Structure-Based Virtual Screening, Molecular Docking, and ADMET Evaluation of Type-II Kinase Inhibitors Targeting BCR-ABL Tyrosine Kinase Using the Crystal Structure 1IEP

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Abstract

Chronic myeloid leukemia (CML) is a myeloproliferative malignancy characterized by the presence of the Philadelphia chromosome, which results in the formation of the constitutively active BCR-ABL fusion tyrosine kinase. The abnormal kinase activity of BCR-ABL drives uncontrolled cellular proliferation and inhibits apoptosis, making it one of the most important molecular targets in cancer therapy. Although tyrosine kinase inhibitors such as imatinib have significantly improved patient outcomes, the emergence of resistance mutations and treatment-associated limitations continues to drive the search for novel inhibitors. Structure-based virtual screening (SBVS) has emerged as a powerful computational approach for identifying potential therapeutic candidates by evaluating the interactions between small molecules and target proteins.

In the present study, a Type-II kinase inhibitor library comprising 2,376 compounds obtained from Asinex was screened against the crystal structure of the ABL kinase domain (PDB ID: 1IEP), which serves as a structural representation of the BCR-ABL target. Molecular docking was performed using AutoDock Vina, and compounds were ranked according to their binding affinities. The top ten compounds exhibiting the lowest binding energies were subjected to detailed interaction analysis using Discovery Studio Visualizer. Among these compounds, Nlig_ligand_2166, Nlig_ligand_2110, and Nlig_ligand_2029 demonstrated favorable binding orientations and hydrogen-bond interactions with key residues located within the active site of the kinase domain. Subsequent pharmacokinetic and drug-likeness evaluation was performed using SwissADME. Comparative ADMET analysis revealed that Nlig_ligand_2110 exhibited the most favorable overall profile, including high gastrointestinal absorption, compliance with Lipinski's Rule of Five, absence of PAINS and Brenk alerts, and acceptable synthetic accessibility.

The integration of molecular docking, interaction analysis, and pharmacokinetic evaluation identified Nlig_ligand_2110 as the most promising lead compound. These findings suggest that the selected compound may serve as a potential candidate for further optimization and experimental validation as a BCR-ABL tyrosine kinase inhibitor.

Keywords: BCR-ABL, chronic myeloid leukemia, tyrosine kinase inhibitor, molecular docking, structure-based virtual screening, AutoDock Vina, ADMET, SwissADME, drug discovery.

1. Introduction

Cancer remains one of the leading causes of mortality worldwide and continues to pose a significant challenge to healthcare systems. The disease is characterized by uncontrolled cellular proliferation resulting from genetic and molecular abnormalities that disrupt normal cellular regulatory mechanisms. Advances in molecular biology have led to the development of targeted therapies that selectively inhibit proteins responsible for tumor progression, offering improved efficacy and reduced toxicity compared with conventional chemotherapy.

Chronic myeloid leukemia (CML) is a myeloproliferative disorder characterized by the excessive proliferation of myeloid cells in the bone marrow and peripheral blood. The disease is strongly associated with the Philadelphia chromosome, a genetic abnormality formed through a reciprocal translocation between chromosomes 9 and 22 [t(9;22)(q34;q11)]. This chromosomal rearrangement results in the fusion of the breakpoint cluster region (BCR) gene with the Abelson tyrosine kinase (ABL1) gene, producing the BCR-ABL fusion protein. The resulting oncoprotein exhibits constitutive tyrosine kinase activity, leading to uncontrolled cell growth and impaired apoptosis.

The BCR-ABL kinase activates several downstream signaling pathways, including PI3K/AKT, JAK/STAT, and RAS/MAPK, which collectively promote cellular proliferation, survival, and leukemic transformation. Because BCR-ABL serves as the primary molecular driver of CML, it has become one of the most important therapeutic targets in cancer research. The development of imatinib mesylate, the first BCR-ABL tyrosine kinase inhibitor (TKI), revolutionized the treatment of CML by specifically targeting the ATP-binding site of the kinase domain. Imatinib demonstrated remarkable clinical success and established targeted kinase inhibition as a highly effective strategy for cancer therapy.

Despite the success of imatinib, resistance remains a significant clinical challenge. Resistance may arise through kinase-domain mutations, gene amplification, altered drug transport, or activation of alternative signaling pathways. Consequently, considerable efforts have been directed toward the identification of new kinase inhibitors capable of overcoming these limitations. The discovery of novel compounds with improved binding affinity, selectivity, and pharmacokinetic properties remains an important objective in anticancer drug development.

Protein kinases represent one of the largest classes of therapeutic targets due to their involvement in regulating cell growth, differentiation, metabolism, and survival. Kinase inhibitors are commonly classified according to their binding mechanisms. Type-I inhibitors bind to the active conformation of the kinase, whereas Type-II inhibitors recognize the inactive DFG-out conformation and occupy both the ATP-binding site and an adjacent hydrophobic pocket. This binding mode often provides enhanced selectivity and reduced off-target activity. Several clinically approved drugs, including imatinib, nilotinib, and sorafenib, belong to the Type-II inhibitor class, highlighting the therapeutic significance of this mechanism.

The application of computational approaches has greatly accelerated the drug discovery process. Structure-based virtual screening (SBVS) enables rapid evaluation of large chemical libraries against a target protein structure, significantly reducing the time and cost associated with experimental screening. Molecular docking, a key component of SBVS, predicts the

orientation and binding affinity of ligands within the active site of a target protein, allowing identification of promising lead compounds. Furthermore, computational prediction of pharmacokinetic properties through ADMET analysis assists in prioritizing compounds with favorable drug-like characteristics before experimental validation.

The crystal structure deposited in the Protein Data Bank under accession code 1IEP represents the ABL kinase domain complexed with imatinib and serves as a validated structural model for studying BCR-ABL inhibition. Because this structure captures the inactive kinase conformation targeted by Type-II inhibitors, it provides an ideal platform for virtual screening of kinase-focused compound libraries.

Therefore, the present study aimed to identify potential BCR-ABL inhibitors through structure-based virtual screening of 2,376 compounds from the Asinex Type-II Kinase Inhibitor Library. Molecular docking was performed using AutoDock Vina against the crystal structure 1IEP, followed by interaction analysis using Discovery Studio Visualizer and pharmacokinetic evaluation using SwissADME. The objective was to identify compounds exhibiting favorable binding interactions and drug-like properties that may serve as potential lead candidates for future development as BCR-ABL tyrosine kinase inhibitors.

