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Synthesis and Anticancer Activity of Some Novel 1h-Pyrazol-5(4*H*)-One Derivatives.

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ABSTRACT

Refluxing pyrazolone with aromatic aldehydes in ethanol and in the presence of catalytic amount of piperidine afforded the Shift's bases **2a-e**. When pyrazolone derivative **1** react with different diazonium salts, compounds **3a-c** were obtained. On the other hand, glycosides **4a-c** were produced when derivative **1** heated under reflux with different aldohexoses and aldopentoses in dioxane and few drops of piperdine. On treatment of compound **1** with phosphorous penta sulfide in dry pyridine, the thione derivative was obtained. When the potassium salt of the latter compound was stirred at room temperature with either semi sugars or tetra acetylated bromo sugars in dry DMF, compounds **7a-d** and **8a,b** were obtained respectively. Six out of the prepared compounds had been directed to anti-tumor activities against three human cancer cell lines using MTT assay. Two compounds showed good anticancer activities.

Keywords: Pyrazolone-aldehydes-diazonium salts- sugars-anticancer

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INTRODUCTION

Cancer still remains a mean threat to human health; it is a major cause of death worldwide. Therefore, the development of highly efficient, selective and less noxious antitumor drug remains an urgent need [1].

Pyrazolone ring have more pharmacological activities as antitubercular [2], antimicrobial [3], anticancer [4,5], antiviral [6], analgesic [7,8], anti-inflammatory [9, 10], antipyretic [10, 11], ulcerogenic [12], lipid peroxidation [12].

The design of new pyrazole derivatives has been an unique position in the drug discovery due to their broad range of biological activities, such as anti-inflammatory, antiangiogenic, antibacterial, antimicrobial, anticancer, antioxidant, ant influenza and analgesic activities [13-17]. Several recent studies suggest pyrazole derivatives as promising anticancer agents [18-22].

Since *N*-substitutions in pyrazolone exhibit biologically active compounds [23], we carried out some reactions on 3-methyl-1*H*-pyrazol-5(4*H*)-one (1) with various electrophilic reagents which expected that the newly obtained compounds possible have anticancer activity.

EXPERIMENTAL

Chemistry

General

Melting points were uncorrected and measured using an Electro-thermal IA 9100 apparatus (Shimadzu, Japan). Micro analytical data were performed by Vario El-Mentar apparatus (Shimadzu, Japan), National Research Centre, Cairo, Egypt. IR spectra were recorded (as potassium bromide pellets) using KBr disc technique on a Perkin-Elmer 1650 Spectrophotometer (υ, cm^{-1}), National Research Centre, Dokki, Egypt. NMR experiments were determined on a JEOL-Ex-300 MHz in deuterated dimethyl sulfoxide (DMSO – d_6) and chemical shifts were expressed as parts per million; ppm (δ values) against TMS as an internal reference (Faculty of Science, Cairo University, Cairo, Egypt). Mass spectra were recorded on Shimadzu GCMS-QP-1000EX mass spectrometer at 70 eV (Cairo University, Cairo, Egypt).

Synthesis of 3-methyl-1*H*-pyrazol-5(4*H*)-one (1)

This compound was prepared according to the reported method (A) by the reaction of ethyl acetoacetate with hydrazine hydrate in presence of ethanol to form 3-methyl-1*H*-pyrazol-5(4H)-one yield 89%; mp 222-225 °C; IR υ 1650 (C=O) 1542 (C=N) 3380cm^{-1} (NH). ^{1}H-NMR (DMSO-d_6) δ: 2.07 (s, -CH_3, 3H), 5.20 (s, -CH, 1H), 10.50 (s, -NH, 2H), MS ($C_4H_6N_2O$) *m/z* = 99 (M^+).

Synthesis of 4-aryylidene-3-methyl-1*H*-pyrazol-5(4*H*)-one (2a-e)

To a mixture of 3-methyl-1*H*-pyrazol-5(4H)-one (**1**) (1 mmol) and the appropriate aldehyde (1 mmol) in dioxan or absolute ethanol (25 ml), few drops of piperidine were added. The reaction mixture was refluxed for 4-6 hrs, and then cooled to room temperature. The precipitate was filtered, dried and washed with ethanol. The product was recrystallized from EtOH to afford compounds **2a-e**, respectively.

