

EFFICIENT SYNTHESIS OF N-PHENYLBENZAMIDES VIA AMIDATION OF VINYL ESTERS OF AROMATIC CARBOXYLIC ACIDS WITH ANILINE

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Abstract: In this work, the amidation reaction of various substituted vinyl esters of aromatic carboxylic acids with aniline was carried out in toluene solution. The effect of substituents on the aromatic ring on the product yield was investigated, and a reaction mechanism was proposed. The following amides of aromatic carboxylic acids were synthesized from the reaction of aniline with the vinyl esters of 4-bromobenzoic acid, 4-chlorobenzoic acid, 4-tert-butylbenzoic acid, 4-nitrobenzoic acid, and benzoic acid: 4-bromo-N-phenylbenzamide (75%), 4-chloro-N-phenylbenzamide (70%), 4-tert-butyl-N-phenylbenzamide (60%), 4-nitro-N-phenylbenzamide (80%), and N-phenylbenzamide (65%). The structures of the synthesized amides were confirmed by ¹H and ¹³C NMR spectral analysis. An *in silico* molecular docking approach was used to evaluate the interaction potential of the synthesized amides with two biological target proteins: the main protease of SARS-CoV-2 (PDB ID: 6LU7) and sphingosine-1-phosphate receptor 3 (S1PR3, PDB ID: 7C4S). Docking calculations were performed using AutoDock Vina 1.2, and the resulting ligand-protein complexes were visualized and analyzed using PyMOL and Discovery Studio software. Binding energy results were used to assess ligand affinity toward both targets. The results showed that 4-tert-butyl-N-phenylbenzamide exhibited the highest affinity for 7C4S, with a binding energy of -10.5 kcal/mol. It also demonstrated strong binding to 6LU7, with a binding energy of -6.7 kcal/mol. Molecular interaction analysis indicated that these compounds occupy key active sites through hydrogen bonding and hydrophobic interactions. Based on these findings, 4-tert-butyl-N-phenylbenzamide may serve as a promising dual-target inhibitor for proteins associated with disease. Further research involving *in vitro* validation and molecular dynamics simulations is recommended.

Key words: Vinyl esters, Amidation, N-phenylbenzamides, Molecular docking, SARS-CoV-2 protease, S1PR3 receptor.

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Introduction

Vinyl esters are widely used compounds in various industries due to their high reactivity, polymerization ability, and unique chemical properties. They find applications in the production of polymers, coatings, paints, composite materials, adhesives, and also serve as intermediate active reagents in organic synthesis. Additionally, vinyl esters are employed in the pharmaceutical industry for drug formulation, in agriculture for the development of pesticides and herbicides, and in the fragrance industry for the synthesis of perfumes [1–3].

Amides represent one of the most

important functional groups in natural products, pharmaceutical drugs, and other bioactive molecules. Polymers containing acid amide functionalities are extensively utilized in industrial applications. In recent years, significant research has focused on the amidation of carboxylic acid vinyl esters with amino-containing compounds. These studies have largely aimed to optimize catalytic methods to improve the efficiency and stability of amidation reactions. Notably, the highly selective and efficient amidation of vinyl esters with various aromatic amines using N-heterocyclic carbene

(NHC)-based catalysts has been reported. Additionally, borate esters have emerged as simple and effective catalysts for facilitating amidation reactions of vinyl esters [4–6].

Amides of carboxylic acids — particularly those derived from aniline—are widely used in the pharmaceutical field. These compounds are

known for their anticancer, anti-inflammatory, and antimicrobial properties. Furthermore, acid amides based on quinazoline derivatives function as ligands in catalytic processes during organic synthesis. Such ligands enhance the selectivity of catalytic reactions and contribute to improving product yields [7,8].

Experimental part

Methods and materials

Chemistry. $^1\text{H-NMR}$ spectra were recorded at a frequency of 400 MHz using a Unity+400 (Varian) instrument in CDCl_3 . GMD (tetramethylsilane) was used as the internal standard for ^1H NMR spectra. For $^{13}\text{C-NMR}$ spectra, the chemical shift of the solvent was used as the internal standard.

