Ф А 2.2.1-32-367

MINISTRY OF HEALTH OF UKRAINE
NATIONAL UNIVERSITY OF PHARMACY
pharmaceutical faculty
department of pharmaceutical chemistry

QUALIFICATION WORK

on the topic: «**DETERMINATION OF THE PROSPECTS OF NEW IMIDAZOLIDINE-2,4-DIONE DERIVATIVES FOR THE TREATMENT OF ALZHEIMER'S DISEASE»**

Prepared by: higher education graduate of group
Фм20(4,10д) англ 01
specialty 226 Pharmacy, industrial pharmacy
educational and professional program Pharmacy
Meriem EL KAHYA
Supervisor: professor of higher education institution of department of pharmaceutical chemistry, DSc, professor
Hanna SEVERINA
Reviewer: professor of higher education institution of Department of clinical pharmacology Institute for Advanced Training of Pharmacy Specialists, DSc, professor
Illya PODOLSKY

Kharkiv 2025

ANNOTATION

The study investigates imidazolidine-2,4-dione derivatives as potential multitarget ligands for the treatment of neurodegenerative diseases. ADMET analysis and molecular docking modelling with AChE and GSK3 were performed. Compound 3426 was found to have high binding affinity and a favourable pharmacokinetic profile. The thesis consists of 3 chapters, general conclusions, and a reference list (87 sources), presented on 51 pages and containing 17 figures, 3 tables.

Keywords: imidazolidine, pyrrolidine, neurodegeneration, in silico, acetylcholinesterase

АНОТАЦІЯ

У роботі досліджено похідні imidazolidine-2,4-dione як потенційні мультитаргетні ліганди для терапії нейродегенеративних захворювань. Проведено ADMET-аналіз, молекулярний докінг до AChE та GSK3. Визначено, що сполука 3426 має високий афінний потенціал та сприятливий фармакокінетичний профіль. Робота складається зі вступу, 3 розділів, загальних висновків та списку використаних джерел (87 найменувань), викладена на 51 сторінці, містить 17 рисунків і 3 таблиці.

Ключові слова: imidazolidine, піролідин, нейродегенерація, *in silico*, ацетилхолінестераза

CONTENTS

LIST OF ABBREVIATIONS

Aβ peptide	Amyloid beta peptide
ACh	Acetylcholine
AChE	Acetylcholinesterase
AD	Alzheimer’s disease
ADMET	Absorption, distribution, metabolism, excretion, and toxicity
AI	Artificial Intelligence
APOE	Apolipoprotein E
APP	Amyloid Precursor Protein
ARIA	Amyloid-Related Imaging Abnormalities
BBB	Blood–Brain Barrier
cAMP	Cyclic adenosine monophosphate
CNS	Central Nervous System
CYP	Cytochrome P450
GLY	Glycine
HIS	Histidine
in silico	Research methods using mathematical calculation methods
in vitro	Research methods using cell cultures
in vivo	Methods of study in a living organism
kcal/mol	Kilocalorie per mole
MCI	Mild Cognitive Impairment
MTDLs	Multitarget-Directed Ligands
NMDA	N-methyl-D-aspartate
PCA	Posterior Cortical Atrophy
PHE	Phenylalanine
PPA	Primary Progressive Aphasia
SAR	Structure–Activity Relationship
SER	Serine
TRP	Tryptophan
TYR	Tyrosine

INTRODUCTION

Relevance of the topic. The search for effective therapeutics for Alzheimer's disease (AD) is currently an urgent and strategically important challenge for global healthcare. Firstly, Alzheimer's disease is the most common form of dementia, affecting millions of people each year. According to the WHO, over 55 million people worldwide live with dementia, with more than 60-70% of these cases attributed to AD. Projections estimate that this number will rise to 139 million by 2050, creating an enormous burden on medical, social, and economic systems.

Secondly, despite significant advances in molecular biology, neuroscience, and pharmacology, no existing drug is capable of curing or halting the progression of AD. Currently approved medications (acetylcholinesterase inhibitors, NMDA receptor antagonists) provide only symptomatic relief, mostly at early stages, and do not target the core mechanisms of neurodegeneration.

Thirdly, AD is one of the most expensive diseases in the world. The cost of care, medications, hospitalization, and social support for AD patients amounts to hundreds of billions of dollars annually, and this figure continues to grow due to the global aging population.

In this context, the search for new, effective neuroprotective agents capable not only of alleviating symptoms but also of modifying key pathogenic pathways of AD – such as amyloidogenesis, tau protein hyperphosphorylation, oxidative stress, and neuroinflammation – remains a high-priority direction in modern medicinal chemistry and pharmacology.

The purpose of the study. Comprehensive evaluation of the therapeutic potential of newly designed imidazolidine-2,4-dione derivatives as promising multifunctional agents for the treatment of Alzheimer's disease, with a focus on their predicted biological activity, pharmacokinetic properties, and interactions with key molecular targets involved in neurodegeneration.

In order to achieve the objective, the following tasks had to be accomplished:

- To analyze and summarize current literature data on the pathogenesis and и pharmacotherapy of Alzheimer’s disease;
- To select virtually generated candidate structures for further study based on logical-structural analysis, identifying the most promising agents for Alzheimer's disease treatment;
- To perform ADMET profiling of the generated candidate compound library and assess the suitability of these structures for further development as potential drug candidates;
- To carry out virtual target prediction for the generated compounds using the SwissTargetPrediction;
- To identify the most relevant biological targets for in silico studies based on the prediction results and the therapeutic importance of the targets;
- To conduct molecular docking of the selected imidazolidine-2,4-dione derivatives with the chosen biological targets;
- To formulate conclusions regarding the possible mechanism of action of the candidate structures and select compounds for further synthesis and *in vitro/in vivo* experimental evaluation.

The object of the study. Molecular modelling, target-oriented molecular docking, and in silico pharmacokinetic evaluation of biologically active compounds as potential neurotropic agents.

The subject of the study. Pharmacokinetic properties, biological targets, and molecular interactions of the generated imidazolidine-2,4-dione derivatives, identified using in silico methods (ADMET analysis, virtual target screening, and molecular docking).

The methods of the study. A The compound design was carried out using BIOVIA Draw 2021. Pharmacokinetic parameters (ADMET) were calculated using the SwissADME online platform. Biological target prediction was performed using SwissTargetPrediction. The *in silico* studies included virtual molecular docking,

conducted with the help of AutoDock Vina, AutoDock Tools, BIOVIA Draw 2021, Discovery Studio Visualizer 2021, and VMD 1.9.3.

The practical value of the results. The use of computer-based modeling (in silico) to investigate and identify the most promising compounds with potential therapeutic properties for the treatment of Alzheimer's disease enables: a) the optimization of subsequent laboratory (in vitro) and clinical (in vivo) studies by saving time, resources, and costs; b) an increased likelihood of identifying compounds with strong inhibitory activity against acetylcholinesterase (AChE) and glycogen synthase kinase 3 (GSK3). Thus, computer modeling may serve as a powerful tool in the development of new and effective therapeutic approaches for Alzheimer's disease.

Elements of scientific research. At the Department of Pharmaceutical Chemistry of the National University of Pharmacy, under the supervision of Prof. Hanna Severina, novel derivatives of imidazolidine-2,4-dione derivatives were developed based on a hybrid pharmacophore concept as potential agents for the treatment of Alzheimer's disease. Their ADMET parameters were evaluated, probable biological targets were predicted, and molecular docking studies were performed. The most promising compound was identified, and its ability to inhibit acetylcholinesterase and glycogen synthase kinase 3 was substantiated.

Structure and scope of the qualification work.

The thesis consists of an introduction, three chapters, conclusions, and a list of references. The work is presented on 51 pages and contains 17 figures, 3 tables. The list of the used literary sources contains 87 titles.

CHAPTER 1
NEURODEGENERATION AND ALZHEIMER'S DISEASE. IMIDAZOLIDINE-2,4-DIONE DERIVATIVES AS BIOLOGICALLY ACTIVE SUBSTANCES
(literature review)

1.1. Dementia – overview, symptoms, causes and diagnosis

Dementia is a broad term used to describe a serious decline in mental abilities – such as memory, language, problem – solving, and thinking – that are severe enough to disrupt a person's everyday life. The most frequent cause of dementia is Alzheimer's disease. Dementia is not a specific illness but a collective term that refers to a group of symptoms that affect cognitive functioning. It often includes memory loss, confusion, and difficulty with communication and decision-making. These symptoms arise due to damage in the brain, and they interfere with daily functioning and independence. Emotional well–being and personal relationships are often impacted as well [1, 2].

Although Alzheimer's disease is responsible for the majority (60% to 80%) of dementia cases, there are other types too. Vascular dementia, caused by small strokes or blood vessel blockages in the brain, is the second most common form. Some individuals may experience mixed dementia, which is a combination of different types [3]. It's important to note that not all memory and thinking problems are due to dementia. Conditions like thyroid disorders or vitamin deficiencies can cause similar symptoms and may be treatable or reversible.

Historically, people referred to dementia as "senility" or "senile dementia," incorrectly assuming that serious mental decline was just a normal part of aging. Understand the difference between normal aging and signs of dementia [4].

Common early symptoms include:

- Forgetting recent events or conversations
- Misplacing items like wallets or keys

- Difficulty handling finances or bills
- Trouble cooking or organizing meals
- Forgetting appointments
- Getting lost in familiar places [5].

These symptoms generally develop slowly and become worse over time. If you or someone you care about starts showing these signs, it's important to consult a doctor. Early diagnosis not only helps manage symptoms better but also opens the door to clinical trials and planning for the future [6].

Dementia results from diseases that damage brain cells, disrupting the communication between them. When brain cells are unable to interact properly, thinking, emotions, and behavior can suffer. Each region of the brain is responsible for different functions – such as memory, movement, or judgment – so damage in a particular area leads to specific difficulties. For instance, in Alzheimer’s disease, abnormal levels of proteins accumulate inside and outside brain cells, making it difficult for them to survive or connect with others. The hippocampus, the brain’s memory center, is often the first region affected, which is why memory loss is one of the earliest symptoms [2, 3].

While most forms of dementia are progressive and irreversible, some causes of memory and thinking problems can be treated:

- Depression
- Medication side effects
- Alcohol misuse
- Thyroid issues
- Lack of certain vitamins

There’s no single test to confirm dementia. Doctors use a combination of medical history, physical and neurological exams, lab tests, and observation of symptoms to make a diagnosis. While they can usually determine whether someone has dementia, identifying the exact type can be more difficult because symptoms can overlap.

Sometimes a general diagnosis of dementia is given without specifying the type. In such cases, seeing a specialist – like a neurologist, psychiatrist, psychologist, or geriatrician – can help clarify the diagnosis [2-4].

1.2. Alzheimer's disease as the main cause of dementia

Alzheimer's disease (AD) is the most common cause of dementia, accounting for an estimated 60% to 80% of all cases [7]. Alzheimer's can be defined as a slowly progressive neurodegenerative disease characterized by neuritic plaques and neurofibrillary tangles due to the accumulation of amyloid-beta peptide (Aβ) in the most affected brain region, the medial temporal lobe and neocortical structures [2]. AD is a specific neurodegenerative disease that leads to this persistent and progressive cognitive impairment.

Here are the main characteristics of Alzheimer's disease as the primary cause of dementia:

1. Progressive and irreversible nature – AD is a disease that slowly progresses over time, and currently, there are no treatments capable of stopping or reversing the underlying brain damage [7].
2. Specific pathological changes in the brain – at the cellular level, AD is characterized by the accumulation of abnormal proteins in the brain, specifically beta-amyloid plaques and neurofibrillary tangles composed of tau protein. These formations interfere with the normal functioning of neurons, leading to their death and the loss of connections between them [7].
3. Gradual onset and worsening of symptoms – typically, the first signs of AD are mild memory problems, especially regarding recent events. As the disease progresses, cognitive impairments become more pronounced, affecting other areas such as language (aphasia), executive functions (planning, decision-making), visuospatial abilities, and orientation [8].
4. Impact on daily life – over time, cognitive impairments become so severe that the person loses the ability to independently perform everyday tasks, requires assistance with self-care, and becomes dependent on others [8].

5. Increased prevalence with age – while AD is not a normal part of aging, the risk of developing it increases significantly with age. Most people diagnosed with AD are older than 65 [7].
6. Distinction from other causes of dementia – it is important to differentiate AD from other causes of dementia, such as vascular dementia (related to impaired blood supply to the brain), Lewy body dementia, frontotemporal dementia, and others, as they may have different mechanisms of development, symptoms, and treatment approaches [9, 10].

Thus, Alzheimer's disease is the primary and devastating force underlying the majority of dementia cases, gradually stripping individuals of their cognitive abilities, independence, and connection to the world. Understanding its characteristics is key to developing effective strategies for diagnosis, treatment, and support for people living with this disease and their families [7].

There is no official data on the prevalence of Alzheimer’s disease among Ukrainians. However, according to a study published in the respected British medical journal *The Lancet*, around 651,000 people were living with dementia in Ukraine in 2019. This number is projected to increase to one million by 2050 [11].

The disease is named after Dr. Alois Alzheimer, who in 1906 observed unusual changes in the brain tissue of a woman who had died from an unknown mental illness. Her symptoms had included memory loss, language difficulties, and erratic behaviour. Upon examining her brain after death, he discovered abnormal clumps (now known as amyloid plaques) and tangled bundles of fibers (now called neurofibrillary or tau tangles) [12].

These plaques and tangles are still considered hallmark features of Alzheimer’s disease. Another key characteristic is the breakdown of connections between neurons in the brain. Neurons are responsible for transmitting signals within the brain and from the brain to muscles and organs. Other complex changes in the brain are also believed to contribute to the progression of Alzheimer’s [12].

Alzheimer’s disease is commonly categorized into two types: sporadic and familial. Sporadic Alzheimer’s typically affects individuals over the age of 65 and

is often referred to as late-onset Alzheimer's. Familial Alzheimer's, on the other hand, results from inherited genetic mutations and tends to affect individuals between the ages of 30 and 65, thus being known as early-onset Alzheimer's. Familial cases are caused by rare autosomal dominant mutations in three genes: amyloid precursor protein, presenilin 1, and presenilin 2 [10].

In some individuals, memory loss is not the first symptom of Alzheimer's. This form is called atypical Alzheimer's disease. Although it is also caused by plaques and tangles, the earliest affected brain region is not the hippocampus. Atypical Alzheimer's is more often diagnosed at a younger age and is less common among individuals over 65.

Atypical variants of AD are characterized by non-amnestic presentations resulting from the predominant degeneration of specific cortical regions outside the medial temporal lobes. These atypical phenotypes include:

Posterior Cortical Atrophy (PCA) – PCA is associated with neurodegeneration in posterior cortical regions, especially the occipital and parietal lobes. These regions are essential for visual processing and visuospatial integration. As a result, early symptoms often include visuoperceptual dysfunction such as difficulties in object recognition, environmental disorientation, or alexia, despite preserved ocular function and visual acuity. These features distinguish PCA from typical amnestic presentations of AD [13, 14].

Primary Progressive Aphasia (PPA) – This variant is attributed to progressive atrophy in left-hemispheric perisylvian language areas. It manifests with early and gradually worsening language impairments, including word-finding difficulties (anomia), pauses in speech, and comprehension deficits. PPA is divided into non-fluent, semantic, and logopenic variants, the latter being most frequently associated with underlying AD pathology [15, 16].

Frontal Variant Alzheimer's Disease (fvAD) – involves predominant degeneration of the frontal cortex, leading to executive dysfunction and behavioral disturbances. Clinical features may include impaired planning, judgment, and disinhibition. Individuals may exhibit socially inappropriate behavior or loss of

insight, resembling frontotemporal dementia (FTD); however, neuroimaging and biomarkers may support an underlying AD etiology [17].

The primary risk factor for the development of sporadic Alzheimer's disease (SAD) is aging. Indeed, increased life expectancy strongly correlates with a higher likelihood of developing neurodegenerative disorders, including AD. Normal aging is associated with structural changes in the brain, such as alterations in membrane fluidity and lipid composition, regional brain volume, cortical thickness and density, as well as the microstructure of gray and white matter. Additionally, there is a progressive loss of neuronal synapses, which ultimately leads to a reduction in overall neuronal density [18].

The prevalence of Alzheimer's dementia increases significantly with age. Approximately 5% of individuals aged 65–74 years, 13.1% of those aged 75–84 years, and 33.3% of people aged 85 and older are affected by Alzheimer's dementia. However, it is important to emphasize that Alzheimer's dementia is not a normal part of aging, and advanced age alone is not sufficient to cause the disease.

While autosomal dominant Alzheimer's disease is caused by mutations in one of the genes involved in amyloid metabolism – such as the amyloid precursor protein (*APP*), presenilin 1 (*PSEN1*), or presenilin 2 (*PSEN2*) – the primary genetic risk factor for sporadic Alzheimer's disease (SAD) is the apolipoprotein E (*APOE*) gene. *APOE* plays a crucial role in lipid transport, including cholesterol, both in peripheral tissues and within the central nervous system [18, 19].

Due to its essential role in the transport of cholesterol from astrocytes to neurons, the *APOE* gene facilitates lipid delivery to neuronal membranes, thereby supporting membrane homeostasis – a critical process in neuronal repair and brain recovery following injury. The *APOE* gene exists in three allelic variants: ε2, ε3, and ε4. Each individual inherits one allele of *APOE* from each parent, resulting in six possible genotypic combinations. These genotypes significantly influence the risk and onset of Alzheimer's disease [12, 20].