3-Methyl-4-(3-phenylallylidene)-1*H*-pyrazol-5(4*H*)-one (2a)

Yield (69%); m.p. 210°C; IR υ 3230 (NH), 1720 (C=O) cm^{-1}; ^{1}H NMR (DSMO-d_6) δ 2.31 (s, 3H, -CH_3), 6.95 (d, *J* = 9 Hz, 2H, ArH), 7.10-7.95 (m, 6H, 4ArH + 2CH=), 11.30 (s, 1H, D_2O exchangeable, NH); MS ($C_{13}H_{12}N_2O$) *m/z* = 212 (M^+).

4-(2-Chlorobenzylidene)-3-methyl-1*H*-pyrazol-5(4*H*)-one (2b)

Yield (67%); m.p. 190°C; IR υ 3235 (NH), 1725 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.32 (s, 3H, -CH_3), 6.95 (d, *J* = 9 Hz, 2H, ArH), 7.39 (s, 1H, -CH=), 7.42 (d, 2H, ArH), 11.3 (s, 1H, D_2O exchangeable, NH); MS ($C_{17}H_{13}BrN_2O_2$) *m/z* = 358 (M^+).

4-(4-(Dimethylamino)benzylidene)-3-methyl-1*H*-pyrazol-5(4*H*)-one (2c)

Yield (87%); m.p. 180°C; IR υ 3230 (NH), 1730 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.34 (s, 3H, -CH_3), 3.08 (s, 6H,-$N(CH_3)_2$), 7.13(d, *J* = 9 Hz, 2H, ArH), 7.20 (s, 1H, -CH=), 7.32 (d, 2H, ArH), 11.09 (s, 1H, D_2O exchangeable, NH); MS ($C_{13}H_{15}N_3O$) *m/z* = 230 (M^+).

4-(4-Hydroxybenzylidene)-3-methyl-1*H*-pyrazol-5(4*H*)-one (2d)

Yield (72%); m.p. 240°C; IR υ 3220 (NH), 1725 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.33 (s, 3H, -CH_3), 6.75 (d, *J* = 9 Hz, 2H, ArH), 7.51 (s, 1H, -CH=), 7.58 (d, *J* = 9 Hz, 2H, ArH), 11.14 (s, 1H, D_2O exchangeable, NH); MS ($C_{11}H_{10}N_2O_2$) *m/z* = 203 (M^+).

4-(3,4-Dimethoxybenzylidene)-3-methyl-1*H*-pyrazol-5(4*H*)-one (2e)

Yield (75%); m.p. 160°C; IR υ 3230 (NH), 1730 (C=O) cm^{-1}; ^1H-NMR (DSMO-d_6) δ 2.36 (s, 3H, -CH_3), 3.83 (s, 3H, -OCH_3), 3.87 (s, 3H, -OCH_3), 6.87 (d, *J* = 9 Hz, 1H, ArH), 7.19 (dd, *J* = 9 Hz, 1H, ArH), 7.50 (s, 1H, -CH=), 7.56 (d, *J* = 9 Hz, 1H, ArH), 11.14 (s, 1H, NH, D_2O exchangeable); MS ($C_{13}H_{14}N_2O_3$) *m/z* = 247 (M^+).

Synthesis of 3-methyl-4-(aryldiazenyl)-1H-pyrazol-5(4*H*)-one (3a-e)

To a solution of the appropriate amine (1 mml) was dissolved in concentrated HCl (15 ml) and water cooled in ice and then $NaNO_2$ (1 mml) in water (10 ml) was added with stirring. A mixture of compound **1** (0.1 mml), NaOAc (5 g), ethanol (50 ml) and water (20 ml) was prepared separately and cooled in ice. The diazonium salt solution was added slowly to the second solution, with ice cooling. The cooled mixture was stirred for 30 min and filtered to give colored crystals, which were crystallized from ethanol to give the corresponding aryldiazenyl **3a-e**, respectively (25).