2. Materials and Methods

2.1 Retrieval and Preparation of Protein Structure

The three-dimensional crystal structure of the ABL kinase domain complexed with imatinib (PDB ID: 1IEP) was retrieved from the Protein Data Bank (PDB). The 1IEP structure was selected because it represents the inactive conformation of the ABL kinase domain and serves as a validated structural model for studying inhibitors of the BCR-ABL tyrosine kinase. Since imatinib is a clinically approved Type-II kinase inhibitor, the structure provides an appropriate binding environment for screening compounds designed to target inactive kinase conformations.

Prior to molecular docking, the protein structure was prepared to ensure compatibility with docking calculations. Water molecules and non-essential heteroatoms present in the crystal structure were removed. Polar hydrogen atoms were added to the protein to account for hydrogen-bond interactions during docking simulations. The prepared structure was subsequently converted into the appropriate file format required for docking analysis. The active site selected for docking corresponded to the ATP-binding pocket occupied by the co-crystallized inhibitor imatinib in the crystal structure.

2.2 Ligand Library Preparation

A Type-II Kinase Inhibitor Library containing 2,376 compounds was obtained from Asinex. This commercially available library consists of structurally diverse compounds specifically designed to target inactive kinase conformations and is widely used for kinase-focused drug discovery studies. The library contains compounds possessing chemical scaffolds commonly associated with Type-II kinase inhibition, making it suitable for screening against the BCR-

ABL kinase domain. The ligand library was downloaded in structure-data format (SDF) and prepared for docking studies. Ligand structures were converted into docking-compatible formats using appropriate molecular preparation tools. During ligand preparation, hydrogen atoms were added where necessary, and molecular geometries were checked to ensure proper structural representation. The prepared ligands were then subjected to virtual screening against the receptor protein.

Table 1. Summary of ligand library preparation.

Step	**Description**
Database	Asinex kinase Type II inhibitor library
Total Compounds	2376
Format	SDF
Processing tool	Open Babel
Output Format	PDBQT
Screening Type	Structure-based virtual screening

2.3 Molecular Docking and Virtual Screening

Structure-based virtual screening was performed using AutoDock Vina, a widely used molecular docking program known for its accuracy and computational efficiency. AutoDock Vina predicts ligand binding poses within a target protein and estimates binding affinities through an empirical scoring function. The program explores multiple ligand conformations and orientations within a defined search space and ranks them according to predicted binding energy.

The docking grid was centered on the ATP-binding region of the ABL kinase domain using the coordinates corresponding to the position of the co-crystallized inhibitor. The grid parameters employed in this study were as follows:

- Center X = 14.105
- Center Y = 72.366
- Center Z = 39.843

The dimensions of the docking box were:

- Size X = 26 Å
- Size Y = 26 Å
- Size Z = 26 Å

These parameters ensured complete coverage of the active-site region and allowed ligands sufficient space to explore possible binding conformations. Each compound in the Asinex Type-II Kinase Inhibitor Library was docked individually against the prepared receptor structure. Following completion of the screening process, compounds were ranked according

to their predicted binding energies. Lower binding energy values indicate stronger predicted interactions between the ligand and the receptor.

The top ten compounds exhibiting the most favorable docking scores were selected for further analysis. These compounds were considered potential lead candidates because of their strong predicted affinity toward the BCR-ABL kinase domain.

To validate the docking protocol, the co-crystallized ligand was redocked into the active site of the prepared 1IEP structure, and the resulting pose was compared with the original crystallographic conformation by RMSD calculation using Open Babel (obrms).

2.4 Protein–Ligand Interaction Analysis

Detailed protein–ligand interaction analysis was performed using Discovery Studio Visualizer. The top ten ligands obtained from molecular docking were imported along with their corresponding docking poses to investigate their interaction patterns within the binding pocket.

Both two-dimensional (2D) and three-dimensional (3D) interaction analyses were conducted. Particular attention was given to hydrogen-bond interactions because hydrogen bonds play an important role in stabilizing protein–ligand complexes and often contribute significantly to binding specificity. Additional interactions, including hydrophobic contacts, π–π interactions, π–alkyl interactions, and van der Waals interactions, were also examined.

Among the top ten docked compounds, three ligands were selected for further study based on their favorable interaction profiles and hydrogen-bonding patterns. These compounds were identified as Nlig_ligand_2166, Nlig_ligand_2110, and Nlig_ligand_2029. The selected ligands demonstrated interactions with key amino acid residues located within the ATP-binding pocket of the kinase domain and exhibited stable binding orientations.

2.5 Pharmacokinetic and Drug-Likeness Evaluation

The three selected compounds were further evaluated using SwissADME, an online platform used for predicting physicochemical properties, pharmacokinetics, drug-likeness, and medicinal chemistry characteristics of small molecules. Canonical SMILES of the selected compounds were generated using Open Babel and submitted to SwissADME

Early assessment of these properties is essential because many compounds with favorable docking scores fail during drug development due to poor pharmacokinetic behavior.

Several parameters were analyzed during ADMET evaluation. These included molecular weight, lipophilicity (LogP), topological polar surface area (TPSA), gastrointestinal absorption, water solubility, and compliance with Lipinski’s Rule of Five. Lipinski’s rule is commonly used to evaluate oral drug-likeness and considers factors such as molecular weight, hydrogen-bond donors, hydrogen-bond acceptors, and lipophilicity.

In addition to drug-likeness assessment, medicinal chemistry filters were examined. Pan-Assay Interference Structure (PAINS) alerts were evaluated to identify substructures that may produce false-positive biological activity during screening assays. Brenk alerts were also

assessed to identify potentially problematic structural features. Synthetic accessibility scores were examined to assess the ease of synthesizing each compound for future experimental studies.

The results obtained from SwissADME were compared among the three selected compounds to identify the candidate exhibiting the most favorable balance of binding affinity, interaction profile, and pharmacokinetic characteristics. Based on this integrated analysis, the most promising lead compound was selected for further consideration.

3. Results and Discussion

3.1 Structure-Based Virtual Screening of the Asinex Type-II Kinase Inhibitor Library

Structure-based virtual screening was performed to identify potential inhibitors of the BCR-ABL tyrosine kinase from the Asinex Type-II Kinase Inhibitor Library. A total of 2,376 compounds were screened against the ATP-binding region of the ABL kinase domain represented by the crystal structure 1IEP. The docking process generated multiple binding conformations for each ligand, and compounds were ranked according to their predicted binding affinities obtained from AutoDock Vina.