Amidation reaction of vinyl esters of aromatic carboxylic acid with aniline: A 50 mL round-bottom flask with three necks and a reflux condenser, dropping funnel, and mechanical stirrer was filled with 0.9 ml (0.93 g, 10 mmol) of aniline that had been dissolved in 10 mL of toluene. Catalytic triethylamine was added to the solution. Then, 10 mmol vinyl ester of aromatic carboxylic acid dissolved in 5 mL toluene was added dropwise. The reaction mixture was warmed to 60 °C and mechanically stirred for 4 hours. After completion, the reaction mixture was brought to room temperature and extracted in dichloromethane (3×15 mL). The organic layer was subsequently washed with a saturated solution of potassium chloride and distilled water. The extract was dried over 5 g of Na_2SO_4 for 12 hours and filtered. The residue was dried to constant weight in a vacuum desiccator after the solvent was evaporated in a vacuum. The amides formed were recrystallized from ethanol and characterized by ^1H and ^{13}C NMR spectroscopy. The spectral data of the synthesized compounds are given below.

N-Phenylbenzamide. Yield 65.0%. $^1\text{H-NMR}$ (600 MHz, CDCl_3): δ 8.56 (s, 1H), 7.95 (dd, $J=7.9$, 1.3 Hz, 2H), 7.68 (dd, $J=7.1$, 1.1 Hz, 2H), 7.50 (t, $J=7.8$ Hz, 1H), 7.44 (t, $J=7.9$ Hz, 2H), 7.32 (t, $J=7.0$ Hz, 2H), 7.14 (t, $J=7.0$ Hz, 1H). $^{13}\text{C-NMR}$ (101 MHz, CDCl_3): δ 166.04, 139.29, 135.20, 132.00, 128.84, 128.54, 127.79, 123.78, 120.50.

4-Bromo-N-phenylbenzamide. Yield 75.0%. $^1\text{H-NMR}$ (400 MHz, CDCl_3): δ 8.81 (s,

1H), 7.87–7.82 (m, 2H), 7.72–7.66 (m, 4H), 7.35–7.29 (m, 2H), 7.14 (tt, $J=6.8$, 1.2 Hz, 1H). $^{13}\text{C-NMR}$ (101 MHz, CDCl_3): δ 165.88, 139.29, 132.71, 131.81, 130.09, 128.84, 125.44, 123.78, 120.50.

4-Nitro-N-phenylbenzamide. Yield 80.0%. $^1\text{H-NMR}$ (400 MHz, CDCl_3): δ 8.40–8.35 (m, 2H), 8.25–8.20 (m, 2H), 7.71–7.66 (m, 2H), 7.35–7.29 (m, 2H), 7.17–7.11 (m, 1H). $^{13}\text{C-NMR}$ (101 MHz, CDCl_3): δ 165.83, 148.78, 140.23, 139.29, 129.52, 128.84, 123.78, 123.69, 120.50.

4-Tert-butyl-N-phenylbenzamide. Yield 60.0%. $^1\text{H-NMR}$ (400 MHz, CDCl_3): δ 8.94 (s, 1H), 7.72–7.66 (m, 4H), 7.46–7.41 (m, 2H), 7.35–7.29 (m, 2H), 7.17–7.11 (m, 1H), 1.34 (s, 12H). $^{13}\text{C-NMR}$ (101 MHz, CDCl_3): δ 165.88, 153.20, 139.29, 132.27, 128.84, 128.73, 125.60, 123.78, 120.50, 34.87, 31.18.

4-Chloro-N-phenylbenzamide. Yield 70.0%. $^1\text{H-NMR}$ (400 MHz, CDCl_3): δ 8.39 (s, 1H), 7.90–7.85 (m, 2H), 7.71–7.66 (m, 2H), 7.55–7.50 (m, 2H), 7.35–7.29 (m, 2H), 7.14 (tt, $J=6.8$, 1.2 Hz, 1H). $^{13}\text{C-NMR}$ (101 MHz, CDCl_3): δ 165.88, 139.29, 136.87, 133.03, 130.11, 129.05, 128.84, 123.78, 120.50.

