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Rapid Microwave Assisted Synthesis and Antiproliferative Evaluation of Novel Steroidal Thiazole Derivatives

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Abstract Three series of novel steroidal thiazole derivatives were synthesized by microwave assisted one-pot reaction from pregnenolone, testosterone and estrone, respectively. Their structures were characterized by IR, NMR and HRMS, and the antiproliferative activities of all the synthesized compounds against human cervical carcinoma (HeLa), human liver carcinoma(HepG2), human lung carcinoma(A549), nasopharyngeal carcinoma(CNE2) and normal kidney epithelial cells(HEK293T) were screened. Among the compounds, 1-estron-17′-ylidene-2-[4′-(*p*-bromophenyl)-2′-thiazol]hydrazone(**12**) displayed distinct antiproliferative activity against all the tested cancer cell lines and was almost inactive to normal kidney epithelial cells.

Keywords Microwave assisted synthesis; Pregnenolone; Testosterone; Estrone; Steroidal thiazole derivative

1 Introduction

Heterocycles can enhance binding affinity and improve *in vitro* potency *via* hydrogen bond interactions. The compounds containing heterocycles have been widely applied to the field of medicine or pesticide because of its good biological activity[1]. Heterosteroids have been paid more attention over the past years by medicinal chemists for drug discovery. The incorporation of a steroid with a heterocycle or a heteroatom will greatly change the chemical property of the steroid and often result in useful alteration of its biological activity[2]. Up to now, a number of different heterocyclic systems have been introduced into the structure of steroids and many of these steroidal heterocycle derivatives have been found to possess potent biological activities, such as anti-cancer, anti-microbial, anti-estrogenic, anti-inflammatory, hypertensive, anabolic and cardiovascular activities[3].

Compounds containing thiazole cycle display various biological functions, such as antimicrobial, antiinflammatory, anticancer, anti-HIV-1 and anticonvulsant functions due to its special chemical structure, and have obtained an extensive application in drugs, pesticides and materials science, etc.[4—9]. For example, compound **1**(Fig.1) is a thiazolyl hydrazonoandrostane which exhibits remarkable cytotoxicity to breast adenocarcinoma(MCF-7), non-small cell lung cancer(NCI-H460) and CNS cancer[10], and compound **2**(Fig.1) shows a distinct antiproliferative activity and owns the concentration of drug yielding 50% cell surrival(IC_{50}) of 15.3 and 10.4 μmol/L against HepG2 and SMMC-7721 liver cancer, respectively[11]. Recently, some steroidal thiazole derivatives possessing a D-ring fused a thiazole were reported as anti-inflammatory and anti-tumor agents[12,13].

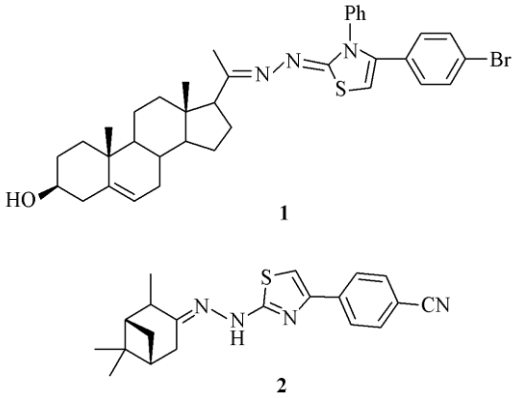

Fig.1 Structures of compounds 1 and 2

In previous studies, we synthesized some novel heterosteroids and determined their anti-tumor activity. The results show that some B-norcholesteryl benzimidazoles and 17-hydrazone aromatic heterocycle derivatives exhibited an excellent antiproliferative activity[14,15]. As a part of our ongoing research outcome on development of new biologically potent anticancer steroids with the structure of heterocycle, this paper introduced the thiazole to steroids and synthesized series of novel steroidal

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thiazole derivatives using a simple microwave assisted green synthesis by one-pot reaction of various steroidal ketones, thiosemicarbazide and 4-substituted-α-bromoacetophenones. The antiproliferative activity of the compounds *in vitro* was further evaluated.

2 Experimental

2.1 Chemistry

All the steroidal ketones were purchased from Sinopharm Chemical Reagent Co., Ltd., Shanghai, China. All the chemicals and solvents were of analytical grade. Melting points were determined on an X_4 apparatus(Beijing Tech Instrument Co., Ltd., Beijing, China) and were uncorrected. The ^{1}H NMR and ^{13}C NMR spectra were recorded in $CDCl_3$ or DMSO-d_6 on a Bruker AV-600 spectrometer at working frequencies of 600 and 150 MHz, and a Bruker AV-300 spectrometer at working frequencies of 300 and 75 MHz, respectively. Coupling constants(J) were expressed in Hz. Infrared spectra were measured with a Thermo Scientific Nicolet IS-10 Spectrophotometer using KBr pellets(Thermo Scientific, America). High resolution electron impact mass spectra(HREIMS) were recorded on an Agilent 6210 time of flight mass spectrometry(TOFMS) instrument(Agilent Technologies, America). The cell proliferation assay was undertaken by 3-(4,5-dimethyl-2-thiazolyl-2,5-diphenyl-*H*-tetrazolium bromide)(MTT) method using 96-well plates on an MLLTISKAN MK3 analysis spectrometer (Thermo Scientific, Shanghai, China). All microwave reactions were carried out in an XH-MC-1(Xianghu, China) microwave reactor and monitored by thin layer chromatography(TLC).

*2.1.1 General Procedure for the Synthesis of Compounds **1**—**5***

Pregnenolone(0.41 mmol), 4-substituted α-bromoacetophenone(0.41 mmol) and thiosemicarbazide(0.42 mmol) were taken in a 100 mL two-net reactive bottle. 15 mL of anhydrous CH_3CH_2OH was added to the reactive bottle to dissolve the mixture. The reaction vessel was put in a microwave reactor and adjusted to the stipulated time at 300 W, 85 °C for 30 min. On completion of the reaction(by TLC), the majority of solvent was evaporated under reduced pressure. 30 mL of water was added to the mixture and the product was extracted with ethyl acetate. The combined extract was washed with water, 5% $NaHCO_3$ and saturated brine, dried over anhydrous Na_2SO_4 and evaporated under reduced pressure to give a crude product. The crude product was purified by flash chromatography on silica gel to afford the corresponding target products **1**—**5**.