In individuals with Down syndrome, a genetic condition characterized by the presence of three copies of chromosome 21 instead of the typical two, there is an

increased risk of developing Alzheimer’s disease. This is because chromosome 21 contains the gene encoding the amyloid precursor protein (APP). The triplication of this chromosome results in overexpression of the APP gene, leading to the accumulation of β-amyloid plaques in the brain – a pathological hallmark of Alzheimer’s disease [17, 22].

A positive family history is not a prerequisite for the development of Alzheimer's disease. However, individuals with a first-degree relative (a parent or sibling) diagnosed with Alzheimer’s are at a higher risk of developing the disease compared to those without such a familial link. Furthermore, the risk significantly increases for individuals who have more than one first-degree relative with the disease [18, 23].

The prevalence of AD and the progression of its symptoms are disproportionately higher in women compared to men. This disparity may be attributed to a range of sex-specific risk factors, including APOE genotype, cardiovascular disease, depression, hormonal depletion, sociocultural variables, and sex-based cognitive traits such as verbal memory performance. To better understand the role of sex differences in AD pathogenesis, research is needed to evaluate the influence of gender on the evolution of Alzheimer’s-related biomarkers across the lifespan, particularly in early stages. This includes assessments of cognitive function, neuroimaging markers, cerebrospinal fluid indicators, and blood-based biomarkers. Furthermore, both preclinical and clinical studies are necessary to develop and tailor AD therapies that address sex-specific biological and clinical profiles [23, 24].

Modifiable risk factors are of considerable interest as they represent actionable targets for preventative strategies. Cardiovascular system impairment constitutes a risk factor for neurodegenerative diseases, and given the brain's extensive vascular network, a healthy cardiovascular system can be considered neuroprotective. This may elucidate why a subset of cardiovascular disease risk factors are shared with AD, including hypertension, dyslipidemia, diabetes mellitus, obesity, dietary factors, smoking, and physical inactivity. Additional modifiable

factors encompass educational attainment, poor sleep quality, stress, air pollution, traumatic brain injury, and hearing loss, among others. Consequently, lifestyle also emerges as a significant risk determinant, wherein intellectual, physical, and social engagement, alongside dietary habits, may contribute to AD prevention [18].

1.3. Pathogenesis and stages of Alzheimer's disease

The healthy adult brain comprises billions of neurons, each possessing long, branching extensions that facilitate connections with other nerve cells. At these points of contact, known as synapses, information transfer occurs through brief bursts of chemical messengers released by one neuron and received by another. These numerous synaptic connections form a complex network underlying memory, thought, sensations, emotions, movements, and acquired skills [19].

In Alzheimer's disease, characteristic pathological changes occur within the brain. Extracellular accumulation of fragments of the beta-amyloid protein in the form of dense formations (amyloid plaques) and intracellular aggregation of abnormally altered tau protein (tau tangles) are two key morphological hallmarks of the disease. These processes are accompanied by damage and death of neurons, known as neurodegeneration, which, together with amyloid and tau pathologies, forms the AT(N) profile of Alzheimer's disease [19].

Beta-amyloid and tau play distinct but interconnected roles in the development of the disease. Amyloid plaques and smaller aggregates of beta-amyloid can exert a toxic effect on neurons, disrupting synaptic transmission and communication between nerve cells. Inside neurons, hyperphosphorylated tau protein forms tangle-like structures that block the intracellular transport of nutrients and other molecules necessary for normal neuronal function and survival. Although the precise sequence of pathological events is still under investigation, it is believed that the primary accumulation of beta-amyloid may precede the appearance of abnormal tau, and an increase in amyloid burden correlates with the subsequent accumulation of tau protein. Other pathological changes associated with Alzheimer's

disease include neuroinflammation and brain atrophy, manifested as a reduction in its volume [19].

It is believed that the presence of toxic beta-amyloid and tau proteins activates immune cells in the brain called microglia (from the ancient Greek words "mikros" – small, "glia" – glue) [25]. These constitute a population of small, elongated, star-shaped cells (microgliocytes) with dense cytoplasm and relatively short, branched processes, which are typically located along the capillaries of the CNS. They originate directly from monocytes (a type of white blood cell necessary for immunity) or perivascular macrophages and belong to the so-called macrophage-monocyte system. Together with macrophages, these cells are active participants in the immune defense of the central nervous system, with microglia attempting to clear the brain of toxic proteins, as well as the widespread debris from dead and dying cells [25].

Significant progress has been made in measuring these changes in the brain. For example, we can now determine abnormal levels of beta-amyloid and tau in cerebrospinal fluid (the fluid surrounding the brain), and a scanning technique known as positron emission tomography can create images showing where beta-amyloid and tau have accumulated. The accumulation of beta-amyloid and tau are biomarkers of Alzheimer's disease. Biomarkers are biological changes that can be measured to indicate the presence or absence of a disease or the risk of developing a disease [19, 26]. The progression of Alzheimer's disease, from subtle changes in the brain unnoticed by the affected individual to alterations that cause memory problems and eventually physical incapacitation, is termed the Alzheimer's disease continuum.

The continuum of AD represents a dynamic process of progressive pathological changes in the brain, manifesting clinically from subsymptomatic stages to pronounced cognitive and functional impairment. Within this continuum, three primary phases are distinguished: preclinical AD, characterized by the presence of disease biomarkers in the absence of overt clinical manifestations; mild cognitive impairment (MCI) due to Alzheimer's disease, defined by objectively

documented cognitive decline insufficient for a dementia diagnosis; and dementia due to AD, also referred to as Alzheimer's dementia. The clinical phase of Alzheimer's dementia is further stratified into mild, moderate, and severe stages [19].

Despite the established sequential progression of the Alzheimer's disease continuum from the asymptomatic preclinical phase to the terminal stage of severe Alzheimer's dementia, the duration individuals spend in each stage demonstrates significant interindividual variability. The temporal characteristics of each phase of the continuum are influenced by biological and genetic determinants such as age of onset, genetic predisposition (particularly the presence of APOE ε4 alleles), biological sex, as well as comorbid conditions and environmental factors [19]. The overall duration of the disease from the onset of the first clinical symptoms to death varies widely, ranging from 2 to 20 years, with an average of approximately 12 years [24, 25].

Preclinical Alzheimer's disease represents the initial phase of the disease continuum, characterized by the presence of subclinical pathomorphological changes in the brain, which serve as early biomarkers of Alzheimer's disease. At this stage, individuals demonstrate positive neuroimaging or biochemical markers of amyloid and/or tau pathology (e.g., elevated amyloid levels in cerebrospinal fluid or its deposition visualized via positron emission tomography, as well as elevated tau levels in cerebrospinal fluid or its pathological conformations detected through appropriate assays) [18, 19].

A defining feature of the preclinical phase is the absence of overt cognitive symptoms, such as memory impairment or deficits in other cognitive domains, indicating the preservation of the brain's functional reserve. During the early stages of pathological processes in Alzheimer's disease, compensatory neuroplastic mechanisms are observed, allowing for the maintenance of normative cognitive system functioning despite the presence of initial structural alterations [18, 19].

It is crucial to emphasize that, while research centres possess the tools and expertise to identify some early neurobiological changes associated with

Alzheimer's disease, further research is critically important for validating the accuracy and specificity of these diagnostic instruments before their routine clinical implementation in hospitals and other medical facilities. An important aspect to note is that not all individuals with evidence of brain changes associated with Alzheimer's disease will necessarily progress to the stages of mild cognitive impairment or dementia. For instance, data exists on individuals with significant amyloid burden detected postmortem who did not exhibit clinical signs of cognitive decline during their lifetime, underscoring the complexity and heterogeneity of the disease's pathogenesis [18, 19].

Mild cognitive impairment (MCI) due to Alzheimer's disease represents an intermediate clinical stage between the preclinical phase and overt Alzheimer's dementia. Individuals at this stage demonstrate confirmed biomarkers, indicating the presence of pathophysiological changes in the brain associated with Alzheimer's disease, specifically amyloid and/or tau pathology detected through neuroimaging or biochemical methods.

Clinically, MCI due to Alzheimer's disease is characterized by the emergence of new but relatively subtle cognitive symptoms, which may include subjective and objective difficulties with memory (predominantly episodic), language (e.g., word-finding difficulties), and thinking (e.g., reduced executive functions or processing speed). These cognitive complaints may be noticeable to the individual, their close family members, and friends, indicating an awareness of the existing changes.

A crucial diagnostic feature of MCI due to Alzheimer's disease is that, despite the presence of cognitive impairments, they typically do not result in a significant impact on the individual's ability to perform most complex everyday activities. It is believed that the manifestation of clinical symptoms at this stage occurs when neuroplastic compensatory mechanisms in the brain become insufficient to counteract the effects of progressive damage and neuronal loss caused by the pathological processes of Alzheimer's disease [18, 19].

Dementia due to Alzheimer's disease, or Alzheimer's dementia, is the clinically manifest stage of the disease, characterized by pronounced and progressive

impairments in cognitive functions, encompassing memory, language, thinking, and/or behaviour. These cognitive-behavioural disturbances reach a degree where they significantly impair the individual's ability to function in daily life, affecting independence in performing routine tasks. The diagnosis of Alzheimer's dementia is established based on the presence of substantial cognitive impairment in conjunction with confirmed biomarkers, indicating the presence of pathophysiological changes in the brain associated with Alzheimer's disease (amyloid and/or tau pathology).

As Alzheimer's disease progresses, patients typically exhibit a variety of cognitive and neuropsychiatric symptoms, the spectrum and severity of which evolve over time. This dynamic symptomatology reflects the progressive damage and loss of neurons in different anatomical regions of the brain, each responsible for specific cognitive and behavioural functions. The rate of dementia progression, that is, the pace of transition from mild to moderate and severe stages, demonstrates significant interindividual variability, dependent on complex interactions of genetic, biological, and environmental factors [18, 19].

The mild stage of Alzheimer's dementia represents the initial phase of clinically manifest dementia due to Alzheimer's disease. At this stage, most individuals retain relative functional independence in many areas of daily living; however, they may experience an increasing need for assistance to optimize their autonomy and ensure safety in certain activities. Specifically, managing financial resources and handling bill payments can pose significant challenges, and the completion of familiar routine tasks may require increased time expenditure. Despite the presence of cognitive impairments, patients at this stage typically retain the ability to operate motor vehicles, fulfil professional responsibilities (depending on their nature), and participate in habitual social and leisure activities [26].

The moderate stage of Alzheimer's dementia, often characterized as the longest phase of clinically manifest dementia, is accompanied by a deepening of cognitive impairments. Individuals at this stage demonstrate more significant difficulties with memory, particularly regarding recent events, and pronounced language disturbances, including an increasing frequency of anomia and reduced

language fluency. A characteristic feature is worsening disorientation in time and space, as well as difficulty in executing sequential, multi-step tasks, such as hygiene procedures (bathing) and dressing. At this stage, urinary incontinence may first manifest, along with significant changes in personality and behaviour, including the emergence of suspiciousness (paranoid ideation) and psychomotor agitation. Furthermore, patients may experience difficulties in recognizing close relatives and acquaintances (agnosia) [26].

The severe stage of Alzheimer's dementia represents the terminal phase of the disease, characterized by a profound impairment of cognitive and functional abilities. At this stage, the individual's capacity for verbal communication is significantly diminished or entirely lost, necessitating around-the-clock palliative care. Progressive damage to brain regions responsible for motor functions leads to progressive physical disability and, ultimately, to a bedridden state (immobility). Prolonged immobility significantly elevates the risk of developing serious physical complications, including thromboembolic events (thromboses), infectious lesions of the skin, and a systemic inflammatory response – sepsis, which can lead to multiple organ failure. Furthermore, damage to cerebral structures controlling the act of swallowing causes dysphagia (swallowing impairment), complicating adequate nutrition and hydration. A consequence of dysphagia can be aspiration – the passage of food boluses into the trachea (airway) instead of the esophagus. Aspiration of food particles into the lungs can trigger severe lung infection – aspiration pneumonia, which is a leading cause of mortality among patients with Alzheimer's disease [26].

1.4. Current therapeutic approaches for AD

Alzheimer's disease exhibits a complex, multifactorial pathophysiology, contributing to the severe course of the illness. Despite significant progress in understanding AD and developing novel treatment modalities, a definitive cure for this condition remains elusive. Nevertheless, contemporary therapeutic approaches

can assist in enhancing patients' quality of life and slowing disease progression in the early stages.

1. Symptomatic Treatment: These therapeutic approaches do not halt the progression of the disease but can help alleviate cognitive and non-cognitive symptoms, thereby improving the quality of life for patients and their caregivers.

- Cholinesterase Inhibitors – these medications (donepezil, galantamine, rivastigmine) increase the levels of acetylcholine, a neurotransmitter crucial for memory and thinking, the levels of which are reduced in AD. They can improve cognitive function in the mild to moderate stages of the disease [27].
- NMDA Receptor Antagonist – Memantine regulates the action of glutamate, another neurotransmitter that, in excessive amounts, can damage brain cells. It may be prescribed in the moderate to severe stages of AD to improve memory, attention, and the ability to perform daily activities, as well as to manage behavioural symptoms [28].
- Treatment of Non-Cognitive Symptoms – antidepressants, anxiolytics, antipsychotics, and other psychotropic medications may be used to manage depression, anxiety, sleep disturbances, agitation, aggression, and other behavioral problems. It is crucial to use these medications cautiously, under close medical supervision, considering their potential side effects.

2. Disease-Modifying Therapy. These treatment approaches aim to impact the underlying pathological processes that drive AD, with the goal of slowing or halting disease progression.

- Anti-Amyloid Monoclonal Antibodies: Several drugs in this class have received approval for treating early stages of AD, including lecanemab [29] and donanemab [30]. They target the removal of amyloid plaques from the brain. Clinical trials have demonstrated their ability to slow cognitive decline, although they are also associated with the risk of side effects such as amyloid-related imaging abnormalities, involving brain edema or microhemorrhages.
- Other investigational drugs – numerous other potential disease-modifying therapies are under active investigation, targeting various pathways including

tau protein, inflammation, metabolic dysfunction, and other pathogenic mechanisms of AD. Some of these agents are in the late stages of clinical trials.

Important Considerations: Early diagnosis is crucial for initiating treatment at stages when it may be most effective, particularly for disease-modifying therapies. The treatment of AD is individualized and depends on the stage of the disease, co-existing medical conditions, and the individual's response to medications. Care and support for patients and their families are integral components of AD therapy.

The drugs approved to date act on a single target (one drug - one target; ODOT), but recently attention has been paid to multiple therapeutic strategies aimed at developing drugs that can affect more than one target [31, 32]. Multipurpose medicines are created by combining two or more pharmacophore structural features of biologically active substances acting on different targets in one molecule. Active scientific research continues to search for new, more effective means to combat this devastating disease

1.5. Imidazolidine-2,4-dione derivatives as biologically active substances

1.5.1. General characterization of imidazolidine-2,4-dione derivatives

Imidazolidine-2,4-dione (hydantoin) is a non-aromatic five-membered heterocycle regarded as an important and privileged scaffold in medicinal chemistry. The significance of the imidazolidine-framework in drug development is supported by the clinical use of several drugs such as phenytoin, nitrofurantoin, and enzalutamide. Imidazolidine possesses five potential substitution sites, including two hydrogen bond acceptors and two hydrogen bond donors. Additional advantages of the hydantoin scaffold include the ease of its synthesis through well-established cyclization reactions and its capacity to accommodate a wide variety of substituents. Due to these properties, numerous hydantoin derivatives with diverse substituents have been designed and synthesized, exhibiting a broad range of biological and

pharmacological activities, including anticancer, antimicrobial, antimetabolic, and antiepileptic effects [33, 34].

In general, the term "hydantoins" refers to a specific group of compounds and typically denotes a class of molecules that share the hydantoin substructure as their core scaffold. 2-Thiohydantoin (2) and 2-selenohydantoin (3) are considered isosteric analogs of hydantoin (Figure 1).

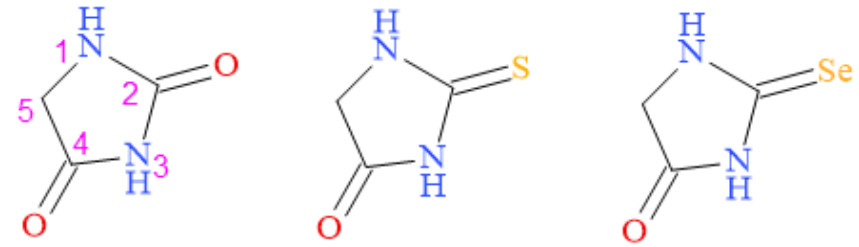


Fig. 1.1. Types of hydantoin [33]

The name "hydantoin" originates from a combination of the terms for the chemical reaction and substrate involved (i.e., hydrogenation and allantoin), as it was first synthesized by the Baeyer group through the hydrogenation of allantoin [33].

1.5.2 General characterization of pharmacological activity of imidazole derivatives

Hydantoins have attracted considerable research interest due to their broad medical and industrial applications, serving as essential pharmacophore fragments or core structural units. Although hydantoin is a relatively small molecule, it features four positions available for substitution as well as four hydrogen bond donors or acceptors. Compounds containing the hydantoin scaffold display a wide range of pharmacological and biological activities, including anticancer [34-41], anti-inflammatory [42, 43], antidiabetic [44], antimicrobial [45, 46], adrenomimetic [47, 48], anticonvulsant [49, 50], antiplatelet [51], and anti-HIV effects [52, 53]. Moreover, some hydantoin-based compounds function as allosteric antagonists of

leukocyte cell adhesion by binding to lymphocyte function-associated antigen-1 (LFA-1) and interfering with protein-protein interactions [54] (Fig.1.2).

