7-(3-Benzyloxypropyl)apigenin, A Novel Cytotoxic Flavone Derivative

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In recent years, there has been a resurgence of scientific interest in flavonoids, which is due to the association of these compounds with a wide range of health promoting effects.[1,2] Among those, much of the interest in flavonoids has recently been focused on their anticancer properties.[3-5] Different mechanisms have been linked to flavonoid-mediated cytotoxicity, including their antioxidant activities, their effects on signal transduction pathways involved in cell proliferation and angiogenesis, as well as their modulation of aromatase activity, a key enzyme involved in estrogen biosynthesis, and the enzymes required for metabolic activation of procarcinogens and the detoxification of carcinogens.[2,6]

Even though multiple mechanisms account for flavonoid-induced cytotoxicity, disruption of the signal transduction pathway for cell proliferation by inhibition of the cyclin-dependent kinases (CDKs) has drawn special attention since the discovery of a synthetic flavonoid flavopiridol as a potent anticancer agent.[7,8] Flavopiridol demonstrated potent in vitro inhibition of all cyclin-dependent kinases (CDKs) tested (CDKs 1, 2, 4 and 7), and clearly blocked cell cycle progression at the G1/S and G2/M boundaries. However, in spite of flavopiridol's potent activity, two major challenges remain: more potent and specific inhibition of the kinase targets.

The crystal structure of the flavopiridol analogue, deschloroflavopiridol, co-crystallized with CDK2,[9] reveals the key hydrogen bonds. In comparison with the other CDK2 inhibitors,[10] desflavopiridol has additional hydrogen bonds around the C8-substituent (Fig. 1). As a result, in order to enhance the selectivity and binding affinity of the flavopiridol analogues against the target kinase, structural modifications at the C2 and C8 positions of the flavone core have been extensively pursued.[10a,11,12]

However, little effort has been made to modify other positions of the flavone scaffold such as the most reactive nearby O7 position.[13] In our previous virtual screening study against CDK2,[14] we found a possible hydrophobic binding pocket composed of Phe80 and Phe146 inside the ATP-binding site (yellow ellipse, Fig. 2). Intriguingly, docking study of several flavonoid derivatives suggested that flavonoids with O7-substituent with optimum chain length would probably fit into this pocket (Fig. 2) to show increases in both selectivity and binding affinity against CDK2.

Thus, in this study, we designed a flexible propyloxybenzyl linker which would locate its terminal aromatic ring in the proposed hydrophobic pocket. Also, an oxygen atom placed in the middle of the propyloxybenzyl linker is expected to have a dual role: formation of an additional hydrogen bonding interaction with backnone NH of Asp145 and enhancement of the solubility of the flavonoid.

For the synthesis of the title compound, commercially available apigenin was initially used as a starting material, but alkylation under basic conditions usually provided dialkylated product at the 7-O and 4-O positions (data not shown). Thus, the synthetic plan was changed to regioselective alkylation of flavanone naringenin (**1**, Fig. 3) followed

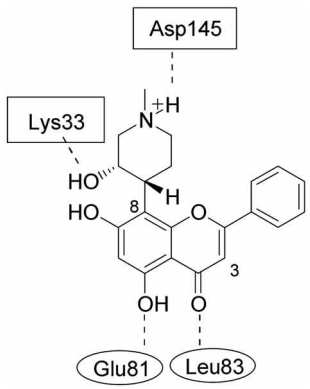

Figure 1. Key (ellipse) and additional (box) H-bonding interactions between CDK2 and deschloroflavopiridol.

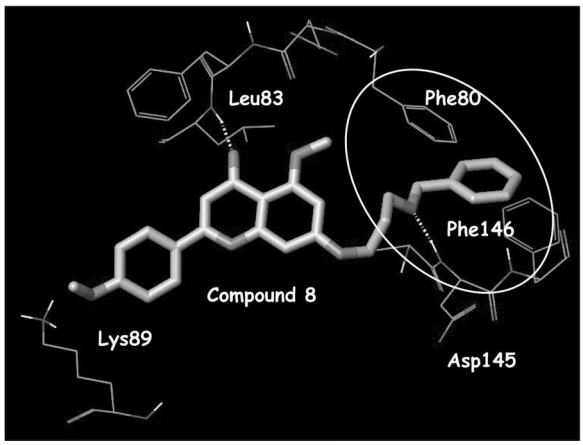

Figure 2. A plausible hydrophobic pocket composed of two amino acids with aromatic side chains (Phe80 and Phe146, inside the ellipse) and docking pose of the title compound **8**.