The docking scores obtained from the screening campaign demonstrated considerable variation among the tested compounds, indicating differences in their ability to interact with the kinase active site. Several compounds exhibited highly favorable binding energies, suggesting strong interactions with the receptor and potential inhibitory activity. The top ten ligands identified through virtual screening displayed binding energies ranging from approximately –9.6 to –10.4 kcal/mol, values generally considered indicative of strong protein–ligand interactions.

Among all screened compounds, Nlig_ligand_2012 exhibited the lowest docking score of –10.359 kcal/mol, followed by Nlig_ligand_2166 with a binding energy of –10.052 kcal/mol. Additional high-ranking compounds included Nlig_ligand_1215, Nlig_ligand_2004, Nlig_ligand_2029, and Nlig_ligand_2110. The presence of multiple compounds with docking scores below –9.5 kcal/mol indicated that the Type-II kinase inhibitor library contains several molecules capable of occupying the ATP-binding pocket of the kinase domain with high predicted affinity.

Table 2. Top ten ligands identified through virtual screening.

Rank	Ligand	Binding Energy (kcal/mol)
1	Nlig_ligand_2012	-10.359
2	Nlig_ligand_2166	-10.052
3	Nlig_ligand_1215	-9.919
4	Nlig_ligand_2004	-9.815
5	Nlig_ligand_2029	-9.781
6	Nlig_ligand_2013	-9.776
7	Nlig_ligand_2133	-9.688

8	Nlig_ligand_1901	-9.650
9	Nlig_ligand_1990	-9.618
10	Nlig_ligand_2110	-9.599

Although Nlig_ligand_2012 exhibited the lowest docking score, subsequent interaction analysis demonstrated that Nlig_ligand_2166, Nlig_ligand_2110, and Nlig_ligand_2029 possessed more favorable hydrogen-bond interactions and binding orientations.

Although docking scores provide valuable information regarding binding potential, it is widely recognized that binding energy alone should not be used as the sole criterion for lead selection. The quality of protein–ligand interactions, the presence of stabilizing hydrogen bonds, binding orientation within the active site, and pharmacokinetic properties must also be considered. Consequently, the top ten compounds were subjected to detailed interaction analysis using Discovery Studio Visualizer.

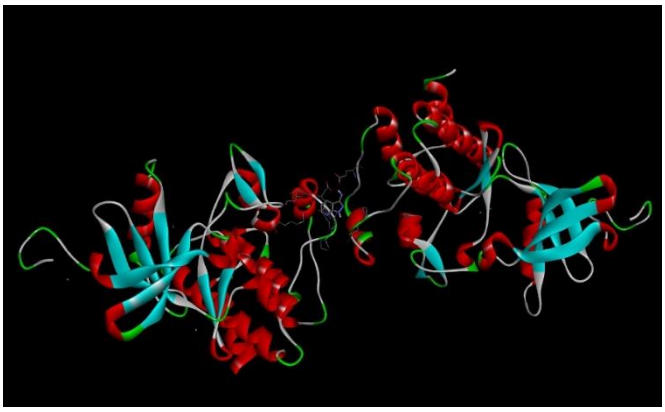
3.2 Protein–Ligand Interaction Analysis

Protein–ligand interaction analysis provides important information regarding the molecular basis of ligand binding and helps identify compounds that establish favorable contacts with residues located within the active site. Hydrogen bonds are particularly important because they contribute to binding specificity and stabilization of the protein–ligand complex. Therefore, the top ten compounds were carefully examined to identify interactions with key residues of the kinase domain.

Following detailed analysis, three compounds were prioritized based on the quality of their interaction profiles. These compounds, namely Nlig_ligand_2166, Nlig_ligand_2110, and Nlig_ligand_2029, demonstrated favorable binding orientations and established hydrogen-bond interactions with residues located within the ATP-binding pocket.

Interaction Analysis of Nlig_ligand_2166

Nlig_ligand_2166 exhibited a binding affinity of −10.052 kcal/mol and formed hydrogen-bond interactions with GLU459 and PRO441. Several hydrophobic interactions further stabilized the ligand within the ATP-binding pocket, indicating a strong binding mode.



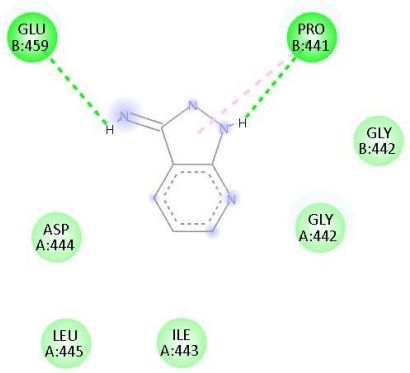


Figure 1. Two-Dimensional and Three-Dimensional interaction diagram of Nlig_ligand_2166 docked within the active site of the receptor.

Interaction Analysis of Nlig_ligand_2110

Nlig_ligand_2110 demonstrated hydrogen-bond interactions with GLU459 and PRO441. Despite ranking tenth in docking score, the ligand exhibited an optimal interaction network and favorable orientation within the binding site. These ex formation and potential inhibitory activity.

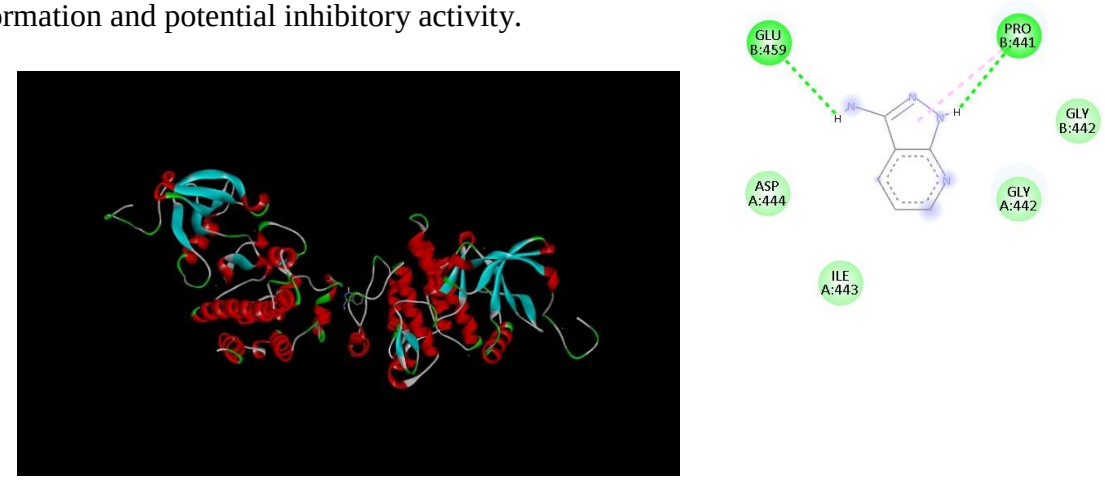


Figure 2. Two-Dimensional and Three-Dimensional interaction diagram of Nlig_ligand_2110 docked within the active site of the receptor.