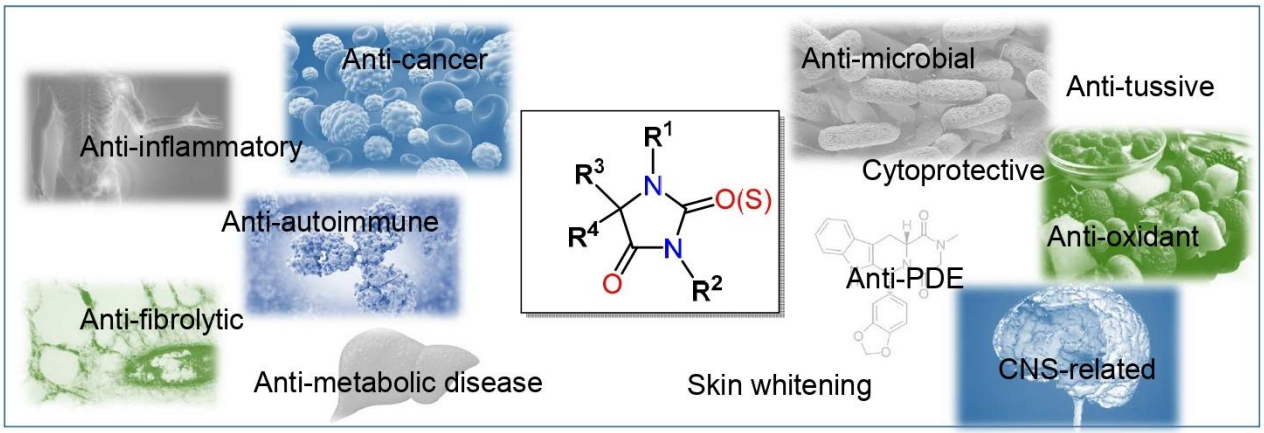


Fig. 1.2. Pharmacological activity of imidazolidine-2,4-dione derivatives

Several clinically approved drugs, including phenytoin, mephenytoin, ethotoin, and fosphenytoin as anticonvulsants (Fig. 1.3) and exert anticonvulsant activity by blocking voltage-gated sodium channels in neurons. They stabilize neuronal membranes, suppress excessive neuronal excitability, and prevent the generation of pathological electrical discharges underlying seizures. Fosphenytoin is a prodrug of phenytoin and, after administration, is rapidly converted into the active compound with an identical mechanism of action. Ethotoin acts similarly but has a milder pharmacological profile with a lower risk of toxic effects. Mephenytoin also acts by blocking sodium channels, but due to the risk of serious adverse effects (e.g., agranulocytosis), its use is limited [55].

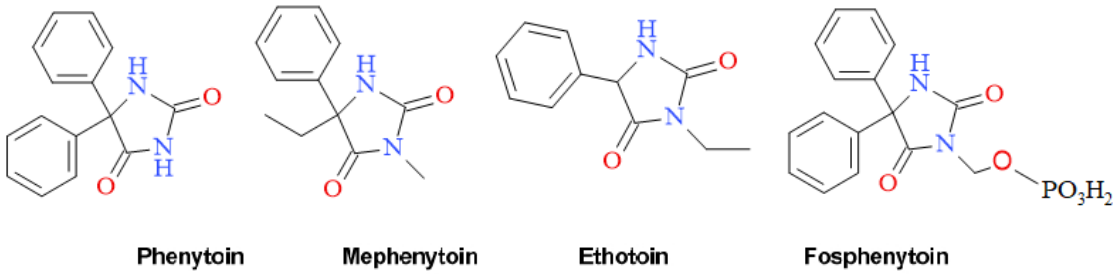


Fig. 1.3. Imidazolidine-2,4-dione derivatives as anticonvulsants

Dantrolene, on the other hand, acts as a skeletal muscle relaxant by interfering with excitation-contraction coupling in muscle fibers. It binds to the ryanodine receptor (RyR1) on the sarcoplasmic reticulum of skeletal muscle cells, inhibiting calcium ion release. This decrease in intracellular calcium concentration reduces muscle contraction strength, making dantrolene effective in conditions like malignant hyperthermia and spasticity (Fig. 1.4).

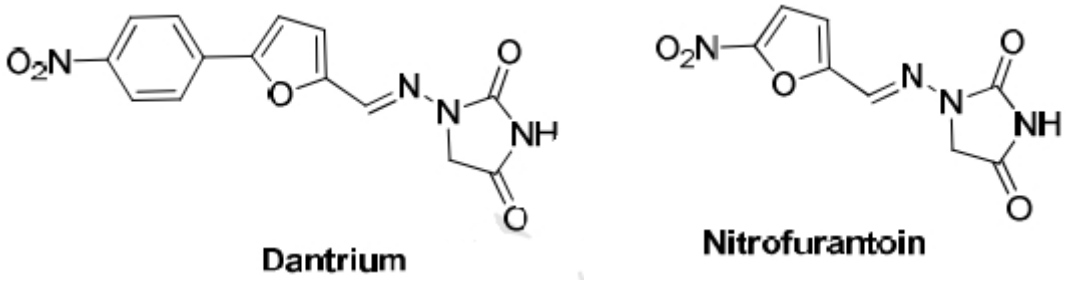


Fig. 1.4. Postsynaptic muscle relaxant and antibacterial agents [33]

Nitrofurantoin is an antibacterial agent that exerts its effect by being enzymatically reduced within bacterial cells to reactive intermediates. These intermediates interact with bacterial ribosomal proteins, DNA, and other macromolecules, causing inhibition of bacterial protein synthesis, DNA damage, and disruption of metabolic processes, ultimately leading to cell death. Its broad-spectrum activity primarily targets Gram-positive and Gram-negative bacteria, making it especially useful for treating urinary tract infections [56].

1.5.3. Characterization of imidazolidine-2,4-dione derivatives derivatives as CNS agents

1.5.3.1. Imidazolidine-2,4-dione derivatives as anticonvulsants

In 2015, Habib and colleagues introduced a new series of benzylidenehydantoin-phenylpiperazine hybrids as potential anticonvulsant agents [57]. These hybrids were designed by combining structural features of two previously known active compounds: a 2-thiohydantoin derivative (3-(alkyl/aryl)-5,5-diphenyl-2-thioxoimidazolidine-4-one and a piperazine-based succinimide (N-

[(4-arylpiperazin-1-yl)-methyl]-3-arylpyrrolidine-2,5-dione. The synthesis involved a two-step procedure, which included the formation of a benzylidene moiety followed by the introduction of an N-phenylpiperazinylmethyl group, resulting in 18 analogs. The anticonvulsant activity of these compounds was evaluated using the strychnine-induced seizure test and the pentylenetetrazole (PTZ)-induced seizure test. In the strychnine model, the synthesized benzylidenehydantoins significantly prolonged survival times compared to strychnine alone, with compound 117a demonstrating the highest potency, nearly matching that of phenytoin. In the PTZ seizure model, most derivatives showed moderate to excellent protective effects (ranging from 33% to 100%), with compound 117b providing complete (100%) protection against seizures.

A partial structure–activity relationship (SAR) study indicated that the presence of lipophilic or bulky halogen substituents, such as chlorine or bromine, at the 4-position of the benzylidene group enhanced anticonvulsant activity in both models. Additionally, ortho-oxygenated phenyl derivatives, containing groups such as OH and OMe, were also associated with improved effects (Fig. 1.5) [57].

The chemical flexibility at the 5-position of the hydantoin ring offers significant advantages in medicinal chemistry and drug discovery. In 2011, Byrtus and colleagues identified HB10 as a promising anticonvulsant candidate [41,58].

Building on this earlier work, in 2016, Czopek and co-workers reported the design, synthesis, and biological evaluation of a new class of hydantoin-based compounds, β-tetralinohydantoins [59]. A total of nine β-tetralinohydantoin derivatives were prepared via a two-step synthetic route, starting with a Bucherer–Bergs reaction of 3,4-dihydronaphthalen-2(1H)-one, followed by a Mannich-type reaction to attach a piperazine moiety.

The anticonvulsant activity of the synthesized compounds was assessed using the maximal electroshock seizure (MES) test and the subcutaneous pentylenetetrazole (scPTZ) seizure test. Among the compounds tested, only one exhibited notable efficacy in both assays, while the others were found to be inactive.

Moreover, it was observed that the introduction of an amide linkage, as seen in compounds B and C (Figure 1.6), led to a loss of anticonvulsant activity [59].

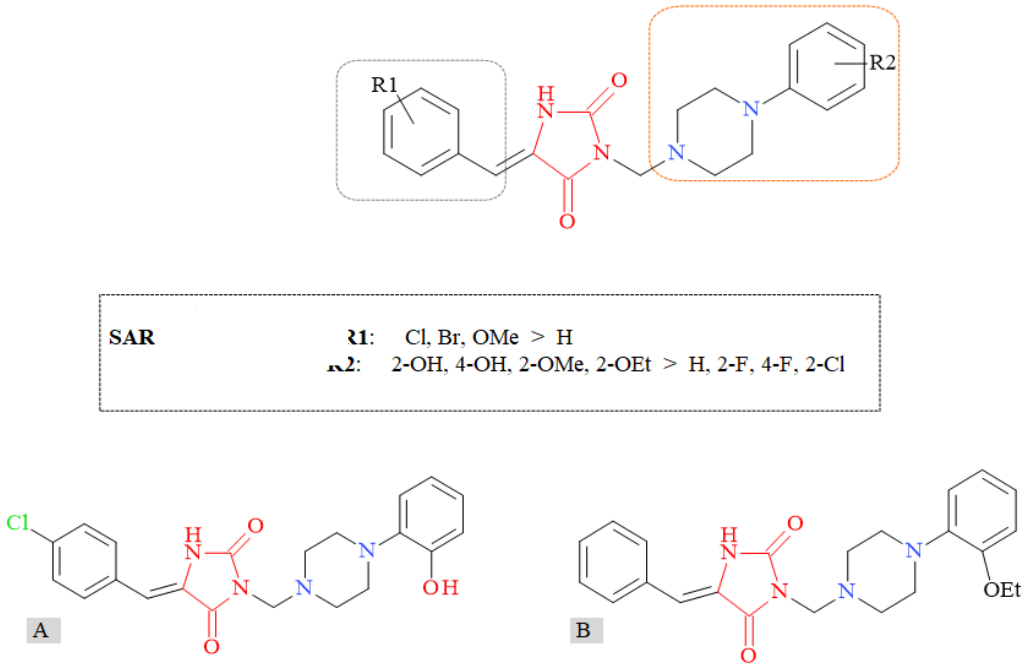


Fig. 1.5. Design and SAR of benzylidenehydantoin-phenylpiperazine hybrids based on known ligands [57]

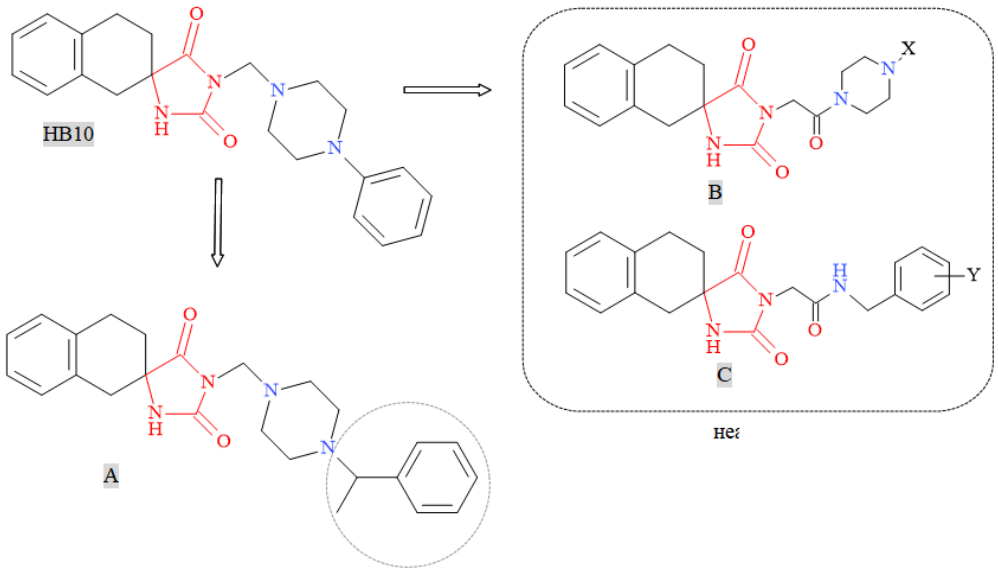
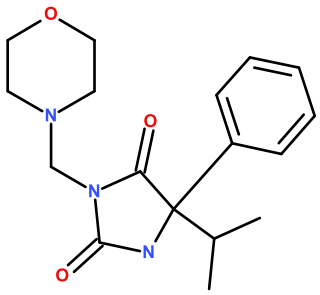


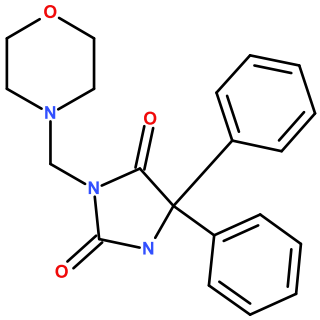
Fig. 1.6. Anticonvulsant activity of β-tetralinohydantoins [59]

As a result of the conducted study, new hybrid compounds containing imidazolidine-2,4-dione and morpholine fragments were synthesized, showing

promising anticonvulsant properties. In particular, compound D – 5-isopropyl-3-(morpholinomethyl)-5-phenylimidazolidine-2,4-dione – demonstrated higher efficacy than phenytoin and levetiracetam in several animal seizure models, including the 6 Hz test (32 and 44 mA). Compound E – 3-(morpholinomethyl)-5,5-diphenylimidazolidine-2,4-dione – also exhibited similar or slightly better activity compared to known anticonvulsant drugs. Both compounds were non-toxic in HepG2 cell lines and did not significantly affect sodium or calcium channels. However, neither of them showed antinociceptive activity in the oxaliplatin-induced neuropathic pain model, indicating their narrow specificity as anticonvulsant agents. The obtained results may serve as a basis for further structural optimization and pharmacological evaluation of new anticonvulsant agents [60].



D



E

1.5.3.2. Imidazolidine-2,4-dione derivatives as serotonin receptor antagonists

In 2014, Handzlik and colleagues reported the development of 5,5-disubstituted hydantoin-phenylpiperazine hybrids (II–IV) as serotonin receptor antagonists. The design of these three compounds was inspired by a previously known ligand. This study is particularly notable because, depending on the nature of the substituents, selective antagonists were identified for the alpha 1-adrenergic receptor (α1-AR), as well as the 5-HT1A, 5-HT6, and 5-HT7 serotonin receptors, highlighting the potential for discovering highly selective drug candidates. The synthesized compounds were classified into three groups based on the substituents at the 5-position of the hydantoin ring: 5,5-diphenyl (III), 5-methyl-5-

phenyl (IV), and 5,5-dimethyl (V). Radioligand binding assays demonstrated that most of the compounds exhibited moderate to strong affinity toward α1-AR, 5-HT1A, and 5-HT7 receptors (with Ki values ranging from 0.8 to 353 nM), while showing weak inhibitory activity against 5-HT3 and 5-HT6 receptors (Fig. 1.7)[61].

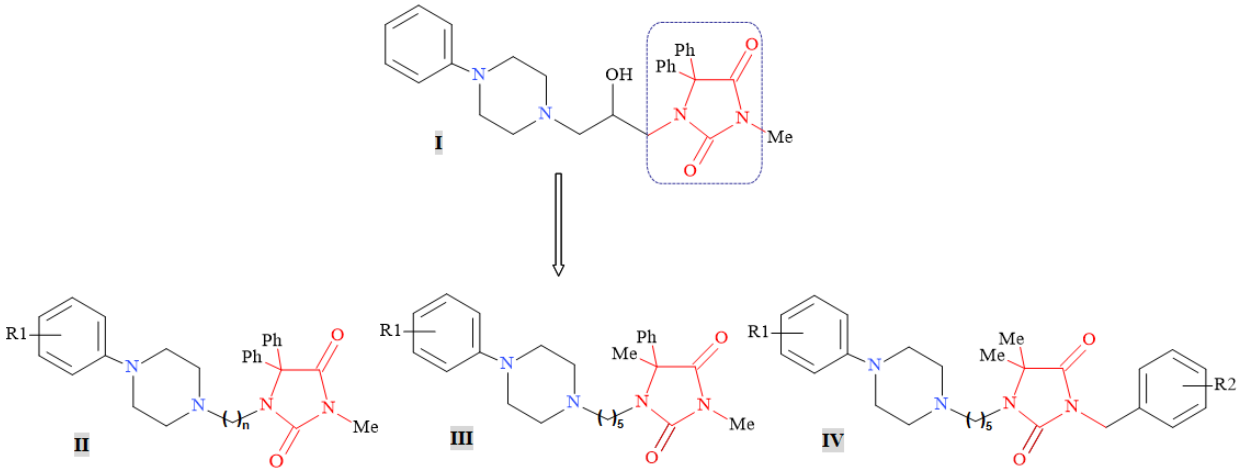


Fig. 1.7 5,5-Disubstituted hybrids of hydantoin with phenylpiperazine with anti-5-HT activity.