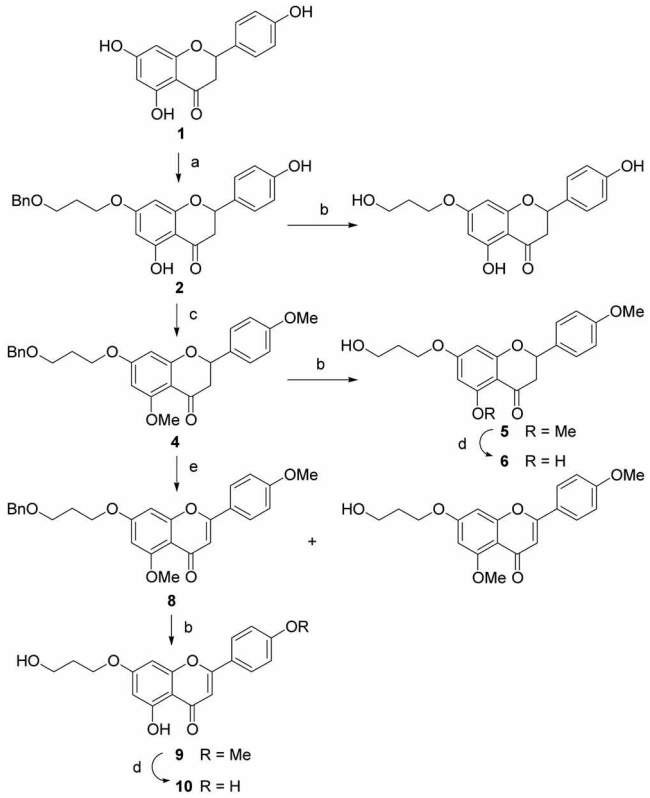

Figure 3. Preparation of naringenin and apigenin ethers. Reagents and Conditions: (a) $I(CH_2)_3OBn$, K_2CO_3, acetone-DMF (1:1), rt; 55%; (b) Pd/C, H_2, MeOH, rt; 46%; (c) Mc_2SO_4, K_2CO_3, acetone, rt; 54%; (d) BBr_3, CH_2Cl_2, 0 °C; 50% for **6**, 62% for **9**, 65% for **10**; (e) DDQ, 1,4-dioxane, acetic acid, rt, 42% for **7**, 41% for **8**.

by DDQ-oxidation to introduce a double bond between C2 and C3 (Fig. 3). 3-Benzyloxypropyl iodide prepared from 1,3-propanediol in three steps was reacted with naringenin in the presence of K_2CO_3 to give O7-alkylated naringenin derivative **2** in 55% yield. However, use of excess base and/or prolonged reaction time always provided a complex mixture of naringenin derivatives and chalcone derivatives formed by ring-opening reaction of the flavanone C-ring. Debenzylation and methylation of **2** provided free naringenin 7-O-propyloxy ether **3** and 5,4′-dimethoxy derivative of **2** (**4**) in 46% and 54% yield, respectively. Hydrogenolysis of the terminal benzyl group of **4** provided naringenin derivative **5** of which 5-OMe group was selectively removed by Lewis acid-mediated demethylation to give **6** in 50% yield. Unsaturation of the C2-C3 bond of **4** by DDQ provided the title compound **8** along with the debenzylated product **7** in 1:1 ratio. Hydrogenolysis of **8** resulted in debenzylation and concurrent demethylation at O5 to give **9** which was converted to the free apigenin derivative **10** by ensuing demethylation (Fig. 3).

To make sure the role of the terminal aromatic ring, it was of interest to compare the biological activities of the title compound **8** with debenzylated analogues **7**, **9** and **10**. First, continuous kinetic monitoring of CDK2 enzyme activity was performed and the result is summarized in Figure 4.

As expected, among the newly prepared flavonoid derivatives, the target molecule **8** showed the best inhibitory activity against the target enzyme (Fig. 4). Intriguingly, the naringenin analogue of **8**, compound **4**, was not active at all, which might result from the conformational differences conferred by the lack of the C2-C3 double bond in naringenin. Compared with the flavopiridol derivatives which show enzyme inhibition at the micromolar range,[10a,11,12] the activities of the flavone derivatives are not good enough. However, it is worth to note that the title compound was more active than apigenin and its 3-benzyloxypropyl linker (**8**) is far better than the 3-propoxy linker (**9** and **10**), which implies that there must be a hydrophobic binding site outside of the ATP binding site to specifically accommodate a benzyl group of **8**.