1-Pregnenolon-20′-ylidene-2-[4′-(*p*-methoxyphenyl)-2′-thiazolyl]hydrazone(**1**): a light yellow solid, yield 48%, m. p. 168—170 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3409, 2927, 1611, 1556, 1486, 1327, 1245, 1170, 1028, 950, 831, 736; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.65(3H, s, 18-CH_3), 1.03(3H, s, 19-CH_3), 1.87(3H, s, 21-CH_3), 3.50—3.61(1H, m, C3-αH), 3.85(3H, s, —OCH_3), 5.38(1H, d, J=4.8 Hz, C6-H), 6.72(1H, s, 5-thiazole-H), 6.93(2H, d, J=8.7 Hz, 3,5-Ph-H), 7.73(2H, d, J=8.7 Hz, 2,6-Ph-H), 8.54 (1H, brs, —NH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 13.3(18-C), 17.0(19-C), 19.4(21-C), 21.1(11-C), 23.3(15-C), 24.2(16-C), 31.6(8-C), 31.8(2-C), 32.1(7-C), 36.5(1-C), 37.3(10-C), 38.9(12-C), 42.3(13-C), 43.9(4-C), 50.2(9-C), 55.3 (17-C), 56.5(—OCH_3), 58.8(14-C), 71.7(3-C), 101.7 (5-thiazole-C), 121.5(6-C), 114.0, 127.1, 128.1, 159.2(Ph-C), 140.8(5-C), 150.8(4-thiazole-C), 151.0(20-C), 170.0 (2-thiazole-C); HREIMS, *m/z*: 520.2998[M+H]$^+$(calcd. for $C_{31}H_{41}N_3O_2S$: 520.2994).

1-Pregnenolon-20′-ylidene-2-[4′-(*p*-trifluoromethylphenyl)-2′-thiazolyl]hydrazone(**2**): a light yellow solid, yield 71%, m. p. 136—138 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3441, 2935, 1693, 1561, 1407, 1324, 1118, 1065, 1013, 950, 846, 751; ^{1}H NMR(600 MHz, $CDCl_3$), δ: 0.62(3H, s, 18-CH_3), 1.01(3H, s, 19-CH_3), 1.84(3H, s, 21-CH_3), 3.51—3.56(1H, m, C3-αH), 5.36(1H, d, J=5.4 Hz, C6-H), 6.95(1H, s, 5-thiazole-H), 7.62(2H, d, J=8.4 Hz, 3,5-PhH), 7.88(2H, d, J=8.4 Hz, 2,6-PhH), 8.66(1H, brs, —NH); ^{13}C NMR(150 MHz, $CDCl_3$), δ: 13.4(18-C), 17.3 (19-C), 19.6(21-C), 21.2(11-C), 23.4(15-C), 24.4(16-C), 31.8 (8-C), 31.9(2-C), 32.2(7-C), 36.7(1-C), 37.4(10-C), 39.0(12-C), 42.4(13-C), 44.0(4-C), 50.3(9-C), 56.6(17-C), 58.9(14-C), 71.9(3-C), 105.6(5-thiazole-C), 121.7(6-C), 123.5(-CF_3), 125.3, 125.7, 126.1, 129.3, 129.5, 138.4(Ph-C), 140.9(5-C), 149.9 (4-thiazole-C), 151.5(20-C), 170.5(2-thiazole-C); HREIMS: *m/z*: 558.2776 [M+H]$^+$(calcd. for $C_{31}H_{38}F_3N_3OS$: 558.2792).

1-Pregnenolon-20′-ylidene-2-[4′-(*p*-nitrophenyl)-2′-thiazolyl]-hydrazone(**3**): a light yellow solid, yield 47%, m. p. 266—270 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3498, 2930, 1594, 1437, 1339, 1225, 1105, 1040, 865, 706; ^{1}H NMR(300 MHz, DMSO-d_6), δ: 0.56(3H, s, 18-CH_3), 0.93(3H, s, 19-CH_3), 1.91(3H, s, 21-CH_3), 3.31—3.21(1H, m, C3-αH), 4.65(1H, brs, —OH), 5.26(1H, d, J=3.3 Hz, C6-H), 7.59(1H, s, 5-thiazole-H), 8.09(2H, d, J=8.4 Hz, 3,5-PhH), 8.25(2H, d, J=8.4 Hz, 2,6-PhH), 10.77(1H, brs, —NH); ^{13}C NMR(75 MHz, DMSO-d_6), δ: 13.4(18-C), 18.8(19-C), 19.6(21-C), 21.1(11-C), 23.5(15-C), 24.3(16-C), 31.7(8-C), 31.8(2-C), 32.1(7-C), 36.6(1-C), 37.4(10-C), 38.6(12-C), 42.6(13-C), 43.6(4-C), 50.1(9-C), 56.3(17-C), 58.4(14-C), 70.5(3-C), 108.9(5-thiazole-C), 120.8(6-C), 124.5, 126.7, 141.7, 146.5(Ph-C), 141.5(5-C), 148.9(4-thiazole-C), 152.9(20-C), 171.4(2-thiazole-C). HREIMS, *m/z*: 535.2743 [M+H]$^+$(calcd. for $C_{30}H_{38}N_4O_3S$: 535.2747).

1-Pregnenolon-20′-ylidene-2-[4′-(*p*-cyanophenyl)-2′-thiazolyl]hydrazone(**4**): a light yellow solid, yield 43%, m. p. 247—249 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3344, 2930, 2224, 1601, 1556, 1439, 1322, 1227, 1118, 1053, 851, 759; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.65(3H, s, 18-CH_3), 1.04(3H, s, 19-CH_3), 1.91(3H, s, 21-CH_3), 3.61—3.50(1H, m, C3-αH), 5.39(1H, d, J=5.1 Hz, C6-H), 7.02(1H, s, 5-thiazole-H), 7.68(2H, d, J=8.1 Hz, 3,5-PhH), 7.89(2H, d, J=8.1 Hz, 2,6-PhH), 8.51(1H, brs, —NH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 13.3(18-C), 17.1(19-C), 19.4(21-C), 21.1(11-C), 23.4(15-C), 24.2(16-C), 31.6(8-C), 31.8(2-C), 32.1(7-C), 36.5(1-C), 37.3(10-C), 38.9(12-C), 42.3(13-C), 43.9(4-C), 50.2(9-C), 56.5(17-C), 58.8(14-C), 71.7(3-C), 106.7(5-thiazole-C), 119.1(—CN), 121.5(6-C), 110.7, 126.2, 132.5, 139.0(Ph-C), 140.8(5-C), 149.4 (4-thiazole-C), 151.6(20-C), 170.3(2-thiazole-C); HREIMS, *m/z*: 515.2845[M+H]$^+$(calcd. for $C_{31}H_{38}N_4OS$: 515.2843).

