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Design and Two New Indol-Steroid Derivatives to Evaluate their Theoretical Activity Against Protein Kinase 2 (CK2) Protein

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Abstract: Several compounds have been developed to evaluate their interaction with CK2-protein surface using some docking models. The objective of this investigation was to prepare two indol-steroid derivatives from 6β-nitroprogesterone using some chemical strategies. In addition, the interaction of both compounds 3 and 6 with CK2-protein was evaluated in a docking model using quinalizarin as tool. The results showed that either compounds 3 or 6 have a higher affinity by 3FL9 protein surface compared with quinalizarin. In conclusion, this phenomenon suggests that either compounds 3 or 6 could exert changes in the biological activity of CK2 protein.

Keywords: Indole; steroid; derivative; 6β-nitroprogesterone; CK2-protein.

1. Introduction

Cancer is a main risk factor of death worldwide [1, 2]; it is noteworthy that some data suggest that protein kinase 2 (CK2) may be related to several types of cancer [3, 4]. For example, some studies showed that CK2 might produce indirectly neoplastic growth through oncogenes activation [5, 6]. Other data showed that CK2 could be involved in some mutations of the cell division cycle via CDC37 (co-chaperone) activation [7]. Another study showed that CK2 could increase breast cancer through nuclear factor-κB phosphorylation [8]. Furthermore, a report indicates that CK2 can regulate Wnt signaling pathways, increasing transcriptional activity; in this way, CK2 can phosphorylating some biological target such as Dvl-protein [9], β-catenin [10], TCF/LEF transcription factors [11] which could be involved in an oncogenesis process [12-14].

On the other hand, to decrease the biological activity of CK2 in patients with cancer have used several CK2- inhibitors such as benzimidazole [15], TBB (4,5,6,7-tetrabromo-2-azabenzimidazole) [16], heparin [17], emodin [18], quinalizarin [19]. Here, it is essential to mention that several compounds have been developed as CK-inhibitors; for example, the preparation of 5-anilinopyrazolo[1,5-a]pyrimidine from 7-oxetan-3-yl amino derivative to

evaluate their biological activity against CK2 *in vitro* [20]. In addition, a 3-cyano-5-aryl-7-aminopyrazolo[1,5-a]pyrimidine was prepared, which showed biological activity against CK2 in HCT-116 cells [21]. Other data showed the synthesis of a pyrroloquinoxaline as CK2-inhibitor on immature lymphocytes [22].

To evaluate the biological activity of several compounds against CK2 a series of theoretical studies have be carried out. For example, a report showed the pharmacophore identification and validation study for some CK2-inhibitors using the CoMFA and CoMSIA methods [23]. Other data showed the preparation of a pharmacophore model for an Indeno[1,2-b]indole derivative as a human protein kinase CK2 Inhibitor using MOE software [24]. Additionally, a study showed the identification of some pharmacophore for CK2 inhibitors using a Bayesian model [25]. Other reports showed the pharmacophore generation for some CK2-inhibitors using LigandScout software [26]. Recently, a pharmacophore was prepared to evaluate the interaction of a steroid derivative with CK2-protein using the LigandScout software [27]. All these data suggest the preparation of several CK2-inhibitors; however, the interaction of some drugs with CK2-protein is very confusing, perhaps this phenomenon could be due to; (1) differences in the chemical structure of each drug; or (2) to different methods used in each theoretical experimental. Analyzing all these data, the objective of this investigation was to prepare two indol-steroid derivatives to evaluate their interaction with CK2-protein, a docking model.

2. Materials and Methods

2.1. General methods.

6β-nitroestrone (compound 3) was prepared using a previously method reported [28]. The other reagents used in this investigation were acquired from Sigma-Aldrich Co., Ltd. The melting point for compounds was evaluated on an Electrothermal (900 model). Infrared spectra (IR) were evaluated with a Thermo Scientific iSOFT-IR spectrometer.^{1}H and ^{13}C NMR spectra were recorded using a Varian VXR300/5 FT NMR spectrometer at 300 MHz in $CDCl_3$ using TMS as internal standard. EIMS spectra were obtained with a Finnigan Trace Gas Chromatography Polaris Q-Spectrometer. Elementary analysis data were acquired from a Perkin Elmer Ser. II CHNS/02400 elemental analyzer.

