Synthesis, characterization and analgesic studies of novel thiazole derivatives of 4-piperidone

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Abstract: In this present study seven novel thiazole derivatives (PM3-PM9) were synthesized by cyclization of key intermediate thiosemicarbazone (PM2), derived from 4-piperidone (PM1). The parent 4-piperidone was synthesized by Mannich condensation reaction with good yield (89%). All the derivatives were characterized by UV, IR, ^{1}HNMR and mass spectral analysis. All the synthesized products were screened for their *in vivo* analgesic activities. Most of the tested compounds exhibited potential to reduce pain and some of them showed good analgesic properties. Thiosemicarbazone derivative showed most significant activity (*p*-value 0.01). All the thiazole derivatives exhibited dose dependent mild to good analgesic activities. Among thiazole derivatives, chloro and nitro substituted compounds (PM3, PM4, and PM5) showed highest analgesic activities.

Keywords: 4-piperidone, thiosemicarbazone, thiazole derivatives, analgesic activity, tail flick method.

INTRODUCTION

Piperidine alkaloid isolated from natural *Piper nigrum* (black peper) constitute a large number of compounds possessed a broad spectrum of pharmacological activities (Bhardwaj *et al*., 2002; Köhler, Rodrigues, Maurelli, & McCormick, 2002; Pae & Patkar, 2007; Rubiralta, Giralt, & Diez, 2013). Among piperidine derivatives, piperidin-4-one or 4-piperidone is mostly used as building block for synthesis of many important marketed bioactive molecules such as piperylone (analgesic, antipyretic), dorastine (anticancer), propiverine (anticholinergic), fentanyl (analgesic, anesthetic), clocapramine (antipsychotic), osanetant (schizophrenia) and pimozide (antipsychotic) (McLeod, Colvin, & Tankanow, 1985; Publishing, 2007; Vardanyan & Hruby, 2014).The organic reaction of thiosemicarbazide with carbonyl group of 4-piperidone is an elegant method to provide highly functionalized intermediate, thiosemicarbazone which is used for preparation of biologically active heterocyclic compounds such as thiazole. Thiazole is present in many natural and synthetic medicinally important compounds such as thiamine, penicillin, sulfazole, ritonavir and tiazofurin (He *et al*., 2016; Jeankumar *et al*.,2013; Lu *et al*., 2012; Reddy *et al*., 2016). Thiazoles have shown antimicrobial, antiretroviral, antifungal, antihistaminic, anti-inflammatory and anti-thyroid activities. Numerous substituted thiazole derivatives are reported to possess significant analgesic properties (A Muhammad, S Masaret, M Amin, A Abdallah, & A Farghaly, 2017; Carter *et al*., 1999; Kalkhambkar, Kulkarni, Shivkumar, & Rao, 2007; Thore, Gupta, & Baheti, 2013). These bioactivities of thiazole, piperidine and their derivatives impelled to design simple methods for the synthesis of piperidine based thiazole derivatives having broad spectrum activities.

MATERIALS AND METHODS

All the reagents used in synthesis were purchased from TCI (Japan) chemical company. Analytical grade ethanol, methanol, acetic acid and ether were purchased from E. Merck. TLC plate Kieselgel 60 (GF-254) was used to monitor the progress of reaction. Melting points was recorded on Buchi 434 instrument and are uncorrected. Shimadzu UV-1600 spectrophotometer was used for ultra violet spectrum in methanol. For infra-red analysis JASCO Fourier transform infrared spectrophotometer was used by making KBr disc method. Mass spectra were recorded by using MAT112 and JEOL electronic impact low resolution mass spectrum (JMS). ^{1}HNMR spectral analysis was performed on Brucker AM 300, and 500MHz spectrophotometer using tetramethylsilane (TMS) as an internal standard.

Synthesis

Synthesis and spectral analysis of 3, 3-dimethyl-2,6-diphenylpiperidin-4-one (PM1)

Compound PM1 was synthesized by adopting one pot total synthesis method as reported by Noller and Baliah (Noller & Baliah, 1948). In reaction flask ammonium acetate (0.1 mol), ketone (0.1 mol) and benzaldehyde (0.2

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mol) were taken in ethanol (30ml) and allowed to reflux. After the completion of reaction confirm by using TLC, the mixture was treated with dilute hydrochloric acid to form precipitates of product. Precipitates were filtered and washed with ethanol-ether (4:1) solution. The precipitates in acetone were treated with aqueous ammonia followed by addition of excess water which librates product in the form of free base (scheme-1). Product was recrystallized from absolute ethanol.