3-Methyl-4-(phenyldiazenyl)-*1H*-pyrazol-5(4*H*)-one (3a)

Yield (75%); m.p. 190°C; IR υ 3235 (NH), 1730 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.91 (s, 3H, -CH_3), 5.80 (s, 1H, -CH=N=), 7.14 (t, *J* = 9 Hz, 1H, ArH), 7.36 (d, *J* = 9 Hz, 2H, AB system, ArH), 7.41 (d, *J* = 9 Hz, 2H, AB system, ArH), 12.41 (s, 1H, D_2O exchangeable, NH); MS ($C_{10}H_{10}N_4O$) *m/z* = 203 (M^+).

3-Methyl-4-(p-tolyldiazenyl)-*1H*-pyrazol-5(4*H*)-one (3b)

Yield (75%); m.p. 195°C; IR υ 3240 (NH), 1735 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.93 (s, 3H, -CH_3), 2.44 (s, 3H, -CH_3), 5.81 (s, 1H, -CH=N=), 7.24 (d, *J* = 9 Hz, 2H,AB system, ArH), 7.73 (d, *J* = 9 Hz, 2H, AB system, ArH), 11.51 (s, 1H, NH, D_2O exchangeable,); MS ($C_{11}H_{12}N_4O$) *m/z* = 217 (M^+).

4-((4-Chlorophenyl)diazenyl)-3-methyl-*1H*-pyrazol-5(4*H*)-one (3c)

Yield (75%); m.p. 220°C; IR υ 3280 (NH), 1750 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.15 (s, 3H, -CH_3), 5.86 (s, 1H, -CH=N=), 7.40 (d, *J* = 9 Hz, 2H, ArH), 7.57 (d, *J* = 9 Hz, 2H, ArH), 11.50 (s, 1H, D_2O exchangeable, NH); MS ($C_{10}H_9ClN_4O$) *m/z* = 237 (M^+).

4-((4-Bromophenyl)diazenyl)-3-methyl-1*H*-pyrazol-5(4*H*)-one (3d)

Yield (75%); m.p. 225°C; IR υ 3270 (NH), 1750 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.08 (s, 3H, -CH_3), 5.78 (s, 1H, -CH=N=), 7.51(d, *J* = 9 Hz, 2H, ArH), 7.71 (d, *J* = 9 Hz, 2H, ArH), 11.41 (s, 1H, D_2O exchangeable, NH); MS ($C_{10}H_9BrN_4O$) *m/z* = 281 (M^+).

4-((2-Chlorophenyl)diazenyl)-3-methyl-1*H*-pyrazol-5(4*H*)-one (3e)

Yield (75%); m.p. 210°C; IR υ 3260 (NH), 1740 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.36 (s, 3H, $-CH_3$), 3.83 (s, 3H, $-OCH_3$), 3.87 (s, 3H, $-OCH_3$), 6.87 (d, J = 9 Hz, 1H, ArH), 7.19 (dd, J = 9 Hz, 1H, ArH), 7.50 (s, 1H, -CH=), 7.56 (d, J = 9 Hz, 1H, ArH), 11.14 (s, 1H, D_2O exchangeable, NH); MS ($C_{10}H_9ClN_4O$) m/z = 237 (M^+).

Synthesis of 4-Glycosyl-3-methyl-1*H*-pyrazol-5(4*H*)-one (4a-e)

General procedure

To a mixture of compound **1** (1 mmol) and the appropriate sugar (1 mmol) in dioxane (25 ml), few drops of piperidine were added. The reaction mixture was refluxed for 4-6 hrs, and then cooled to room temperature. The precipitate was filtered, dried and washed with methanol. The products were recrystallized from MeOH/H_2O to afford compounds **4a-e**, respectively.

4-Glucosyl-3-methyl-1*H*-pyrazol-5(4*H*)-one (4a)

Yield (75%); m.p. 290°C; IR υ 3270 (NH), 1745 (C=O) cm^{-1}; ^1H-NMR (DSMO-d_6) δ 2.05 (s, 3H, $-CH_3$), 2.65-4.01 (m, 10H, glucose), 7.90 (s, 1H,anomeric H of glucose); MS ($C_{10}H_{16}N_2O_6$) m/z = 261 (M^+).

4-Galactosyl-3-methyl-1*H*-pyrazol-5(4*H*)-one (4b)

Yield (75%); m.p. 290°C; IR υ 3270 (NH), 1740 (C=O) cm^{-1}; ^1H-NMR (DSMO-d_6) δ 2.05 (s, 3H, $-CH_3$), 2.65-4.20 (m, 10H, galactose), 7.92 (s, 1H,anomeric H of galactose); MS ($C_{10}H_{16}N_2O_6$) m/z = 261 (M^+).