2.2. Synthesis of a steroid-pirrol derivative.

1-[(3aS,6S)-6-ethyl-3-[1-(3-ethynylphenyl)-5-phenyl-2,3-dihy-dropyrrol-4-yl]-3a,6-dime-thyl-2,3,4,5,5a,7,8,9,9a,9b-decahy-dro-1H-cyclopenta[a]naphthalen-7-yl]propan-2-one (2)

In a round bottom flask (10 ml), progesterone (200 mg, 0.64 mmol), 3-ethynylaniline (100 µl, 0.90 mmol), Copper(II) chloride, iodine (170 mg, 0.67 mmol) and 5 ml of dimethyl sulfoxide were stirred at reflux for 12 h. Then, the solvent was evaporated under reduced pressure and following the product was purified via crystallization using the methanol:hexane:water (4:2:1) system; yielding 60% of product; m.p. 70-72 ^{o}C; IR (V_{max}, cm^{-1}) 2110 and 1712: ^{1}H NMR (300 MHz, $CDCl_3$-*d*) δ_H: 0.58 (s, 3H), 0.86 (s, 3H), 1.10-1.82 (m, 13H), 2.08-2.82 (m 6H), 2.88 (s, 1H), 2.94 (m, 1H), 2.98 (m, 1H), 3.02-4.04 (m, 3H), 4.94 (d, 1H, J = 1.90 Hz), 6.88-7.48 (m, 8H) ppm. ^{13}C NMR (300 Hz, $CDCl_3$) δ_C: 12.42, 19.06, 21.32, 24.60, 25.59, 31.27, 32.00, 32.62, 37.15, 37.34, 37.50, 38.06, 42.83, 46.92, 48.36, 53.44, 56.22, 56.44, 78.22, 84.00, 117.22, 122.62, 123.41, 123.59, 126.94, 127.22, 128.00,

129.00, 129.16, 129.80, 130.94, 138.66, 143.40, 145.20, 208.90 ppm. EI-MS m/z: 515.31. Anal. Calcd. for $C_{37}H_{41}NO$. C, 86.17; H, 8.01; N, 2.72; O, 3.10. Found: C, 86.14; H, 8.00.

2.3. Synthesis of an indol-steroid-pentacosa derivative.

(1S,22S)-21-[1-(3-ethynylphenyl)-5-phenyl-2,3-dihydropyrrol-4-yl]-1,22-dimethyl-10-azahexacyclo[12.11.0.03,12.04,9.017,25.018,22]pentacosa-3(12),4(9),5,7-tetraene (3)

In a round bottom flask (10 ml), compound 2 (200 mg, 0.39 mmol) phenylhydrazine (50 µl, 0.50 mmol), and 5 ml of acetic acid were stirred at reflux for 12 h. Then, the solvent was evaporated under reduced pressure and following the product was purified via crystallization using the methanol:water (4:1) system; yielding 55% of product; m.p. 80-82 ºC; IR (V_{max}, cm^{-1}) 3410 and 2112: ^{1}H NMR (300 MHz, $CDCl_3$-*d*) δ_H: 0.58 (s, 3H), 1.00 (s, 3H), 1.12-1.82 (m, 12H), 2.04-2.46 (m, 4H), 2.88 (s, 1H), 2.94-3.02 (m, 2H), 3.62-3.70 (m, 2H), 3.98-4.04 (m, 2H), 5.42 (d, 1H, J = 1.90 Hz), 6.88-6.98 (m, 2H), 7.12 (m, 1H), 7.14-7.20 (m, 3H), 7.22-7.42 (m, 3H), 7.44-7.48 (m, 4H), 7.80 (broad, 1H) ppm. ^{13}C NMR (300 Hz, $CDCl_3$) δ_C: 12.42, 18.34, 21.12, 22.40, 24.62, 25.59, 31.27, 31.84, 32.22, 34.12, 38.06, 40.54, 42.80, 46.92, 52.36, 56.22, 56.44, 78.22, 84.02, 111.42, 111.62, 117.22, 117.43, 118.50, 119.00, 120.96, 123.41, 123.59, 126.98, 127.16, 128.00, 128.14, 129.00, 129.16, 129.80, 130.96, 134.12, 136.44, 143.36, 143.76, 145.20 pp. EI-MS m/z: 588.35. Anal. Calcd. for $C_{43}H_{44}N_2$. C, 87.71; H, 7.53; N, 4.76. Found: C, 87.70; H, 7.50.