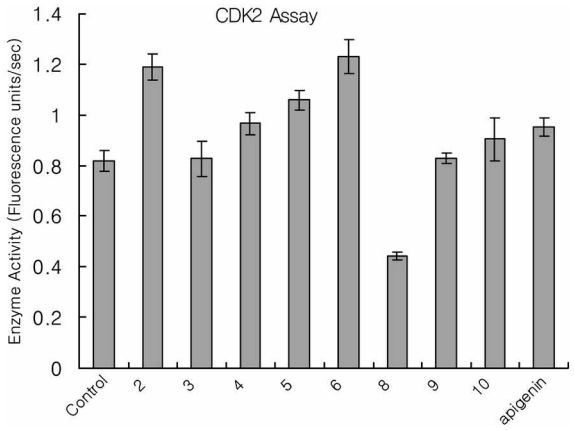

Figure 4. CDK2 activity after treatment with compounds **2-10** (20 μM) and apigenin. Due to the low solubility, the CDK2-inhibitory activity of the compound **7** was not estimated.

The cell viability of the newly synthesized flavonoid derivatives against Huh-7 cell were then determined using the MTT assay,[15] and the results are summarized in Figure 5.

As anticipated from the CDK2 inhibition assay, the fully protected apigenin ether **8** showed potent cytotoxicity whereas other derivatives did not give significant influence on the cell viability. The most surprising result in this study is that the flavone derivative **8** showed potent cytotoxic effect in spite of its moderate inhibitory activity against CDK2. The result is even more surprising when the com-

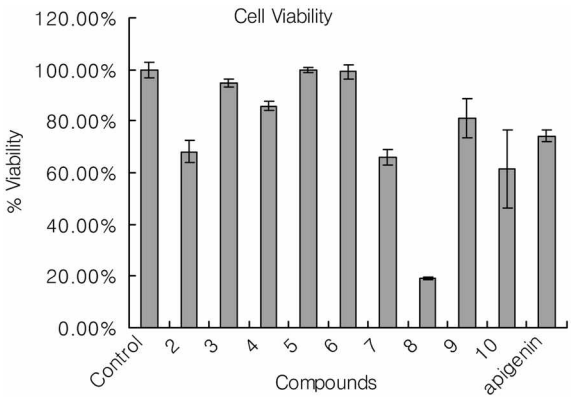

Figure 5. Cell viability assay results for compounds **2-10** (50 μM) and apigenin.

pounds **7** and **8** which differ only in the terminal benzyl group are compared. These two compounds show two folds difference in activity in the enzyme assay (Fig. 4), but the difference becomes six folds in the cell-based assay (Fig. 5). Thus, as many other cellular kinases have the similar ATP-binding sites, it is rational to suspect the possible involvement of other enzymes as secondary targets, and enzyme inhibition assays targeting against various kinases such as GSK-IIIβ, MAPK and PKC are in progress. Also, as the O7-substitution employs simple and straightforward synthetic routes, in order to optimize biological activity of the title compound **8**, construction of a compound library with various O7-linkers is underway.

Experimental Section

Syntheses and Characterization of 7-(3-Benzyloxy-propoxy)-5-methoxy-2-(4-methoxy-phenyl)-chromen-4-one (8) and Its Analogues.

7-(3-Benzyloxy-propoxy)-5-hydroxy-2-(4-hydroxy-phenyl)-chroman-4-one (2): To a solution of naringenin (**1**, 710 mg, 2.6 mmol) in a 1:1 mixture of acetone/DMF (6 mL) were added K_2CO_3 (432 mg, 3.1 mmol) and $I(CH_2)_3OBn$ (546 mg, 3.1 mmol). The reaction mixture was stirred for 6 h at 50 °C. After a 6 h, the mixture was filtered, concentrated and purified by column chromatography (2:1 = Hex:EtOAc) to afford **2** (600 mg, 1.43 mmol, 55% yield) as a yellow powder: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 2.05 (quint, J = 6.1 Hz, 2H), 2.75 (dd, J = 17.2, 3.0 Hz, 1H), 3.08 (dd, J = 17.2, 13.0 Hz, 1H), 3.62 (t, J = 6.1 Hz, 2H), 4.07 (t, J = 6.2 Hz, 2H) 4.51 (s, 2H), 5.30 (dd, J = 13.0 Hz, 2.9 Hz, 1H), 6.02 (d, J = 2.2 Hz, 1H), 6.05 (d, J = 2.2 Hz, 1H), 6.85 (d, J = 8.6 Hz, 2H), 7.24-7.32 (m, 7H), 12.0 (s, 1H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 195.9, 167.1, 163.6, 162.6, 156.2, 137.6, 129.6, 128.0, 127.5, 127.3, 115.3, 102.7, 95.2, 94.2, 78.5, 72.7, 66.0, 65.0, 42.6, 28.9.