Interaction Analysis of Nlig_ligand_2029

Nlig_ligand_2029 formed hydrogen bonds with GLY442 and LYS404. The ligand occupied the active site effectively and demonstrated a stable docking pose supported by multiple non-covalent interactions.

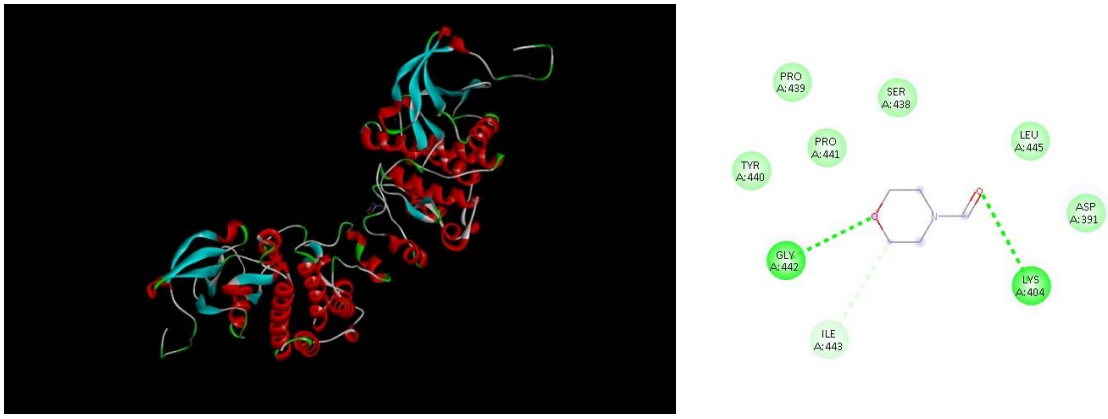


Figure 3. Two-Dimensional and Three-Dimensional interaction diagram of Nlig_ligand_2029 docked within the active site of the receptor.

Table 3. Interaction analysis of top-ranked ligands docked within the ABL kinase domain active site

Ligands	Score	H bonds	Residues
Nlig_ligand_2012	-10.359	1	TYR440
Nlig_ligand_2166	-10.052	2	GLU459, PRO441
Nlig_ligand_1215	-9.919	1	GLU459

Nlig_ligand_2004	-9.815	1	PRO441
Nlig_ligand_2029	-9.781	3	GLY442, ILE443, LYS404
Nlig_ligand_2013	-9.776	0	(none)
Nlig_ligand_2133	-9.688	2	LYS404, ILE443
Nlig_ligand_1901	-9.650	1	TYR440
Nlig_ligand_1990	-9.618	2	TYR440, GLY442
Nlig_ligand_2110	-9.599	2	PRO441, GLU459

3.3 RMSD Validation of Docking Protocol

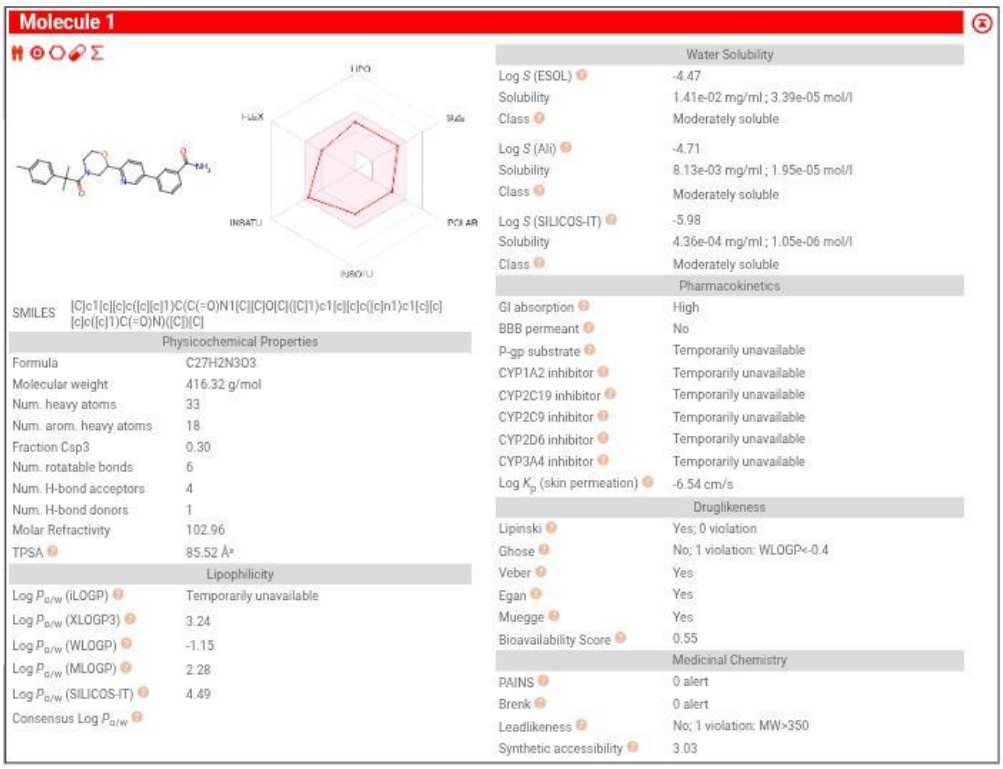
To assess the reliability of the docking protocol, root mean square deviation (RMSD) analysis was performed by comparing the pose of the co-crystallized reference ligand with the docked ligand pose using the obrms command in Open Babel. The calculated RMSD value was 1.2 Å, indicating a close agreement between the experimental binding conformation and the redocked pose. In molecular docking studies, an RMSD value below 2.0 Å is generally considered acceptable for successful reproduction of the native binding mode. This low RMSD value indicates that AutoDock Vina effectively reproduced the native binding mode of imatinib, including the characteristic hydrogen-bond interactions with key residues GLU459 and PRO441 within the active site. Therefore, the obtained result confirms that the selected docking parameters and protocol were sufficiently reliable for virtual screening and docking analysis of the Type-II kinase inhibitor library against the BCR-ABL kinase domain.

3.4 ADMET Evaluation of Selected Ligands

Following interaction analysis, Nlig_ligand_2166, Nlig_ligand_2110, and Nlig_ligand_2029 were subjected to pharmacokinetic evaluation using SwissADME. The objective of this analysis was to identify compounds possessing favorable drug-like characteristics in addition to strong binding affinity.