Compound PM1($C_{19}H_{21}NO_2$); %yield: 82; m.p: 247±01; $UV_{\lambda max}$ (MeOH)nm:262, 250, 204; $IR\upsilon_{max}$(KBr)cm^{-1}:3352, 2901, 1725, 1579, 1135, 856, 706; EIMS m/z (%): 279 M^+ (87), 194(100), 147(50), 106(47), 70(47.7); ^{1}H-NMR ($CDCI_3$, 300MHz) δ(ppm): 0.950 (s, 3H, H-14), 1.100 (s, 3H, H-13), 2.452-2.445 (d. J=2.8 Hz, 3H, H-5b), 2.487-2.479 (d, J=3.2 Hz, 3H, H-5a), 3.815 (s, 1H, H-2), 4.067-4.031 (t, 3H, H-6), 7.500-7.263 (m, 10H, H-8, 9, 10, 11, 12, 17, 18, 19, 20, 21)

Synthesis and spectral analysis of (Z)-2-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene) hydrazinecarbo-thioamide(PM2)

Compound PM2 was prepared by refluxing a mixture of 3, 3-dimethyl-2,6-diphenylpiperidin-4-one (0.01 mol) and thiosemicarbazide (0.01 mol) in mild acidic ethanol (50 ml) for three hours with continuous stirring (scheme-1). Progress of reaction was monitored with TLC. Precipitates of products were obtained by pouring the content of reaction flask in ice cold water. The precipitated product was purified by washing with cold distilled water and recrystallization from hot absolute ethanol.

Compound PM2($C_{20}H_{24}N_4S$); %yield: 86.5; m.p: 157± 02; $UV_{\lambda max}$(MeOH)nm: 273, 207; $IR\upsilon_{max}$(KBr)cm^{-1}: 3411, 3253, 2906, 1590, 1137, 861; EIMS m/z (%): 352 M^+ (51), 277(29), 247(36), 230(23), 194(100), 172(40), 131(24), 104(58), 82(47); ^{1}H-NMR ($CDCI_3$, 300MHz) δ(ppm): 1.174 (s, 6H, H-13, 14), 2.422-2.410 (m, 3H, H-5b), 2.621-2.479 (m, 3H, H-1, 5a), 3.327 (s, 1H, H-2), 3.803-3.769 (t, 3H, H-6), 7.787-7.260 (m, 10ArH, H-8, 9, 10, 11, 12, 20, 21, 22, 23, 24), 8.195 (s, 2H, H-25), 10.420 (s, 1H, H-16).

Synthesis of 3, 3-dimethyl-2,6-diphenylpiperidin-4-one thiazole derivatives (PM3-PM9)

Compounds PM3-PM9 were synthesized by reacting an equimolar amount (0.01 mol) of (Z)-2-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene) hydrazine carbothioamide (PM2) and substituted phenacyl halides in ethanol (30 ml) with 2-3drops of tri ethyl amine (TEA) at room temperature with continuous stirring for two hours. After completion of reaction the content were poured in ice cold water and precipitate obtained were purified by washing and recrystallization from hot ethanol.

(Z)-4-(4-chlorophenyl)-2-(2-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene)hydrazinyl) thiazole (PM3)

Compound PM3($C_{28}H_{27}ClN_4S$);%yield: 69; m.p: 202±04; $UV_{\lambda max}$(MeOH)nm: 273, 252, 207; $IR\upsilon_{max}$(KBr)cm^{-1}: 3422, 2920, 2363, 1585, 1471, 1193 ; EIMS m/z (%): 486 M^+(54), 381(85), 348(12), 312(64), 277(20), 210(45), 194(100), 168(21), 104(26), 91(14), 77(10); ^{1}H-NMR ($CDCI_3$, 500MHz)δ(ppm); 1.103 (s ,6H, H-20, 21), 2.316-2.281 (m, 3H, H-13b), 2.672-2.641 (m, 4H, H-11, 13a), 3.684 (s, 1H, H-10), 3.922-3.913 (m, 4H, H-12), 7.431-7.320 (m, 11H, H-23, 24, 25, 26, 27, 29, 30, 31, 32, 33, 5), 7.720-7.717 (d, J=1.5Hz, 2H, H-16, 18), 8.213-8.197 (d, J= 8Hz, 2H, H-15, 19), 10.710 (s, 1H, H-6).