5-Hydroxy-2-(4-hydroxy-phenyl)-7-(3-hydroxy-propoxy)-chroman-4-one (3): To a solution of **2** (100 mg, 0.24 mmol) in MeOH (3 mL) was added 10% Pd/C (10 mg), and the flask was charged with H_2 gas. This mixture was stirred for 12 h at room temperature and filtered through a short celite pad washing with ether. The filtrate was concentrated and purified by column chromatography (1:1 = Hex:EtOAc) to afford **3** (36.3 mg, 0.11 mmol, 46% yield) as a pale yellow powder: ^{1}H NMR (CD_3OD, 400 MHz) δ 1.98 (quint, J = 6.2 Hz, 2H), 2.73 (dd, J = 17.2, 3.0 Hz, 1H), 3.15 (dd, J = 17.2, 12.9 Hz, 1H), 3.71 (t, J = 6.2 Hz, 2H), 4.10 (t, J = 6.2 Hz, 2H), 5.37 (dd, J = 12.9, 2.8 Hz, 1H), 6.04 (d, J = 2.2 Hz, 1H), 6.06 (d, J = 2.2 Hz, 1H), 6.82 (d, J = 8.5, 2H), 7.32 (d, J = 8.5 Hz, 2H).

7-(3-Benzyloxy-propoxy)-5-methoxy-2-(4-methoxy-phenyl)-chroman-4-one (4): To a solution of **2** (1.5 g, 3.57 mmol) in acetone (40 mL) were added K_2CO_3 (1.4 mL, 14.3 mmol) and dimethyl sulfate (2.5 g, 17.9 mmol), and the mixture was stirred for 5 h at room temperature. The reaction mixture was filtered, concentrated, and purified by column chromatography (4:1 = Hex:EtOAc) to afford **4** (865 mg, 1.93 mmol, 54% yield) as a pale yellow powder: ^{1}H NMR ($(CD_3)_2CO$, 400 MHz) δ 2.06 (quint, J = 6.1 Hz, 2H), 2.75 (dd, J = 16.5, 2.8 Hz, 1H), 3.03 (dd, J = 16.5 , 13.1 Hz, 1H), 3.62 (t, J = 6.0 Hz, 2H), 3.80 (s, 3H), 3.86 (s, 3H), 4.09 (t, J = 6.2 Hz, 2H), 4.50 (s, 2H), 5.33 (dd, J = 13.0, 2.6 Hz, 1H), 6.06 (d, J = 2.1 Hz, 1H), 6.13 (d, J = 2.1 Hz, 1H), 6.92 (d, J = 8.6 Hz, 2H), 7.24-7.31 (m, 5H), 7.36 (d, J = 8.6 Hz, 2H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 189.3, 165.3, 164.9, 162.2, 160.0, 138.2, 130.8, 128.3, 127.6, 127.5, 114.0, 105.8, 94.0, 93.3, 78.8, 73.0, 66.3, 65.2, 56.0, 55.2, 45.3, 29.4.