Molecular docking. Five molecules of N-phenylbenzamide derivative were taken into molecular docking analysis in this study. Sphingosine-1-phosphate receptor 3 (S1PR3, PDB ID: 7C4S) and SARS-CoV-2 main protease (PDB ID: 6LU7) were the targets selected. Protein structures were downloaded from the Protein Data Bank, and AutoDock Vina 1.2 was used to perform the docking calculations. Grid box parameters were located in the center of active sites of target proteins, and exhaustiveness was set to 8. Lowest binding energy conformations were used for further evaluation. PyMOL and Discovery Studio programs were employed for visualizing obtained complexes, where ligand-protein interactions (hydrogen bonds, van der Waals contacts, and π - π stacking)

were examined. The binding energies (ΔG) were determined to be kcal/mol and were used to

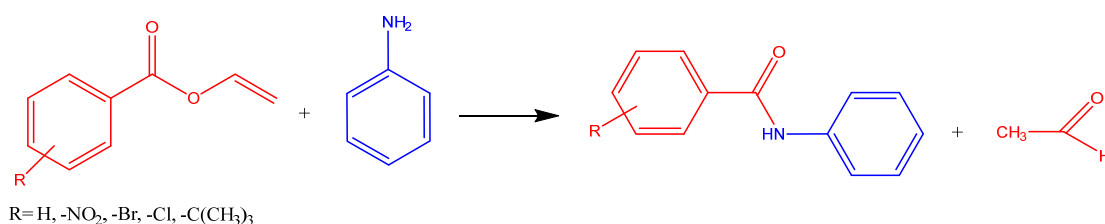
examine the binding affinities of the ligands.

Results and discussion

Various derivatives of N-phenylbenzamide are biologically active compounds [9]. These compounds are used as medicinal substances in the pharmaceutical industry. In medicine, they are developed as anticancer drugs that inhibit the growth of cancer cells [10]. Additionally, these compounds are antibiotics or antifungals against some strains of bacteria and fungi. Some N-phenylbenzamide are anti-inflammatory [11]. Aniline derivatives of aromatic carboxylic acids, such as N-phenylbenzamide, also find application in agriculture as herbicides or pesticides because they have biological activity against weeds and pests [12].

In this study, the amidation reaction of

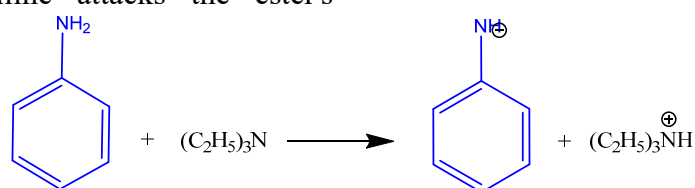
vinyl esters of various substituted aromatic carboxylic acids with aniline was carried out in a toluene solution in the presence of a catalytic amount of triethylamine. Using this method, the reaction of aniline with vinyl esters of benzoic acid, vinyl esters of 4-bromobenzoic acid, vinyl esters of 4-nitrobenzoic acid, vinyl esters of 4-tert-butylbenzoic acid, and vinyl esters of 4-chlorobenzoic acid led to the synthesis of amides of aromatic carboxylic acids — N-phenylbenzamide, 4-bromo-N-phenylbenzamide, 4-nitro-N-phenylbenzamide, 4-tert-butyl-N-phenylbenzamide, and 4-chloro-N-phenylbenzamide. The general reaction scheme is presented below (Scheme 1).



Scheme 1

Aniline reacts with vinyl esters of aromatic carboxylic acids by a nucleophilic substitution process. An amide bond is created when the amino group of aniline attacks the ester's

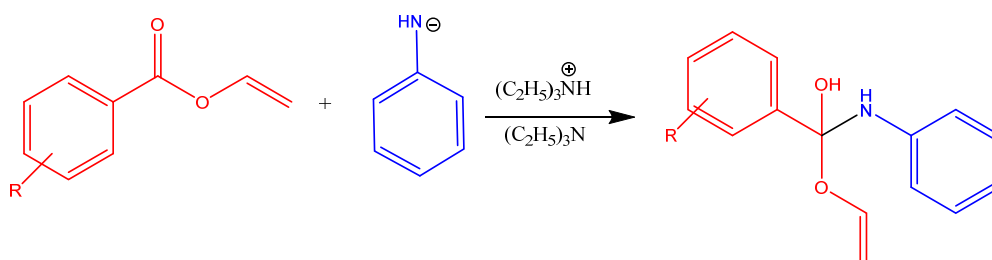
carbonyl atom. Triethylamine first increases aniline's nucleophilicity by deprotonating its amino group (Scheme 2).