Conclusions to Chapter 1

In Chapter 1, a comprehensive literature review was conducted on the current understanding of Alzheimer's disease. Key aspects of the disease's etiology were analyzed, including genetic, environmental, and social factors contributing to its development. Epidemiological data were reviewed, highlighting the global increase in disease prevalence associated with an aging population. Special attention was given to the analysis of risk factors such as heredity, cardiovascular diseases, diabetes, obesity, lifestyle, and educational level.

The pathogenic mechanisms underlying the progression of Alzheimer's disease were discussed in detail, focusing on the role of amyloid plaques, neurofibrillary tangles, oxidative stress, neurotransmitter system imbalances, and inflammatory processes in the CNS.

Furthermore, current approaches to drug therapy were summarized, including symptomatic treatments (such as acetylcholinesterase inhibitors and NMDA

receptor antagonists) and promising directions for the development of disease-modifying therapies.

The role of hydantoin derivatives as potential CNS agents was also addressed. It was shown that, due to their structural flexibility and chemical derivatizability, hydantoin-based compounds can exhibit a broad range of biological activities, including neuroprotective, anticonvulsant, and antioxidant properties. This highlights the potential of hydantoin derivatives for further investigation in the context of therapies for neurodegenerative diseases, particularly Alzheimer's disease.

Thus, the analysis underscores the relevance of searching for new biologically active molecules based on the hydantoin scaffold for the development of effective therapeutics for Alzheimer's disease.

CHAPTER 2
DETERMINATION OF DRUG-LIKE PARAMETERS AND PREDICTION OF TARGETED EFFECTS OF IMIDAZOLIDINE-2,4-DIONE DERIVATIVES

2.1. Characteristics of research objects

Derivatives of imidazolidine-2,4-dione have already demonstrated activity toward the CNS, as discussed in detail in Chapter 1. A significant body of experimental research in this area has been conducted by scientists at the National University of Pharmacy under the supervision of Doctor of Pharmaceutical Sciences, Professor Hanna Severina, who studied the synthesis and neurotropic properties of various azole and azine derivatives.

Continuing the series of studies aimed at identifying new compounds capable of targeting CNS-related receptors, several virtual molecules were designed based on SAR analysis of known active structures. During the creation of the target derivatives, a hybrid design concept was employed – combining multiple pharmacophoric fragments using an appropriate alkyl linker.

The core structural fragment was selected as imidazolidine-2,4-dione, a key component of the well-known anticonvulsant phenytoin (see Fig. 2.1). To increase lipophilicity, this fragment was connected to a cyclohexane ring via a spiro atom, resulting in the formation of the diazaspiro[4.5]decane-2,4-dione system.

Thus, the resulting structure represents a spirocyclic system, consisting of:

- a five-membered imidazolidine ring containing two nitrogen atoms;
- a six-membered cyclohexane ring, joined through a central spiro atom;
- two keto groups located at positions 2 and 4 of the imidazolidine moiety.

The second pharmacophoric fragment of the hybrid molecule was selected as 2-oxopyrrolidine, the key structural unit of piracetam – a nootropic drug that modulates glutamatergic neurotransmission. The fragment in the first position of the pyrrolidine cycle is a substituted benzyl, as in an Aniracetam. Four compounds have

been constructed. The general design scheme of the compounds is presented in Figure 2.1.

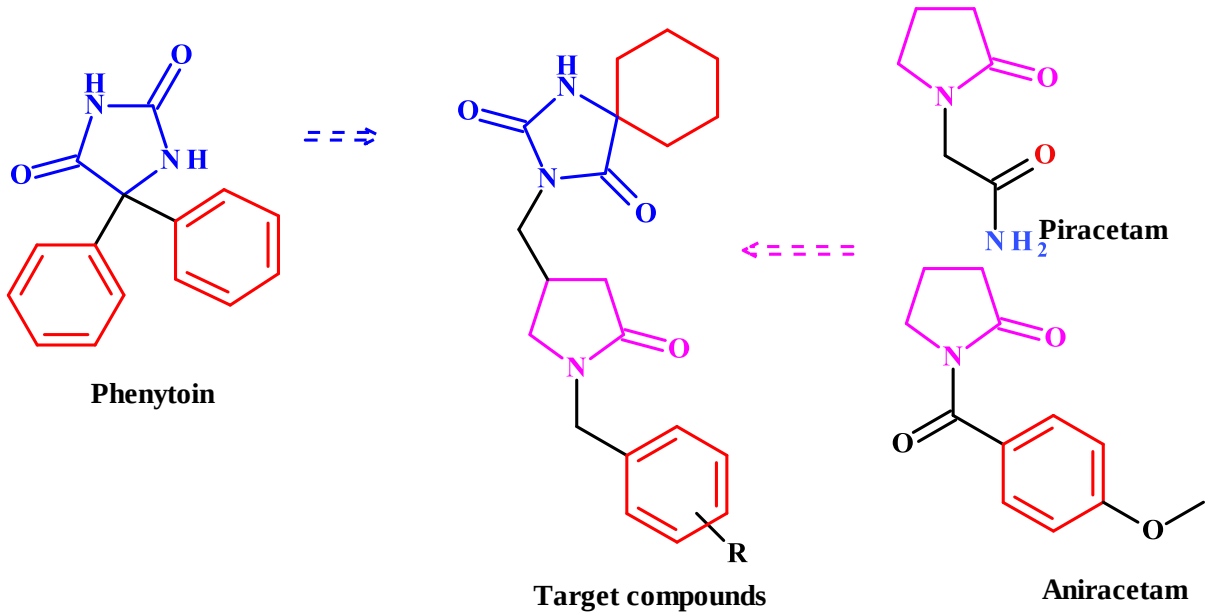


Fig. 2.1 Design of a virtual compound database

2.2. Calculation of parameters of absorption, distribution, metabolism, excretion and toxicity of predicted compounds

ADMET (Absorption, Distribution, Metabolism, Excretion, and Toxicity) represents a comprehensive set of parameters that describe both the pharmacokinetic and pharmacodynamic behavior of a drug molecule. This profile plays a crucial role in determining the therapeutic efficacy and safety of a biologically active compound. A well-balanced ADMET profile is essential for the successful progression of a drug candidate through all stages of development – from preclinical studies to clinical application [62].

Poor absorption, rapid metabolism, undesirable tissue distribution, or pronounced toxicity are often the main reasons for failures in clinical trials, despite high in vitro activity of novel chemical compounds. Therefore, early prediction of ADMET properties has become a mandatory step in the rational design of drug candidates.

In recent decades, modern computational technologies, particularly machine learning techniques and Quantitative Structure-Activity Relationship (QSAR) analysis, have been actively incorporated into ADMET modeling. These approaches enable rapid and accurate estimation of key parameters that define the biopharmaceutical behavior of a compound within the human body [62].

In this study, the ADMET profile was evaluated using the online platform SwissADME, which provides a bioavailability radar – a visual tool for assessing the drug-likeness of small molecules (Fig. 2.2). This radar graphically displays six critical molecular descriptors that significantly affect oral bioavailability: lipophilicity (LIPO), molecular size (SIZE), polarity (POLAR), water solubility (INSOLU), unsaturation (INSATU), and molecular flexibility (FLEX). A comprehensive assessment of these parameters allows for a well-grounded evaluation of the compound's potential as a future therapeutic agent [62].

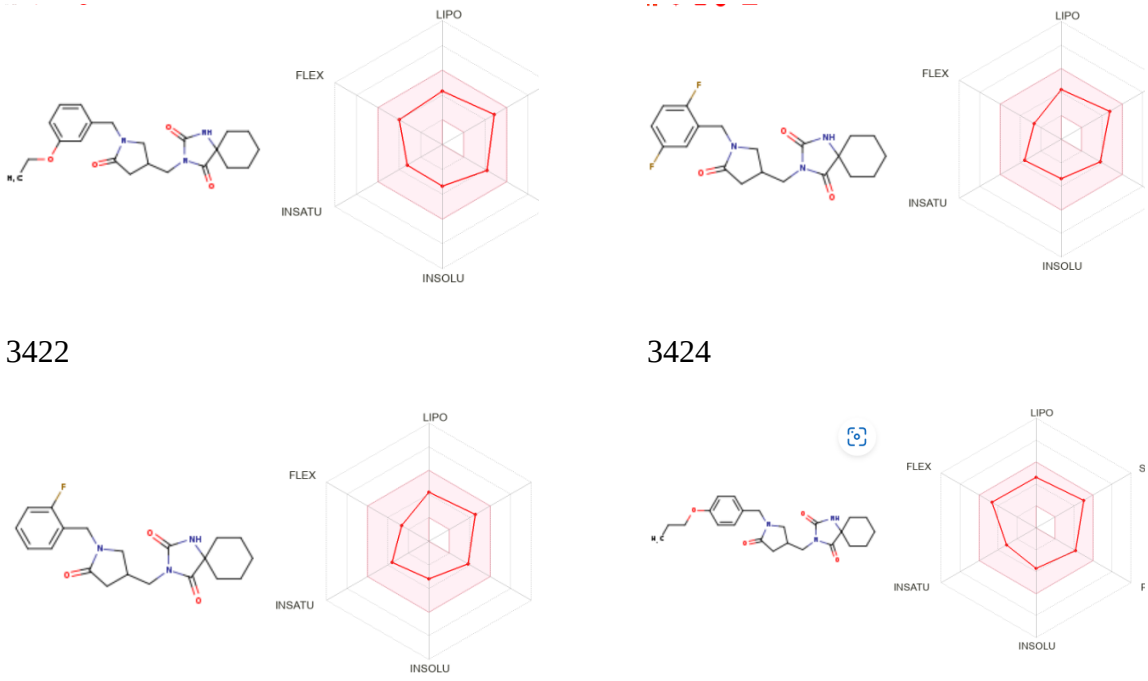


Fig. 2.2. Visualization of the SwissADME bioavailability radar of the studied derivatives

The optimal molecular weight range is between 150 and 500 g/mol [64], and all four studied compounds fall within this parameter. They also meet other key criteria: topological polar surface area (TPSA) within the acceptable range of 20 – 130 Å² [65], molecular flexibility (no more than 9 rotatable bonds) [64], and molar refractivity (within the range of 40–130). Additionally, the fraction of sp^3-hybridized carbon atoms, which reflects the degree of molecular saturation, should be no less than 0.25 [66], and all the compounds comply with this requirement. The number of heavy atoms can influence a molecule's bioavailability and lipophilicity; the number of aromatic heavy atoms may affect its binding interactions with proteins and enzymes; the number of rotatable bonds contributes to the molecule's conformational flexibility; and the number of hydrogen bond donors and acceptors plays a role in both solubility and intermolecular binding (Fig. 2.3).

Physicochemical Properties	
Formula	C22H29N3O4
Molecular weight	399.48 g/mol
Num. heavy atoms	29
Num. arom. heavy atoms	6
Fraction Csp3	0.59
Num. rotatable bonds	6
Num. H-bond acceptors	4
Num. H-bond donors	1
Molar Refractivity	120.22
TPSA	78.95 Å²

Compound 3422

Physicochemical Properties	
Formula	C20H23F2N3O3
Molecular weight	391.41 g/mol
Num. heavy atoms	28
Num. arom. heavy atoms	6
Fraction Csp3	0.55
Num. rotatable bonds	4
Num. H-bond acceptors	5
Num. H-bond donors	1
Molar Refractivity	108.84
TPSA	69.72 Å²

Compound 3424

Physicochemical Properties	
Formula	C20H24FN3O3
Molecular weight	373.42 g/mol
Num. heavy atoms	27
Num. arom. heavy atoms	6
Fraction Csp3	0.55
Num. rotatable bonds	4
Num. H-bond acceptors	4
Num. H-bond donors	1
Molar Refractivity	108.88
TPSA	69.72 Å²

Compound 3426

Physicochemical Properties	
Formula	C23H31N3O4
Molecular weight	413.51 g/mol
Num. heavy atoms	30
Num. arom. heavy atoms	6
Fraction Csp3	0.61
Num. rotatable bonds	7
Num. H-bond acceptors	4
Num. H-bond donors	1
Molar Refractivity	125.03
TPSA	78.95 Å²

Compound 3430

Fig. 2.3. Physicochemical properties of the studied compounds

The values of lipophilicity are important for predicting the behavior of compounds in biological systems, particularly their ability to penetrate biological membranes. The optimal range for iLOGP is from -3.93 to 6.46. Regarding MLOGP, it should be < 4.15, XLOGP3 should be between -0.7 and +5.0 [67, 68], and WLOGP should be < 5.88 [63]. Therefore, all compounds fit within the lipophilicity criteria (Fig. 2.4).

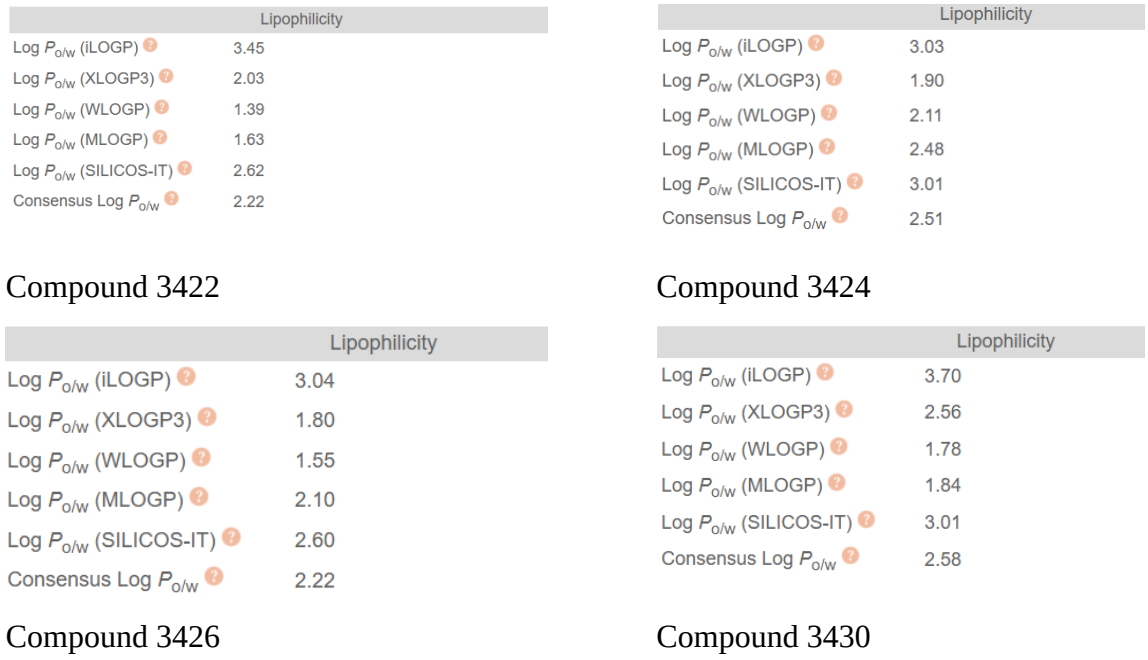


Fig. 2.4 Lipophilicy of the studied compounds

An important aspect for parenteral drugs is their water solubility (log S), as it ensures a sufficient amount of the API in a small volume of the therapeutic dose. The qualitative assessment of solubility class is proposed according to the following scale for log s [63]. This scale helps to determine how effectively a drug will dissolve in water, which is crucial for its absorption and efficacy in the body when administered parenterally.