2.4. Synthesis of an indol-steroid-pentacosa derivative.

(10R,13S,17S)-17-[1-(3-ethynylphenyl)-5-phenyl-2,3-dihy-dropyrrol-4-yl]-10,13-dime-thyl-6-nitro-1,2,4,7,8,9,11,12,14, 15,16,17-dodecahydrocyclopenta[a]phenanthren-3-one (5)

In a round bottom flask (10 ml), 6β-nitroprogesterone (200 mg, 0.56 mmol), 3-ethynylaniline (100 µl, 0.90 mmol), Copper(II) chloride, iodine (170 mg, 0.67 mmol) and 5 ml of dimethyl sulfoxide were stirred at reflux for 12 h. Then, the solvent was evaporated under reduced pressure and following the product was purified via crystallization using the methanol:hexane:water (4:1:1) system; yielding 58% of product; m.p. 138-140 ºC; IR (V_{max}, cm^{-1}) 2110, 1712and 1538: ^{1}H NMR (300 MHz, $CDCl_3$-*d*) δ_H: 0.56 (s, 3H), 1.10 (m, 1H), 1.20 (s, 3H), 1.26-1.94 (m, 11H), 2.10-2.60 (m, 5H), 2.86 (s, 1H), 2.90 (m, 1H), 2.92-3.00 (m, 2H), 3.30-3.42 (m, 2H), 3.98-4.04 (m, 2H), 6.86-7.48 (m, 9H) ppm. ^{13}C NMR (300 Hz, $CDCl_3$) δ_C: 12.42, 18.40, 23.72, 24.63, 25.56, 30.52, 30.72, 31.27, 33.32, 37.36, 38.09, 40.50, 42.83, 44.20, 46.92, 54.70, 56.06, 56.46, 78.20, 84.02, 117.22, 123.40, 123.59, 126.96, 127.22, 128.04, 129.00, 129.16, 129.70, 129.80, 130.98, 131.94, 143.36, 145.20, 205.70 ppm. EI-MS m/z: 560.30. Anal. Calcd. for $C_{37}H_{40}N_2O_3$. C, 79.25; H, 7.19; N, 5.00; O, 8.56. Found: C, 79.22; H, 5.00.

2.5. Preparation of a nitro-indol-steroid derivative.

(1R,5S,6S)-6-[1-(3-ethynylphenyl)-5-phenyl-2,3-dihydropy-rrol-4-yl]-1,5-dimethyl-12-nitro-16-azahexacyclo[11.11.0.02,10.05,9.015,23.017,22]tetracosa-12,15(23), 17(22),18,20-penta-ene (6)

In a round bottom flask (10 ml), compound 5 (200 mg, 0.36 mmol) phenylhydrazine (50 µl, 0.50 mmol), and 5 ml of acetic acid were stirred at reflux for 12 h. Then, the solvent