All three compounds demonstrated acceptable physicochemical properties and satisfied major drug-likeness criteria. Furthermore, all selected ligands complied with Lipinski's Rule of Five, indicating favorable molecular characteristics commonly associated with orally active drugs. The absence of PAINS alerts among the selected compounds represented an important finding. PAINS alerts are associated with structural motifs capable of generating false-positive results during biological screening. Compounds lacking such alerts are generally considered more reliable candidates for further development. Similarly, the absence of Brenk alerts indicated that the compounds did not contain structural features commonly associated with toxicity or poor pharmacological behavior.

Nlig_ligand_2110 exhibited the most favorable profile among the three compounds. The molecule possessed a molecular weight of approximately 390 Da, high gastrointestinal absorption, acceptable lipophilicity, compliance with Lipinski's rule of five, and no PAINS or Brenk alerts. Nlig_ligand_2166 also demonstrated favorable properties but possessed a higher molecular weight and slightly less desirable overall drug-likeness profile. Nlig_ligand_2029 exhibited acceptable pharmacokinetic characteristics but showed comparatively higher lipophilicity and lower lead-likeness.



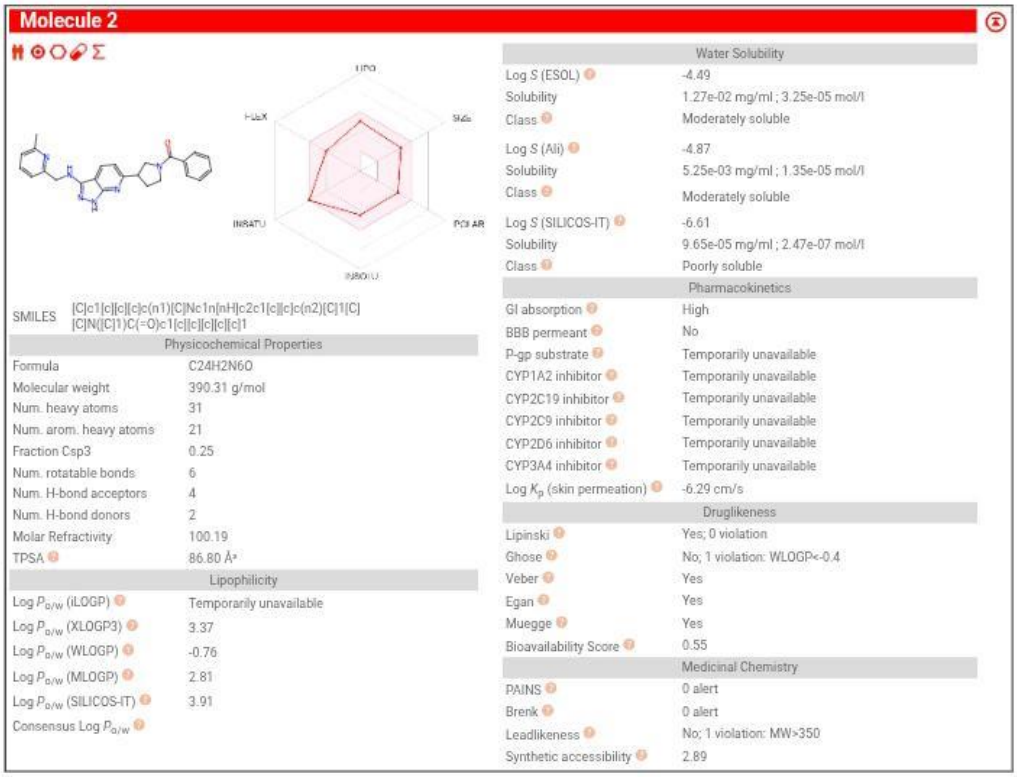
Molecule 1

SMILES [C]c1[c][c]c([c][c]1)C(C(=O)N1[C][C]O[C]([C]1)c1[c][c]c([c]n1)c1[c][c][c]c([c]1)C(=O)N)([C])[C]

Physicochemical Properties	
Formula	C27H2N3O3
Molecular weight	416.32 g/mol
Num. heavy atoms	33
Num. arom. heavy atoms	18
Fraction Csp3	0.30
Num. rotatable bonds	6
Num. H-bond acceptors	4
Num. H-bond donors	1
Molar Refractivity	102.96
TPSA	85.52 Å²
Lipophilicity	
Log $P_{o/w}$ (iLOGP)	Temporarily unavailable
Log $P_{o/w}$ (XLOGP3)	3.24
Log $P_{o/w}$ (WLOGP)	-1.15
Log $P_{o/w}$ (MLOGP)	2.28
Log $P_{o/w}$ (SILICOS-IT)	4.49
Consensus Log $P_{o/w}$	

Water Solubility	
Log *S* (ESOL)	-4.47
Solubility	1.41e-02 mg/ml ; 3.39e-05 mol/l
Class	Moderately soluble
Log *S* (Ali)	-4.71
Solubility	8.13e-03 mg/ml ; 1.95e-05 mol/l
Class	Moderately soluble
Log *S* (SILICOS-IT)	-5.98
Solubility	4.36e-04 mg/ml ; 1.05e-06 mol/l
Class	Moderately soluble
Pharmacokinetics	
GI absorption	High
BBB permeant	No
P-gp substrate	Temporarily unavailable
CYP1A2 inhibitor	Temporarily unavailable
CYP2C19 inhibitor	Temporarily unavailable
CYP2C9 inhibitor	Temporarily unavailable
CYP2D6 inhibitor	Temporarily unavailable
CYP3A4 inhibitor	Temporarily unavailable
Log K_p (skin permeation)	-6.54 cm/s
Druglikeness	
Lipinski	Yes; 0 violation
Ghose	No; 1 violation: WLOGP<-0.4
Veber	Yes
Egan	Yes
Muegge	Yes
Bioavailability Score	0.55
Medicinal Chemistry	
PAINS	0 alert
Brenk	0 alert
Leadlikeness	No; 1 violation: MW>350
Synthetic accessibility	3.03

