014; 35:

602–647. https://doi.org/10.1210/er.2013-1121.

[66] Welch WJ, Howard M. Antagonists to the rescue. The Journal of Clinical Investigation. 2000; 105: 853–854. https://doi.org/10.1172/JCI9158.

[67] Morello JP, Salahpour A, Laperrière A, Bernier V, Arthus MF, Lonergan M, *et al*. Pharmacological chaperones rescue cell-surface expression and function of misfolded V2 vasopressin receptor mutants. The Journal of Clinical Investigation. 2000; 105: 887–895. https://doi.org/10.1172/JCI8688.

[68] Marinko JT, Huang H, Penn WD, Capra JA, Schlebach JP, Sanders CR. Folding and Misfolding of Human Membrane Proteins in Health and Disease: From Single Molecules to Cellular Proteostasis. Chemical Reviews. 2019; 119: 5537–5606. https://doi.org/10.1021/acs.chemrev.8b00532.

[69] structu PF, Park C. Selective stabilization of a partially unfolded protein by a metabolite. Journal of Molecular Biology. 2012; 422: 403–413. https://doi.org/10.1016/j.jmb.2012.05.044.

[70] Kasper JR, Park C. Ligand binding to a high-energy partially unfolded protein. Protein Science: a Publication of the Protein Society. 2015; 24: 129–137. https://doi.org/10.1002/pro.2596.

[71] Mahley RW, Huang Y. Small-molecule structure correctors target abnormal protein structure and function: structure corrector rescue of apolipoprotein E4-associated neuropathology. Journal of Medicinal Chemistry. 2012; 55: 8997–9008. https://doi.org/10.1021/jm3008618.

[72] Germain DP, Hughes DA, Nicholls K, Bichet DG, Giugliani R, Wilcox WR, *et al*. Treatment of Fabry's Disease with the Pharmacologic Chaperone Migalastat. The New England Journal of Medicine. 2016; 375: 545–555. https://doi.org/10.1056/NEJMoa1510198.

[73] Thonhofer M, Gonzalez Santana A, Fischer R, Torvisco Gomez A, Saf R, Schalli M, *et al*. 5-Fluoro derivatives of 4-epi-isofagomine as D-galactosidase inhibitors and potential pharmacological chaperones for GM1-gangliosidosis as well as Fabry's disease. Carbohydrate Research. 2016; 420: 6–12. https://doi.org/10.1016/j.carres.2015.10.009.

[74] Yam GHF, Bosshard N, Zuber C, Steinmann B, Roth J. Pharmacological chaperone corrects lysosomal storage in Fabry disease caused by trafficking-incompetent variants. American Journal of Physiology. Cell Physiology. 2006; 290: C1076–C1082. https://doi.org/10.1152/ajpcell.00426.2005.

[75] Bulawa CE, Connelly S, Devit M, Wang L, Weigel C, Fleming JA, *et al*. Tafamidis, a potent and selective transthyretin kinetic stabilizer that inhibits the amyloid cascade. Proceedings of the National Academy of Sciences of the United States of America. 2012; 109: 9629–9634. https://doi.org/10.1073/pnas.1121005109.

[76] Prosperi F, Suzumoto Y, Marzuillo P, Costanzo V, Jelen S, Iervolino A, *et al*. Characterization of five novel vasopressin V2 receptor mutants causing nephrogenic diabetes insipidus reveals a role of tolvaptan for M272R-V2R mutation. Scientific Reports. 2020; 10: 16383. https://doi.org/10.1038/s41598-020-73089-x.

[77] Schrier RW, Gross P, Gheorghiade M, Berl T, Verbalis JG, Czerwiec FS, *et al*. Tolvaptan, a selective oral vasopressin V2-receptor antagonist, for hyponatremia. The New England Journal of Medicine. 2006; 355: 2099–2112. https://doi.org/10.1056/NEJMoa065181.

[78] Noorwez SM, Malhotra R, McDowell JH, Smith KA, Krebs MP, Kaushal S. Retinoids assist the cellular folding of the autosomal dominant retinitis pigmentosa opsin mutant P23H. The Journal of Biological Chemistry. 2004; 279: 16278–16284. https://doi.org/10.1074/jbc.M312101200.

[79] Petäjä-Repo UE, Hogue M, Bhalla S, Laperrière A, Morello JP, Bouvier M. Ligands act as pharmacological chaperones and increase the efficiency of delta opioid receptor maturation. The EMBO Journal. 2002; 21: 1628–1637. https://doi.org/10.1093/emboj/21.7.1628.

[80] Gorvin CM, Hannan FM, Cranston T, Valta H, Makitie O, Schalin-Jantti C, *et al*. Cinacalcet Rectifies Hypercalcemia in a Patient With Familial Hypocalciuric Hypercalcemia Type 2 (FHH2) Caused by a Germline Loss-of-Function $G\alpha_{11}$ Mutation. Journal of Bone and Mineral Research: the Official Journal of the American Society for Bone and Mineral Research. 2018; 33: 32–41. https://doi.org/10.1002/jbmr.3241.

[81] Leach K, Wen A, Cook AE, Sexton PM, Conigrave AD, Christopoulos A. Impact of clinically relevant mutations on the pharmacoregulation and signaling bias of the calcium-sensing receptor by positive and negative allosteric modulators. Endocrinology. 2013; 154: 1105–1116. https://doi.org/10.1210/en.2012-1887.