4-Mannosyl-3-methyl-1*H*-pyrazol-5(4*H*)-one (4c)

Yield (75%); m.p. 290°C; IR υ 3260 (NH), 1735 (C=O) cm^{-1}; ^1H- NMR (DSMO-d_6) δ 2.05 (s, 3H, $-CH_3$), 2.65-4.30 (m, 10H, mannose), 7.90 (s, 1H,anomeric H of mannose); MS ($C_{10}H_{16}N_2O_6$) m/z = 261 (M^+).

4-Xylosyl -3-methyl-1*H*-pyrazol-5(4*H*)-one (4d)

Yield (75%); m.p. 290°C; IR υ 3240 (NH), 1750 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.06 (s, 3H, $-CH_3$), 2.65-4.34 (m, 9H, xylose), 7.95 (s, 1H,anomeric H of xylose); MS ($C_9H_{14}N_2O_5$) m/z = 231 (M^+).

4-Ribosyl -3-methyl-1*H*-pyrazol-5(4*H*)-one (4e)

Yield (75%); m.p. 290°C; IR υ 3260 (NH), 1750 (C=O) cm^{-1}; ^1H NMR (DSMO-d_6) δ 2.06 (s, 3H, $-CH_3$), 2.65-4.31 (m, 9H, ribose), 7.98 (s, 1H,anomeric H of ribose); MS ($C_9H_{14}N_2O_5$) m/z = 231 (M^+).

Synthesis of 3-methyl-1*H*-pyrazole-5(4*H*)-thione (5)

To a mixture of compound **1** (0.98 g, 1 mmol) and phosphorus pentasulfide (2.22 ml, 1 mmol) in dry pyridine (25 ml). The reaction mixture was refluxed for 6-8 hrs, and then cooled to room temperature and poured over crushed ice. The precipitate was filtered off, washed with cold water several time and dryed. The product was recrystallized from a mixture of dioxane/EtOH(3:1) to afford compound **5** as yellow powder in 70% yield; mp 283-5°C; IR(υ, cm^{-1})3340(NH), 2980(aliphatic CH_3 and CH_2), and 1640(C=); ^1H-NMR (DSMO-d_6) δ 2.0 (s, 3H, CH_3), 1.45(s, 2H, CH_2), and 9.12 (s, 1H, -NH); MS ($C_4H_6N_2S$) m/z (M^+) .114(100%).

Synthesis of 5-Methyl-4*H*-pyrazole-3-thio-(dimethyl thioether, or methyl ethyl ether) (7a,b) and 3-(2',3',4',6'-Tetra-*O*-acetyl-*β*-D-glucopyanosyl or galacto- pyanosylthio)-5-methyl-4-*H*-pyrazole (8a,b)

General procedure

To a solution of **5** (1.14g, 10mmol) in dry DMF (25mL) , sodium hydride(0.26g, 15mmole) was added portion wise through 20 min. Solution was stirred at room temperature for another 45 min. Then, a solution of chloromethyl methyl thioether(0.96mL, 10mmol) or cholоromethyl ethylether (0.94mL, 10mmol) or

2',3',4',6'-tetra-*O*-acetyl-*α*-D-glucopyanosyl bromide(4.11g, 10mmol) or 2',3',4', 6'-tetra-*O*-acetyl-*α*-D-galactopyanosyl bromide(4.11g, 10mmol) in dry DMF(15mL) was dropped within 30 min. and the reaction mixture was stirred at room temperature until the completion 3-6hr. After completion, the reaction mixture was poured on water and neutralized with few drops of concentrated hydrochloric acid. The aqueous phase was extracted with ethyl acetate (3x20mL) while the combined organic phase was washed with water and dried over anhydrous sodium sulfate. The needed product was obtained after the removal of the residual solvent under vacuum whereby a precipitate was formed. The formed precipitate was collected upon and recrystallized from suitable solvent to give the corresponding S-glycoside derivatives **7a,b** and **8a,b** in a good yields, respectively.