was evaporated under reduced pressure and following the product was purified via crystallization using the methanol:water (4:1) system; yielding 53% of product; m.p. 172-174 ^{o}C; IR (V_{max}, cm^{-1}) 3412, 2110 and 1538: ^{1}H NMR (300 MHz, $CDCl_3$-*d*) δ_H: 0.56 (s, 3H), 0.98 (m, 1H), 1.10 (s, 3H), 1.22-1.92 (m, 10H), 2.36-2.80 (m, 7H), 2.86 (s, 1H), 2.88 (m, 1H), 2.92-3.00 (m, 2H), 3.10 (broad, 1H), 3.30 (m, 1H), 3.98-4.06 (m, 2H), 4.80-6.70 (m, 3H), 6.86-6.98 (m, 2H), 7.00 (m, 1H), 7.12-7.48 (m, 8H) ppm. ^{13}C NMR (300 Hz, $CDCl_3$) δ_C: 12.42, 19.96, 23.70, 24.63, 25.59, 25.96, 30.22, 30.70, 31.27, 35.60, 38.09, 41.12, 42.80, 46.02, 46.90, 56.04, 56.44, 56.60, 60.34, 78.22, 84.02, 108.36, 116.27, 117.22, 122.24, 123.40, 123.59, 126.98, 127.04, 127.20, 128.02, 128.92, 129.00, 129.00, 129.16, 129.80, 130.84, 130.90, 130.98, 143.40, 145.20, 150.20 ppm. EI-MS m/z: 635.35. Anal. Calcd. for $C_{43}H_{45}N_3O_2$. C, 81.23; H, 7.13; N, 6.61; O, 5.03. Found: C, 81.20; H, 7.10.

2.6. Pharmacophore model.

A pharmacophore for both compounds 3 and 6 were developed using LigandScout software [29].

2.7. Protein-ligand interaction.

The interaction of both compounds 3 and 6 with CK2-protein surface was evaluated using 3FL5-protein [30] as a tool. Furthermore, both Chimerax [31] and Achilles Blind Docking Server models [32] were used to calculate both binding energy and distance between amino acid residues of 3FL5-protein and both compounds 3 and 6.

2.8. Pharmacokinetics parameter.

To evaluate some pharmacokinetic factors involved in the chemical structure of either compounds 3 or 6, the SwissADME software was used [33].

3. Results and Discussion

Have been prepared several compounds as CK2-inhibitors; however, some of the protocols use some reagents which are dangerous and require special conditions [15-22]. Also, the interaction with CK2 protein is very confusing; perhaps, this phenomenon could be to different structure chemical of each compound. Analyzing these data, in this investigation, two indol-steroid derivatives were synthesized to evaluate their interaction with CK2 protein using several strategies as follows.

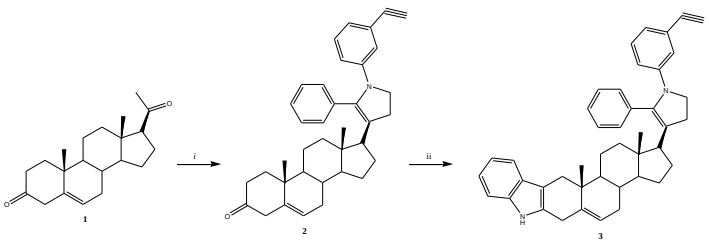

Figure 1. Synthesis of an indol-steroid-pentacosa derivative (**3**). Reagents and conditions: *i* = progesterone, 3-ethynylaniline, Copper(II) chloride, iodine, dimethyl sulfoxide; *ii* = phenylhydrazine, acetic acid.

3.1. Preparation of a steroid-pirrol derivative.

There are some reports for preparation of pirrol derivatives using several reagents such as tetrakis(triphenylphosphine)palladium(0) [34], $K_2S_2O_8$/(2,2,6,6/Tetramethyl-1-pi-peridin-

yloxy) [35], $Cu(Oac)_2$ [36], phosphoric acid [37] and others. In this investigation, the synthesis of a steroid-pirrol derivative (compound 2) was prepared from progesterone, 3-ethynylaniline, acetone in the presence of dimethyl sulfoxide (Figure 1).