(Z)-2-(2-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene)hydrazinyl)-4-(3-nitrophenyl) thiazole (PM4)

Compound PM4($C_{28}H_{27}N_5O_2S$);%yield: 65;m.p: 199± 0.5; $UV_{\lambda max}$(MeOH)nm:271, 239, 211; $IR\upsilon_{max}$(KBr) cm^{-1}: 3312, 2910, 2315, 1558, 1480, 1085, 820; EIMS m/z (%): 497 M^+ (40), 465(15), 392(42), 323(20), 221(14), 194(100), 172(21), 104(14), 91(17), 77(9); ^{1}H-NMR ($CDCI_3$, 500MHz) δ(ppm): 1.125 (s, 6H, H-20, 21), 2.300-2.288 (m, 3H, H-13b), 2.665-2.652 (m, 4H, H-11, 13a), 3.601 (s, 1H, H-10), 3.914-3.910 (m, 4H, H-12), 7.450-7.481 (m, 11H, H-23, 24, 25, 26, 27. 29, 30, 31, 32, 33, 5), 7.766-7.760 (t, 3H, H-18), 8.202-8.152 (d, J= 25 Hz, 2H, H-19), 8.422-8.478 (d, J= 5, Hz, 2H, H-17), 8.675 (s, 1H, H-15), 11.853 (s, 1H, H-6).

(Z)-2-(2-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene)hydrazinyl)-4-(4-nitrophenyl) thiazole (PM5)

Compound PM5($C_{28}H_{27}N_5O_2S$); %yield: 75; m.p: 197±01;$UV_{\lambda max}$(MeOH)nm:292, 263; $IR\upsilon_{max}$(KBr) cm^{-1}: 3336, 2912, 2316, 1565, 1452, 1134 ; EIMS m/z (%): 497 M^+ (52), 392(90), 323(46), 221(24), 194(100), 172(16), 104(28), 91(16), 77(21); ^{1}H-NMR ($CDCI_3$, 500MHz) δ(ppm): 1.170 (s, 6H, H-20, 21), 2.325-2.290 (m, 3H, H-13b), 2.591-2.549 (m, 4H, H-11, 13a), 3.567 (s, 1H, H-10), 3.903-3.897 (m, 4H, H-12), 7.642-7.631 (m, 11H, H-23, 24, 25, 26, 27. 29, 30, 31, 32, 33, 5), 7.931-7.922 (d *J*= 4.5Hz, 2H, H-15, 19), 8.229-8.215 (d, J=7 Hz, 2H, H-16,18), 10.967 (s, 1H, H-6)

(Z)-4-(4-bromophenyl)-2-(2-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene)hydrazinyl) thiazole (PM6)

Compound PM6($C_{28}H_{27}BrN_4S$); %yield: 72; m.p:180± 0.5;$UV_{\lambda max}$(MeOH)nm: 272, 253, 209; $IR\upsilon_{max}$(KBr) cm^{-1}: 3407, 2919, 2321, 1581, 1402, 1202; EIMS m/z (%): 530 M^+(51), 425(57), 392(18), 358(33), 277(16), 255(38), 194(100), 172(14), 130(15), 104(32), 91(16); ^{1}H-NMR ($CDCI_3$, 500MHz) δ(ppm); 1.121 (s ,6H, H-20, 21), 2.296-2.282 (m, 3H, H-13b), 2.424-2.411 (m, 4H, H-11, 13a), 3.486 (s, 1H, H-10), 3.893-3.882 (m, 4H, H-12), 7.531-7.520 (m, 11H, H-23, 24, 25, 26, 27, 29, 30, 31, 32, 33, 5), 7.723-7.718 (d, J=2.5Hz, 2H, H-16, 18), 7.823-7.185 (d, J= 4Hz, 2H, H-15, 19), 11.044 (s, 1H, H-6).