Scheme 2

A tetrahedral intermediate complex is created when the deprotonated amine attacks the

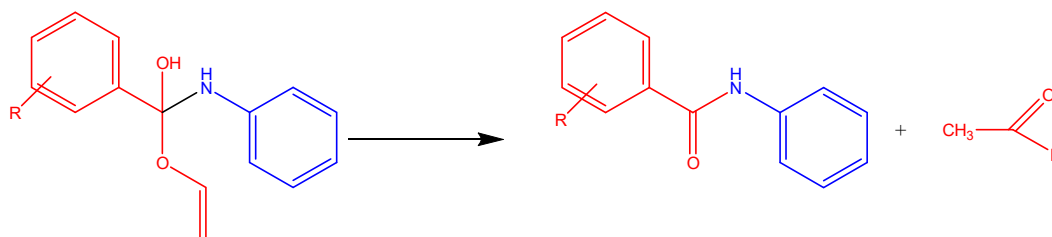
vinyl ester's carbonyl atom nucleophilically (Scheme 3).



Scheme 3

The equivalent aniline amide of the carboxylic acid is then formed as a result of the

release of acetic aldehyde from the tetrahedral intermediate complex (Scheme 4).



Scheme 4

Triethylamine not only deprotonates the amine but also neutralizes additional acidic by-products formed during the reaction intermediates. The structures of the synthesized amides obtained from the reaction of vinyl esters of carboxylic acids with aniline were confirmed

by ^1H and ^{13}C NMR spectral analysis. Below are the ^1H and ^{13}C NMR spectra of N-phenylbenzamide, which was obtained from the reaction of vinyl ester of benzoic acid with aniline.

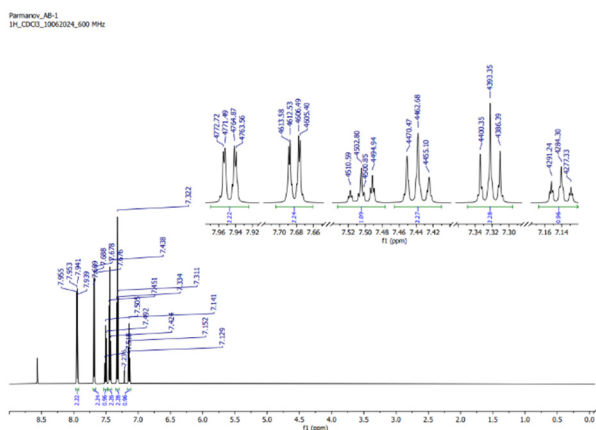


Fig. 1. ^1H -NMR spectrum of N-phenylbenzamide

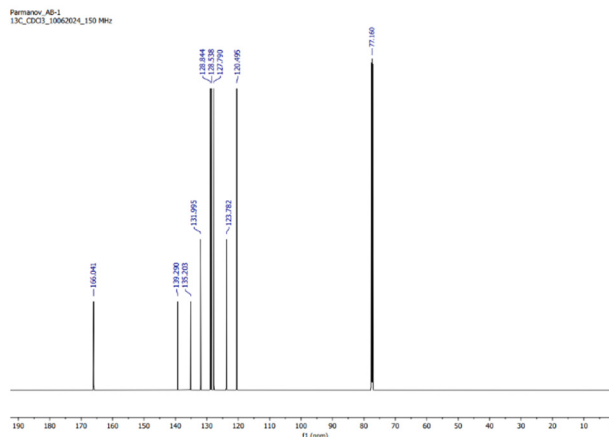


Fig. 2. ^{13}C -NMR spectrum of N-phenylbenzamide

In the ^1H -NMR spectrum of N-phenylbenzamide (Fig. 1), the signals of the benzoic acid aromatic ring protons appear in the 7.51–7.95 ppm region, while the signals of the protons in the aniline ring are observed in the 7.06–7.5 ppm region.

In the ^{13}C -NMR spectrum of N-phenylbenzamide (Fig. 2), the signal of the carbonyl carbon from the benzoic acid group appears at 166.6 ppm, while the signals of the

carbon atoms in the aromatic ring are observed in the 126–132 ppm region. The signal of the first carbon atom in the aniline ring is detected at 137.1 ppm, whereas the remaining carbon atoms appear in the 119–128 ppm region.

The reaction was carried out in a toluene solution at 60 °C for 4 hours. The starting vinyl esters of carboxylic acids and aniline were taken in a 1:1 molar ratio. The obtained results are presented in Table 1.