The ^1H NMR spectrum of 2 showed several signals at 0.58-0.86 ppm for methyl groups bound to steroid nucleus; at 1.10-2.82, 2.98, and 4.94 ppm for steroid moiety; at 2.88 ppm for alkyne group; at 2.94, 3.02-4.04 ppm for 2,3-Dihydro-1*H*-pyrrole ring; at 6.88-7.48 ppm for phenyl groups. Besides, the ^{13}C NMR spectra display chemical shifts at 12.42-19.06 ppm for methyl groups linked to steroid nucleus; at 21.32-25.59, 32.00-56.22, 122.62 and 138.66 ppm for steroid moiety; at 31.27, 56.44, 129.16 and 143.20 ppm for 2,3-Dihydro-1H-pyrrole ring; at 78.22-84.00 ppm for alkyne group; at 117.22, 123.41-129.00, 129.80-130.94 and 145.20 ppm for phenyl groups. Additionally, the mass spectrum from 2 showed a molecular ion (m/z) 208.90.

3.2. Synthesis of an indol-steroid-pentacosa derivative.

Several indol derivatives have been prepared using some reagents such as $CoCl_2/Ag_2CO_3/Et_3N$ [38], $Cu(Oac)_2$/1,1'-Bis(diphenylphosphino)ferrocene [39], CuI/N,N'-Dimethylethyl-enediamine [40], $Rh_2(Oac)_4$ [41], tetramethyl thiourea [42] and others. In this study, the compound 2 reacted with phenylhydrazine in the presence of acetic acid to form indol-steroid-pentacosa derivative (3). The ^1H NMR spectrum of 3 (Figure 1) showed several signals at 0.58-1.00 ppm for methyl groups bound to steroid nucleus; at 1.12-2.46, 3.62-3.70 and 5.42 ppm for steroid moiety; at 2.88 ppm for alkyne group; at 2.94-3.02 and 3.98-4.04 ppm for 2,3-Dihydro-1H-pyrrole ring; at 6.88-6.98, 7.14-7.20 and 7.44-7.48 ppm for phenyl groups; at 7.12, 7.22-7.42 ppm for indol ring. Besides, the ^{13}C NMR spectra display chemical shifts at 12.42-18.34 ppm for methyl groups linked to steroid nucleus; at 21.12-25.59, 31.84-56.22, 117.43 and 143.76 ppm for steroid moiety; at 31.27, 56.44, 129.16 and 143.36 ppm for 2,3-Dihydro-1H-pyrrole ring; at 78.22-84.02 ppm for alkyne group; at 111.42-11.62, 118.50-120.96, 128.14 and 134.12-136.44 ppm for indole ring; at 117.22, 123.41-128.00, 129.00, 129.80-130.96 and 145.20 ppm for phenyl groups. Finally, the mass spectrum from **3** showed a molecular ion (m/z) 588.35.

3.3. Synthesis of a 2,3-dihydropyrrolyl-steroid-3-one derivative.

The 2,3-dihydropyrrolyl-steroid-3-one analog was prepared from 6β-nitroprogesterone, 3-ethynylaniline, Copper(II) in the presence of dimethyl sulfoxide (Figure 2).

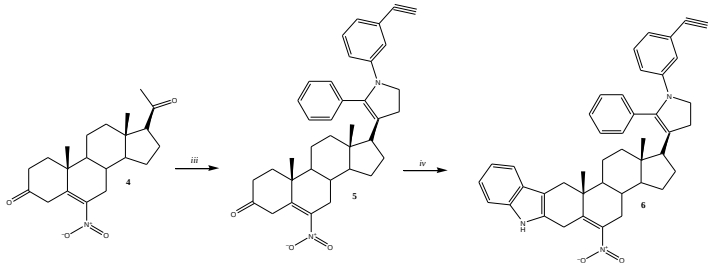

Figure 2. Preparation of a nitro-indol-steroid derivative (**6**). reagents and conditions: *iii* = 6β-nitroprogesterone, 3-ethynylaniline, Copper(II) chloride, iodine, dimethyl sulfoxide; *iv* = phenylhydrazine, acetic acid.