Table 1: Analgesic activity of PM1-PM9 at 15 mg/kg in comparison with aspirin and control

Comp.	Dose	LATENCY TIME (seconds)						
		0min	30 min	60 min	90 min	120 min	150 min	180 min
PM1	15mg/kg	0.90±0.04 *(*)	1.46±0.04 **(*)	2.03±0.07 **(**)	2.58±0.25 **(**)	2.12±0.15 **(**)	1.46±0.11 **(**)	1.11±0.08 **(*)
PM2		0.85±0.04 *(**)	1.79±0.08 **(*)	2.15±0.09 **(*)	2.37±0.08 **(**)	2.01±0.05 **(**)	1.26±0.06 **(**)	0.93±0.05 **(**)
PM3		0.92±0.08 *(*)	1.42±0.09 **(*)	1.82±0.04 **(**)	2.23±0.08 **(**)	1.94±0.11 **(**)	1.40±0.07 **(**)	0.99±0.05 **(**)
PM4		0.82±0.07 *(**)	1.31±0.11 **(**)	1.68±0.15 **(**)	1.84±0.26 **(**)	1.60±0.23 **(**)	1.15±0.07 **(**)	0.90±0.03 **(**)
PM5		0.85±0.04 *(**)	1.17±0.05 **(**)	1.67±0.05 **(**)	2.14±0.13 **(**)	1.65±0.04 **(**)	1.08±0.05 **(**)	0.90±0.04 **(**)
PM6		0.86±0.04 *(**)	1.11±0.04 **(**)	1.57±0.10 **(**)	1.98±0.04 **(**)	1.58±0.05 **(**)	1.16±0.05 **(**)	1.01±0.03 **(**)
PM7		0.84±0.05 *(**)	1.06±0.04 **(**)	1.57±0.04 **(**)	1.85±0.10 **(**)	1.49±0.16 **(**)	1.11±0.03 **(**)	0.87±0.05 **(**)
PM8		0.85±0.04 *(**)	1.16±0.04 **(**)	1.54±0.11 **(**)	1.92±0.05 **(**)	1.46±0.08 **(**)	1.20±0.08 **(**)	1.00±0.08 **(**)
PM9		0.81±0.05 *(**)	1.12±0.08 **(**)	1.46±0.04 **(**)	1.73±0.07 **(**)	1.29±0.09 **(**)	1.05±0.04 **(**)	0.67±0.29 **(**)
Control	10mg/kg	0.86 ±0.03	0.89 ±0.03	0.98±0.03	1.02±0.05	1.01±0.06	0.99 ±0.04	0.97 ±0.04
Aspirin		0.93±0.02	1.98±0.03	2.91± 0.03	3.71±0.07	3.15±0.02	2.13±0.04	1.68±0.03

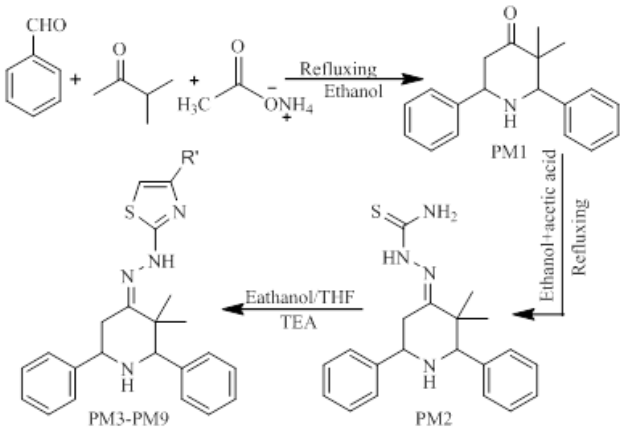
Table 2: Analgesic activity of PM1-PM9 at 30 mg/kg in comparison with aspirin and control