3-Methyl-5-(dimethylthio)-4*H*-pyrazole (7a)

Compound **7a** was recrystalized from absolute ethanol dioxane mixture(3:1) as a yellow powder. Yield (65%); m.p. 188-2°C; IR(υ, cm^{-1}) 2990, 2986(aliphatic CH_3 and CH_2), and 1640(C=N); ^{1}H-NMR (DSMO-*d*$_6$)(δ, ppm) 1.4 (s, 2H, CH_2), 1.8(s,2H,CH_2), 2.06 (s, 3H, CH_3), 3.04 (s, 3H, CH_3); MS ($C_6H_{10}N_2S_2$) m/z: 174 (M^+)100%.

3-((Ethoxymethy) thio)-5-methyl-4*H*-pyrazole (7b)

Compound **7b** was recrystalized from absolute ethanol as a yellowish brown powder. Yield (65%); m.p. 193-5-2°C; IR(υ, cm^{-1}) 2990, 2986(aliphatic CH_3 and CH_2), and 1640(C=N); ^{1}H-NMR (DSMO-*d*$_6$)(δ, ppm) 1.25 (t, 3H, CH_3), 1.46 (s,2H,CH_2), 2.06 (s, 3H, CH_3), 3.30 (s, 2H, CH_2), 3.85 (q, 2H, CH_2); MS ($C_7H_{12}N_2SO$) m/z: 172 (M^+)100%.

3-(2',3',4',6'-Tetra-*O*-acetyl-*β*-D-glucopyanosylthio)-5-methyl-4*H*-pyrazole(8a):

Compound **8a** was recrystalized from absolute ethanol as a pall yellow powder. Yield (75%); m.p. 202-2°C; IR(υ, cm^{-1}) 2993, 2986 and 2880(aliphatic CH_3 and CH_2), 1770,1765 (C=O), and 1638(C=N); ^{1}H-NMR (DSMO-*d*$_6$)(δ, ppm) 1.46(s,2H,CH_2), 1.95-2.0(4s, 12H, 4 ((-CO$\underline{CH_3}$)), 2.08 (s, 3H, CH_3), 3.68 (d, 10Hz, 2H, H-6', H-6''), 4.22 (q, 1H, 5'-H), 4.84 (t,10Hz, 1H, H-4'), 5.32 (t, 10Hz, 1H, H-3'), 5.62 (t, 10Hz, 2H, H-2`), 5.87 (d, 10Hz, 1H, H-1`); MS ($C_{18}H_{24}N_2SO_7$) m/z: 444 (M^+)100%.

3-(2',3',4',6'-Tetra-*O*-acetyl-*β*-D-galactopyanosylthio)-5-methyl-*4H*-pyrazole(8b):

Compound **8a** was recrystalized from absolute ethanol as a pall yellow powder. Yield (80%); m.p. 211-2°C; IR(υ, cm^{-1}) 2990, 2985 and 2881(aliphatic CH_3 and CH_2), 1768,1765 (C=O), and 1644(C=N); ^{1}H-NMR (DSMO-*d*$_6$)(δ, ppm)1.46 (s,2H,CH_2), 1.99-2.06(4s, 12H, 4 ((-CO$\underline{CH_3}$)), 2.08 (s, 3H, CH_3), 4.0 (d, 10Hz, 2H, H-6', H-6''), 4.26 (q, 1H, H-5'), 4.88 (t, 10Hz, 1H, H-4'), 5.55 (t, 10Hz, 1H, H-3'), 5.65 (t, 10Hz, 1H, H-2'), 5.92 (d, 9.95Hz, 1H, H-1'); MS ($C_{18}H_{24}N_2SO_7$) m/z: 444 (M^+)100%.

Biological Evaluation

In-vitro anticancer activity

Cell culture of HepG-2 (human liver carcinoma), PC-3 (human prostate adenocarcinoma) and HCT116 (human colorectal carcinoma) cell lines were purchased from the American Type Culture Collection (Rockville, MD) and maintained in RPMI-1640 medium which was supplemented with 10% heat-inactivated FBS (fetal bovine serum), 100U/ml penicillin and 100U/ml streptomycin. The cells were grown at 37°C in a humidified atmosphere of 5% CO_2.