The ^1H NMR spectrum of 5 showed several signals at 0.56-1.20 ppm for methyl groups bound to steroid nucleus; at 1.10, 1.24-1.94, 2.10-2.60, 2.90 and 3.30-3.42 ppm for steroid moiety; at 2.86 ppm for alkyne group; at 2.92-3.00 and 3.98-4.04 ppm for 2,3-Dihydro-1H-

pyrrole ring; at 6.86-7.48 ppm for phenyl groups; at 7.12, 7.22-7.42 ppm for indol ring. Also, the ^{13}C NMR spectra display chemical shifts at 12.42-18.40 ppm for methyl groups linked to steroid nucleus; at 23.72-30.72, 33.32-56.06, 129.70 and 131.94 ppm for steroid moiety; at 31.27, 56.46, 129.16 and 143.36 ppm for 2,3-Dihydro-1H-pyrrole ring; at 78.20-84.02 ppm for alkyne group; at 117.22-129.00, 129.80-130.98 and 145.20 ppm for phenyl groups; at 205.70 ppm for ketone group. In addition, the mass spectrum from 5 showed a molecular ion (m/z) 588.35.

3.4. Preparation of a nitro-indol-steroid derivative.

Finally, a nitro-indol-steroid derivative (compound 6) was synthesized via the reaction of 5 with phenylhydrazine in the presence of acetic acid to form 6 (Figure 2). The ^{1}H NMR spectrum of 6 (Figure 1) showed several signals at 0.58-1.10 ppm for methyl groups bound to steroid nucleus; at 0.98 and 1.22-2.80 ppm for steroid moiety; at 2.88 ppm for alkyne group; at 2.92-3.00 and 3.98-4.04 ppm for 2,3-Dihydro-1H-pyrrole ring; at 3.10 ppm for the amino group; at 3.30-4.80-6.70 and 7.00 ppm for indole ring; at 6.86-6.98 and 7.12-7.48 ppm for phenyl groups. The ^{13}C NMR spectra display chemical shifts at 12.42-19.96 ppm for methyl groups linked to steroid nucleus; at 23.70-30.70, 35.60-56.04, 56.60-60.34 and 130.84-130.90 ppm for steroid moiety; at 31.27, 56.44, 129.16 and 143.40 ppm for 2,3-Dihydro-1H-pyrrole ring; at 78.22-84.02 ppm for alkyne group; at 108.36-116.27, 122.24, 127.04, 128.92 and 150.20 ppm for indole ring; at 117.22, 123.40-126.98, 127.20-128.02, 129.00, 129.80-130.96 and 145.20 ppm for phenyl groups. Finally, the mass spectrum from **6** showed a molecular ion (m/z) 6.35.35.

3.5. Pharmacophore evaluation.

Several pharmacophore models have developed to describe the three-dimensional orientation adopted by the functional groups of some drugs, which could be bound to different biomolecules [43].

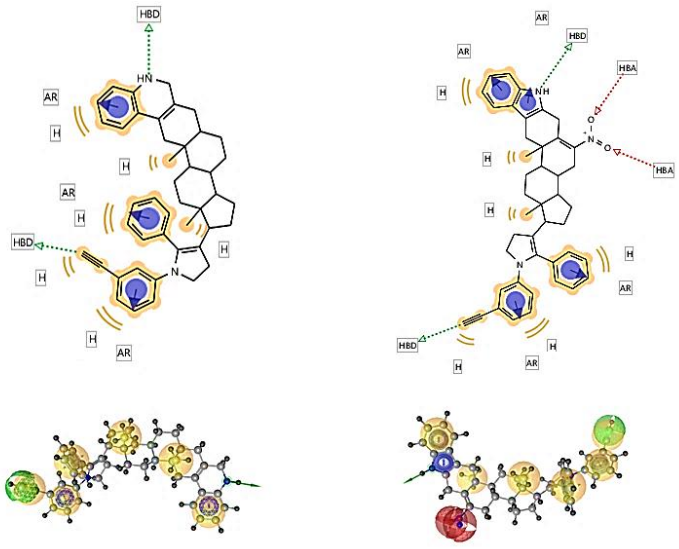

Figure 3. Pharmacophore from both compounds **3** (left) and **6** (right) using the LigandScout software. The model involves a hydrogen bond acceptor (HBA, red) and hydrogen bond donor (HBD, green).