Gastrointestinal absorption of drugs describes the process by which the active components of drugs are absorbed through the stomach and intestinal wall into the circulatory system. P-glycoprotein is a crucial membrane protein responsible for exporting many foreign substances from cells, such as from the gastrointestinal tract into the lumen or from the brain [69]. It also plays a protective role for the CNS

against xenobiotic effects [70]. All drugs are substrates for P-glycoprotein. Additionally, key compounds involved in drug elimination through metabolic biotransformation include five major isoforms of cytochrome P450, such as CYP1A2, CYP2C19, CYP2C9, etc [71-73]. A more negative log Kp (cm/s) indicates lower permeability of a molecule through the skin [74]. The results of the study are shown in Fig. 2.6

Water Solubility	
Log *S* (ESOL)	-3.35
Solubility	1.77e-01 mg/ml ; 4.44e-04 mol/l
Class	Soluble
Log *S* (Ali)	-3.32
Solubility	1.93e-01 mg/ml ; 4.83e-04 mol/l
Class	Soluble
Log *S* (SILICOS-IT)	-5.14
Solubility	2.91e-03 mg/ml ; 7.28e-06 mol/l
Class	Moderately soluble

Compound 3422

Water Solubility	
Log *S* (ESOL)	-3.36
Solubility	1.72e-01 mg/ml ; 4.38e-04 mol/l
Class	Soluble
Log *S* (Ali)	-2.99
Solubility	4.03e-01 mg/ml ; 1.03e-03 mol/l
Class	Soluble
Log *S* (SILICOS-IT)	-5.17
Solubility	2.64e-03 mg/ml ; 6.75e-06 mol/l
Class	Moderately soluble

Compound 3424

Water Solubility	
Log *S* (ESOL)	-3.19
Solubility	2.41e-01 mg/ml ; 6.46e-04 mol/l
Class	Soluble
Log *S* (Ali)	-2.88
Solubility	4.89e-01 mg/ml ; 1.31e-03 mol/l
Class	Soluble
Log *S* (SILICOS-IT)	-4.91
Solubility	4.64e-03 mg/ml ; 1.24e-05 mol/l
Class	Moderately soluble

Compound 3426

Water Solubility	
Log *S* (ESOL)	-3.70
Solubility	8.20e-02 mg/ml ; 1.98e-04 mol/l
Class	Soluble
Log *S* (Ali)	-3.87
Solubility	5.63e-02 mg/ml ; 1.36e-04 mol/l
Class	Soluble
Log *S* (SILICOS-IT)	-5.53
Solubility	1.22e-03 mg/ml ; 2.95e-06 mol/l
Class	Moderately soluble

Compound 3430

Fig. 2.5 Water solubility of the studied compounds

Pharmacokinetics	
GI absorption	High
BBB permeant	No
P-gp substrate	Yes
CYP1A2 inhibitor	No
CYP2C19 inhibitor	No
CYP2C9 inhibitor	No
CYP2D6 inhibitor	Yes
CYP3A4 inhibitor	Yes
Log K_p (skin permeation)	-7.30 cm/s

Compound 3422

Pharmacokinetics	
GI absorption	High
BBB permeant	Yes
P-gp substrate	Yes
CYP1A2 inhibitor	No
CYP2C19 inhibitor	No
CYP2C9 inhibitor	No
CYP2D6 inhibitor	Yes
CYP3A4 inhibitor	Yes
Log K_p (skin permeation)	-7.34 cm/s

Compound 3424

Pharmacokinetics	
GI absorption	High
BBB permeant	Yes
P-gp substrate	Yes
CYP1A2 inhibitor	No
CYP2C19 inhibitor	Yes
CYP2C9 inhibitor	No
CYP2D6 inhibitor	Yes
CYP3A4 inhibitor	Yes
Log K_p (skin permeation)	-7.30 cm/s

Compound 3426

Pharmacokinetics	
GI absorption	High
BBB permeant	No
P-gp substrate	Yes
CYP1A2 inhibitor	No
CYP2C19 inhibitor	No
CYP2C9 inhibitor	Yes
CYP2D6 inhibitor	Yes
CYP3A4 inhibitor	Yes
Log K_p (skin permeation)	-7.00 cm/s

Compound 3430

Fig. 2.6 Pharmacokinetics of the studied compounds

All tested compounds do not show any alerts according to the PAINS rules. This indicates that they do not contain structural fragments typically found in inactive or toxic compounds. However, all tested compounds have one alert according to the Brenk rule, related to the guanidino fragment. Since guanidine can be toxic, its presence in these compounds requires further investigation. None of the studied compounds meet all the criteria for a "leader" compound, and their synthetic accessibility is moderate (Fig. 2.7).

Druglikeness	
Lipinski	Yes; 0 violation
Ghose	Yes
Veber	Yes
Egan	Yes
Muegge	Yes
Bioavailability Score	0.55

Medicinal Chemistry	
PAINS	0 alert
Brenk	1 alert: hydantoin
Leadlikeness	No; 1 violation: MW>350
Synthetic accessibility	3.80

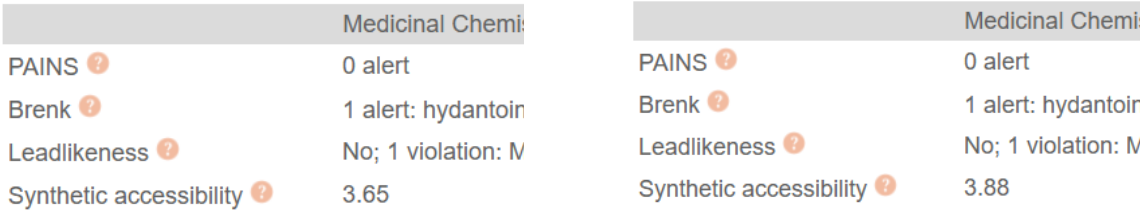
Compound 3422

Druglikeness	
Lipinski	Yes; 0 violation
Ghose	Yes
Veber	Yes
Egan	Yes
Muegge	Yes
Bioavailability Score	0.55

Medicinal Chemistry	
PAINS	0 alert
Brenk	1 alert: hydantoin
Leadlikeness	No; 1 violation: MW>350
Synthetic accessibility	3.71

Compound 3424

Druglikeness	
Lipinski	Yes; 0 violation
Ghose	Yes
Veber	Yes
Egan	Yes
Muegge	Yes
Bioavailability Score	0.55



Compound 3426 Compound 3430

Fig. 2.7 Druglikeness and Medicinal Chemistry parameters of the studied compounds

A summarized comparison table of all four compounds by key ADMET parameters is presented in Table 2.1

Table 2.1

Comparative table of key ADMET parameters of the studied compounds

Parameter	Compound 3422	Compound 3424	Сполука 3426	Сполука 3430
Molecular weight	399.48	391.41	373.42	413.51
TPSA (Å²)	78.95	69.72	69.72	78.95
Fraction Csp3	0.59	0.55	0.55	0.61
Num. rotatable bonds	6	4	4	7
HBA / HBD	4 / 1	5 / 1	4 / 1	4 / 1
LogP (Consensus)	2.22	2.51	2.22	2.58
Water Solubility (ESOL, LogS)	-3.35 (soluble)	-3.36 (soluble)	-3.19 (soluble)	-3.70 (soluble)
GI absorption	Hi	Hi	Hi	Hi
BBB permeant	No	Yes	Yes	No
P-gp csubstrate	Yes	Yes	Yes	Yes
CYP1A2 inhibitor	No	No	No	No
CYP2C19 inhibitor	No	No	Yes	Hi
CYP2C9 inhibitor	No	No	No	Yes
CYP2D6 inhibitor	Yes	Yes	Yes	Yes
CYP3A4 інгібітор	Yes	Yes	Yes	Yes
Log Kp (skin permeation)	-7.30	-7.34	-7.30	-7.00

Bioavailability Score	0.55	0.55	0.55	0.55
PAINS/Brenk alerts	0/1 (hydantoin)	0/1 (hydantoin)	0/1 (hydantoin)	0/1 (hydantoin)
Leadlikeness	Hi (MW > 350)	Hi (MW > 350)	Hi (MW > 350)	Hi (MW > 350)
Synthetic accessibility	3.80	3.71	3.65	3.88

The best BBB permeability is observed in compounds 2424 and 2426, making them potentially suitable for CNS targeting. Compound 2426 also has the highest water solubility, with the least negative LogS value. Compound 2424 shows the lowest CYP inhibition (inhibiting only CYP2D6 and CYP3A4). Compound 2426 has the lowest molecular weight and the best CNS accessibility. Compound 2430 has the highest Csp^3 fraction and the greatest molecular complexity, which may indicate synthetic challenges but also offers good three-dimensionality.

Based on a comprehensive analysis of the ADMET profiles, the most promising compound for further investigation – especially if the target is CNS-related neurodegeneration modulation – is derivative 3426.

Its advantages include:

- High gastrointestinal absorption;
- Ability to cross the blood–brain barrier (BBB) – crucial for CNS-targeting drugs;
- No violations of Lipinski, Ghose, Veber, Egan, or Muegge rules – indicating good drug-likeness;
- Best water solubility – beneficial for bioavailability and formulation;
- Moderate LogP (~2.22) – providing an optimal hydrophilic–lipophilic balance;
- Lower CYP enzyme inhibition – suggesting a reduced risk of metabolic interactions.

Minor drawbacks include:

- Inhibition of CYP2C19, 2D6, and 3A4 – thus potential drug–drug interactions should be considered;

- Non-compliance with lead-likeness (MW > 350) – though this is less relevant during the hit optimization stage.

Therefore, compound 3426 is the most optimal candidate for continued in silico, in vitro, and pharmacological testing, especially for CNS targets. As such, it has been selected for further affinity studies with CNS-related targets.

2.3. Predicting the targeted impact of a promising derivative 3426

A wide range of small molecules, including metabolites, signaling molecules, and drugs, exhibit strong biological activity in various living systems. This activity is often mediated through physical interactions with proteins or other macromolecules. Therefore, understanding the targets of biologically active molecules is essential for comprehending, predicting, and modulating their activity. Specifically, this information can help predict adverse side effects due to off-target interactions, potentially reducing participant dropout rates in clinical trials due to toxicity [75, 76] or it can be used to identify new targets for existing drugs and repurpose them for the treatment of different diseases [77,78].

Ligand-based target prediction has proven to be highly effective and fast in predicting the correct protein targets of compounds in the context of drug development [79, 80]. Quantitative assessment of similarity between compounds using various methods has confirmed the intuitive "molecular similarity hypothesis," which postulates the presence of common protein targets for similar molecules [81,82].

The program SwissTargetPrediction was used to predict the most likely targets for the studied derivative 3426. The visualization of the results is presented as a pie chart (Fig. 2.8), illustrating the distribution of protein target classes in percentages.

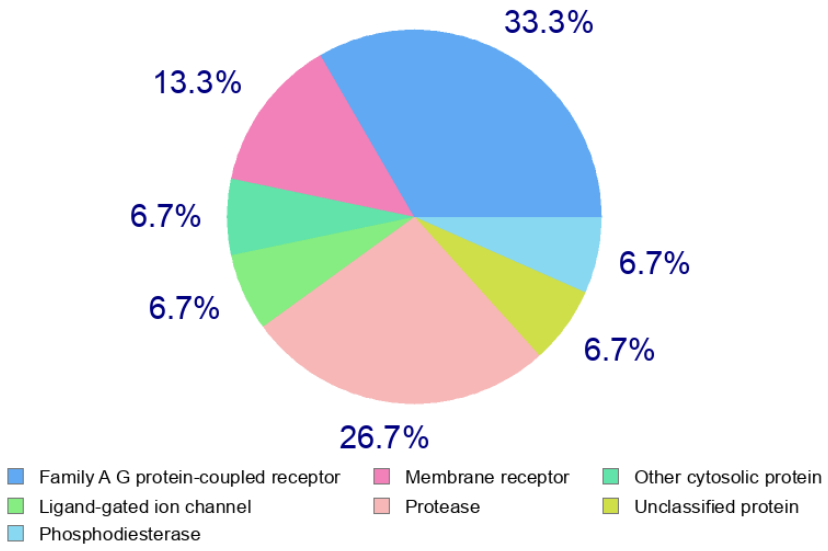


Fig. 2.8. Visualization of the most probable classes of biotargets in percentage for compound 3426

Among the general classes of targets, we highlighted those that may influence and modulate neurodegenerative processes.

- Family A G protein-coupled receptors (GPCRs) – 33.3% – targets include serotonin, dopamine, adenosine, and opioid receptors. GPCRs are key regulators of neurotransmission, cognition, mood, and arousal.
- Membrane receptors – 13.3% – include proteins such as Sigma-1 receptor and translocator protein (TSPO), which exhibit neuroprotective or pro-inflammatory activity.
- Proteases – 6.7% – for example, gamma-secretase, responsible for β-amyloidogenesis, a central mechanism in the pathogenesis of Alzheimer's disease.
- Ligand-gated ion channels – 6.7% – represented by GABA-A receptors, which are the main inhibitory channels in the CNS, critical for excitation-inhibition balance.

- Phosphodiesterases – 6.7% – regulate cAMP/cGMP levels, participate in neurotransmission and memory formation; considered promising targets in Alzheimer’s disease.
- Other cytosolic protein / Unclassified protein / Enzymes – 6.7% each – include proteins with less defined classification, but involved in apoptosis regulation, protein phosphorylation, and metabolism.

Thus, GPCRs dominate the prediction, which aligns with their leading role in regulating neurophysiology and the structural similarity of the studied compound to known ligands of these targets. The presence of proteases, phosphodiesterases, GABA channels, and membrane receptors in the prediction highlights the potential for a multitarget effect on neurodegeneration.

The analysed targets represent key molecular mechanisms underlying neurodegenerative diseases, including β-amyloid formation, oxidative stress, neuroinflammation, and neurotransmitter imbalance. Particular attention is given to gamma-secretase, MAO-B, CDK5, and GABA-A receptors, which are directly implicated in Alzheimer's and Parkinson's pathogenesis (Table 2.2).

The inclusion of acetylcholinesterase highlights the importance of addressing cognitive decline through classical and complementary pathways. Overall, the diversity of target classes supports the potential of multitarget-directed ligands in developing more effective neuroprotective therapies.

Acetylcholinesterase (AChE), although not appearing in the prediction results (likely due to a lack of structural similarity to known ligands), remains a key pharmacological target in the treatment of cognitive disorders.
It should be considered in multitarget drug design, especially for compounds with potential nootropic activity. Although GSK3, a key kinase involved in tau hyperphosphorylation in Alzheimer’s disease, was not among the predicted targets, it remains a highly relevant candidate for further evaluation.

Table 2.2

Top targets most likely involved in neurodegeneration

№	Target	Relevance to Neurodegeneration
1	Gamma-secretase complex *(PSEN1/2, PSENEN)*	Generates β-amyloid from APP – a central mechanism in Alzheimer's disease.
2	Monoamine oxidase B (MAOB)	Oxidizes dopamine, generating free radicals; inhibition reduces neurotoxicity (Parkinson's disease, AD).
3	Cyclin-dependent kinase 5 (CDK5)	Abnormal activation leads to tau protein hyperphosphorylation and neuronal death.
4	Indoleamine 2,3-dioxygenase (IDO1)	Activates the kynurenine pathway, producing neurotoxic metabolites; promotes inflammation.
5	Translocator protein (TSPO)	Marker of activated microglia; target for modulating and imaging neuroinflammation.
6	Sigma-1 receptor (SIGMAR1)	Modulates cellular survival and calcium homeostasis; provides neuroprotective effects.
7	Adenosine A2A receptor (ADORA2A)	Involved in neuroinflammation and glutamate toxicity; therapeutic target in Parkinson's disease.
8	GABA-A receptors (α1–α5, β3, γ2)	Dysregulation contributes to cognitive decline and seizure disorders.
9	P2X7 receptor (P2RX7)	Triggers release of pro-inflammatory cytokines by microglia; drives chronic CNS inflammation.
10	DYRK1A	Overexpressed in down syndrome; linked to memory loss and Alzheimer-like pathology.

Conclusions to the Chapter 2

1. The design strategies for target imidazolidine-2,4-dione derivatives as hybrid compounds have been described.
2. A comprehensive ADMET analysis of the four investigated compounds was carried out, allowing for the evaluation of their pharmacokinetic, physicochemical, and medicinal chemistry properties in order to identify the most promising candidate for further research.
3. All compounds meet the basic criteria for molecular weight (150–500 g/mol), topological polar surface area, molecular refractivity, fraction of sp^3-hybridized carbon atoms, and number of rotatable bonds, indicating their potential drug-likeness and compliance with modern drug evaluation guidelines.

4. The best blood-brain barrier permeability was shown by compounds 2424 and 2426, making them potentially suitable for targeting the CNS. Among them, compound 3426 proved to be the most balanced across several key parameters: it has the best water solubility, high gastrointestinal absorption, an acceptable lipophilicity level (LogP ≈ 2.22), no violations of Lipinski, Veber, Egan, Ghose, or Muegge rules, and a moderate CYP inhibition profile, reducing the risk of metabolic interactions.
5. Minor drawbacks were identified – a violation of the lead-likeness criterion (molecular weight > 350 g/mol) and inhibition of CYP2C19, CYP2D6, and CYP3A4 enzymes, which requires further attention in *in vitro* studies of potential drug interactions.
6. Compound 3426 is considered the most promising candidate for further *in silico*, *in vitro*, and pharmacological research, particularly for addressing neurodegenerative diseases with CNS targets.
7. The prediction results for the investigated ligand 3426 highlight a diverse range of neurodegeneration-related targets – such as gamma-secretase, MAO-B, CDK5, GABA-A receptors, and the Sigma-1 receptor – emphasizing the potential for multitarget therapeutic strategies that address amyloidogenesis, oxidative stress, neurotransmitter imbalance, and neuroinflammation.

CHAPTER 3
PREDICTION OF THE AFFINITY OF IMIDAZOLIDINE-2,4-DIONE DERIVATIVES FOR TARGETS REGULATING NEURODEGENERATION

One of the promising approaches to the treatment of neurodegenerative diseases is multitarget drug design, aimed at simultaneously inhibiting several pathogenic mechanisms. In this context, acetylcholinesterase (AChE) and glycogen synthase kinase 3 (GSK3) are key targets: the former is a classical enzyme implicated in the pathogenesis of cognitive impairment, while the latter plays a critical role in tau protein hyperphosphorylation in Alzheimer's disease. Considering the neuroactive potential of imidazolidine-2,4-dione derivative 3426, an *in silico* prediction of its binding affinity to the active sites of ACHE and GSK3 was carried out to assess its suitability for further pharmacological investigation.

3.1 Molecular docking into the AChE inhibitor site

One of the leading approAChEs in the study of cognitive impairment is the cholinergic hypothesis, which suggests that the decline in cognitive function is associated with a deficit of the neurotransmitter acetylcholine. It has been shown that acetylcholinesterase (AChE) can bind to amyloid-β peptides and may contribute to the formation of amyloid plaques [83]. Although acetylcholine can also be degraded by butyrylcholinesterase, its activity progressively increases in patients with Alzheimer's disease, whereas AChE activity remains unchanged or decreases [84].

To date, no treatment has been found that effectively halts the progression of AD. For mild to moderate stages of the disease, cholinesterase inhibitors such as donepezil, galantamine, and rivastigmine, as well as memantine – an NMDA receptor antagonist that modulates glutamatergic activity – are prescribed. While acetylcholinesterase inhibitors do not prevent disease progression, they offer symptomatic relief of cognitive decline and contribute to improved quality of life for patients and caregivers alike [85].