Figure 4. SwissADME prediction results for Nlig_ligand_2166



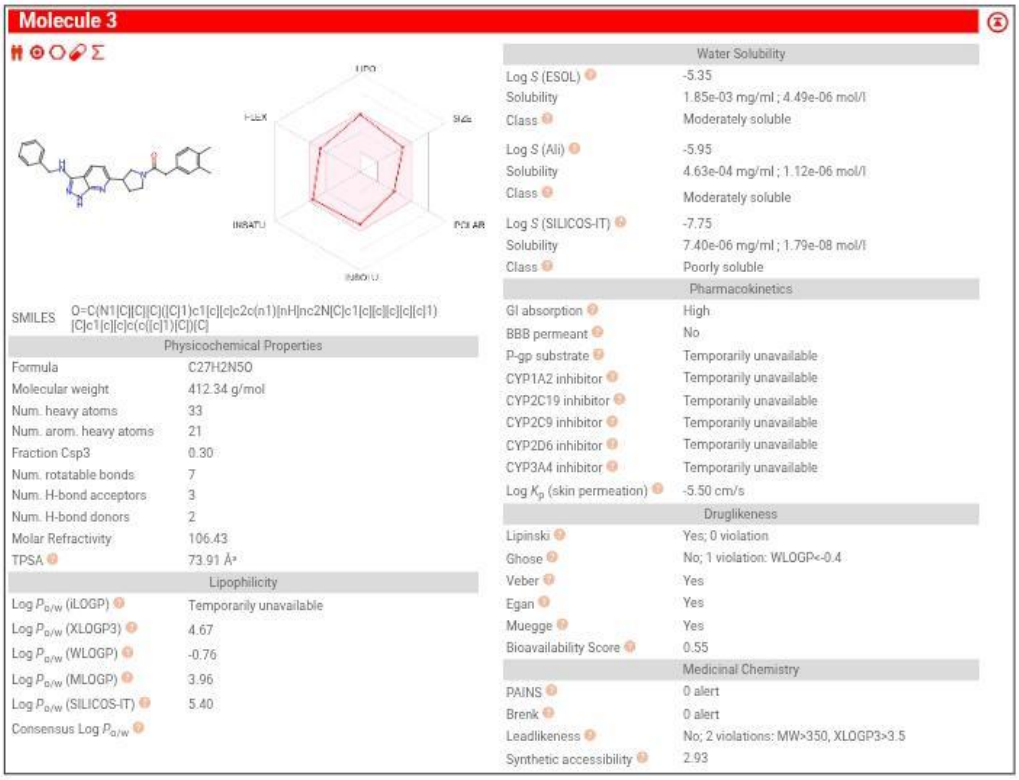
Molecule 2

SMILES [C]c1[c][c][c]c(n1)[C]Nc1n[nH]c2c1[c][c]c(n2)[C]1[C][C]N([C]1)C(=O)c1[c][c][c][c][c]1

Physicochemical Properties	
Formula	C24H2N6O
Molecular weight	390.31 g/mol
Num. heavy atoms	31
Num. arom. heavy atoms	21
Fraction Csp3	0.25
Num. rotatable bonds	6
Num. H-bond acceptors	4
Num. H-bond donors	2
Molar Refractivity	100.19
TPSA	86.80 Å²
Lipophilicity	
Log $P_{o/w}$ (iLOGP)	Temporarily unavailable
Log $P_{o/w}$ (XLOGP3)	3.37
Log $P_{o/w}$ (WLOGP)	-0.76
Log $P_{o/w}$ (MLOGP)	2.81
Log $P_{o/w}$ (SILICOS-IT)	3.91
Consensus Log $P_{o/w}$	

Water Solubility	
Log *S* (ESOL)	-4.49
Solubility	1.27e-02 mg/ml ; 3.25e-05 mol/l
Class	Moderately soluble
Log *S* (Ali)	-4.87
Solubility	5.25e-03 mg/ml ; 1.35e-05 mol/l
Class	Moderately soluble
Log *S* (SILICOS-IT)	-6.61
Solubility	9.65e-05 mg/ml ; 2.47e-07 mol/l
Class	Poorly soluble
Pharmacokinetics	
GI absorption	High
BBB permeant	No
P-gp substrate	Temporarily unavailable
CYP1A2 inhibitor	Temporarily unavailable
CYP2C19 inhibitor	Temporarily unavailable
CYP2C9 inhibitor	Temporarily unavailable
CYP2D6 inhibitor	Temporarily unavailable
CYP3A4 inhibitor	Temporarily unavailable
Log K_p (skin permeation)	-6.29 cm/s
Druglikeness	
Lipinski	Yes; 0 violation
Ghose	No; 1 violation: WLOGP<-0.4
Veber	Yes
Egan	Yes
Muegge	Yes
Bioavailability Score	0.55
Medicinal Chemistry	
PAINS	0 alert
Brenk	0 alert
Leadlikeness	No; 1 violation: MW>350
Synthetic accessibility	2.89

Figure 5. SwissADME prediction results for Nlig_ligand_2110



Molecule 3

SMILES O=C(N1[C][C][C]([C]1)c1[c][c]c2c(n1)[nH]nc2N[C]c1[c][c][c][c][c]1)[C]c1[c][c]c(c([c]1)[C])[C]

Physicochemical Properties	
Formula	C27H2N5O
Molecular weight	412.34 g/mol
Num. heavy atoms	33
Num. arom. heavy atoms	21
Fraction Csp3	0.30
Num. rotatable bonds	7
Num. H-bond acceptors	3
Num. H-bond donors	2
Molar Refractivity	106.43
TPSA	73.91 Å²

Lipophilicity	
Log $P_{o/w}$ (iLOGP)	Temporarily unavailable
Log $P_{o/w}$ (XLOGP3)	4.67
Log $P_{o/w}$ (WLOGP)	-0.76
Log $P_{o/w}$ (MLOGP)	3.96
Log $P_{o/w}$ (SILICOS-IT)	5.40
Consensus Log $P_{o/w}$	

Water Solubility	
Log S (ESOL)	-5.35
Solubility	1.85e-03 mg/ml ; 4.49e-06 mol/l
Class	Moderately soluble
Log S (Ali)	-5.95
Solubility	4.63e-04 mg/ml ; 1.12e-06 mol/l
Class	Moderately soluble
Log S (SILICOS-IT)	-7.75
Solubility	7.40e-06 mg/ml ; 1.79e-08 mol/l
Class	Poorly soluble

Pharmacokinetics	
GI absorption	High
BBB permeant	No
P-gp substrate	Temporarily unavailable
CYP1A2 inhibitor	Temporarily unavailable
CYP2C19 inhibitor	Temporarily unavailable
CYP2C9 inhibitor	Temporarily unavailable
CYP2D6 inhibitor	Temporarily unavailable
CYP3A4 inhibitor	Temporarily unavailable
Log K_p (skin permeation)	-5.50 cm/s

Druglikeness	
Lipinski	Yes; 0 violation
Ghose	No; 1 violation: WLOGP<-0.4
Veber	Yes
Egan	Yes
Muegge	Yes
Bioavailability Score	0.55

Medicinal Chemistry	
PAINS	0 alert
Brenk	0 alert
Leadlikeness	No; 2 violations: MW>350, XLOGP3>3.5
Synthetic accessibility	2.93

Figure 6. SwissADME prediction results for Nlig_ligand_2029

Table 4. Comparative ADMET assessment.