1-Pregnenolon-20′-ylidene-2-[4′-(*p*-bromophenyl)-2′-thiazolyl]hydrazone(**5**): a light yellow solid, yield 35%, m. p.

155—157 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3384, 2930, 1556, 1429, 1359, 1314, 1230, 1038, 906, 816, 726; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.59(3H, s, 18-CH_3), 1.02(3H, s, 19-CH_3), 1.73(3H, s, 21-CH_3), 3.57—3.49(1H, m, C3-αH), 5.35(1H, d, J=4.5 Hz, C6-H), 6.83(1H, s, 5-thiazole-H), 7.49(2H, d, J=8.1 Hz, 3,5-PhH), 7.64(2H, d, J=8.1 Hz, 2,6-PhH), 9.35(1H, brs, —NH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 13.3(18-C), 17.4(19-C), 19.4(21-C), 21.2(11-C), 23.3(15-C), 24.3(16-C), 31.6(8-C), 31.8(2-C), 32.1(7-C), 36.6(1-C), 37.3(10-C), 38.8(12-C), 42.3(13-C), 43.9(4-C), 50.2(9-C), 56.5(17-C), 58.8(14-C), 71.7(3-C), 103.8(5-thiazole-C), 121.4(6-C), 121.4, 127.5, 131.7, 134.0(Ph-C), 140.8(5-C), 150.1(4-thiazole-C), 151.5(20-C), 170.9(2-thiazole-C); HREIMS, *m/z*: 568.1997[M+H]$^+$(calcd. for $C_{30}H_{38}BrN_3OS$: 568.1999).

*2.1.2 Synthesis of 1-Pregnenolon-20′-ylidene-2-[4′-nathphalenyl-2′- thiazolyl]hydrazone(**6**)*

The reaction of pregnenolone(106 mg, 0.41 mmol), β-bromoacetylnathphalene(101 mg, 0.41 mmol) and thiosemicarbazide(39 mg, 0.42 mmol) in anhydrous ethanol as described in Section 2.1.1 afforded 92 mg of compound **6**(51%), a yellow solid, m. p. 157—158 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3416, 2932, 1556, 1474, 1392, 1319, 1230, 1043, 911, 831, 726; ^{1}H NMR (300 MHz, $CDCl_3$), δ: 0.58(3H, s, 18-CH_3), 1.01(3H, s, 19-CH_3), 1.79(3H, s, 21-CH_3), 3.59—3.49(1H, m, C3-αH), 5.36(1H, d, J=4.2 Hz, C6-H), 6.97(1H, s, 5-thiazole-H), 7.50—7.43(2H, m, Ph-H), 7.89—7.81(4H, m, Ph-H), 8.30(1H, s, 1-Naph-H), 9.16(1H, brs, —NH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 13.3(18-C), 17.3(19-C), 19.4(21-C), 21.1(11-C), 23.4(15-C), 24.2(16-C), 31.6(8-C), 31.8(2-C), 32.1(7-C), 36.5(1-C), 37.3(10-C), 38.8(12-C), 42.3(13-C), 43.8(4-C), 50.1 (9-C), 56.5(17-C), 58.8(14-C), 71.7(3-C), 104.0(5-thiazole-C), 121.5(6-C), 124.1, 124.7, 125.9, 126.2, 127.6, 128.2, 128.4, 133.0, 132.4, 133.7(Naph-C), 140.8(5-C), 151.2(4-thiazole-C), 151.3(20-C), 170.6(2-thiazole-C); HREIMS, *m/z*: 540.3049 [M+H]$^+$(calcd. for $C_{34}H_{41}N_3OS$: 540.3043).

*2.1.3 Synthesis of Compounds 7—**12***

Compounds **7**—**12** were prepared similarly according to the procedure of Section 2.1.1, but using estrone as a reactive substrate to replace the pregnenolone.

1-Estron-17′-ylidene-2-[4′-(*p*-methoxyphenyl)-2′-thiazolyl]-hydrazone(**7**): a light yellow solid, yield 49%, m. p. 179—181 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3431, 2925, 1698, 1599, 1499, 1247, 1177, 1095, 841, 741; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.94(3H, s, 18-CH_3), 2.88—2.80(2H, m, C16-H), 3.85(3H, s, —OCH_3), 6.60(1H, d, J=2.4 Hz, C4-H), 6.67(1H, dd, J=8.1, 2.4 Hz, C2-H), 6.72(1H, s, 5-thiazole-H), 6.93(2H, d, J=8.7 Hz, 3,5-PhH), 7.18(1H, d, J=8.1 Hz, C1-H), 7.73(2H, d, J=8.7 Hz, 2,6-PhH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 17.2(18-C), 23.3(15-C), 25.8(11-C), 26.2(16-C), 27.2(7-C), 29.5(6-C), 34.1(12-C), 38.2(8-C), 44.2(13-C), 44.8(9-C), 52.6(14-C), 55.3(—OCH_3), 101.5(5-thiazole-C), 112.9(2-C), 115.4(4-C), 127.2(1-C), 132.2(10-C), 138.0(5-C), 150.8(4-thiazole-C), 153.7(3-C), [114.0, 126.5, 127.8, 159.3](Ph-C), 165.4(17-C), 170.0(2-thiazole-C); HREIMS, *m/z*: 474.2215 [M+H]$^+$(calcd. for $C_{28}H_{31}N_3O_2S$: 474.2226).

1-Estron-17′-ylidene-2-[4′-(*p*-nitrophenyl)-2′-thiazolyl]-hydrazone(**8**): a yellow solid, yield 53%, m. p. 163—164 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3451, 2927, 1556, 1339, 1282, 1108, 851, 788,714; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.97(3H, s, 18-CH_3), 2.90—2.80(2H, m, C16-H), 6.60(1H, d, J=2.4 Hz, C4-H), 6.67(1H, dd, J=8.4, 2.4 Hz, C2-H), 7.08(1H, s, 5-thiazole-H), 7.18(1H, d, J=8.4 Hz, C1-H), 7.94(2H, d, J=9.0 Hz, 3,5-PhH), 8.25(2H, d, J=9.0 Hz, 2,6-PhH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 17.2(18-C), 23.3(15-C), 25.9(11-C), 26.2(16-C), 27.2(7-C), 29.5(6-C), 34.1(12-C), 38.2(8-C), 44.1(13-C), 45.0(9-C), 52.6(14-C), 107.4(5-thiazole-C), 112.9(2-C), 115.4(4-C), 126.5(1-C), 132.2(10-C), 138.0(5-C), 146.9(4-thiazole-C), 153.7(3-C), 124.1, 126.3, 140.7, 148.9(Ph-C), 166.1(17-C), 170.4(2-thiazole-C); HREIMS, *m/z*: 489.1960[M+H]$^+$(calcd. for $C_{27}H_{28}N_4O_3S$: 489.1973).