7-(3-Hydroxy-propoxy)-5-methoxy-2-(4-methoxy-phenyl)-chroman-4-one (5): To a solution of **4** (60 mg, 0.13 mmol) in MeOH (3 mL) was added 10% Pd/C (10 mg), and the flask was charged with H_2 gas. The reaction mixture was stirred for 12 h at room temperature and filtered through a short celite pad washing with ether. The filtrate was concentrated and purified by column chromatography (2:1 = Hex:EtOAc) to afford **5** (20 mg, 0.06 mmol, 45% yield) as a white powder: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 2.01 (quint, J = 5.9 Hz, 2H), 2.63 (m, 1H), 2.71 (m, 1H), 3.79 (s, 3H), 3.81 (s, 3H), 3.84 (t, J = 5.8 Hz, 2H), 4.06 (t, J = 5.8 Hz, 2H), 4.92 (d, J = 9.1 Hz, 1H), 6.07 (d, J = 1.6 Hz, 1H), 6.12 (d, J = 1.6 Hz, 1H), 6.91(d, J = 8.2 Hz, 2H), 7.34 (d, J = 8.4 Hz, 2H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 159.7, 159.0, 158.8, 156.8, 134.2, 127.8, 114.3, 104.0, 94.5, 92.0, 66.3, 61.1, 55.9, 55.7, 53.9, 32.3, 29.8, 19.8.

5-Hydroxy-7-(3-hydroxy-propoxy)-2-(4-methoxy-phenyl)-chroman-4-one (6): To a stirred solution of **5** (100 mg, 0.23 mmol) in CH_2Cl_2 (7 mL) at 0 °C, 1.0 M solution of BBr_3 in CH_2Cl_2 (0.46 mL, 0.46 mmol) was added slowly. The reaction mixture was stirred for 1 h at 0 °C, and then quenched with a 1:1 mixture of MeOH/H_2O (6 mL). After evaporation of the volatiles under reduced pressure, the residue was purified by column chromatography in silica gel (3:1 = Hex:EtOAc) to afford **6** (40 mg, 0.12 mmol, 50% yield) as a pale yellow powder: ^{1}H NMR ($(CD_3)_2CO$, 400 MHz) δ 1.95 (quint, J = 6.1 Hz, 2H), 2.74 (dd, J = 17.1, 2.8 Hz, 1H), 3.18 (dd, J = 17.1. 12.8 Hz, 1H), 3.70 (t, J = 6.0 Hz, 2H), 3.88 (s, 3H), 4.13 (t, J = 6.0 Hz, 2H), 5.48 (dd, J = 12.9, 3.0 Hz, 1H), 6.01 (d, J = 2.0 Hz, 1H), 6.04 (d, J = 2.0 Hz, 1H), 6.90 (d, J = 8.4 Hz, 2H), 7.40 (d, J = 8.44 Hz, 2H).

7-(3-Hydroxy-propoxy)-5-methoxy-2-(4-methoxy-phenyl)-chromen-4-one (7) and 7-(3-Benzyloxy-propoxy)-5-methoxy-2-(4-methoxy-phenyl)-chromen-4-one (8): To a solution of **4** (50 mg, 0.12 mmol) dissolved in 1,4-dioxnae (5 mL) were added DDQ (54.5 mg, 0.24 mmol) and acetic acid (0.1 mL). The mixture was stirred for 12 h at 80 °C and then concentrated under reduced pressure. The residue was dissolved in CH_2Cl_2 and filtered. The filtrate was washed with H_2O, dried over $MgSO_4$, concentrated, and purified by column chromatography on silica gel (40:1 = CH_2Cl_2:MeOH) to afford **7** (18 mg, 0.05 mmol, 42% yield) and **8** (22 mg, 0.049 mmol, 41% yield) as pale brown oil and a dark yellow powder, respectively. For compound **7**: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 2.10 (quint, J = 6.1 Hz, 2H), 3.87 (s, 3H), 3.89-3.95 (m, 5H), 4.21 (t, J = 6.0 Hz, 2H), 6.31 (d, J = 2.1 Hz, 1H), 6.52 (d, J = 2.1 Hz, 1H), 6.58 (s, 1H), 6.97 (d, J = 8.9

Hz, 2H), 7.78 (d, J = 8.9 Hz, 2H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 207.4, 178.2, 163.6, 162.5, 161.2, 161.1, 160.1, 128.0, 124.1, 114.7, 109.4, 107.8, 96.7, 93.8, 66.0, 59.8, 56.7, 55.8, 32.3: For compound **8**: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 2.13 (m, 2H), 3.69 (t, J = 5.9 Hz, 2H), 3.88 (s, 3H), 3.95 (s, 3H), 4.20 (t, J = 6.2 Hz, 2H), 4.55 (s, 2H), 6.35 (d, J = 2.1 Hz, 1H), 6.57 (d, J = 2.1 Hz, 1H), 6.60 (s, 1H), 7.00 (d, J = 8.9 Hz, 2H), 7.48 (m, 5H), 7.81 (d, J = 8.9 Hz, 2H) ; ^{13}C NMR ($CDCl_3$, 100 MHz) δ 177.7, 163.3, 162.0, 160.8, 160.7, 159.8, 138.2, 129.7, 128.2, 127.6, 126.6, 123.8, 114.3, 109.1, 107.6, 96.4, 93.4, 73.1, 66.4, 65.4, 56.4, 55.5, 30.9; HRMS (MALDI-TOF): ($M + H^+$) calcd. for $C_{27}H_{27}O_6$, 447.181; found, 447.037.