Therefore, the search for new biologically active compounds with similar pharmacological effects but reduced side effects and broader multitarget potential remains highly relevant. To evaluate the binding affinity to the cholinesterase active site and predict potential pharmacological activity, an in silico molecular docking study was performed using AutoDock Vina.

The crystallographic structure of acetylcholinesterase in complex with a donepezil, derived from *Tetronarce californica* was used, as reported by Caliandro R. et al. in 2018 [86]. The native ligand occupies the enzyme's active-site gorge, extending from the catalytic anionic site near Trp84 to the peripheral anionic site near Trp279 (PDB ID: 5NAU), like the binding mode of donepezil (PDB ID: 1EVE). The analysis of ligand–protein complexes revealed key structural features that influence inhibitory activity, providing a foundation for structure-based design of more selective and potent AChE inhibitors.

The calculated binding energy of donepezil was -11.0 kcal/mol. The affinity of the investigated ligand 3426 was similarly high, at -10.9 kcal/mol, showing no significant difference compared to the reference ligand (Fig. 3.1, Table 3.1).

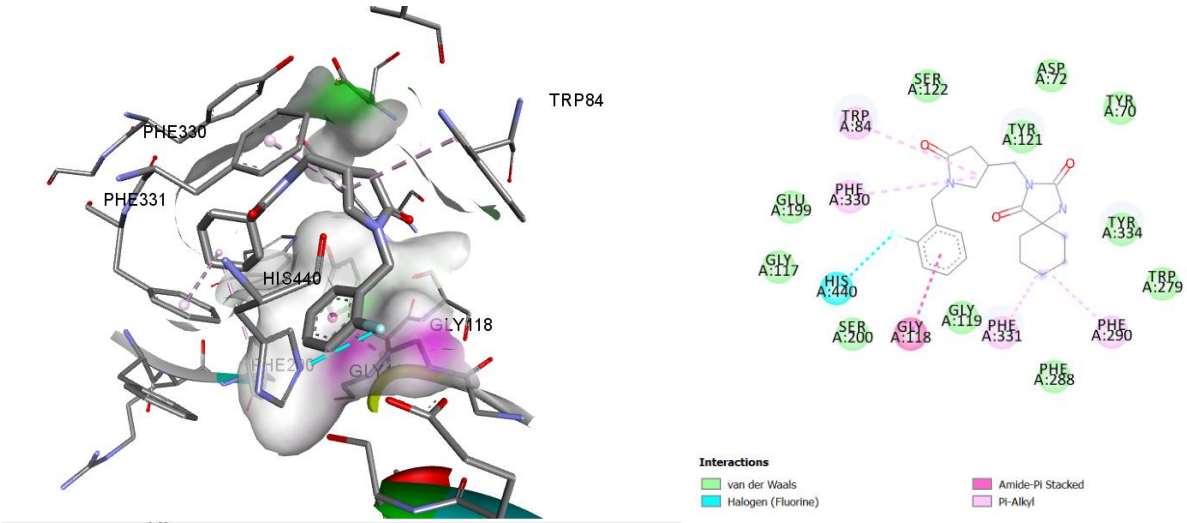


Fig. 3.1. 3D and 2D visualization of the conformational arrangement of ligand 3426 in the active site of acetylcholinesterase inhibitor

The molecular docking results indicate significant contributions from hydrophobic interactions, π-stacking, and even halogen bonding, supporting the formation of a stable ligand–enzyme complex (Table 3.1).

Table 3.1

Results of molecular docking of derivative 3426 into the active site of AChE and GSK3

AChE				GSK3			
Residues	Distance	Category	Types	Residues	Distance	Category	Types
HIS440	3,38884	Halogen	Halogen (Fluorine)	ALA26	3,68572	Hydrogen Bond	Carbon Hydrogen Bond
GLY118, GLY119	3,88894	Hydrophobic	Amide-Pi Stacked	PHE31	4,27852	Hydrophobic	Pi-Pi Stacked
TRP84	4,68498	Hydrophobic	Pi-Alkyl	VAL34	4,20496	Hydrophobic	Alkyl
PHE290	5,30241	Hydrophobic	Pi-Alkyl	ALA47	4,16668	Hydrophobic	Alkyl
PHE330	4,45315	Hydrophobic	Pi-Alkyl	LEU158	4,73144	Hydrophobic	Alkyl
PHE331	5,16441	Hydrophobic	Pi-Alkyl	–	–	–	–

The ligand 3426 forms several characteristic interactions within the active site of AChE:

A halogen bond is formed between the fluorine atom of the ligand (as a halogen donor) and the NE2 atom of HIS440 (as a halogen acceptor) at a distance of 3.39 Å. This specific interaction contributes to the directional binding of the ligand and enhances its selectivity for the enzyme's active site.

An amide–π stacking interaction is observed between the ligand and the backbone atoms of GLY118 and GLY119 at a distance of 3.89 Å. This type of interaction stabilizes the positioning of the ligand near the main chain of the enzyme and is a common mechanism for anchoring compounds within the binding pocket.

Multiple π–alkyl hydrophobic interactions occur between the ligand and key aromatic residues: TRP84 (4.68 Å), PHE290 (5.30 Å), PHE330 (4.45 Å), and PHE331 (5.16 Å). These residues, particularly tryptophan and phenylalanine, are

part of the anionic and peripheral anionic sites of AChE. Their π-orbitals interact with the alkyl regions of the ligand, contributing to hydrophobic anchoring within the active gorge.

Overall, ligand 3426 demonstrates a network of hydrophobic and specific interactions with crucial AChE residues, notably TRP84 (a key anchoring residue in the catalytic gorge) and HIS440 (part of the catalytic triad). The presence of a halogen bond further enhances binding affinity and selectivity, suggesting high stability and promising bioactivity of the complex.

3.2. Molecular docking into the GSK3 inhibitor site

Glycogen Synthase Kinase 3 (GSK3), particularly its β-isoform (GSK3β), plays a central role in the pathogenesis of neurodegenerative diseases, especially Alzheimer's disease. In the context of neurodegeneration, its overactivation leads to hyperphosphorylation of tau protein, resulting in neurofibrillary tangles, a hallmark of Alzheimer's disease pathology.

Additionally, GSK3β contributes to amyloid-β production, synaptic dysfunction, oxidative stress, and neuroinflammation. Because of its involvement in multiple pathological mechanisms, GSK3 is considered a high-value therapeutic target. Selective inhibition of GSK3β is being explored as a strategy to prevent tau aggregation, restore synaptic function, and slow down neuronal loss.

Hence, GSK3 inhibitors are being actively investigated as potential disease-modifying agents for Alzheimer's disease and other tauopathies.

The crystal structure of Glycogen Synthase Kinase 3 from *Leishmania infantum* (PDB ID: 7S6U) complexed with its native ligand AZD5438 [4-(2-methyl-3-isopropylimidazol-4-yl)-*N*-(4-methylsulfonylphenyl)pyrimidin-2-amine] was used as the target for molecular docking [87].

The binding affinity of the investigated ligand to GSK3 was –9.0 kcal/mol, which is slightly higher than that of the reference inhibitor AZD5438 (–8.5 kcal/mol), further supporting its promise as a GSK3-targeting agent.

The docking analysis of ligand 3426 with the enzyme protein Glycogen Synthase Kinase 3 (GSK3) revealed the potential for several key interactions (Fig. 3.2, Table 3.1):

- π–π Stacked interaction with PHE31 at a distance of 4.28 Å – this is a significant aromatic interaction between the ligand and the phenylalanine residue, which helps anchor the ligand within the hydrophobic pocket through parallel alignment of π-orbitals.
- Hydrophobic alkyl–alkyl interactions with VAL34 (4.20 Å), ALA47 (4.17 Å), and LEU158 (4.73 Å) – interactions with the alkyl chains of aliphatic residues form a hydrophobic environment that stabilizes the ligand's positioning within the binding site and promotes effective anchoring.
- Carbon–hydrogen bond with ALA26, observed at 3.69 Å – although weaker than conventional hydrogen bonds, C–H···O interactions can still contribute to ligand stabilization, particularly when located in proximity to other binding forces.

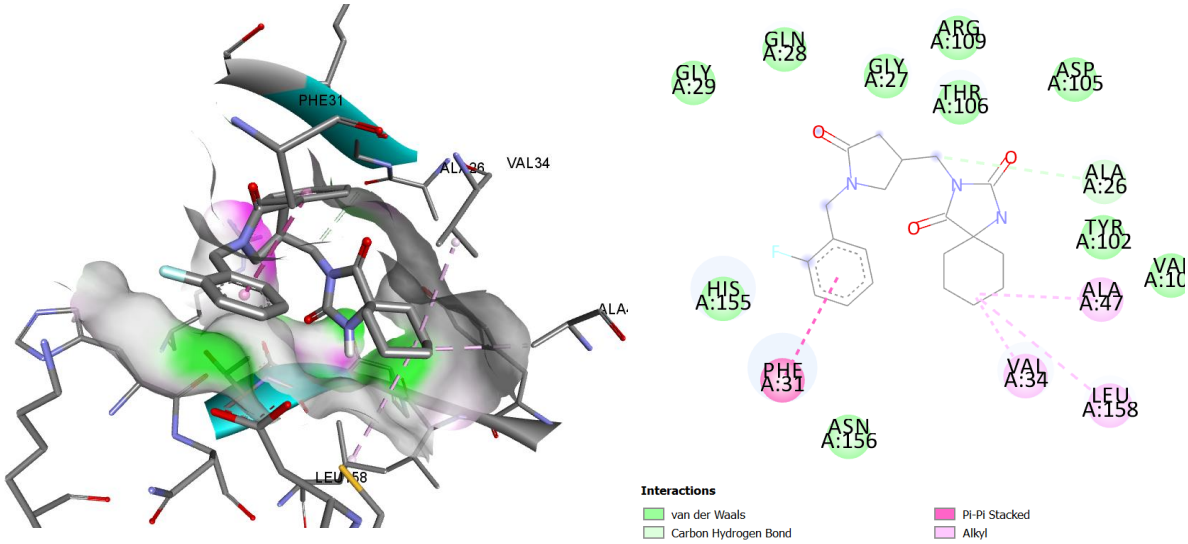


Fig. 3.2. 3D and 2D visualization of the conformational arrangement of ligand 3426 in the active site of GSK3

Ligand 3426 forms a combination of hydrophobic interactions and a weak hydrogen bond with key residues of GSK3. The π–π stacking with PHE31 is

especially notable. This set of interactions supports stable ligand binding and indicates strong potential for the development of a selective GSK3 inhibitor.

Conclusions for Chapter 3

1. Molecular modeling of the interaction between ligand 3426 and acetylcholinesterase (AChE) as well as glycogen synthase kinase 3 (GSK3) revealed the formation of stable complexes with key amino acid residues in the active sites of both targets.
2. Ligand 3426 exhibits strong and specific binding to AChE through a combination of halogen bonding, amide–π stacking, and multiple hydrophobic π–alkyl interactions, particularly with key residues TRP84 and HIS440, indicating a stable complex with high affinity and potential biological activity – comparable to the reference drug donepezil. The calculated binding energy (-10.9 kcal/mol) is nearly equivalent to that of the reference drug donepezil (-11.0 kcal/mol), indicating a high affinity potential.
3. For GSK3, π–π stacking with PHE31, hydrophobic interactions with VAL34, ALA47, and LEU158, as well as a weak hydrogen bond with ALA26 were identified. This combination of interactions ensures favourable complex stability, confirming the ligand's potential to bind effectively with GSK3.
4. The obtained results indicate the potential of the imidazolidine-2,4-dione derivative 3426 as a multitarget ligand capable of simultaneously modulating cholinergic transmission and tau phosphorylation, which represents a relevant approach in the treatment of neurodegenerative diseases.

CONCLUSIONS

1. An analysis and synthesis of the literature on the pathogenesis and pharmacotherapy of Alzheimer's disease was carried out, along with an overview of the pharmacological effects of imidazolidine-2,4-dione derivatives on the central nervous system.
2. The design of hybrid imidazolidine-2,4-dione derivatives was successfully carried out, and their structural features were optimized to meet modern medicinal chemistry criteria.
3. ADMET profiling of the four synthesized compounds confirmed their drug-likeness, with all meeting key parameters such as molecular weight, TPSA, molecular refractivity, and rotatable bonds. Compound 3426 exhibited the most favourable pharmacokinetic profile, including excellent BBB permeability and high gastrointestinal absorption.
4. Despite minor limitations – such as CYP enzyme inhibition and exceeding the lead-likeness molecular weight threshold – compound 3426 emerged as the most promising candidate for further development in CNS-related pharmacology.
5. Target prediction and molecular docking results revealed strong binding affinity of compound 3426 toward multiple neurodegeneration-relevant targets, including AChE, GSK3, MAO-B, CDK5, and GABA-A receptors, highlighting its potential as a multitarget-directed therapeutic agent for neurodegenerative diseases.
6. Molecular docking studies demonstrated that ligand 3426 forms stable and specific complexes with both acetylcholinesterase and glycogen synthase kinase 3, showing high binding affinities (-10.9 kcal/mol and -9.0 kcal/mol, respectively) that are comparable or superior to known reference inhibitors such as donepezil and AZD5438.
7. The observed network of interactions – including hydrogen bonding, π–π stacking, and hydrophobic contacts with key active-site residues—supports

the potential of imidazolidine-2,4-dione derivative 3426 as a multitarget-directed ligand, offering a promising approach for modulating both cholinergic dysfunction and tau-related pathology in neurodegenerative disease therapy.

REFERENCES

1. Dementia subtypes and suicidality: A systematic review and meta-analysis / E. Nuzum et al. *Neurosci. Biobehav. Rev.* 2025. Vol. 169. P. 105995.
2. Arvanitakis Z., Bennett D. A. What Is Dementia? *JAMA*. 2019. Vol. 322, № 17. P. 1728.
3. What Is Dementia? Alzheimer's Disease and Dementia. URL: https://www.alz.org/alzheimers-dementia/what-is-dementia (Date of access: 09.12.2024).
4. Dementia / World Health Organization. URL: https://www.who.int/news-room/fact-sheets/detail/dementia (Date of access: 03.01.2025).
5. Comprehensive Dementia Care Models: State of the Science and Future Directions / K. P. Murali et al. *Res. Gerontol. Nurs.* 2025. Vol. 18, № 1. P. 7–16.
6. Adaptación cultural de una terapia de estimulación cognitiva / N. Medel et al. *Vertex*. 2025. Vol. 35. P. 47–55.
7. Alzheimer's disease / P. Scheltens et al. *Lancet*. 2021. Vol. 397, № 10284. P. 1577–1590.
8. Khan S., Barve K. H., Kumar M. S. Recent Advancements in Pathogenesis, Diagnostics and Treatment of Alzheimer's Disease. *Curr Neuropharmacol*. 2020. Vol. 18, № 11. P. 1106–1125.
9. What Is Alzheimer's Disease? URL: https://www.nia.nih.gov/health/alzheimers-and-dementia/what-alzheimers-disease (Date of access: 10.01.2025).
10. Athar T., Al Balushi K., Khan S. A. Recent advances on drug development and emerging therapeutic agents for Alzheimer’s disease. *Mol Biol Rep.* 2021. Vol. 48, № 7. P. 5629–5645.
11. Share of the population with neurodegenerative diseases. *Our World in Data.* URL: https://ourworldindata.org/grapher/alzheimers-parkinsons-prevalence?time=latest (Date of access: 06.01.2025).

12. Alzheimer's Disease Genetics Fact Sheet. *National Institute on Aging*. URL: https://www.nia.nih.gov/health/genetics-and-family-history/alzheimers-disease-genetics-fact-sheet#three (Date of access: 12.01.2025).
13. Posterior cortical atrophy / S. J. Crutch et al. *The Lancet Neurology*. 2012. Vol. 11, № 2. P. 170–178.
14. Mendez M. F., Ghajarania M. Posterior cortical atrophy: A rare visual variant of Alzheimer disease. *Neurologist.* 2001. Vol. 7, № 3. P. 145–149
15. Classification of primary progressive aphasia and its variants / M. L. Gorno-Tempini. *Neurology*. 2011. Vol. 76, № 11. P. 1006–1014.
16. Mesulam M. M. Primary progressive aphasia. *Annals of Neurology*. 2001. Vol. 49, № 4. P. 425–432.
17. Atrophy patterns in early clinical stages across distinct phenotypes of Alzheimer's disease / R. Ossenkoppele et al. *Human Brain Mapping*. 2015. Vol. 36, № 11. P. 4421–4437.
18. Alzheimer’s Disease: Treatment Strategies and Their Limitations / E. Passeri et al. *International Journal of Molecular Sciences*. 2022. Vol. 23, № 22. P. 13954.
19. Alzheimer's association report. 2023 Alzheimer's disease facts and figures. *Alzheimer's & Dementia*. 2023. Vol. 19, № 4. P. 1598-1695.
20. Mahley R. W., Huang Y. Apolipoprotein E sets the stage: response to injury triggers neuropathology. *Neuron*. 2012. Vol. 76, № 5. P. 871–885.
21. Alzheimer's Disease in People With Down Syndrome. *National Institute on Aging*. URL: https://www.nia.nih.gov/health/alzheimers-causes-and-risk-factors/alzheimers-disease-people-down-syndrome (Date of access: 12.01.2025).
22. A genetic cause of Alzheimer’s disease: mechanistic insights from Down syndrome / F. K. Wiseman et al. *Nature Reviews Neuroscience*. 2015. Vol. 16, № 9. P. 564–574.
23. Bertram L., Tanzi R. E. The genetic epidemiology of neurodegenerative disease. *Journal of Clinical Investigation*. 2005. Vol. 115, № 6. P. 1449–1457.