1-Estron-17′-ylidene-2-[4′-(*p*-trifluoromethylphenyl)-2′-thiazolyl]hydrazone(**9**): a yellow solid, yield 58%, m. p. 164—165 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3366, 2922, 1706, 1616, 1556, 1491, 1327, 1162, 1063, 841 701; ^{1}H NMR(600 MHz, $CDCl_3$), δ: 0.93(3H, s, 18-CH_3), 2.41—2.35(2H, m, C6-H), 2.89—2.79 (2H, m, C16-H), 6.57(1H, d, J=2.4 Hz, C4-H), 6.65(1H, dd, J=8.4, 2.4 Hz, C2-H), 6.95(1H, s, 5-thiazole-H), 7.17(1H, d, J=9.0 Hz, C1-H), 7.63(2H, d, J=7.8 Hz, 3,5-PhH), 7.88(2H, d, J=7.8 Hz, 2,6-PhH); ^{13}C NMR(150 MHz, $CDCl_3$), δ: 17.3(18-C), 23.4(15-C), 26.0(11-C), 26.3(16-C), 27.3(7-C), 29.6(6-C), 34.2(12-C), 38.3(8-C), 44.2(13-C), 45.0(9-C), 52.7(14-C), 105.5(5-thiazole-C), 113.0(2-C), 115.5(4-C), 123.5(—CF_3), 126.7(1-C), 132.4(10-C), 138.2(5-C), 149.8 (4-thiazole-C), 153.7(3-C), 125.7, 125.8, 126.2, 138.2(Ph-C), 165.9(17-C), 170.3(2-thiazole-C); HREIMS, *m/z*: 512.1983 [M+H]$^+$(calcd. for $C_{28}H_{28}F_3N_3OS$: 512.1976).

1-Estron-17′-ylidene-2-[4′-(*p*-cyanophenyl)-2′-thiazolyl]-hydrazone(**10**): a yellow solid, yield 44%, m. p. 188—190 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3399, 2922, 2224, 1654, 1556, 1494, 1342, 1282, 1103, 1043, 841, 739; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.94(3H, s, 18-CH_3), 2.87—2.81(2H, m, C16-H), 6.60(1H, d, J=2.1 Hz, C4-H), 6.68(1H, dd, J=8.4, 2.1 Hz, C2-H), 7.01(1H, s, 5-thiazole-H), 7.17(1H, d, J=8.7 Hz, C1-H), 7.66(2H, d, J=8.4 Hz, 3,5-PhH), 7.88(2H, d, J=8.4 Hz, 2,6-PhH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 17.2(18-C), 23.3(15-C), 25.9 (11-C), 26.2(16-C), 27.2(7-C), 29.5(6-C), 34.1(12-C), 38.2 (8-C), 44.1(13-C), 44.9(9-C), 52.6(14-C), 106.6(5-thiazole-C), 113.0 (2-C), 115.5(4-C), 119.0(—CN), 126.5(1-C), 132.0 (10-C), 138.0(5-C), 149.1(4-thiazole-C), 153.9(3-C), 110.7, 126.3, 132.6, 138.9(Ph-C), 166.3(17-C), 170.5(2-thiazole-C); HREIMS, *m/z*: 469.2062[M+H]$^+$(calcd. for $C_{28}H_{28}N_4OS$: 469.2068).

1-Estron-17′-ylidene-2-[4′-(*p*-chlorophenyl)-2′-thiazolyl]-hydrazone(**11**): a light yellow solid, yield 43%, m. p. 146—147 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3324, 2927, 1721, 1551, 1494, 1399, 1245, 1088, 916 ,831, 726; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.94(3H, s, 18-CH_3), 2.87—2.83(2H, m, C16-H), 6.59(1H, d, J=2.4 Hz, C4-H), 6.67(1H, dd, J=8.4, 2.4 Hz, C2-H), 6.83(1H, s, 5-thiazole-H), 7.17(1H, d, J=8.4 Hz, C1-H), 7.36(2H, d, J=8.7 Hz, 3,5-PhH), 7.72(2H, d, J=8.7 Hz, 2,6-PhH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 17.2(18-C), 23.3(15-C), 25.8 (11-C), 26.2(16-C), 27.2(7-C), 29.5(6-C), 34.1(12-C), 38.2 (8-C), 44.1(13-C), 44.9(9-C), 52.6(14-C), 103.7(5-thiazole-C),

113.0(2-C), 115.5(4-C), 126.5(1-C), 132.2(10-C), 138.0(5-C), 149.9(4-thiazole-C), 153.7(3-C), 127.2, 128.8, 133.4, 133.4 (Ph-C), 165.8(17-C), 170.3(2-thiazole-C); HREIMS, *m/z*: 478.1720[M+H]$^+$(calcd. for $C_{27}H_{28}ClN_3OS$: 478.1723).

1-Estron-17′-ylidene-2-[4′-(*p*-bromophenyl)-2′-thiazolyl]-hydrazone(**12**): a light yellow solid, yield 34%, m. p. 235—236 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3320, 2927, 1609, 1556, 1496, 1394, 1282, 1068, 1003, 828, 729; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.92(3H, s, 18-CH_3), 2.85—2.75(2H, m, C16-H), 6.60(1H, d, *J*=1.5 Hz, C4-H), 6.68(1H, d, *J*=8.4 Hz, C2-H), 6.83(1H, s, 5-thiazole-H), 7.16(1H, d, *J*=8.7 Hz, C1-H), 7.50(2H, d, *J*=8.4 Hz, 3,5-PhH), 7.65(2H, d, *J*=8.4 Hz, 2,6-PhH); ^{13}C NMR (75 MHz, $CDCl_3$), δ: 17.2(18-C), 23.2(15-C), 25.9(11-C), 26.2(16-C), 27.2(7-C), 29.5(6-C), 34.1(12-C), 38.2(8-C), 44.1(13-C), 44.9(9-C), 52.6(14-C), 103.9(5-thiazole-C), 113.1(2-C), 115.6(4-C), 126.5(1-C), 132.0(10-C), 138.0(5-C), 149.8(4-thiazole-C), 154.0(3-C), 121.6, 127.6, 131.8, 133.7(Ph-C), 166.3(17-C), 170.8(2-thiazole-C); HREIMS, *m/z*: 522.1215[M+H]$^+$(calcd. for $C_{27}H_{28}BrN_3OS$: 522.1226).