Comp.	Dose	LATENCY TIME (seconds)						
		0min	30 min	60 min	90 min	120 min	150 min	180 min
PM1	30mg/kg	0.88±0.02 **(**)	3.23±0.31 **(**)	4.16±0.17 **(**)	5.00±0.06 **(**)	4.16±0.19 **(**)	2.97±0.15 **(**)	2.09±0.12 **(**)
PM2		0.84±0.04 **(**)	3.44±0.45 **(**)	4.14±0.18 **(**)	4.95±0.20 **(**)	3.95±0.09 **(**)	2.57±0.13 **(**)	1.64±0.21 **(*)
PM3		0.83±0.06 **(**)	2.94±0.11 **(**)	3.39±.25 **(**)	4.28±0.24 **(**)	3.96±0.22 **(**)	2.38±.34 **(**)	1.55±0.23 **(**)
PM4		0.84±0.05 **(**)	2.40±0.32 **(**)	3.29±0.30 **(**)	3.59±0.48 **(*)	2.93±0.21 **(*)	2.37±0.17 **(**)	1.64±0.14 **(*)
PM5		0.88±0.03 **(**)	2.26±0.16 **(**)	3.26±0.12 **(**)	3.96±0.06 **(*)	3.26±0.11 **(*)	2.22±0.07 **(*)	1.52±0.09 **(**)
PM6		0.85±0.04 **(**)	2.13±0.09 **(**)	3.24±0.15 **(**)	3.97±0.07 **(*)	3.23±0.13 **(*)	2.25±0.12 **(*)	1.44±0.08 **(**)
PM7		0.87±0.04 **(**)	2.10±0.09 **(**)	3.27±0.16 **(**)	3.78±0.20 **(*)	2.82±0.11 **(**)	2.06±0.09 **(*)	1.450±.07 **(**)
PM8		0.84±0.04 **(**)	2.19±0.11 **(**)	3.07±0.17 **(**)	3.47±0.20 **(*)	2.48±0.05 **(**)	1.56±0.09 **(**)	1.15±0.07 **(**)
PM9		0.88±0.04 *(**)	2.10±0.07 **(**)	2.57±0.10 **(**)	3.24±0.08 **(*)	2.59±0.08 **(**)	1.54±0.16 **(**)	1.15±0.03 **(**)
Control	10mg/kg	0.86 ±0.03	0.89 ±0.03	0.98±0.03	1.02±0.05	1.01±0.06	0.99 ±0.04	0.97 ±0.04
Aspirin		0.93±0.02	1.98±0.03	2.9± 0.03	3.71±0.07	3.15±0.02	2.13±0.04	1.68±0.03

n=7 values are mean±2SEM
*= $p<0.05$ significant difference as compared to control (10%DMSO)
**= $p<0.01$ highly significant difference as compared to control
(*)=$p<0.05$ significant difference as compared to standard (aspirin)
(**)=$p<0.01$ highly significant difference as compared to standard (aspirin)

(Z)-1-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene)-2-(4-(4-methoxyphenyl)thiazole-2-yl)hydrazine (PM7)

Compound PM7($C_{28}H_{30}N_4OS$);%yield: 63;m.p:172± 01; $UV_{\lambda max}$ (MeOH) nm: 276, 212, 205; $IR\upsilon_{max}$(KBr) cm^{-1}: 3413, 2905, 2391, 1594, 1140, 859; EIMS m/z (%): 482 M^+(21), 391(12), 377(26), 358(33), 279(71), 208(11), 194(100), 147(70), 135(35), 104(73), 91(25); ^{1}H-NMR ($CDCI_3$, 500MHz) δ(ppm); 1.085 (s ,6H, H-20, 21), 2.245-2.240 (m, 3H, H-13b), 2.521-2.510 (m, 4H, H-11, 13a), 3.415 (s, 1H, H-10), 3.564(s, 3H,H-35), 3.957-3.950 (m, 4H, H-12), 7.022-7.008(d, J=7Hz, 2H, H-16, 18), 7.511-7.508 (m, 11H, H-23, 24, 25, 26, 27, 29, 30, 31, 32, 33, 5), 7.651-7.648 (d, J=1.5Hz, 2H, H-15, 19), 10.565(s, 1H, H-6).