24. APOE genotype and neuroimaging markers of Alzheimer's disease: systematic review and meta-analysis / Y. Liu et al. *Journal of Neurology, Neurosurgery & Psychiatry*. 2014. Vol. 86, № 2. P. 127–134.
25. Wang Q., Xie C. Microglia activation linking amyloid-β drive tau spatial propagation in Alzheimer's disease. *Frontiers in Neuroscience*. 2022. Vol. 16. P. 1128.
26. 2024 Alzheimer's disease facts and figures. *Alzheimer's Dementia*. 2024. Vol. 20, № 5. P. 3708–3821.
27. Zhang N., Gordon M. L. Clinical efficacy and safety of donepezil in the treatment of Alzheimer's disease in Chinese patients. *Clin Interv Aging*. 2018. Vol. 13. P. 1963–1970.
28. Balázs N., Bereczki D., Kovács T. Cholinesterase inhibitors and memantine for the treatment of Alzheimer and non-Alzheimer dementias. *Ideggyogy Sz*. 2021. Vol. 74, № 11–12. P. 379–387.
29. Lecanemab in Early Alzheimer's Disease / C. H. Dyck et al. *N Engl J Med*. 2023. Vol. 388, № 1. P. 9–21.
30. Donanemab in Early Symptomatic Alzheimer Disease: The TRAILBLAZER-ALZ 2 Randomized Clinical Trial / J. R. Sims et al. *JAMA*. 2023. Vol. 330, № 6. P. 512–527.
31. Coumarin linked heterocyclic hybrids: A promising approach to develop multi target drugs for Alzheimer's disease / A. Husain et al. *Journal of Molecular Structure*. 2021. Vol. 1241. P. 130618.
32. Benek O., Korabecny J., Soukup O. A Perspective on Multi-target Drugs for Alzheimer’s Disease. *Trends in Pharmacological Sciences*. 2020. Vol. 41, № 7. P. 434–445.
33. Cho S., Kim S.-H., Shin D. Recent applications of hydantoin and thiohydantoin in medicinal chemistry. *European Journal of Medicinal Chemistry*. 2019. Vol. 164. P. 517–545.

34. Novel derivatives of spirohydantoin induce growth inhibition followed by apoptosis in leukemia cells / C. V. Kavitha et al. *Biochemical Pharmacology*. 2009. Vol. 77, № 3. P. 348–363.
35. Suzen S., Buyukbingol E. Anti-cancer activity studies of indolalthiohydantoin (PIT) on certain cancer cell lines. *Il Farmaco*. 2000. Vol. 55, № 4. P. 246–248.
36. 5-Benzylidene-hydantoins as new EGFR inhibitors with antiproliferative activity / C. Carmi et al. *Bioorganic & Medicinal Chemistry Letters*. 2006. Vol. 16, № 15. P. 4021–4025.
37.
38. Synthesis, biological evaluation and molecular modeling study of novel tacrine–carbazole hybrids as potential multifunctional agents for the treatment of Alzheimer's disease / S. Thiratmatrakul et al. *European Journal of Medicinal Chemistry*. 2014. Vol. 75. P. 21–30.
39. Anti-tumor and anti-angiogenic activity of novel hydantoin derivatives: Inhibition of VEGF secretion in liver metastatic osteosarcoma cells / Basappa et al. *Bioorganic & Medicinal Chemistry*. 2009. Vol. 17, № 14. P. 4928–4934.
40. Structure−Activity Relationship for Thiohydantoin Androgen Receptor Antagonists for Castration-Resistant Prostate Cancer (CRPC) / M. E. Jung et al. *Journal of Medicinal Chemistry*. 2010. Vol. 53, № 7. P. 2779–2796.
41. Identification of MK-5710 ((8aS)-8a-methyl-1,3-dioxo-2-[(1S,2R)-2-phenylcyclopropyl]-N-(1-phenyl-1H-pyrazol-5-yl)hexahydroimid azo[1,5-a]pyrazine-7(1H)-carboxamide), a potent smoothened antagonist for use in Hedgehog pathway dependent malignancies. Part 1 / S. Malancona et al. *Bioorganic & Medicinal Chemistry Letters*. 2011. Vol. 21, № 15. P. 4422–4428.
42. Synthesis of amidine and amide derivatives and their evaluation for anti-inflammatory and analgesic activities / S. M. Sondhi et al. *European Journal of Medicinal Chemistry*. 2009. Vol. 44, № 3. P. 1010–1015.

43. Anti-inflammatory and antinociceptive activities of indole–imidazolidine derivatives / A. S. H. da Silva Guerra et al. *International Immunopharmacology*. 2011. Vol. 11, № 11. P. 1816–1822.

44. Synthesis of hydantoin analogues of (2S,3R,4S)-4-hydroxyisoleucine with insulinotropic properties / D. Sergent et al. *Bioorganic & Medicinal Chemistry Letters*. 2008. Vol. 18, № 15. P. 4332–4335.

45. Amine–alkyl derivatives of hydantoin: New tool to combat resistant bacteria / J. Handzlik et al. *European Journal of Medicinal Chemistry*. 2011. Vol. 46, № 12. P. 5807–5816.

46. Antibacterial Activity of 5-Dialkylaminomethylhydantoins and Related Compounds / F. Fujisaki et al. *Chemical & Pharmaceutical Bulletin*. 2010. Vol. 58, № 8. P. 1123–1126.

47. The discovery of long-acting saligenin β2 adrenergic receptor agonists incorporating hydantoin or uracil rings / P. A. Procopiou et al. *Bioorganic & Medicinal Chemistry*. 2011. Vol. 19, № 14. P. 4192–4201.

48. Pharmacophore models based studies on the affinity and selectivity toward 5-HT1A with reference to α1-adrenergic receptors among arylpiperazine derivatives of phenytoin / J. Handzlik et al. *Bioorganic & Medicinal Chemistry*. 2011. Vol. 19, № 3. P. 1349–1360.

49. Synthesis and potential anticonvulsant activity of new N-3-substituted 5,5-cyclopropanespirohydantoins / Q. Zhu et al. *European Journal of Medicinal Chemistry*. 2009. Vol. 44, № 1. P. 296–302.

50. Synthesis and Anticonvulsant Activity of New N-Mannich Bases Derived from 5-Cyclopropyl-5-phenyl-hydantoins / H. Byrtus et al. *Archiv der Pharmazie*. 2010. Vol. 344, № 4. P. 231–241.

51. Discovery of an Orally Active Non-Peptide Fibrinogen Receptor Antagonist Based on the Hydantoin Scaffold / H. U. Stilz et al. *Journal of Medicinal Chemistry*. 2001. Vol. 44, № 8. P. 1158–1176.

52. Design, synthesis, and SAR of heterocycle-containing antagonists of the human CCR5 receptor for the treatment of HIV-1 infection / D. Kim et al.

Bioorganic & Medicinal Chemistry Letters. 2001. Vol. 11, № 24. P. 3099-3102.

53. Inhibitors of HCV NS5A: From Iminothiazolidinones to Symmetrical Stilbenes / J. L. Romine et al. *ACS Medicinal Chemistry Letters*. 2011. Vol. 2, № 3. P. 224–229.
54. Binding Site Elucidation of Hydantoin-Based Antagonists of LFA-1 Using Multidisciplinary Technologies: Evidence for the Allosteric Inhibition of a Protein−Protein Interaction / K. Last-Barney et al. *Journal of the American Chemical Society*. 2001. Vol. 123, № 24. P. 5643–5650.
55. Phenytoin – An anti-seizure drug: Overview of its chemistry, pharmacology and toxicology / J. Patocka et al. *Food Chem Toxicol*. 2020. Vol. 142. P. 111393.
56. Nitrofurantoin: properties and potential in treatment of urinary tract infection: a narrative review / M. Mahdizade Ari et al. *Front Cell Infect Microbiol*. 2023. Vol. 13. P. 1148603.
57. Habib M. M. W., Abdelfattah M. A. O., Abadi A. H. Design and Synthesis of Novel Phenylpiperazine Derivatives as Potential Anticonvulsant Agents. *Archiv der Pharmazie*. 2015. Vol. 348, № 12. P. 868–874.
58. Synthesis and anticonvulsant activity of new N-Mannich bases derived from 5-cyclopropyl-5-phenyl- and 5-cyclopropyl-5-(4-chlorophenyl)imidazolidine-2,4-diones / H. Byrtus et al. *Bioorganic & Medicinal Chemistry*. 2011. Vol. 19, № 20. P. 6149–6156.
59. Design, synthesis, anticonvulsant, and antiarrhythmic properties of novel N-Mannich base and amide derivatives of β-tetralinohydantoin / A. Czopek et al. *Pharmacological Reports*. 2016. Vol. 68, № 5. P. 886–893.
60. Synthesis, anticonvulsant, antinociceptive activity and preliminary safety evaluations of novel hybrid imidazolidine-2,4-dione derivatives with morpholine moiety / A. Czopek et al. *ChemMedChem*. 2025. Vol. 20, № 7. P. 202400612.

61. SAR-studies on the importance of aromatic ring topologies in search for selective 5-HT7 receptor ligands among phenylpiperazine hydantoin derivatives / J. Handzlik et al. *European Journal of Medicinal Chemistry*. 2014. Vol. 78. P. 324–339.
62. Jia L., Gao H. Machine dearning for prediction. *Artificial intelligence in drug design*. 2021. Vol. 2390. P. 447–460.
63. Daina A., Michielin O., Zoete V. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. *Scientific Reports.* 2017. Vol. 7. P. 42717.
64. Open Babel: An open chemical toolbox / N. M. O'Boyle et al. *Journal of Cheminformatics*. 2011. Vol. 3, № 1. P. 33.
65. Ertl P., Rohde B., Selzer P. Fast Calculation of Molecular Polar Surface Area as a Sum of Fragment-Based Contributions and Its Application to the Prediction of Drug Transport Properties. *Journal of Medicinal Chemistry*. 2000. Vol. 43, № 20. P. 3714–3717.
66. Lovering F., Bikker J., Humblet C. Escape from Flatland: Increasing Saturation as an Approach to Improving Clinical Success. *Journal of Medicinal Chemistry*. 2009. Vol. 52, № 21. P. 6752–6756.
67. Simple Method of Calculating Octanol/Water Partition Coefficient. / I. Moriguchi et al. *Chemical & Pharmaceutical Bulletin*. 1992. Vol. 40, № 1. P. 127–130.
68. Computation of Octanol−Water Partition Coefficients by Guiding an Additive Model with Knowledge / T. Cheng et al. *Journal of Chemical Information and Modeling*. 2007. Vol. 47, № 6. P. 2140–2148.
69. Montanari F., Ecker G. F. Prediction of drug–ABC-transporter interaction – Recent advances and future challenges. *Advanced Drug Delivery Reviews*. 2015. Vol. 86. P. 17–26.
70. The role of ABC transporters in drug absorption, distribution, metabolism, excretion and toxicity (ADME–Tox) / G. Szakács et al. *Drug Discovery Today*. 2008. Vol. 13, № 9-10. P. 379–393.

71. Testa B., Krämer S. The Biochemistry of Drug Metabolism - An Introduction. *Chemistry & Biodiversity*. 2008. Vol. 5, № 11. P. 2171–2336.

72. Wolf C. R. Science, medicine, and the future: Pharmacogenetics. *BMJ*. 2000. Vol. 320, № 7240. P. 987–990.

73. Di L. The role of drug metabolizing enzymes in clearance. *Expert Opinion on Drug Metabolism & Toxicology*. 2014. Vol. 10, № 3. P. 379–393.

74. Potts R. O. Guy R. H. Predicting Skin Permeability. *Pharmaceutical Research*. 1992. Vol. 9. P. 663–669.

75. Kola I., Landis J. Can the pharmaceutical industry reduce attrition rates?. *Nature Reviews Drug Discovery*. 2004. Vol. 3, № 8. P. 711–716.

76. Large-scale prediction and testing of drug activity on side-effect targets / E. Lounkine et al. *Nature*. 2012. Vol. 486, № 7403. P. 361–367.

77. Ashburn T. T., Thor K. B. Drug repositioning: identifying and developing new uses for existing drugs. *Nature Reviews Drug Discovery*. 2004. Vol. 3, № 8. P. 673–683.

78. Predicting new molecular targets for known drugs / M. J. Keiser et al. *Nature*. 2009. Vol. 462, № 7270. P. 175–181.

79. Tools for in silico target fishing / A. Cereto-Massagué et al. *Methods*. 2015. Vol. 71. P. 98–103.

80. Similarity-based machine learning methods for predicting drug–target interactions: a brief review / H. Ding et al. *Briefings in Bioinformatics*. 2013. Vol. 15, № 5. P. 734–747.

81. Willett P., Winterman V. A Comparison of Some Measures for the Determination of Inter-Molecular Structural Similarity Measures of Inter-Molecular Structural Similarity. *Quantitative Structure-Activity Relationships*. 1986. Vol. 5, № 1. P. 18–25.

82. Johnson M., Lajiness M., Maggiora G. Molecular similarity: a basis for designing drug screening programs. *Prog. Clin. Biol. Res.* 1989. Vol. 291. P. 167–171.

83. A Structural Motif of Acetylcholinesterase That Promotes Amyloid β-Peptide Fibril Formation† / G. V. De Ferrari et al. *Biochemistry*. 2001. Vol. 40, № 35. P. 10447–10457.

84. Selective butyrylcholinesterase inhibition elevates brain acetylcholine, augments learning and lowers Alzheimer -amyloid peptide in rodent / N. H. Greig et al. *Proceedings of the National Academy of Sciences*. 2005. Vol. 102, № 47. P. 17213–17218.

85. Multitargeted drugs discovery: Balancing anti-amyloid and anticholinesterase capacity in a single chemical entity / M. L. Bolognesi et al. *Bioorganic & Medicinal Chemistry Letters.* 2011. Vol. 21, № 9. P. 2655–2658.

86. Kinetic and structural studies on the interactions of Torpedo californica acetylcholinesterase with two donepezil-like rigid analogues / R. Caliandro et al. *Journal of Enzyme Inhibition and Medicinal Chemistry*. 2018. Vol. 33, № 1. P. 794–803.

87. Structural basis for achieving GSK-3β inhibition with high potency, selectivity, and brain exposure for positron emission tomography imaging and drug discovery / V. Bernard-Gauthier et al. *Journal of Medicinal Chemistry*. 2019. Vol. 62(21). P. 9600-9617.

APPLICATIONS

THEORETICAL STUDIES ON THE ANTIOXIDANT ACTIVITY OF ISOVITEXIN WITH THE XANTHINE OXIDASE ENZYME

Maslov O.Yu., Antonenko O.V., Koval A.O.
Scientific supervisor: Kolisnyk S.V.
National University of Pharmacy, Kharkiv, Ukraine
alexmaslov392@gmail.com

Introduction. Xanthine oxidase (XOD) are single-gene products that exist in separate but interconvertible forms. XOD utilizes hypoxanthine or xanthine as a substrate and O2 as a cofactor to produce superoxide and uric acid. XOD is reported to play an important role in cellular oxidative status, detoxification of aldehydes, oxidative injury in ischemia-reperfusion, and neutrophil mediation. XOD may serve as a messenger or mediator in the activation of neutrophil, T cell, cytokines, or transcription in defense mechanisms rather than as a free radical generator of tissue damage.

Aim. To perform molecular docking of isovitexin with the xanthine oxidase enzyme.

Material and methods. A molecular docking study was conducted using the tool known as AutoDockTools 1.5.6. Genetic algorithm parameters were applied for ligand interaction, with 10 runs of this criterion. XOD (PDB ID: 1fiq) structure was obtained from PDB database. The resolution of 1svc was 2.5 Å. The ligand structures of isovitexin (CID_162350) was obtained from PubChem database. The active site of the docking protein was identified utilizing the Computed Atlas for Surface Topography of Proteins. As a standard was taken diclofenac sodium. We applied the following classification of selectivity: inhibition concentration (IC)50<0.001 mM (high selective); 0.05>IC50>0.01 (medium selective); IC50>0.05 mM (low selective).

Results and discussion. The isovitexin had a high value of free energy value (- 11.31 kcal/mol), whereas IC50 was 0.00000515 mmol, so isovitexin belong to high selective inhibitor. Comparing result with diclofenac sodium standard, the affinity of isovitexin was 62% more than of diclofenac sodium (-4.17 kcal/mol, IC50 – 0.88 mmol).

Conclusions. It was established that isovitexin is a potentially medium selective inhibitor of xanthine oxidase. So, the extract with isovitexin can be applied for developing a new antioxidant drug for preventing oxidative stress.

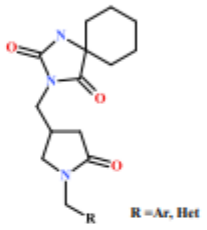
DETERMINATION OF ADMET PARAMETERS OF NEW 3-[1-R-5-OXO-PYRROLIDIN-3-YL]METHYL]-1,3-DIAZASPIRO[4.5]DECANE-2,4-DIONE DERIVATIVES

Elkahya M., [1]Saifudinova R.P.
Scientific supervisor: Severina H.I.
National University of Pharmacy, Kharkiv, Ukraine
[1]Cherkasy Medical Academy, Cherkasy, Ukraine
elkahyameriem@gmail.com

Introduction. Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by irreversible loss of cognitive functions and memory, representing a significant medical and social challenge in the context of an aging population. Current treatment methods for AD are mostly symptomatic and unable to halt or significantly slow the progression of the disease. The pathogenesis of AD is complex and multifactorial, involving the accumulation of amyloid

plaques, neurofibrillary tangles, neuronal inflammation, oxidative stress, and cholinergic dysfunction. Given the increasing prevalence of AD and the lack of effective treatments, there is an urgent need for fundamental and translational research aimed at developing new therapeutic strategies. The critical importance of identifying innovative approaches that target key pathogenic mechanisms of AD is evident, in order to develop disease-modifying therapies capable of improving patients' quality of life and reducing the societal burden of the disease.