Property	2166	2110	2029
GI Absorption	High	High	High
Lipinski Compliance	Yes	Yes	Yes
PAINS Alerts	0	0	0
Brenk Alerts	0	0	0
Overall Drug-Likeness	Good	Excellent	Moderate
Final Ranking	2	1	3

3.5 Selection of the Final Lead Compound

An important objective of virtual screening studies is the identification of compounds that demonstrate not only strong binding affinity but also favorable drug-like properties. For this reason, lead selection should be based on a comprehensive evaluation rather than docking score alone.

Although Nlig_ligand_2012 exhibited the most favorable binding energy among all screened compounds, detailed interaction analysis and ADMET evaluation identified Nlig_ligand_2110 as the superior overall candidate. The compound demonstrated stable

interactions within the ATP-binding pocket, formed hydrogen bonds with key residues, exhibited favorable pharmacokinetic characteristics, and satisfied multiple drug-likeness criteria.

The findings of this study emphasize the importance of integrating molecular docking with pharmacokinetic evaluation during lead identification. Compounds displaying moderate docking scores may ultimately prove more suitable for development than compounds exhibiting exceptionally strong binding affinity but poor pharmacological properties.

Based on the combined results of virtual screening, protein–ligand interaction analysis, and ADMET evaluation, Nlig_ligand_2110 was selected as the final lead compound and represents the most promising candidate identified in the present investigation.

Overall, the integration of docking and ADMET results suggests that Nlig_ligand_2110 is the most promising candidate for further optimization and experimental validation as a potential BCR-ABL inhibitor.

4. Discussion

The present study employed structure-based virtual screening of the Asinex Type-II Kinase Inhibitor Library against the BCR-ABL tyrosine kinase using the crystal structure 1IEP to identify potential inhibitory compounds. Molecular docking analysis revealed that the screened ligands exhibited varying binding affinities toward the ATP-binding pocket of the kinase domain. Several compounds demonstrated favorable docking scores, with the top-ranked ligands showing binding energies below –9.5 kcal/mol, indicating stable protein–ligand complex formation and potential inhibitory activity.

BCR-ABL is a well-established therapeutic target in chronic myeloid leukemia (CML) because of its constitutive tyrosine kinase activity, which promotes uncontrolled cell proliferation and survival. Inhibition of BCR-ABL remains the primary therapeutic strategy for the treatment of CML. Although currently available tyrosine kinase inhibitors such as imatinib have significantly improved patient outcomes, resistance and adverse effects continue to motivate the search for novel inhibitors with improved pharmacological properties.

Among the selected compounds, Nlig_ligand_2166, Nlig_ligand_2110, and Nlig_ligand_2029 demonstrated favorable interaction profiles within the kinase active site. Hydrogen-bond analysis revealed that Nlig_ligand_2166 and Nlig_ligand_2110 formed interactions with GLU459 and PRO441, while Nlig_ligand_2029 interacted with GLY442 and LYS404. These interactions are likely to contribute to stabilization of the ligand within the ATP-binding pocket and support effective inhibition of kinase activity.

Although Nlig_ligand_2166 exhibited the most favorable docking score among the three selected compounds, ADMET analysis indicated that Nlig_ligand_2110 possessed the most balanced pharmacokinetic profile. The compound demonstrated favorable drug-likeness properties, high gastrointestinal absorption, compliance with Lipinski’s Rule of Five, and the absence of PAINS and Brenk alerts. These characteristics suggest that Nlig_ligand_2110 may have a greater likelihood of success during subsequent stages of drug development.

Type-II kinase inhibitors bind preferentially to the inactive conformation of kinases and extend into an adjacent hydrophobic pocket, resulting in improved selectivity and enhanced binding stability. Since the crystal structure 1IEP represents the inactive conformation of the ABL kinase domain bound to imatinib, the favorable binding behavior observed for the selected ligands is consistent with the expected mechanism of Type-II kinase inhibition.

Overall, the combined docking, interaction, and ADMET results suggest that Nlig_ligand_2110 is the most promising candidate identified in this study. The compound demonstrated a favorable balance between binding affinity, interaction quality, and pharmacokinetic properties, making it a suitable candidate for further optimization and experimental validation as a potential BCR-ABL tyrosine kinase inhibitor.

5. Limitations

The present study was conducted entirely using computational methods, including molecular docking and ADMET prediction, which possess certain inherent limitations. Molecular docking provides a theoretical estimate of protein–ligand interactions and represents a static binding model that may not fully account for the dynamic nature of proteins under physiological conditions.

Additionally, molecular dynamics (MD) simulations were not performed to assess the long-term stability and conformational behavior of the docked complexes. Such analyses could provide a more comprehensive understanding of ligand binding and complex stability.

Furthermore, the pharmacokinetic properties predicted through ADMET analysis are based on computational models and may not accurately reflect biological behavior in experimental systems. The findings of this study have not been validated through in vitro or in vivo investigations.

Therefore, although the selected compounds demonstrated favorable binding affinities and promising drug-like characteristics, further computational studies and experimental validation are necessary to confirm their potential as BCR-ABL tyrosine kinase inhibitors.

6. Conclusion

The present study employed an integrated computational drug discovery approach involving structure-based virtual screening, molecular docking, protein–ligand interaction analysis, and ADMET evaluation to identify potential BCR-ABL tyrosine kinase inhibitors from the Asinex Type-II Kinase Inhibitor Library. The crystal structure of the ABL kinase domain complexed with imatinib (PDB ID: 1IEP) was utilized as the receptor model because it represents the inactive kinase conformation targeted by clinically successful Type-II inhibitors.

A total of 2,376 compounds were screened against the ATP-binding pocket of the kinase domain using AutoDock Vina. Several compounds demonstrated strong predicted binding affinities, with docking scores below –9.5 kcal/mol. The top ten ligands were further

investigated through protein–ligand interaction analysis using Discovery Studio Visualizer. Among these compounds, Nlig_ligand_2166, Nlig_ligand_2110, and Nlig_ligand_2029 exhibited favorable interaction patterns characterized by hydrogen-bond formation and stable binding orientations within the active site.