Table 1. Amidation reaction of vinyl esters of aromatic carboxylic acids with aniline

N $^{\circ}$	Vinyl esters	Product yield, %
1	$\text{C}_6\text{H}_5\text{COOCH}=\text{CH}_2$	65
2	$4\text{-Cl-C}_6\text{H}_4\text{COOCH}=\text{CH}_2$	70
3	$4\text{-Br-C}_6\text{H}_4\text{COOCH}=\text{CH}_2$	75
4	$4\text{-NO}_2\text{-C}_6\text{H}_4\text{COOCH}=\text{CH}_2$	80
5	$4\text{-(CH}_3)_3\text{C-C}_6\text{H}_4\text{COOCH}=\text{CH}_2$	60

A number of variables, such as the kind of substituents on the aromatic ring and the reaction conditions, affect the yield of amides produced by the amidation reaction of vinyl esters of different substituted aromatic carboxylic acids with aniline. The yield of the resultant amides is usually impacted by electron-donating groups (like tert-butyl) and electron-withdrawing groups (like bromine, chlorine, and nitro). It is evident from the data that a 65% yield of N-phenylbenzamide was produced when unsubstituted vinyl ester of benzoic acid reacted with aniline. Cl, Br, and NO₂ are electron-withdrawing groups that raise the positive charge on the carbonyl carbon, which makes it easier for the amino group to attack nucleophilically. Consequently, 4-bromo-N-phenylbenzamide yielded 75%, 4-chloro-N-phenylbenzamide yielded 70%, and 4-nitro-N-phenylbenzamide yielded 80%. Conversely, groups that donate electrons, such as tert-butyl, lessen the carbonyl carbon's electrophilicity, which lowers the yield of the result. As a result, 60% of 4-tert-butyl-N-phenylbenzamide was produced.

The molecular docking method is an essential tool in contemporary pharmaceutical research for examining the interactions between biologically active substances and their target proteins. This method makes it possible to assess

a ligand's thermodynamic stability and capacity for binding to the protein. The stability of the ligand-protein complex is reflected in the binding free energy ($\Delta G_{\text{binding}}$) determined by docking software; the stronger the binding interaction, the more negative the value [13].

This work investigated how synthetic amide derivatives interacted with two distinct target proteins, 7C4S and 6LU7, both associated with SARS-CoV-2, using the AutoDock Vina algorithm. The 7C4S protein reflects some of the S (spike) protein of SARS-CoV-2, which is required for viral access into host cells. Conversely, 6LU7, the main protease of SARS-CoV-2, is absolutely essential for breaking down viral polyproteins during replication.

Using molecular docking, the binding energies of the synthetic compounds with each target protein—7C4S (sphingosine-1-phosphate receptor 3, S1PR3) and 6LU7 (SARS-CoV-2 main protease) - are shown below. Negative ΔG values indicate the thermodynamic stability of the ligand-protein complex formation; the energy values are stated in kilocalories per mole (kcal/mol). For every chemical, Table 2 methodically compiles the relative binding energy values.

Table 2. Binding energies of synthesized N-phenylbenzamides with 7C4S (S1PR3) and 6LU7

No	Names of N-phenylbenzamides	7C4S (S1PR3) Binding Energy (kcal/mol)	6LU7 Binding energy (kcal/mol)
1	N-Phenylbenzamide	-9.2	-6.2
2	4-Nitro-N-phenylbenzamide	-9.4	-6.8
3	4-Bromo-N-phenylbenzamide	-9.5	-6.3
4	4-Chloro-N-phenylbenzamide	-6.8	-6.6
5	4-Tert-butyl-N-phenylbenzamide	-10.5	-6.7

With a value of -10.5 kcal/mol, 4-tert-butyl-N-phenylbenzamide displayed the lowest (i.e., strongest) binding energy among the binding energies of the compounds with the 7C4S (S1PR3) receptor. This outcome suggests that with S1PR3, this molecule can create the most energetically favorable and stable complex. Among high-affinity ligands are also 4-bromo-N-phenylbenzamide (-9.5 kcal/mol) and 4-nitro-

N-phenylbenzamide (-9.4 kcal/mol). Their binding energies are rather higher than those of 4-tert-butyl-N-phenylbenzamide, yet they still show good binding. By contrast, 4-chloro-N-phenylbenzamide displayed the lowest binding with the 7C4S receptor at -6.8 kcal/mol. This value indicates lesser interactions between the ligand and the receptor since it is much higher than that of the other compounds.

In terms of binding energy with the 6LU7 enzyme, 4-nitro-N-phenylbenzamide exhibited the lowest energy at -6.8 kcal/mol. Additionally, 4-tert-butyl-N-phenylbenzamide (-6.7 kcal/mol) and 4-chloro-N-phenylbenzamide (-6.6 kcal/mol) also showed relatively strong binding with 6LU7. These values are close to each other, indicating that these compounds have very similar affinity for the enzyme. N-Phenylbenzamide showed the weakest binding

energy to 6LU7 at -6.2 kcal/mol. The -6.3 kcal/mol value of 4-bromo-N-phenylbenzamide also indicates low affinity.