2.1.4 Synthesis of Compounds 13—18

Compounds **13**—**18** were prepared similarly according to the procedure of Section 2.1.1, but using testosterone as a reactive substrate to replace the pregnenolone.

1-Testosteron-3′-ylidene-2-[4′-(*p*-trifluoromethylphenyl)-2′-thiazolyl]hydrazone(**13**): a light yellow solid, yield 45%, m. p. 254—256 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3244, 2935, 1614, 1554, 1409, 1319, 1272, 1165, 1031, 841, 749, 704; ^{1}H NMR(600 MHz, $CDCl_3$), δ: 0.75(3H, s, 18-CH_3), 0.87(3H, s, 19-CH_3), 2.29—2.21(2H, m, C2-H and C6-αH), 2.44(1H, br d, *J*=14.4 Hz, C6-βH), 3.63(1H, t, *J*=8.4 Hz, C17-H), 5.90(1H, s, C4-H), 6.95(1H, s, 5-thiazole-H), 7.60(2H, d, *J*=7.8 Hz, 3,5-PhH), 7.82(2H, d, *J*=7.8 Hz, 2,6-PhH), 9.93(1H, brs, —NH); ^{13}C NMR(150 MHz, $CDCl_3$), δ: 11.2(18-C), 17.6(19-C), 21.0(11-C), 21.1(15-C), 23.5(16-C), 30.6(6-C), 31.8(2-C), 32.3(1-C), 34.7(7-C), 35.9(8-C), 36.6(12-C), 37.7(10-C), 42.9 (13-C), 50.6(14-C), 53.8(9-C), 81.9(17-C), 105.4(5-thiazole-C), 120.4(4-C), 123.4(—CF_3), 125.2, 125.8, 126.2, 129.4, 129.6, 138.3(Ph-C), 149.9(4-thiazole-C), 150.8(5-C), 155.4(3-C), 170.7(2-thiazole-C); HREIMS, *m/z*: 530.2447[M+H]$^+$(calcd. for $C_{29}H_{34}F_3N_3OS$: 530.2463).

1-Testosteron-3′-ylidene-2-[4′-(*p*-cyanophenyl)-2′-thiazolyl]-hydrazone(**14**): a light yellow solid, yield 21%, m. p. 253—254 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3498, 2930, 2219, 1601, 1549, 1432, 1329, 1265, 1098, 941, 846, 761; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.80(3H, s, 18-CH_3), 1.03(3H, s, 19-CH_3), 2.40—2.25(2H, m, C2-H and C6-αH), 2.52(1H, dd, *J*=13.5, 2.7 Hz, C6-βH), 3.67(1H, t, *J*=8.7 Hz, C17-H), 5.94(1H, s, C4-H), 7.02(1H, s, 5-thiazole-H), 7.67(2H, d, *J*=8.4 Hz, 3,5-PhH), 7.87(2H, d, *J*=8.4 Hz, 2,6-PhH), 9.07(1H, brs, —NH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 11.1(18-C), 17.8(19-C), 20.7(11-C), 21.0(15-C), 23.4(16-C), 30.5(6-C), 31.7(2-C), 32.3(1-C), 34.7(7-C), 35.8 (8-C), 36.5(12-C), 37.7(10-C), 42.8 (13-C), 50.6(14-C), 53.7 (9-C), 81.7(17-C), 106.6(5-thiazole-C), 119.0(—CN), 120.3 (4-C), 110.7, 126.3, 132.5, 139.0(Ph-C), 149.4(4-thiazole-C), 150.0(5-C), 155.4(3-C), 169.8 (2-thiazole-C); HREIMS, *m/z*: 487.2532[M+H]$^+$(calcd. for $C_{29}H_{34}N_4OS$: 487.2544).

1-Testosteron-3′-ylidene-2-[4′-(*p*-nitrophenyl)-2′-thiazolyl]-hydrazone(**15**): a yellow solid, yield 42%, m. p. 198—200 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3436, 2925, 1599, 1447, 1342, 1105, 1045, 853, 716; ^{1}H NMR(300 MHz, DMSO-d_6), δ: 0.65(3H, s, 18-CH_3), 1.01(3H, s, 19-CH_3), 2.78(1H, d, *J*=16.5 Hz, C6-βH), 3.50— 3.40(1H, m, C17-H), 4.48(1H, d, *J*=4.5 Hz, —OH), 5.77(1H, s, C4-H), 7.60(1H, s, 5-thiazole-H), 8.08(2H, d, *J*=8.7 Hz, 3,5-PhH), 8.25(2H, d, *J*=8.7 Hz, 2,6-PhH), 11.02(1H, brs, —NH); ^{13}C NMR(75 MHz, DMSO-d_6), δ: 11.6(18-C), 17.9(19-C), 21.1(11-C), 21.7(15-C), 23.5(16-C), 30.3(6-C), 32.0(2-C), 32.1(1-C), 35.0(7-C), 35.8(8-C), 36.9(12-C), 37.6(10-C), 42.8(13-C), 50.6(14-C), 53.9(9-C), 80.4(17-C), 108.8(5-thiazole-C), 120.8(4-C), 124.5, 126.7, 141.4, 146.5(Ph-C), 148.9(4-thiazole-C), 150.4(5-C), 154.9(3-C), 170.4(2-thiazole-C); HREIMS, *m/z*: 507.2430[M+H]$^+$(calcd. for $C_{28}H_{34}N_4O_3S$: 507.2427).

1-Testosteron-3′-ylidene-2-[4′-(*p*-methoxyphenyl)-2′-thiazolyl]hydrazone(**16**): a yellow solid, yield 43%, m. p. 201—203 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3461, 2920, 1703, 1604, 1554, 1491, 1399, 1297, 1175, 1025, 836, 736; ^{1}H NMR(300 MHz, DMSO-d_6), δ: 0.67(3H, s, 18-CH_3), 1.03(3H, s, 19-CH_3), 2.78(1H, t, *J*=16.2 Hz, C6-βH), 3.46—3.39(1H, m, C17-H), 3.77(3H, s, —OCH_3), 4.47(1H, d, *J*=4.8 Hz, —OH), 5.75(1H, s, C4-H), 6.95(2H, d, *J*=8.7 Hz, 3,5-PhH), 7.05(1H, s, 5-thiazole-H), 7.77(2H, d, *J*=8.7 Hz, 2,6-PhH), 10.88(1H, brs, —NH); ^{13}C NMR(75 MHz, DMSO-d_6), δ: 11.7(18-C), 18.0(19-C), 21.1(11-C), 21.6(15-C), 23.5(16-C), 30.3(6-C), 32.0(2-C), 32.2(1-C), 35.0(7-C), 35.9(8-C), 36.9(12-C), 37.6(10-C), 42.8(13-C), 50.6(14-C), 53.9(9-C), 55.5(—OCH_3), 80.4(17-C), 101.7(5-thiazole-C), 121.0(4-C), 114.4, 127.3, 128.7, 159.1(Ph-C), 149.8(4-thiazole-C), 150.8(5-C), 154.3 (3-C), 169.9(2-thiazole-C); HREIMS, *m/z*: 492.2679[M+H]$^+$ (calcd. for $C_{29}H_{37}N_3O_2S$: 492.2679).