Where R' = 4-Cl-phenyl, 4-NO_2-phenyl, 3-NO_2-phenyl, 4-Br-phenyl, 4-OCH_3-phenyl, naphthalene ring and 3,4-dihydoxy phenyl

Scheme 1: synthetic procedures of compounds PM1-PM9

(Z)-2-(2-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene)hydrazinyl)-4-naphthalen-2-yl) thiazole (PM8)

Compound PM8 ($C_{32}H_{30}N_4S$);%yield: 169;m.p: 81±02;$UV_{\lambda max}$(MeOH)nm: 287, 247; $IR\upsilon_{max}$(KBr)cm^{-1}: 3445, 2932, 2315, 1575, 1215; EIMS m/z (%): 502 M^+(51), 397(79), 362(11), 328(36), 314(29), 226(79), 194(100), 184(48), 172(15), 155(29), 127(27), 91(17); ^{1}H-NMR ($CDCI_3$, 500MHz) δ(ppm); 1.085 (s ,6H, H-20, 21), 2.306-2.230 (m, 3H, H-13b), 2.508-2.496 (m, 4H, H-11, 13a), 3.536 (s, 1H, H-10), 3.934-3.926 (m, 4H, H-12), 7.624-7.615 (m, 9H, H- 27, 29, 30, 31, 32, 33, 36, 37, 5), 7.712-7.706 (m, 2H, H-21.22), 8.022-8.016 (m, 4H, H-18, 19, 20, 23), 8.322(s,1H,H-15), 10.254 (s, 1H, H-6).

(Z)-4-(2-(2-(3,3-dimethyl-2,6-diphenylpiperidin-4-ylidene)hydrazinyl)thiazole benzene-1,2-diol(PM9)

Compound PM9 ($C_{28}H_{28}N_4O_2S$); %yield: 55;m.p: 150± 01;$UV_{\lambda max}$(MeOH)nm: 263; $IR\upsilon_{max}$ (KBr) cm^{-1}: 3353, 2932, 2955, 1564, 1256; EIMS m/z (%): 484 M^+(24), 379(52), 344(20), 296(63), 277(34), 224(47), 194(162), 171(71), 145(14), 131(100), 104(75); ^{1}H-NMR ($CDCI_3$, 500MHz) δ(ppm); 1.087 (s ,6H, H-20, 21), 2.198-2.177 (m, 3H, H-13b), 2.397-2.387 (m, 4H, H-11, 13a), 3.516 (s, 1H, H-10), 3.946-3.936 (m, 4H, H-12), 6.186(s, 1H, H-16), 6.920-6.918(m, 2H, H-15, 19), 7.431-7.429(m, 11H, H-23, 24, 25, 26, 27, 29, 30, 31, 32, 33, 5), 9.485 (s, 2H, H-34, 35), 10.963 (s, 1H, H-6).