[82] Newton CL, Whay AM, McArdle CA, Zhang M, van Koppen CJ, van de Lagemaat R, *et al*. Rescue of expression and signaling of human luteinizing hormone G protein-coupled receptor mutants with an allosterically binding small-molecule agonist. Proceedings of the National Academy of Sciences of the United States of America. 2011; 108: 7172–7176. https://doi.org/10.1073/pnas.1015723108.

[83] Jean-Alphonse F, Perkovska S, Frantz MC, Durroux T, Méjean C, Morin D, *et al*. Biased agonist pharmacochaperones of the AVP V2 receptor may treat congenital nephrogenic diabetes insipidus. Journal of the American Society of Nephrology: JASN. 2009; 20: 2190–2203. https://doi.org/10.1681/ASN.2008121289.

[84] Liu HL, Zhong HY, Zhang YX, Xue HR, Zhang ZS, Fu KQ, *et al*. Structural basis of tolvaptan binding to the vasopressin V_2 receptor. Acta Pharmacologica Sinica. 2024; 45: 2441–2449. https://doi.org/10.1038/s41401-024-01325-5.

[85] Böselt I, Römpler H, Hermsdorf T, Thor D, Busch W, Schulz A, *et al*. Involvement of the V2 vasopressin receptor in adaptation to limited water supply. PloS One. 2009; 4: e5573. https://doi.org/10.1371/journal.pone.0005573.

[86] Bernier V, Morello JP, Zarruk A, Debrand N, Salahpour A, Lonergan M, *et al*. Pharmacologic chaperones as a potential treatment for X-linked nephrogenic diabetes insipidus [published erratum in Journal of the American Society of Nephrology. 2006; 17: 591]. Journal of the American Society of Nephrology: JASN. 2006; 17: 232–243. https://doi.org/10.1681/ASN.2005080854.

[87] Bernier V, Lagacé M, Lonergan M, Arthus MF, Bichet DG, Bouvier M. Functional rescue of the constitutively internalized V2 vasopressin receptor mutant R137H by the pharmacological chaperone action of SR49059. Molecular Endocrinology (Baltimore, Md.). 2004; 18: 2074–2084. https://doi.org/10.1210/me.2004-0080.

[88] Robben JH, Sze M, Knoers NVAM, Deen PMT. Rescue of vasopressin V2 receptor mutants by chemical chaperones: specificity and mechanism. Molecular Biology of the Cell. 2006; 17: 379–386. https://doi.org/10.1091/mbc.e05-06-0579.

[89] Robben JH, Sze M, Knoers NVAM, Deen PMT. Functional rescue of vasopressin V2 receptor mutants in MDCK cells by pharmacochaperones: relevance to therapy of nephrogenic diabetes insipidus. American Journal of Physiology. Renal Physiology. 2007; 292: F253–F260. https://doi.org/10.1152/ajprenal.00247.2006.

[90] Takahashi K, Makita N, Manaka K, Hisano M, Akioka Y, Miura K, *et al*. V2 vasopressin receptor (V2R) mutations in partial nephrogenic diabetes insipidus highlight protean agonism of V2R antagonists. The Journal of Biological Chemistry. 2012; 287: 2099–2106. https://doi.org/10.1074/jbc.M111.268797.

[91] Szalai L, Sziráki A, Erdélyi LS, Kovács KB, Tóth M, Tóth AD, *et al*. Functional Rescue of a Nephrogenic Diabetes Insipidus Causing Mutation in the V2 Vasopressin Receptor by

Specific Antagonist and Agonist Pharmacochaperones. Frontiers in Pharmacology. 2022; 13: 811836. https://doi.org/10.3389/fphar.2022.811836.

[92] Ranadive SA, Ersoy B, Favre H, Cheung CC, Rosenthal SM, Miller WL, *et al*. Identification, characterization and rescue of a novel vasopressin-2 receptor mutation causing nephrogenic diabetes insipidus. Clinical Endocrinology. 2009; 71: 388–393. https://doi.org/10.1111/j.1365-2265.2008.03513.x.

[93] Rochdi MD, Vargas GA, Carpentier E, Oligny-Longpré G, Chen S, Kovoor A, *et al*. Functional characterization of vasopressin type 2 receptor substitutions (R137H/C/L) leading to nephrogenic diabetes insipidus and nephrogenic syndrome of inappropriate antidiuresis: implications for treatments. Molecular Pharmacology. 2010; 77: 836–845. https://doi.org/10.1124/mol.109.061804.

[94] Erdem Tuncdemir B, Mergen H, Saglar Ozer E. Evaluation of pharmacochaperone-mediated rescue of mutant V2 receptor proteins. European Journal of Pharmacology. 2019; 865: 172803. https://doi.org/10.1016/j.ejphar.2019.172803.

[95] Avcu EM, Erdem Tuncdemir B, Saglar Ozer E. Effects of YM087 and VPA985 on the T237M mutant receptor functionality in nephrogenic diabetes insipidus. Turkish Journal of Biochemistry. 2024; 49: 685–690. https://doi.org/10.1515/tjb-2024-0024.

[96] Mighell TL, Lehner B. A small molecule stabilizer rescues the surface expression of nearly all missense variants in a GPCR [published erratum in Nature Structural & Molecular Biology. 2025; 32: 2633. doi: 10.1038/s41594-025-01734-y]. Nature Structural & Molecular Biology. 2025; 32: 2429–2440. https://doi.org/10.1038/s41594-025-01659-6.

[97] Zhang T, Liu H, You C, Zhang Y, Xu Y, Pan B, *et al*. Structural insights into antagonist recognition by the vasopressin V2 receptor. Nature Communications. 2025; 16: 9734. https://doi.org/10.1038/s41467-025-64735-x.