MTT cytotoxicity assay

The antitumor activity against HepG-2, PC-3 and HCT-116 human cancer cell lines was estimated using the 3-[4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2H-tetrazolium bromide (MTT) assay, which is based on the cleavage of the tetrazolium salt by mitochondrial dehydrogenases in viable cells [24-26]. Cells were dispensed in a 96 well sterile micro plate (5 x 10^4 cells/well), and incubated at 37°C with series of different concentrations, in DMSO, of each tested compound or Doxorubicin® (positive control) for 48 h in a serum free medium prior to the MTT assay. After incubation, media were carefully removed, 40 µL of MTT (2.5 mg/mL) were added to each well and then incubated for an additional 4 h. The purple formazan dye crystals were

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solubilized by the addition of 200 µL of DMSO. The absorbance was measured at 590 nm using a Spectra Max® Paradigm® Multi-Mode micro plate reader. The relative cell viability was expressed as the mean percentage of viable cells compared to the untreated control cells.

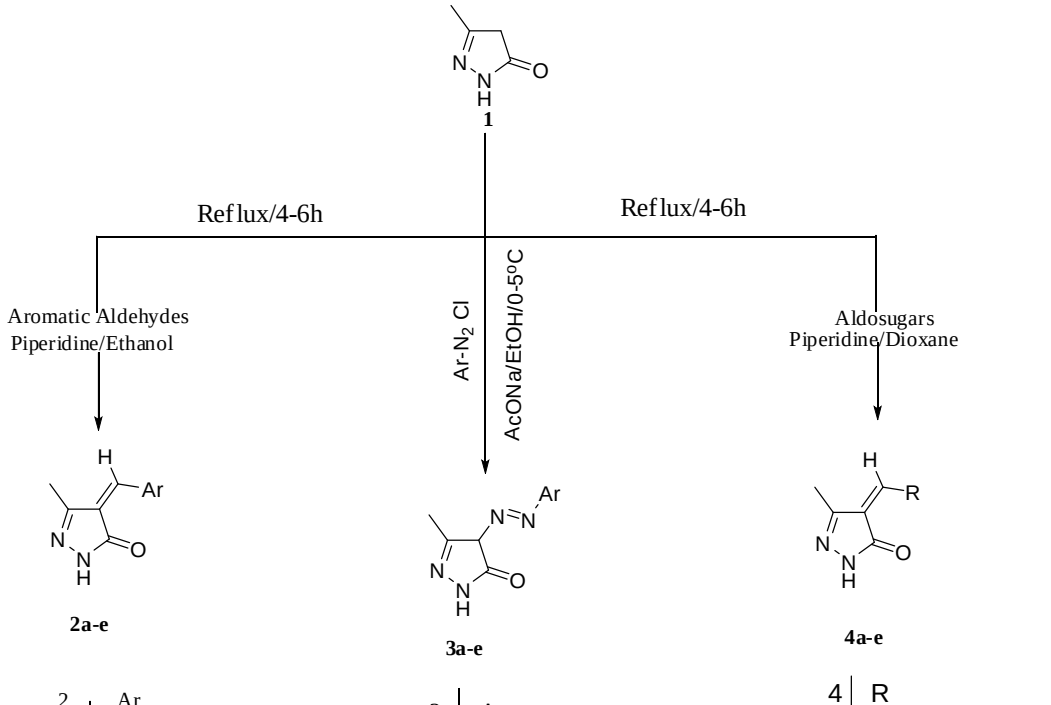
Statistical analysis

All experiments were conducted in triplicate and repeated in three different days. All the values were represented as mean ± SD. IC_{50}s were determined by probit analysis using SPSS software program (SPSS Inc., Chicago, IL).

RESULTS AND DISCUSSION

Chemistry

The synthetic approach was confined to two general schemes to obtain the target compounds. The reaction of ethyl acetoacetate with with hydrazine hydrates in presence of EtOH to form compound **1**. The active methylene group of pyrazolone ring is a favorable units to react with electrophiles usually result in the formation of C=C in case of aldehydes, sugar or C=N bond in case of dizonium chloride of anilines. Therefore, the reactivity of compound **1** towards substituted aldehydes as carbon electrophiles and toward substituted dizonium chloride of anilines has attention. The reaction of compound **1** with a series of aldehydes in refluxed absolute ethanol in the presence of piperidine as basic catalyst afforded the corresponding 4-aryylidene-3-methyl-*1H*-pyrazol-5(*4H*)-one (**2a-e**) (Scheme **1**).