1-Testosteron-3′-ylidene-2-[4′-(*p*-chlorophenyl)-2′-thiazolyl]-hydrazone(**17**): a light yellow solid, yield 42%, m. p. 187—189 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3394, 2927, 1723, 1551, 1427, 1324, 1265, 1088, 828, 724; ^{1}H NMR(300 MHz, DMSO-d_6), δ: 0.78(3H, s, 18-CH_3), 0.94(3H, s, 19-CH_3), 2.78(1H, d, *J*=16.2 Hz, C6-βH), 3.65(1H, t, *J*=8.4 Hz, C17-H), 5.92(1H, s, C4-H), 6.82(1H, s, 5-thiazole-H), 7.33(2H, d, *J*=8.4 Hz, 3,5-PhH), 7.67(2H, d, *J*=8.4 Hz, 2,6-PhH), 9.87(1H, brs, —NH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 11.1(18-C), 17.6(19-C), 21.0 (11-C), 21.1(15-C), 23.4(16-C), 30.5(6-C), 31.8(2-C), 32.3 (1-C), 34.6(7-C), 35.8(8-C), 36.5(12-C), 37.6(10-C), 42.8 (13-C), 50.6(14-C), 53.7(9-C), 81.7(17-C), 103.7(5-thiazole-C), 120.4(4-C), 127.3, 128.8, 133.3, 133.5(Ph-C), 150.1(4-thiazole-C), 150.4(5-C), 155.1(3-C), 170.4(2-thiazole-C); HREIMS, *m/z*: 496.2189[M+H]$^+$(calcd. for $C_{28}H_{34}ClN_3OS$: 496.2188).

1-Testosteron-3′-ylidene-2-[4′-(*p*-bromophenyl)-2′-thiazolyl]-hydrazone(**18**): a light yellow solid, yield 12%, m. p. 235—237 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3441, 2936, 1624, 1551, 1472, 1392, 1260, 1068, 906, 826, 729; ^{1}H NMR(300 MHz, $CDCl_3$), δ: 0.78(3H, s, 18-CH_3), 0.96(3H, s, 19-CH_3), 2.46(1H, d, *J*=16.2 Hz, C6-βH), 3.65(1H, t, *J*=8.4 Hz, 17-CH), 5.92(1H, s, C4-H), 6.84(1H, s, 5-thiazole-H), 7.48(2H, d, *J*=8.4 Hz, 3,5-PhH), 7.61(2H, d, *J*=8.4 Hz, 2,6-PhH), 9.81(1H, brs, —NH); ^{13}C NMR(75 MHz, $CDCl_3$), δ: 11.0(18-C), 17.6(19-C),

18.4(11-C), 21.0(15-C), 23.4(16-C), 30.5(6-C), 31.7(2-C), 32.2 (1-C), 34.6(7-C), 35.8(8-C), 36.5(12-C), 37.6(10-C), 42.8 (13-C), 50.6(14-C), 53.7(9-C), 81.7(17-C), 103.8(5-thiazole-C), 120.4(4-C), 121.5, 127.6, 131.7, 133.9(Ph-C), 150.1 (4-thiazole-C), 150.4(5-C), 155.1(3-C), 170.3(2-thiazole-C); HREIMS, *m/z*: 540.1684[M+H]$^+$(calcd. for $C_{28}H_{34}BrN_3OS$: 540.1672).

*2.1.5 General Procedure for the Syntheses of Compounds **19** and **20***

To a solution of stannous chloride(220 mg) in 0.1 mL of concentrated hydrochloric acid, compound **3** or **8**(0.2 mmol) and ethyl acetate(4.0 mL) were added. The reaction mixture was heated under reflux for about 2 h until no starting material was observed. Then, the reaction was terminated and 4 mL of water was added to the reaction mixture after cooling to room temperature. The mixture was adjusted to pH=8—9 with 1 mol/L solution of NaOH. The product was extracted with ethyl acetate. The combined extract was washed with saturated brine, dried with anhydrous sodium sulfate, and evaporated under reduced pressure. The crude product was purified by silica gel chromatography with ethyl acetate/CH_2Cl_2(1/10, volume ratio) to give the corresponding target products.

1-Pregnenolon-20′-ylidene-2-[4′-(*p*-aminophenyl)-2′-thiazolyl]hydrazone(**19**): a crimson solid, yield 44%, m. p. 265—267 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3334, 2932, 1634, 1556, 1491, 1329, 1290, 1177, 1040, 826, 734; ^{1}H NMR(300 MHz, DMSO-d$_6$), δ: 0.57(3H, s, 18-CH$_3$), 0.94(3H, s, 19-CH$_3$), 1.90(3H, s, 21-CH$_3$), 3.23—3.31(1H, m, C3-αH), 4.61(1H, d, *J*=4.2 Hz, —OH), 5.19(2H, brs, —NH$_2$), 5.28(1H, d, *J*=3.6 Hz, C6-H), 6.55(2H, d, *J*=8.4 Hz, Ph-H), 6.77(1H, s, Ar-H), 7.51(2H, d, *J*=8.4 Hz, Ph-H), 10.49(1H, brs, —NH); ^{13}C NMR(75 MHz, DMSO-d$_6$), δ: 13.4(18-C), 18.7(19-C), 19.6 (21-C), 21.2(11-C), 23.5(15-C), 24.3(16-C), 31.7(2-C), 31.9 (7-C), 32.1(8-C), 36.6(12-C), 37.4(1-C), 38.7(10-C), 42.7(4-C), 43.6(13-C), 50.2(9-C), 56.3(14-C), 58.5(17-C), 70.5(3-C), 99.0(Ar-C), 120.8(6-C), 114.1, 123.8, 126.9, 132.5(Ph-C), 141.7(5-C), 148.6(Ar-C), 151.6(20-C), 170.5(C═N); HREIMS, *m/z*: 505.3001[M+H]$^+$(calcd. for $C_{30}H_{40}N_4OS$: 505.3001).