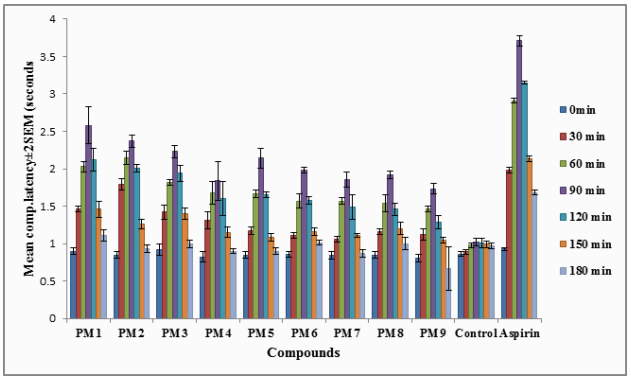


Fig. 1: Analgesic activity of PM1-PM9 at 15 mg/kg

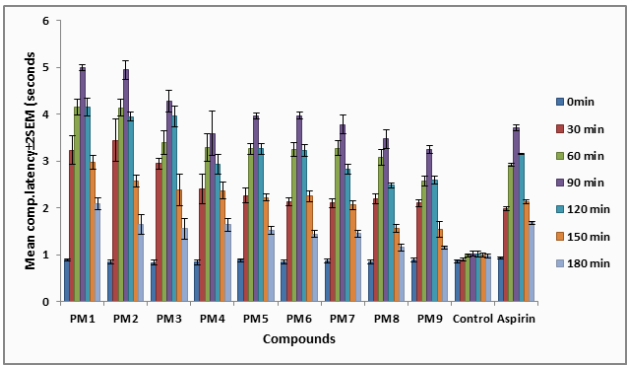


Fig. 2: Analgesic activity of PM1-PM9 at 30 mg/kg

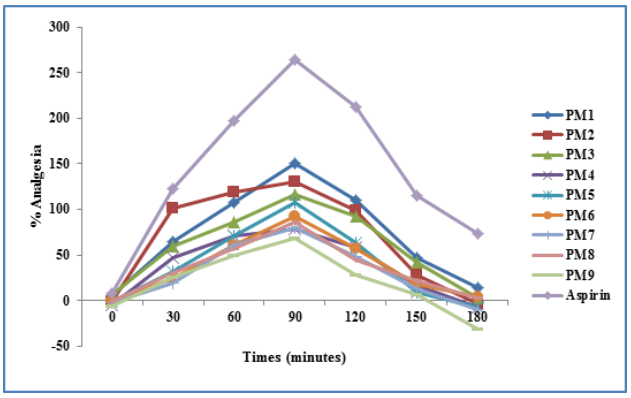


Fig. 3: Percentage pain protection of PM1-PM9 at dose 15 mg/kg

Analgesic activity

Animals

Healthy mice (Swiss albino) of either sex of 20-25g were taken from animal house of department of pharmacology, University of Karachi. These mice were acclimatized for a weak before procedure under maintained laboratory conditions of temperature 25±04°C and humidity 45-50%. They were placed in proper plastic cages and exposed to alternate 12 hours light and dark cycles and supplied with

standard specified food and water. The entire procedure was maintained in compliance with the animal care and use ethics as accepted internationally(Naderi, Sarvari, Milanifar, Boroujeni, & Akhondi, 2012; Retnam *et al*., 2016).

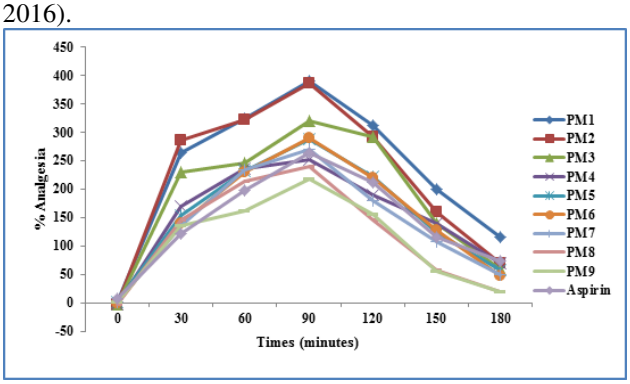


Fig. 4: Percentage pain protection of PM1-PM9 at dose 30 mg/kg

Acute toxicity test

Before the selection of doses for procedure acute toxicity test were performed on a group of mice (n=5) by administrating orally a single dose of 100 mg/kg of each synthesized compounds (PM1-PM9). After two days of administration no mortality was observed all animals were conscious and showed normal behavior without any symptom of allergy.

Dose preparation

Animals were divided into three groups having seven mice in each groups (n=7). Dose of 10 ml/kg of DMSO (10%) was used for control groups. Aspirin was used in dose of 10 mg/kg for standard group. Test groups were treated with two selected dose of 15 and 30 mg/kg of PM1-PM9. All the drugs were administered orally.

Tail flick method

To evaluate the analgesic potential of synthesized compounds D' Amour and Smith hot water tail immersion method (D'Amour & Smith, 1941) was used. Water bath maintained at constant temperature of 55±01°C was used to record the reaction time i.e., flicking of tail immersed in hot water bath in seconds after 0, 30, 60, 90, 150 and180 minutes of administration of drugs. To avoid any damage to tail cut off time for immersion for all groups were maintained at 30 seconds (Ganeshpurkar & Rai, 2013; Reyes, Abdulla, Jaime Jr Bisa, & Solidum). This reaction time is called as latency time. The percent pain protection against thermal stimuli was calculated by applying following formula:

$$\text{Percent pain protection} = \frac{\text{Drug mean latency time} - \text{Control mean latency time}}{\text{Control mean latency time}} \times 100$$

STATISTICAL ANALYSIS

For statistical analysis of recorded latency time software program statistical package for social science (SPSS) was used. Results were expressed as mean standard error (mean ± 2SEM). To compare the mean differences among the independent groups one way ANOVA was performed and Tukey's test was applied to compare the differences among individual groups. The significant difference value ($p<0.05$) and highly significant difference value ($p<0.01$) was shown by * and ** for control group while (*) and (**) for aspirin.