2	Ar
a	$C_6H_4CH=CH-$
b	2-Cl-C_6H_4-
c	4-OH-3-OCH_3-C_6H_4-
d	4-N$(CH_3)_2$-C_6H_4-
e	3,4-OCH_3-C_6H_4-

3	Ar
a	C_6H_5-
b	4-CH_3-C_6H_4-
c	4-Cl-C_6H_4-
d	4-Br-C_6H_4-
e	2-Cl-C_6H_4-

4	R
a	R= -glucose
b	R= -galactose
c	R= -mannose
d	R= -xylose
e	R= -ribose

Scheme (1): synthesis of compounds 2a-4e

Compound **1** react with dizonium chloride salts of anilines in the presence of anhydrous sodium acetate in ice bath yielded 3-methyl-4-(aryldiazenyl)-1*H*-pyrazol-5(*4H*)-ones (**3a-e**), respectively. The treatment of compound **1** with sugars in dioxane and few drops of piperdine yielded aldo-sugar pyrazolone (**4a-e**) (**Scheme 1**). Compound **1** refluxed with phosphorous penta sulfide in dry pyridine in 2:1 molar ratio for eight

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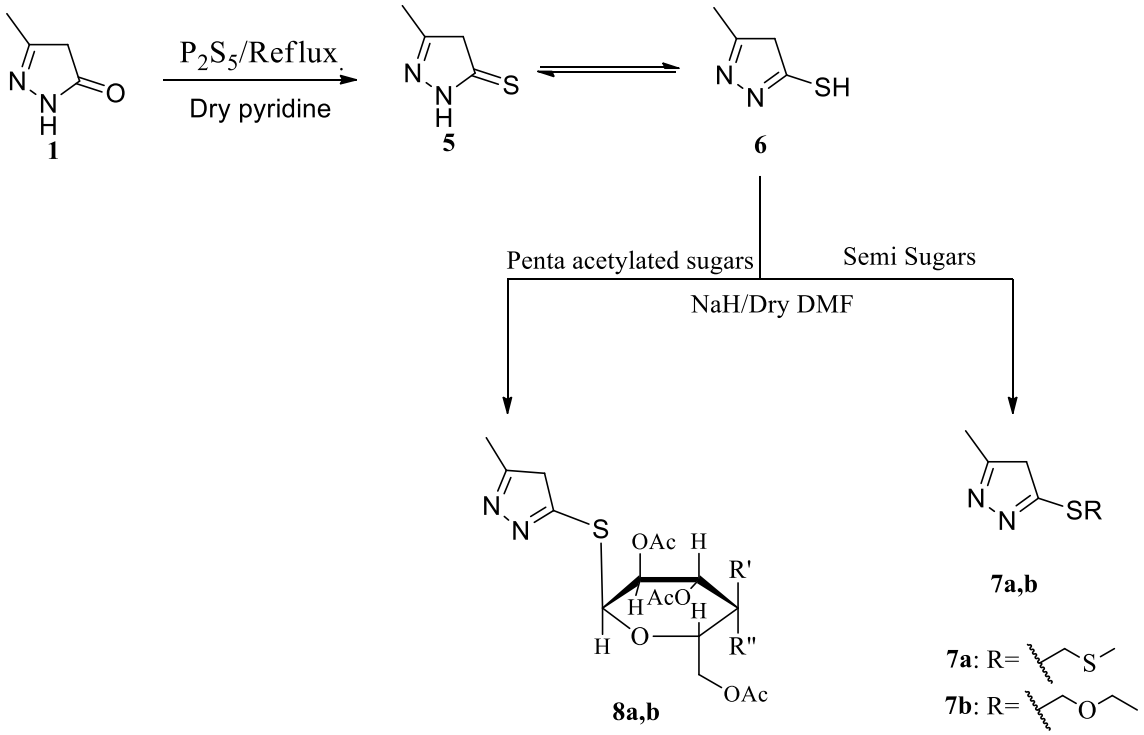
hours, the thione derivative **5** was obtained in good yield. In liquid state, the thione structure **5** was 100% converted to its enol-form (–SH) **5**, so it gives its potassium salt **6** on refluxing with ethanolic potassium hydroxide for four hours. When the potassium salt of derivative **5** coupled with the acyclic activated sugar analogues as chloromethyl methylthioether , or chloromethyl ethyl ether and halosugars namely, cyclic activated sugar as 2`,3`,4`,6`-tetra-*O*-acetyl-α-D-glucopyranosyl bromide or 2`,3`,4`,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide, in the presence of NaH in dry DMF to give the corresponding *S*-glycoside derivatives **7a,b** and **8a,b** in a good yields, respectively. The ^{1}H-NMR spectrum of glycosides **8a,b** were showed the anomeric proton as a doublet at δ = 5.87 ppm with a spin-spin coupling constant ($J_{1`,\ 2`}$ = 9.95Hz) corresponding to a trans orientation of H-1` and H-2` protons indicating the β-configuration. The structures of the synthesized compounds were confirmed on the basis of their IR and ^{1}H-NMR spectroscopic analysis. (See experimental).Scheme (**2**)