Subsequent pharmacokinetic and drug-likeness evaluation using SwissADME revealed that all three compounds possessed acceptable physicochemical properties and complied with major drug-likeness criteria. However, Nlig_ligand_2110 demonstrated the most favorable overall profile, combining stable protein–ligand interactions with high gastrointestinal absorption, compliance with Lipinski's Rule of Five, absence of PAINS and Brenk alerts, and acceptable synthetic accessibility. Although Nlig_ligand_2166 exhibited a lower docking score, the integrated assessment indicated that Nlig_ligand_2110 possessed the most balanced combination of binding affinity, interaction quality, and pharmacokinetic suitability.

The findings of this study highlight the value of combining molecular docking with pharmacokinetic evaluation during lead identification. The results suggest that Nlig_ligand_2110 may serve as a promising lead compound for the development of novel BCR-ABL tyrosine kinase inhibitors. Nevertheless, molecular docking represents only an initial stage of drug discovery, and additional studies are required to validate the therapeutic potential of the identified compound. Future investigations should include molecular dynamics simulations, binding free-energy calculations, in vitro kinase inhibition assays, and cellular studies to confirm biological activity and further optimize the compound for potential therapeutic application.

Overall, this study demonstrates the effectiveness of structure-based virtual screening as a cost-effective strategy for identifying novel kinase inhibitors and provides a foundation for the future development of improved therapeutic agents targeting BCR-ABL-driven malignancies.

References

1. Berman HM, Westbrook J, Feng Z, Gilliland G, Bhat TN, Weissig H, et al. The Protein Data Bank. *Nucleic Acids Research*. 2000;28(1):235–242. DOI: https://doi.org/10.1093/nar/28.1.235
2. Druker BJ, Talpaz M, Resta DJ, Peng B, Buchdunger E, Ford JM, et al. Efficacy and safety of a specific inhibitor of the BCR-ABL tyrosine kinase in chronic myeloid leukemia. *New England Journal of Medicine*. 2001;344(14):1031–1037. DOI: https://doi.org/10.1056/NEJM200104053441401
3. Druker BJ. Translation of the Philadelphia chromosome into therapy for CML. *Blood*. 2008;112(13):4808–4817. DOI: https://doi.org/10.1182/blood-2008-07-077958
4. Goldman JM, Melo JV. Chronic myeloid leukemia—advances in biology and new approaches to treatment. *New England Journal of Medicine*. 2003;349(15):1451–1464. DOI: https://doi.org/10.1056/NEJMra020777
5. Faderl S, Talpaz M, Estrov Z, O'Brien S, Kurzrock R, Kantarjian HM. The biology of chronic myeloid leukemia. *New England Journal of Medicine*. 1999;341(3):164–172. DOI: https://doi.org/10.1056/NEJM199907153410306
6. Zhang J, Yang PL, Gray NS. Targeting cancer with small molecule kinase inhibitors. *Nature Reviews Cancer*. 2009;9(1):28–39. DOI: https://doi.org/10.1038/nrc2559

7. Cohen P. Protein kinases—the major drug targets of the twenty-first century? *Nature Reviews Drug Discovery*. 2002;1(4):309–315. DOI: https://doi.org/10.1038/nrd773
8. Roskoski R Jr. Properties of FDA-approved small molecule protein kinase inhibitors. *Pharmacological Research*. 2019;144:19–50. DOI: https://doi.org/10.1016/j.phrs.2019.03.006
9. Liu Y, Gray NS. Rational design of inhibitors that bind to inactive kinase conformations. *Nature Chemical Biology*. 2006;2(7):358–364. DOI: https://doi.org/10.1038/nchembio799
10. Hantschel O, Superti-Furga G. Regulation of the c-Abl and Bcr-Abl tyrosine kinases. *Nature Reviews Molecular Cell Biology*. 2004;5(1):33–44. DOI: https://doi.org/10.1038/nrm1280
11. Trott O, Olson AJ. AutoDock Vina: improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. *Journal of Computational Chemistry*. 2010;31(2):455–461. DOI: https://doi.org/10.1002/jcc.21334
12. Forli S, Huey R, Pique ME, Sanner MF, Goodsell DS, Olson AJ. Computational protein–ligand docking and virtual drug screening with AutoDock. *Nature Protocols*. 2016;11(5):905–919. DOI: https://doi.org/10.1038/nprot.2016.051
13. Kitchen DB, Decornez H, Furr JR, Bajorath J. Docking and scoring in virtual screening for drug discovery. *Nature Reviews Drug Discovery*. 2004;3(11):935–949. DOI: https://doi.org/10.1038/nrd1549
14. Lionta E, Spyrou G, Vassilatis DK, Cournia Z. Structure-based virtual screening for drug discovery: principles, applications and recent advances. *Current Topics in Medicinal Chemistry*. 2014;14(16):1923–1938. DOI: https://doi.org/10.2174/1568026614666140929124445
15. Meng XY, Zhang HX, Mezei M, Cui M. Molecular docking: a powerful approach for structure-based drug discovery. *Current Computer-Aided Drug Design*. 2011;7(2):146–157. DOI: https://doi.org/10.2174/157340911795677602
16. 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;7:42717. DOI: https://doi.org/10.1038/srep42717
17. Lipinski CA, Lombardo F, Dominy BW, Feeney PJ. Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings. *Advanced Drug Delivery Reviews*. 2001;46(1–3):3–26. DOI: https://doi.org/10.1016/S0169-409X(00)00129-0
18. Veber DF, Johnson SR, Cheng HY, Smith BR, Ward KW, Kopple KD. Molecular properties that influence oral bioavailability of drug candidates. *Journal of Medicinal Chemistry*. 2002;45(12):2615–2623. DOI: https://doi.org/10.1021/jm020017n
19. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. *Cell*. 2011;144(5):646–674. DOI: https://doi.org/10.1016/j.cell.2011.02.013
20. Jabbour E, Kantarjian H. Chronic myeloid leukemia: 2020 update on diagnosis, therapy and monitoring. *American Journal of Hematology*. 2020;95(6):691–709. DOI: https://doi.org/10.1002/ajh.25792
21. O'Hare T, Zabriskie MS, Eiring AM, Deininger MW. Pushing the limits of targeted therapy in chronic myeloid leukaemia. *Nature Reviews Cancer*. 2012;12(8):513–526. DOI: https://doi.org/10.1038/nrc3317
22. Weisberg E, Manley PW, Cowan-Jacob SW, Hochhaus A, Griffin JD. Second generation inhibitors of BCR-ABL for the treatment of imatinib-resistant chronic myeloid leukemia. *Nature Reviews Cancer*. 2007;7(5):345–356. DOI: https://doi.org/10.1038/nrc2126