RESULTS

All the compounds were synthesized with good yield in pure form. Synthesized compounds were evaluated for analgesic activity. The results of analgesic potential of compounds are presented in tables 1-2 and in figs. 1-4.

DISCUSSION

Chemistry

Structures of synthesized compounds were well confirmed by spectrometric techniques. Characteristic absorption band in UV spectrum of PM1 of benzene and carbonyl group at 204 and 250 nm and λ_{max} at 273 nm in PM2 showed the formation of piperidone and thiosemicarbazone derivative. Thiazole derivatives showed absorption band at 252 and 273 nm due to transition of π electron (π-π*) in the phenyl rings and transition within the imine group of thiazole. The characteristic strong absorption band at 1725 cm^{-1} of carbonyl group (C=O) of PM1 in FTIR spectrum was replaced by band of carbon nitrogen group at 1590 cm^{-1} (C=N). Strong stretching bands at 3411 and 3253 cm^{-1} showed the presence of primary (NH_2) and secondary (NH) amino groups of thiosemicarbazone compound. The broad stretching and strong bending bands between 3445-3314 cm^{-1} and at 1549-1593 cm^{-1} in thiazole derivatives (PM3-PM9) also confirmed the presence of C-N bond in compounds(Gosavi & Rao, 1967; Santhakumari, Ramamurthi, Vasuki, Yamin, & Bhagavannarayana, 2010). The EIMS of compounds were found to be consistent with projected molecular structures. The ^{1}HNMR spectra of compounds displayed characteristic peaks between 1.032-0.821ppm and 7.481-7.218 ppm for hydrogen of dimethyl group and diaryl ring of 4-piperidone respectively. Characteristic singlet around 11.51, 8.07 and 10.50 ppm also present in ^{1}HNMR spectra for primary and secondary amines.

Analgesic activity

For the treatment of all type of pain various analgesic agents are available but these agents showed serious side effects such as nausea, gastrointestinal (GI) bleeding, heartburn and physical dependence. COX-2 inhibitors protect the GI tract but have more risk of cardiovascular effects. Therefore, it is required to design new more potent analgesic agents with fewer side effects; in this regard various efforts have been reported including

piperidine derivatives. Recently designed piperidin-4-one derivatives also showed significant analgesic activities.

Synthesized compounds showed dose dependent analgesic activity, latency time increases with the increase in dose. At 30 mg/kg dose all the compounds showed significant analgesic activity ($p<0.01$) throughout the period of evaluation as compared to standard aspirin and control. Compounds displayed rapid onset of analgesic activity, at 30 minutes percent analgesia as compared to group of control and standard was 136-280% which increases with time and showed maximum activity after 90 minutes of administration of drug that is 200-390%. Latency time decreases gradually with time.

During structure activity relationship study of synthesized compounds it was observed that when carbonyl group of piperidone was replaced with thiosemicarbazide group analgesic potential increases as compared to standard aspirin and other derivatives. Analgesic activity decreases when this thiosemicarbazone compounds cyclized into thiazole derivatives. Among thiazole derivatives the most significant activity observed in compound having substituted chloro(Cl) and nitro(NO_2) group at para position with the dose of 30 mg/kg.

Sulphur and nitrogen heteroatoms in synthesized thiosemicarbazone, provide binding sites and hence influence the pharmacokinetics and activity of compound. Activity of synthesized substituted thiazole derivatives is also attributed to the heteroatoms, electron donating and withdrawing effects, and this can be displayed through interaction with receptor at electronic level.

CONCLUSIONS

Synthesis is a dynamic process and the searching for new biologically active heterocyclic analogues continues to be a demanding research in medicinal chemistry to combat the new challenges. Slight modification in structure may produce tremendous change in activity level. By using simple synthetic procedures series of compounds were efficiently synthesized. It is clear from the foregoing study that the newly synthesized compounds constitute an interesting template for future designing of synthetic analgesic drug after detailed pharmacological studies.

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