1-Estron-17′-ylidene-2-[4′-(*p*-aminophenyl)-2′-thiazolyl]-hydrazone(**20**): a crimson solid, yield 50%, m. p. 206—208 °C. IR(KBr), $\tilde{\nu}$/cm^{-1}: 3399, 2927, 1701, 1556, 1444, 1337, 1180, 916; ^{1}H NMR(300 MHz, CDCl$_3$), δ: 0.96(3H, s, 18-CH$_3$), 2.85—2.91(2H, m, 16-CH$_2$), 6.60(1H, d, *J*=1.8 Hz, C4-H), 6.64(1H, s, Ar-H), 6.64—6.68(1H, m, C2-H), 6.72(2H, d, *J*=8.4 Hz, Ph-H), 7.19(1H, d, *J*=8.7 Hz, C1-H), 7.60(2H, d, *J*=8.4 Hz, Ph-H); ^{13}C NMR(75 MHz, CDCl$_3$), δ: 17.2(18-C), 23.3(15-C), 25.8(11-C), 26.2(16-C), 27.2(7-C), 29.5(6-C), 34.1(12-C), 38.2(8-C), 44.1(13-C), 44.8(9-C), 52.6(14-C), 100.5(Ar-C), 112.8(2-C), 115.3(4-C), 126.5(1-C), 132.4(10-C), 138.1(5-C), 146.0(Ar-C), 115.1, 125.9, 127.1, 151.2(Ph-C), 153.5(3-C), 165.0(17-C), 169.6(C═N); HREIMS, *m/z*: 459.2219[M+H]$^+$ (calcd. for $C_{27}H_{30}N_4OS$: 459.2227).

2.2 Biological Assays

2.2.1 Materials

Stock solutions of the compounds were prepared in sterile dimethyl sulphoxide(DMSO, Sigma) at a concentration of 10 mg/mL and afterward diluted with complete nutrient medium RPMI-1640(HyClone) supplemented with 10%(volume fraction) heat inactivated fetal bovine serum and 0.1 g/L streptomycin sulfate+0.1 g/L penicillin G.

2.2.2 Cell Culture

HeLa, A549, HEPG2, CNE-2 cancer cells and HEK293T cells were grown in the RPMI-1640 medium supplemented with 10% heat inactivated fetal bovine serum and 0.1 g/L penicillin G+0.1 g/L streptomycin sulfate in a humidified atmosphere of 5% CO_2 at 37 °C.

2.2.3 Assay for Cell Viability

The anticancer activity *in vitro* was measured using the MTT assay. First, different concentrations of the compounds were added to the cells after cells(1×10^4—2×10^4 cell per well) were seeded in 96-well plates for 24 h. An equal amount of DMSO was added to cells as a negative control and *cis*-platinum as a positive control. For each individual dose, triplicate wells were prepared. Next, 190 μL of RPMI-1640 and 10 μL of MTT solution(5 mg/mL) were added to each well after reincubated for 72 h and the cells were washed with sterile phosphate buffer saline(PBS), and cells were incubated for additional 4 h. After the supernatant was discarded, the purple formazan crystals were dissolved in 200 μL of DMSO. Last, the plates were read on an automated microplate spectrophotometer(MLLTISKAN MK3, Thermo Scientific) at 492 nm after 10 min.

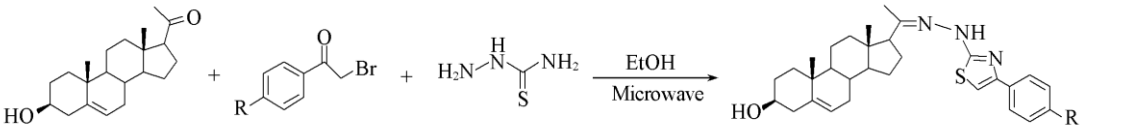
3 Results and Discussion

3.1 Chemistry

Microwave assisted synthesis has many advantages such as easy operation, simple post-treatment, low cost, high yield and no pollution, and has been applied widely in the field of organic synthesis. Ahluwalia *et al*.[16] reported the synthesis of some thiazolyl aromatic hydrazones *via* microwave irradiation method. Zhang *et al*.[17] prepared a series of *N*-[4-(coumarin-3-yl)thiazol-2-yl] aromatic aldehyde hydrazones *via* the condensation of thiosemicarbazone with α-bromoacetyl coumarin, using microwave heating method. Here, we synthesized a series of 4-substitutedphenyl thiazol steroidal ketone hydrazones *via* one-pot reaction under microwave irradiation using various steroidal ketones, thiosemicarbazide and 4-substituted-α-bromoacetophenones as reactive materials.

The conditions of microwave reaction were determined as microware power: 300 W, temperature: 85 °C, time: 30 min, solvent: EtOH by our investigation[18]. First, the microwave irradiation of a mixtuer of pregnenolone, thiosemicarbazide and 4-substituted-α-bromoacetophenones for 30 min afforded target compounds **1**—**5**(Scheme 1). The structures of products were characterized from their spectroscopic and analytical data. The ^{1}H NMR spectrum of compound **1** shows a characteristic aromatic singlet signal of 5-thiazole-H at δ 6.72, and the ^{13}C NMR spectrum of compound **1** exhibits signals for three aromatic carbons of thiazole at δ 101.7, 150.8 and 170.0. These data show that thiazole ring has been formed in the molecule of

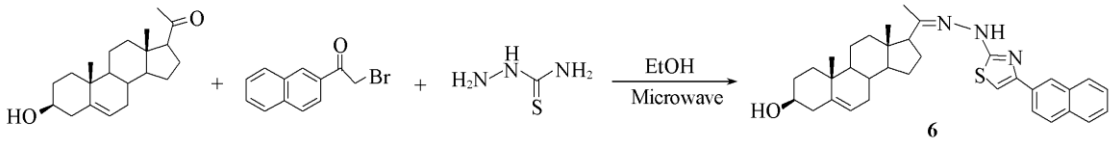
pregnenolone. The resonances signals of Ph-H at δ 6.93(2H, d, J=8.7 Hz), 7.73(2H, d, J=8.7 Hz) and Ph-C at δ 114.0, 127.1, 128.1, 159.2 prove the presence of a benzene ring in compound **1**. The HREIMS of compound **1** exhibits molecular ion peak at m/z 520.2998. All spectra demonstrate the structure of compound **1**.