Based on the hybrid-pharmacophore concept, we have designed new 3-[1-R-5-oxo-pyrrolidin-3-yl]methyl]-1,3-diazaspiro[4.5]decane-2,4-dione (1) derivatives as potential agents for the treatment of Alzheimer's disease, exhibiting anticholinesterase activity and antagonism towards NMDA receptors.



1

Aim. Calculation of ADMET parameters for the generated structures in the series of derivatives of 3-[1-R-5-oxo-pyrrolidin-3-yl]methyl]-1,3-diazaspiro[4.5]decane-2,4-dione as potential biologically active substances.

Materials and methods. The program BIOVIA Draw 2021 was used for compound construction. SwissADME Webtool was applied to calculate ADMET parameters.

Results and discussion. SwissADME is a free web-based tool developed by the Swiss Institute of Bioinformatics that allows you to evaluate pharmacokinetic properties, drug-likeness, and molecular descriptors of small molecules. A comprehensive in silico analysis of ADMET parameters – encompassing absorption, distribution, metabolism, excretion, and toxicity – was conducted to evaluate the pharmacokinetic behavior and drug-likeness of the synthesized 3-[1-R-5-oxo-pyrrolidin-3-yl]methyl]-1,3-diazaspiro[4.5]decane-2,4-dione. These characteristics are crucial for predicting the efficacy and safety of drug candidates, particularly at early stages of drug discovery.

To assess oral bioavailability, the bioavailability radar was employed, providing a visual representation of six key physicochemical properties: lipophilicity, molecular size, polarity, water solubility, molecular flexibility, and saturation. Notably, all investigated compounds fell within the optimal region of the radar, suggesting favorable profiles for oral administration.

The lipophilicity of each molecule was estimated using five different logP calculation models – iLOGP, XLOGP3, WLOGP, MLOGP, and SILICOS-IT – with results confirming compliance with the recommended range for compounds targeting the central nervous system (CNS). Alongside this, aqueous solubility was predicted, indicating acceptable levels for potential bioavailability.

Further pharmacokinetic predictions included: passive gastrointestinal absorption: high for all tested structures; blood–brain barrier (BBB) permeability: the majority of compounds demonstrated a capacity to cross the BBB, a critical parameter for CNS-active agents; P-glycoprotein substrate status: investigated to estimate potential efflux and bioavailability constraints; metabolism via cytochrome P450 (CYP450) enzymes: predictions suggested that the compounds are likely to undergo phase I biotransformation, with minimal risk of CYP-related interactions.

Ф А 2.2.1–32-366

National University of Pharmacy

Faculty for foreign citizens' education
Department pharmaceutical chemistry
Level of higher education master
Specialty 226 Pharmacy, industrial pharmacy
Educational and professional program Pharmacy

APPROVED
The Head of
Department of
pharmaceutical
chemistry

Victoriya GEORGIYANTS
" 3 " ____September___ 2024 year

ASSIGNMENT
FOR QUALIFICATION WORK OF
AN APPLICANT FOR HIGHER EDUCATION

Meriem EL KAHYA

1. Topic of qualification work: «Determination of the prospects of new imidazolidine-2,4-dione derivatives for the treatment of Alzheimer's disease », supervisor of qualification work: Hanna Severina, DSc, professor

approved by order of NUPh from "27th" of September 2024 № 237

2. Deadline for submission of qualification work by the applicant for higher education: May 2025

3. Outgoing data for qualification work: The study investigates imidazolidine-2,4-dione derivatives as potential multitarget ligands for the treatment of neurodegenerative diseases. ADMET analysis and molecular docking modelling with AChE and GSK3 were performed.

4. Contents of the settlement and explanatory note (list of questions that need to be developed): to analyze current literature data on the pathogenesis and и pharmacotherapy of Alzheimer's disease; to select virtually generated candidate structures for further study based on logical-structural analysis, identifying the most promising agents for Alzheimer's disease treatment; to perform ADMET profiling of the generated candidate compound library and assess the suitability of these structures for further development as potential drug candidates; to carry out virtual target prediction for the generated compounds using the SwissTargetPrediction; to identify the most relevant biological targets for in silico studies based on the prediction results and the therapeutic importance of the targets; to conduct molecular docking of the selected imidazolidine-2,4-dione derivatives with the chosen biological targets; to formulate conclusions regarding the possible mechanism of action of the candidate structures.
5. List of graphic material (with exact indication of the required drawings): 17 figures, 3 tables

6. Consultants of chapters of qualification work

Chapter	Name, SURNAME, position of consultant	Signature, date	
		assignment was issued	assignment was received
1	Hanna SEVERINA, professor of higher education institution of the Department of pharmaceutical chemistry	04.09.2024	04.09.2024
2	Hanna SEVERINA, professor of higher education institution of the Department of pharmaceutical chemistry	25.10.2024	25.10.2024
3	Hanna SEVERINA, professor of higher education institution of the Department of pharmaceutical chemistry	25.12.2024	25.12.2024
4	Hanna SEVERINA, professor of higher education institution of the Department of pharmaceutical chemistry	29.01.2025	29.02.2025

7. Date of issue of the assignment: "_3st_" of September 2024

CALENDAR PLAN

№	Name of stages of qualification work	Deadline for the stages of qualification work	Notes
1.	Neurodegeneration and Alzheimer's disease. Imidazolidine-2,4-dione derivatives as biologically active substances (Literature review)	**September-November 2024**	**done**
2.	Determination of drug-like parameters and prediction of targeted effects of imidazolidine-2,4-dione derivatives	**October- December 2024**	**done**
3.	Prediction of the affinity of imidazolidine-2,4-dione derivatives for targets regulating neurodegeneration	**January 2025– February 2025**	**done**
4.	Preparation of qualification work and submission to the Examination Commission	**February-April 2025**	**done**

An applicant of higher education _____ Meriem EL KAHYA

Supervisor of qualification work _____ Hanna SEVERINA

ВИТЯГ З НАКАЗУ № 237

По Національному фармацевтичному університету

від 27 вересня 2024 року

Затвердити теми кваліфікаційних робіт здобувачам вищої освіти 5-го курсу Фм20(4.10д) 2024-2025 навчального року, освітньо-професійної програми – Фармація, другого (магістерського) рівня вищої освіти, спеціальності 226 – Фармація, промислова фармація, галузь знань 22 Охорона здоров'я, денна форма здобуття освіти (термін навчання 4 роки 10 місяців), які навчаються за контрактом (мова навчання англійська та українська) згідно з додатком № 1.

Прізвище, ім'я здобувача вищої освіти	Тема кваліфікаційної роботи		Посада, прізвище та ініціали керівника	Рецензент кваліфікаційної роботи
• **по кафедрі фармацевтичної хімії**				
Ель Ках'я Меріем	Визначення перспективності нових похідних імідазолідин-2,4-діону для лікування хвороби Альцгеймера	Determination of the prospects of new imidazolidine-2,4-dione derivatives for the treatment of Alzheimer's disease	д.ф.н., проф. Северіна Г.І.	проф. Подольський І.М.

Ректор

Вірно. Секретар

ФА2.8-03-317

ВИСНОВОК

експертної комісії про проведену експертизу
щодо академічного плагіату у кваліфікаційній роботі

здобувача вищої освіти

«06» травня 2025 р. № 331123371

Проаналізувавши кваліфікаційну роботу здобувача вищої освіти Ель Ках'я Меріем, групи Фм20(4,10д) англ 01, спеціальності 226 Фармація, промислова фармація, освітньої програми «Фармація» навчання на тему: «Визначення перспективності нових похідних імідазолідин-2,4-діону для лікування хвороби Альцгеймера / Determination of the prospects of new imidazolidine-2,4-dione derivatives for the treatment of Alzheimer's disease», експертна комісія дійшла висновку, що робота, представлена до Екзаменаційної комісії для захисту, виконана самостійно і не містить елементів академічного плагіату (компіляції).

Голова комісії,
проректор ЗВО з НПР,
професор **Інна ВЛАДИМИРОВА**

Ф А 2.2.1-32-353

REVIEW

of scientific supervisor for the qualification work of the master's level of higher education of the specialty 226 Pharmacy, industrial pharmacy

Meriem EL KAHYA

on the topic: «Determination of the prospects of new imidazolidine-2,4-dione derivatives for the treatment of Alzheimer's disease »

Relevance of the topic. The topic of the master's thesis, “Determination of the Prospects of New Imidazolidine-2,4-dione Derivatives for the Treatment of Alzheimer’s Disease,” is highly relevant in the context of contemporary pharmaceutical science. Alzheimer's disease remains one of the most urgent global health challenges, and the need for new multitarget therapeutic agents is of great importance. The choice of imidazolidine-2,4-dione derivatives as the research focus is scientifically justified, given their proven neuroactive potential and pharmacophoric value. The work addresses both fundamental and applied aspects of medicinal chemistry and pharmacology, making it pertinent for ongoing drug discovery initiatives.

Practical value of conclusions, recommendations and their validity. The study provides practically significant results based on comprehensive in silico methods, including ADMET analysis, virtual target prediction, and molecular docking. The identification of compound 3426 as a promising multitarget ligand with high blood–brain barrier permeability and affinity toward neurodegeneration-related targets (such as AChE and GSK3) offers a rational basis for its further experimental investigation. The recommendations are grounded in well-validated computational tools and reflect a high degree of predictive reliability. These findings may serve as a foundation for future synthesis and biological evaluation of novel agents for Alzheimer’s disease treatment.

Assessment of work. The thesis is well-structured, logically coherent, and demonstrates a high theoretical level. The author has shown a solid understanding of neuropharmacology, modern trends in CNS drug development, and the methodology of in silico modeling. The literature review is comprehensive and up to date, the research objectives are clearly defined, and the methodology is adequately selected. The graphical and tabular presentation of results is illustrative and supports the conclusions. The scientific language and academic style meet the requirements for a master's thesis. The student has demonstrated independence, diligence, and analytical thinking throughout the research process.

General conclusion and recommendations on admission to defend. The qualification work of Meriem EL KAHYA is performed at a high level with scientific novelty and practical significance of the results obtained. In terms of relevance, level of implementation and validity of conclusions, the work meets the requirements for graduate qualification works and can be submitted for defense in the Examination Commission.

Scientific supervisor _____ Hanna SEVERINA
«13 » May 2025 year

Ф А 2.2.1-32-356

REVIEW

for qualification work of the master's level of higher education, specialty 226 Pharmacy, industrial pharmacy

Meriem EL KAHYA

on the topic: «Determination of the prospects of new imidazolidine-2,4-dione derivatives for the treatment of Alzheimer's disease »

Relevance of the topic. The selected topic is highly relevant, given the global prevalence of neurodegenerative diseases, particularly Alzheimer's disease, and the lack of effective curative treatments. The search for novel multifunctional compounds capable of modulating key pathogenic mechanisms remains a top priority in modern pharmaceutical science. The focus on imidazolidine-2,4-dione derivatives is well-founded, as these structures have demonstrated neuroactive potential in literature.

Theoretical level of work. The thesis demonstrates a high level of theoretical preparation. The author has thoroughly analyzed scientific sources related to the pathogenesis, therapy, and modern strategies for drug development against Alzheimer's disease. The literature review is well-structured, comprehensive, and up to date, covering both clinical aspects and molecular targets of neurodegeneration. The reasoning is logical, the arguments are well-developed, and the scientific language is appropriate.

Author's suggestions on the research topic. The author proposed a rational hybrid design strategy for new imidazolidine-2,4-dione derivatives, applying pharmacophore combination principles. In silico modeling was performed, including ADMET analysis, target prediction, and molecular docking, to evaluate the binding affinity of the compounds toward AChE and GSK3. Compound 3426 was identified as a promising candidate, which reflects the scientific novelty and practical direction of the research.

Practical value of conclusions, recommendations and their validity. The practical value of the results obtained lies in the substantiation of further research directions for new neuroprotective compounds. The recommendation to proceed with the synthesis and experimental verification of compound 3426 is logical, convincing, and supported by reliable in silico tools. The conclusions are fully consistent with the objectives and are backed by the presented data.

Disadvantages of work. The thesis contains minor drawbacks related to stylistic inconsistencies and typographical errors. Occasional misuse of articles and syntactic structures is noted in the English-language sections. However, these issues do not significantly affect the overall scientific value of the work.

General conclusion and assessment of the work. The qualification work of Meriem EL KAHYA in terms of relevance, scientific novelty of the obtained results, methodological level, theoretical and practical significance, volume of performed research meets the requirements of the Regulation on the Procedure for the Preparation and Defence of Qualification Works at the National Pharmaceutical University and can be recommended for defence at the Examination Commission.

Reviewer _____ prof. Illya PODOLSKY

«15» May 2025 year

Ф А2.2.1-91-287

ВИТЯГ

з протоколу засідання кафедри фармацевтичної хімії

№ 14 від 16 травня 2025 р.

Засідання проводилось з використанням ZOOM технологій з 12 год. 05 хв. по 12 год. 50 хв.
Чисельний склад кафедри: 16 науково педагогічних працівників, з них присутні – 16 осіб.
ПРИСУТНІ: зав.каф. проф. Георгіянц В.А., професори: Баюрка С.В., Перехода Л.О., Северіна Г.І., Сидоренко Л.В., доценти: Амжад Абу Шарк І., Бевз Н.Ю., Віслоус О.О., Головченко О. С., Гриненко В.В., Кобзар Н.П., Михайленко О.О., Петрушова Л.О., Рахімова М.В., Яременко В.Д., ас. Григорів Г.В.; аспіранти: Асмолов В. Є., Гончар О.О., Гуріна В. О., Коптєлов А. С., Куцанян А. А., Мураль Д. В., Сайфудінова Р. П., Сулейман Р. М., Суржиков І.О.
ПОРЯДОК ДЕННИЙ:
Звіт про стан виконання кваліфікаційної роботи здобувача вищої освіти фармацевтичного факультету, Фм20(4,10д) англ 01 групи, спеціальності «226 Фармація, промислова фармація», освітньої програми «Фармація» Меріем Ель КАХ'Я на тему: «Визначення перспективності нових похідних імідазолідин-2,4-діону для лікування хвороби Альцгеймера».
СЛУХАЛИ: доповідь здобувача вищої освіти здобувача вищої освіти фармацевтичного факультету, Фм20(4,10д) англ 01 групи, спеціальності «226 Фармація, промислова фармація», освітньої програми «Фармація» Меріем ЕЛЬ КАХ'Я на тему: «Визначення перспективності нових похідних імідазолідин-2,4-діону для лікування хвороби Альцгеймера», керівник – професор кафедри фармацевтичної хімії, д.фарм.н., проф. Ганна СЕВЕРІНА.
УХВАЛИЛИ: рекомендувати кваліфікаційну роботу Меріем ЕЛЬ КАХ'Я до офіційного захисту в Екзаменаційній комісії.

Голова

зав. кафедри, доктор фарм. наук,

професор **Вікторія ГЕОРГІЯНЦ**

Секретар

доцент, канд. фарм. наук **Марина РАХІМОВА**

Ф А2.2.1-32-042

НАЦІОНАЛЬНИЙ ФАРМАЦЕВТИЧНИЙ УНІВЕРСИТЕТ

ПОДАННЯ
ГОЛОВІ ЕКЗАМЕНАЦІЙНОЇ КОМІСІЇ
ЩОДО ЗАХИСТУ КВАЛІФІКАЦІЙНОЇ РОБОТИ

Направляється здобувач вищої освіти Меріем Ель Ках'я до захисту кваліфікаційної роботи
за галуззю знань 22 Охорона здоров'я
спеціальністю 226 Фармація, промислова фармація
освітньо-професійною програмою Фармація

на тему: «Визначення перспективності нових похідних імідазолідин-2,4-діону для лікування хвороби Альцгеймера»

Кваліфікаційна робота і рецензія додаються.

Декан факультету _____ / Микола ГОЛІК /

Висновок керівника кваліфікаційної роботи

Здобувачка вищої освіти Меріем Ель Ках'я виконала роботу на сучасному рівні. За період виконання кваліфікаційної роботи проявила високий рівень теоретичної підготовки. Кваліфікаційна робота викладена послідовно, грамотно, висновки коректні та логічні, витікають зі змісту роботи. Кваліфікаційна робота Меріем Ель Ках'я може бути рекомендована до захисту в Екзаменаційній комісії.

Керівник кваліфікаційної роботи

_____ Ганна СЕВЕРІНА

«13» травня 2024 р.

Висновок кафедри про кваліфікаційну роботу

Кваліфікаційну роботу розглянуто. Здобувачка вищої освіти Меріем Ель Ках'я допускається до захисту даної кваліфікаційної роботи в Екзаменаційній комісії.

Завідувачка кафедри
фармацевтичної хімії

_____ Вікторія ГЕОРГІЯНЦ

«16» травня 2025 року

Qualification work was defended

of Examination commission on

« __ » __June__ 2025 year

With the grade _____

Head of the State Examination commission,

DPharmSc, Professor

_____ / Volodymyr YAKOVENKO /