Scheme (2)

Biological activity:

Anti-tumor activity

Six compounds were tested *in vitro* for their anti-tumor activities against HepG-2, PC-3 and HCT-116 human carcinoma cell lines using MTT assay. The percentage of the intact cells was measured and compared to the control (**Figure 1**). The activities of these compounds against the three carcinoma cells were compared with that of Doxorubicin®.

The obtained results showed that all compounds showed dose-dependent anticancer activities against the three cancer cells. It is obvious that **2a**, **3a** of the tested synthesized compounds showed significant antitumor activity. The IC_{50} values are shown in **Table 1**.

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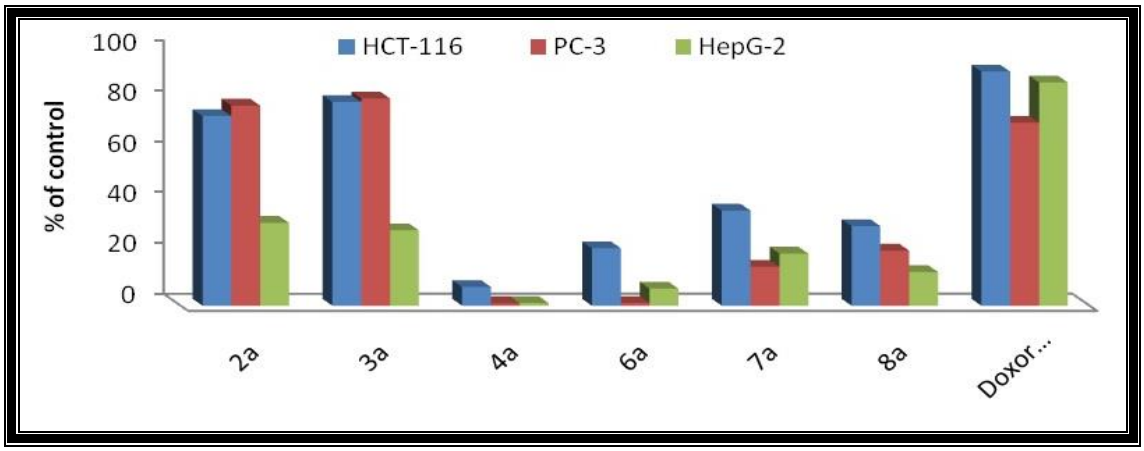

Figure 1: Anticancer activity of the six compounds from Scheme 1 against three cancer types, using MTT assay at 100 ppm.

Table 1: The anticancer IC_{50} values of the six compounds using MTT assay against the three cancer types.

Compound	HCT-116	PC-3	HepG-2
	IC_{50} (µg/mL)		
2a	66.79062	63.28074	152.8435
3a	62.1357	61.07867	168.0342
4a	677.2166	> 1000	> 1000
6a	220.4001	> 1000	742.9739
7a	133.203	325.3678	242.819
8a	159.1739	230.1665	376.3914

CONCLUSION

In summary, we synthesized a series of pyrazolone derivatives and their structures confirmed by IR, Mass and NMR spectroscopic analysis. These compounds were examined for their anticancer activities and IC_{50}. The results showed that the compounds **2a, 3a** showed good anticancer activities.

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