Thus, when considering all the docking results collectively, 4-tert-butyl-N-phenylbenzamide stands out as the most noteworthy compound. It binds most strongly to the S1PR3 receptor and also demonstrates a high binding affinity to the 6LU7 protease (Fig. 3).

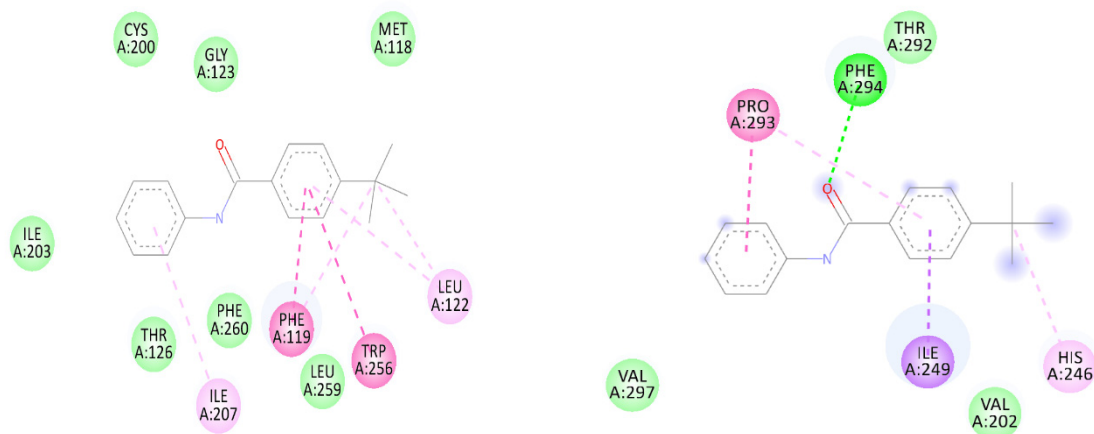


Fig. 3. Comparison of molecular docking poses of 4-tert-butyl-N-phenylbenzamide with (A) S1PR3 receptor (PDB ID: 7C4S) and (B) SARS-CoV-2 main protease (PDB ID: 6LU7). Interactions include hydrogen bonding and hydrophobic contacts. Ligands are represented in stick model

Conclusion

The amidation reaction between aniline and several substituted vinyl esters of aromatic carboxylic acids was examined in this work. The reaction was conducted for four hours at 60 °C in a toluene solution with a catalytic quantity of triethylamine present. The results showed that the product yield is greatly influenced by the type of substituents on the aromatic ring. The carbonyl group's electrophilicity was enhanced by electron-withdrawing groups (Br, Cl, and NO_2), which made amide production easier. Consequently, large yields of 4-nitro-N-phenylbenzamide (80%), 4-chloro-N-phenylbenzamide (70%), and 4-bromo-N-phenylbenzamide (75%) were produced. Conversely, the electron-donating tert-butyl group reduced the electrophilicity of the carbonyl carbon, decreasing reaction efficiency, which was confirmed by the 60% yield of 4-tert-butyl-N-phenylbenzamide. In the amidation reaction of unsubstituted vinyl ester of benzoic acid, N-phenylbenzamide was obtained with a 65% yield.

By using ^1H and ^{13}C NMR spectral analysis, the structures of the synthesized amides were verified. This process offers a viable way to synthesize amide derivatives of aromatic carboxylic acids efficiently. Because of its high efficiency and ease of use, it may find use in both pharmaceutical synthesis and practical chemistry. If the research goal is to select the best ligand for a single target, the following compounds are preferred: for S1PR3 – 4-tert-butyl-N-phenylbenzamide, for 6LU7 – 4-nitro-N-phenylbenzamide.

As a future direction, it is advisable to conduct in-depth experimental studies on the biochemical activity, toxicological profile, and pharmacokinetic properties of these compounds. The docking calculations suggest that the structure of 4-tert-butyl-N-phenylbenzamide is highly adaptable to the selected target proteins and capable of performing functional interactions, making it a promising candidate for practical use as an inhibitor.

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Conflicts of Interest: The authors have disclosed that they have no conflicts of interest.

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