1: R=OCH_3; **2**: R=CF_3; **3**: R=NO_2; **4**: R=CN; **5**: R=Br

Scheme 1 Synthetic route of compounds 1—5

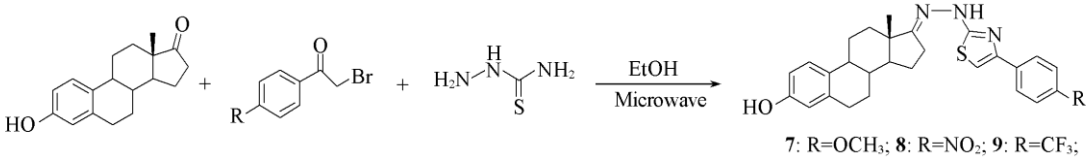
The microwave reaction of β-bromoacetylnathphalene with pregnenolone, thiosemicarbazide afforded compound **6**(Scheme 2). The structure of compound **6** was confirmed by analytical and spectral data.



6

Scheme 2 Synthetic route of compound 6

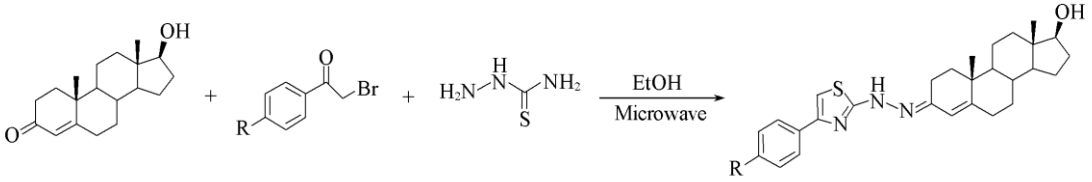
Similarly, series of new steroidal thiazole derivatives **7**—**12** were obtained by one-step microwave reaction *via* the reaction of estrone with the α-bromoacetophenone possessing different substituents such as OCH_3, CF_3, NO_2, CN etc., and thiosemicarbazide(Scheme 3). The structures of all compounds were characterized by IR, NMR and HRMS spectra.



7: R=OCH_3; **8**: R=NO_2; **9**: R=CF_3;
10: R=CN; **11**: R=Cl; **12**: R=Br

Scheme 3 Synthetic routes of compounds 7—12

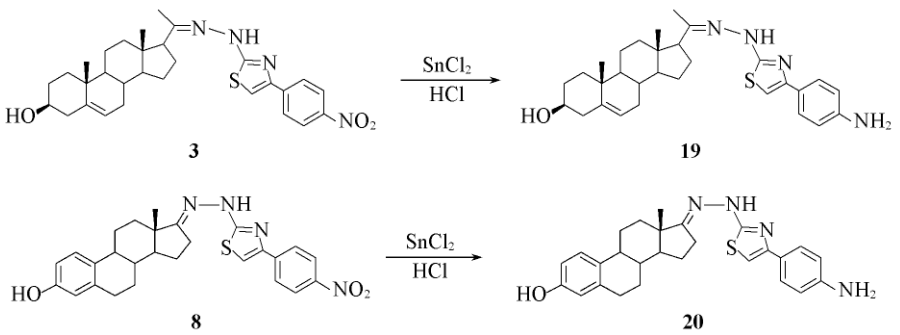
Our previous studies indicated that some 3, 6-disubstituted steroidal compounds with an oxime group, hydroxyl, oxime ether or thiosemicarbazone group showed a good cytotoxicity against some cancer cells[19—21]. To investigate the effect of thiazolyl at different positions of steroidal nucleus on biological activity, compounds **13**—**18** with a structure of 3-substituted thiazole were prepared by microwave assisted synthesis using testosterone as starting material(Scheme 4).



13: R=CF_3; **14**: R=CN; **15**: R=NO_2;
16: R=OCH_3; **17**: R=Cl; **18**: R=Br

Scheme 4 Synthetic routes of compounds 13—18

Furthermore, 4-aminophenyl steroidal thiazole derivatives (**19** and **20**) were obtained by the reduction of —NO_2 using stannous chloride as reductant(Scheme 5).



3 **19**

8 **20**

Scheme 5 Synthetic routes of compounds 19 and 20

3.2 Biological Evaluations

To determine the antiproliferative effect of the target steroidal thiazole compounds, we evaluated the cytotoxicity of compounds **1**—**20** *in vitro* against human cervical carcinoma (HeLa), human liver carcinoma(HepG2), human lung carcinoma(A549) and CNE 2(nasopharyngeal carcinoma) cell lines using MTT assay, and non-cancer cells HEK293T(normal kidney epithelial cells) were chosen as control. The results are shown as IC_{50} values in Table 1.

Table 1 ***In vitro*** **antiproliferative activities(IC_{50} in μmol/L) of compounds 1—20***

Compd.	HeLa	HepG2	A549	CNE-2	HEK293T
1	>100	>100	>100	ND	>100
2	>100	ND	61.0	ND	>100
3	>100	ND	>100	ND	>100
4	>100	>100	ND	>100	>100
5	>100	>100	ND	33.5	>100
6	>100	74.1	ND	24.1	>100
7	>100	>100	25.3	ND	>100
8	>100	>100	>100	ND	>100
9	53.9	>100	50.2	ND	>100
10	>100	>100	>100	ND	>100
11	20.9	>100	>100	ND	>100
12	54	5.7	82	11.5	>100
13	75.5	47.2	>100	ND	>100
14	>100	>100	ND	>100	>100
15	67.1	>100	>100	ND	>100
16	>100	81.4	75.3	ND	>100
17	>100	>100	>100	ND	>100
18	>100	>100	ND	46.3	>100
19	>100	ND	89.3	ND	>100
20	>100	>100	>100	ND	>100
cis-Platin	10.1	67	2.3	16.8	10.3

* ND: not determined.

From the data shown in Table 1, these steroidal thiazole derivatives show a poor antiproliferative activity against the tested cancer cells and normal kidney epithelial cells except for compound **12**. Compound **12** shows distinct cytotoxicity on all tested cancer cells and is almost inactive against HEK293T cells, and compound **11** possess a moderate cytotoxicity against HeLa cells. Compared with the result of our previous work[15], we think that a main cause of the lower activity of these compounds may be that a greater size of 5-phenyl group on thiazole ring blocks the interaction of compounds with the cells. We will study it further.

4 Conclusions

Series of novel steroidal thiazole derivatives of pregnenolone, estrone and testosterone were respectively synthesized and their structures were characterized by IR, NMR and HRMS. The anticancer activity of the compounds against human carcinoma cell lines and normal kidney epithelial cells(HEK293T) was screened. It was found that estrone thiazole derivative **12** displayed distinct antiproliferative activity against all the tested cancer cell lines and was almost inactive to normal kidney epithelial cells. However, other compounds didn't exhibit obvious antiproliferative activity to all the tested cells.

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