Analyzing these data, in this investigation, the LigandScout software [29] was used to prepare a pharmacophore model for both compounds 3 and 6. The results found (Figure 3) showed that functional groups involved in the chemical structure of both compounds 3 and 6

could interact through hydrophobic contacts or as hydrogen bond acceptors or as hydrogen bond donor with the CK2-protein surface (Table 1). However, it is essential to mention that some studies suggest that the interaction of several drugs with some proteins could be conditioned by the different types of amino acid residues involved in the protein surface [30].

Table 1. Physicochemical parameters involved in the chemical structure of both compounds **3** and **6**.

Parameter	Compound 3 ($C_{44}H_{48}N_2$)	Compound 6 ($C_{43}H_{43}N_3O_2$)
cLogP	10.84	10.04
TPSA	20.72	72.53
HBA	0	2
HBD	2	2

3.6. Protein-ligand interaction.

There are studies that indicate that CK2 protein can be the target of several drugs [44]. To predict these interactions, some methods have been used, such as Autodock [45], Dock 6.1 [46], Dockingserver [47], and others. Analyzing these data in this study, the theoretical interaction of both compounds 3 and 6 with CK2-protein surface was evaluated using both 3FL5-protein and quinalizarin (an CK2-inhibitor) [48] as theoretical tools. Furthermore, Chimerax software [31] and Achilles' blind docking server [32] were used to evaluate the interaction of both compounds 3 and 6 with the 3FL5-protein surface. The results showed a different type of amino acid residues involved in the interaction of quinalizarin and both compounds 3 and 6 with 3FL5-protein surface (Figures 4 and 5; Tables 2-7); it is noteworthy that probably the Arg47 aminoacid residue could interact with the amino group of indole for compound 3 via a hydrogen bond. Furthermore, the Val45 aminoacid residue could interact with both amino and nitro groups through a hydrogen bond for compound 6. These phenomena could be translated as low activation energy (-9.80 Kcal/mol) for compound 3 compared with 6 (-9.20 Kcal/mol) and quinalizarin (-6.80 Kcal/mol). All these data suggest that both compounds 3 and 6 could induce greater changes in the biological activity of CK2-protein compared with quinalizarin.

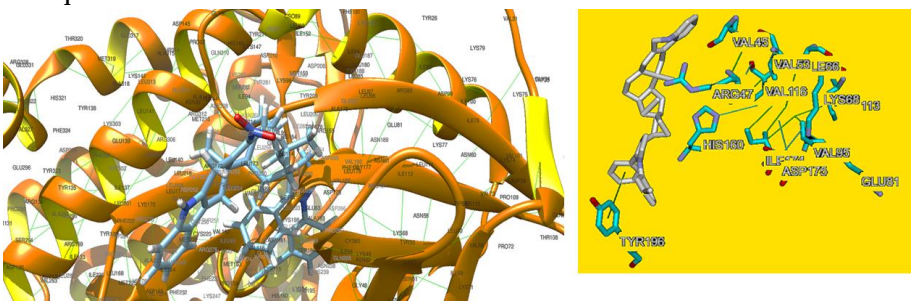

Figure 4. Interaction of compound **3** with 3FL5-protein surface using Chimerax software (left) and Achilles blind docking server (right).