5-Hydroxy-7-(3-hydroxy-propoxy)-2-(4-methoxy-phenyl)-chromen-4-one (9): To a solution of **8** (70 mg, 0.2 mmol) dissolved in CH_2Cl_2 (8 mL) at 0 °C was slowly added 1.0 M solution of BBr_3 in CH_2Cl_2 (0.2 mL, 0.2 mmol). The reaction mixture was stirred for 1 h, quenched with a 1:1 mixture of MeOH and H_2O (6 mL), concentrated under reduced pressure, and then purified by column chromatography on silica gel (20:1 = CH_2Cl_2:MeOH) to afford **9** (42 mg, 0.12 mmol, 62% yield) as a pale yellow powder: ^{1}H NMR ($CDCl_3$, 400 MHz) δ 2.05 (quint, J = 6.2 Hz, 2H), 3.79 (t, J = 6.2 Hz, 2H), 3.89 (s, 3H), 4.18 (t, J = 6.2 Hz, 2H), 6.33 (d, J = 2.1 Hz, 1H), 6.55 (d, J = 2.1 Hz, 1H), 6.57 (s, 1H), 7.03 (d, J = 8.9 Hz, 2H), 7.87 (d, J = 9.0 Hz, 2H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 183.5, 166.1, 165.5, 163.8, 162.3, 158.7, 128.9, 123.9, 115.3, 106.0, 104.3, 99.4, 93.8, 66.2, 59.1, 56.0, 32.6.

5-Hydroxy-2-(4-hydroxy-phenyl)-7-(3-hydroxy-propoxy)-chromen-4-one (10): To a stirred solution of **9** (130 mg, 0.38 mmol) dissolved in CH_2Cl_2 (12 mL) at 0 °C was slowly added 1.0 M solution of BBr_3 in CH_2Cl_2 (0.76 mL, 0.76 mmol). The reaction mixture was stirred for 1 h, quenched with a 1:1 mixture of MeOH and H_2O (8 mL), concentrated under reduced pressure, and then purified by column chromatography on silica gel (20:1 = CH_2Cl_2:MeOH) to afford **10** (82 mg, 0.25 mmol, 65% yield) as a pale yellow powder: ^{1}H NMR ($(CD_3)_2CO$, 400 MHz) δ 1.63 (quint, J = 6.2 Hz, 2H), 3.39 (t, J = 6.1 Hz, 2H), 3.88 (t, J = 6.3 Hz, 2H), 5.96 (d, J = 1.8 Hz, 1H), 6.31 (s, 1H), 6.33 (d, J = 1.7 Hz, 1H), 6.67 (d, J = 8.7 Hz, 2H), 7.60 (d, J = 8.7 Hz, 2H); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 184.0, 166.8, 166.0, 163.8, 162.8, 159.5, 130.1, 124.0, 117.7, 106.7, 105.0, 100.9, 95.4, 67.3, 59.5, 55.8, 33.8.

CDK2 Assay. The CDK2-Cyclin A enzyme activity was measured by a fluorescence kinetic assay using the Biosource Omnia Ser/Thr recombinant kit 7 (Invitrogen Corporation, CA, USA). The assay was carried our in a volume of 30 μL at room temperature in 384-well black polystyrene plates. The final concentrations of the assay constituents were 1 μM CDK2-Cyclin A, 10 μM peptide substrate, and 1 mM ATP. The compounds were solubilized in DMSO and added to the reaction mixture at fixed concentrations of 20 μM for flavone derivatives. The final concentration of DMSO in the mixture was 1%. Continuous kinetic monitoring of enzyme activity was performed on Spectramax Gemini reader (λ_{ex} 355 nm and λ_{em} 460 nm). The experiment was carried out in triplicate and the percent inhibition of enzyme activity was calculated.

Cell Viability Assay. A cell suspension with 100,000 cells/mL was prepared and 100 µL of the cell suspension was added to each well of a 96-well plate. Plate was cultured at 37 °C for 24 h and culture media (DMEM) were replaced with media containing inhibitors of fixed concentration at 50 μM. After further 48 h incubation, 10 μL of CCK-8 solution was added to each well and the plate was incubated at 37 °C for 1 h. The optical densities (OD) at a wavelength of 450 nm were measured using the microplate reader (Spectramax Plus, Molecular Devices, UK). Cells incubated in culture media alone served as a control for cell viability (untreated wells).

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References

1. Havsteen, B. H. *Pharmacol. Ther.* **2002**, *96*, 67.
2. Middleton Jr., E.; Kandaswami, C.; Theoharides, T. C. *Pharmacol. Rev.* **2000**, *52*, 673.
3. Buchler, P.; Gukovskaya, A. S.; Mouria, M.; Buchler, M. C.; Buchler, M. W.; Friess, H.; Pandol, S. J.; Reber, H. A.; Hines, O. J. *Pancreas* **2003**, *26*, 264.
4. Kohno, H.; Tanaka, T.; Kawabata, K.; Hirose, Y.; Sugie, S.; Tsuda, H.; Mori, H. *Int. J. Cancer* **2002**, *101*, 461.
5. Rice, L.; Samedi, V. G.; Medrano, T. A.; Sweeney, C. A.; Baker, H. V.; Stenstrom, A.; Furman, J.; Shiverick, K. T. *Prostate* **2002**, *52*, 201.
6. Kellis Jr., J. T.; Vickery, L. E. *Science* **1984**, *225*, 1032.
7. Aklilu, M.; Kindler, H. L.; Donehower, R. C.; Mani, S.; Vokes, E. E. *Ann. Oncol.* **2003**, *14*, 1270.
8. Kaur, G.; Stetler-Stevenson, M.; Sebers, S.; Worland, P.; Sedlacek, H.; Myers, C.; Czech, J.; Naik, R.; Sausville, E. *J. Nat. Cancer Inst.* **1992**, *84*, 1736.
9. (a) Filgueira de Azevedo Jr., W.; Mueller-Dieckmann, H.-J.; Schulze-Gahmen, U.; Worland, P. J.; Sausville, E.; Kim, S.-H. *Proc. Natl. Acad. Sci. USA* **1996**, *93*, 2735. (b) Kim, S.-H. *Pure Appl. Chem.* **1998**, *70*, 555. (c) Kim, S.-H.; Schulze-Gahmen, U.; Brandsen, J.; Filgueira de Azevedo Jr., W. *Prog. Cell Cycle Res.* **1996**, *2*, 137.
10. (a) Sielecki, T. M.; Boylan, J. F.; Benfield, P. A.; Trainor, G. L. *J. Med. Chem.* **2000**, *43*, 1; (b) Crews, C. M.; Mohan, R. *Curr. Opin. Chem. Biol.* **2000**, *4*, 47.
11. (a) Dancey, J.; Sausville, E. A. *Nat. Rev. Drug Discov.* **2003**, *2*, 296. (b) McGovern, S. L.; Shoichet, B. K. *J. Med. Chem.* **2003**, *46*, 1478.
12. Ahn, Y. M.; Vogeti, L.; Liu, C.-J.; Santhapuram, H. K. R.; White, J. M.; Vasandani, V.; Mitscher, L. A.; Lushington, G. H.; Hanson, P. R.; Powell, D. R.; Himes, R. H.; Roby, K. F.; Ye, Q.; Georg, G. I. *Bioorg. Med. Chem.* **2007**, *15*, 702.
13. Kim, J.; Lee, Y.; Kim, H.; Kang, S.-Y.; Park, K.-S.; Cho, J.-H.; Lee, Y.-Y.; Kim, B.-S.; Lim, Y.; Chong, Y. *Bull. Korean Chem. Soc.* **2005**, *26*, 2065.
14. (a) Park, K.-S.; Kim, J.; Chong, Y.; Choo, H. *Bull. Korean Chem. Soc.* **2007**, *28*, 211. (b) Park, K.-S.; Kim, J.; Chong, Y. *Mol. Simul.* **2007**, in press.
15. Lee, M. *Bull. Korean Chem. Soc.* **2007**, *28*, 25.