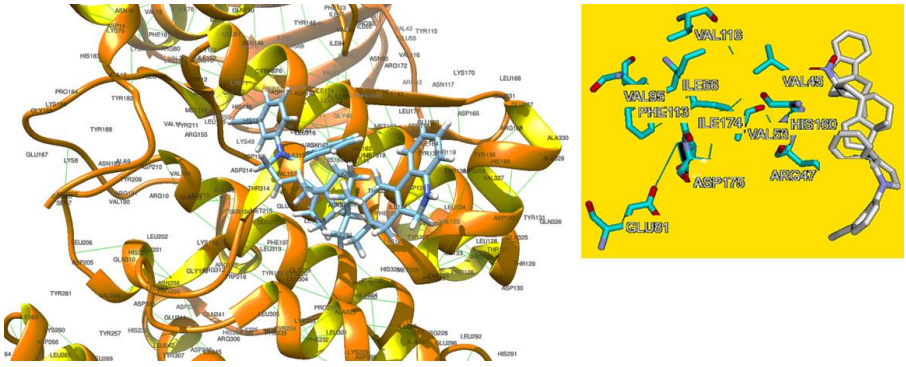

Figure 5. Binding of compound **6** with 3FL5-protein surface using Chimerax software (left) and Achilles blind docking server (right).

Table 2. Interaction of hydrophobic involved between compound **3** and CK2 protein.

Aminoacid residue	Ligand Carbon	Distance
Lys_{44}	35	3,69
Phe_{121}	42	3.64
Pro_{159}	43	3.74
Tyr_{196}	8	3.55
Tyr_{196}	6	3.62
Phe_{197}	44	3.95
Phe_{197}	43	3.56

.

Table 3. Hydrogen bonds of compound **3** and CK2 protein (3FL5).

Aminoacid residue	Distance H-A	Distance D-A	Don angle
Arg_{47}	2.15	2.01	121.43

Table 4. Interaction of hydrophobic involved between compound **6** and CK2 protein surface (3FL5).

Aminoacid residue	Ligand Carbon	Distance
Lys_{49}	5	3.96
Phe_{121}	43	3.21
Phe_{121}	22	3.34
Lys_{158}	45	3.36
Pro_{159}	44	3.82
Tyr_{196}	9	3.54

Table 5. Hydrogen bonds of compound **6** and CK2 protein surface (3FL5).

Aminoacid residue	Distance H-A	Distance D-A	Don angle
Val_{45}	2.15	2.81	121.43

Table 6. Interaction of hydrophobic involved between quinalizarin and CK2 protein surface.

Aminoacid residue	Ligand Carbon	Distance
Leu_{249}	1	3.29
Val_{256}	13	3.10
Tyr_{307}	6	3.07

Table 7. Hydrogen bonds of compound quinalizarin and CK2 protein surface (3FL5).

Aminoacid residue	Distance H-A	Distance D-A	Don angle
Leu_{249}	3.47	3.90	109.02
Arg_{278}	2.35	2.81	106.65
Arg_{278}	3.34	3.70	102.43
Tyr_{307}	2.93	3.40	109.31
Asp_{308}	3.20	4.00	144.59
Asp_{308}	2.49	3.49	166.47

3.7. Pharmacokinetic evaluation.

There are several studies to evaluate some pharmacokinetic parameters of several drugs using theoretical models such as PKQuest [49], PharmPK [50, 51] Gitub [52], SwissADME [33]. In this way, in this study, some pharmacokinetic parameters involved in both compounds **3** and **6** were evaluated using SwissADME software. The results showed in Table 8 indicate that these compounds could have low gastrointestinal absorption and, consequently, low metabolism exerted by the cytochrome P450 system. These data suggest that these compounds should be administered using other vias in some biological models, such as happening with other drugs [52].

Table 8. Pharmacokinetic parameters.

Parameter	Compound 3	Compound 6
GI absortion	Low	Low
BBB permeant	No	No
Pg-substrate	No	No
CYP1A2	No	No
CYP2C19	No	No
CYP2C9	No	No
CYP2D6	No	No
CYP3A4	No	No
Log K_p (skyn permeation)	-2.65 cm/seg	-3.18 cm/seg

4. Conclusions

In this study, the facile synthesis of two indol-steroid derivatives using several chemical strategies is reported. In addition, Theoretical analysis of the interaction between two indol-steroid derivatives showed a higher affinity of compounds **3** and **6** by the 3FL5 protein compared with quinalizarin, which is translated as a possible inhibition of the biological activity of CK2 protein.

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Conflicts of Interest

The authors declare no conflict of interest.

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