

SYNTHESIS AND EVALUATION OF NOVEL THIAZOLIDINEDIONES FOR ANTI DIABETIC ACTIVITY

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ABSTRACT

The aim of our study was to review on synthesis and various pharmacological activities associated with thiazolidinediones. Thiazolidinediones serve as a boon in the antidiabetic therapy by increasing the sensitivity towards insulin. 1, 3-Thiazolidine-2, 4-dione contains basic skeleton of thiazole or thiazolidine. Presence of one carbonyl group in thiazole at 4th position makes it thiazolidine-4-one which is known for various activities and presence of another carbonyl group at 2nd position makes it thiazolidine-2, 4-dione which is basically known for its antidiabetic activity. The thiazolidinediones used in oral combination therapy in management of patients with type II diabetes are ligand-activated transcription factors belonging to the nuclear receptor super family. The two clinically used PPAR γ agonists pioglitazone and rosiglitazone have made a great contribution to the therapy of type II diabetes. However, weight gain and hepatotoxicity are side effects of thiazolidinedione. For lesser side effects, researchers are focusing on modification of side chain at C-5 position of thiazolidinedione nucleus and its derivatisation. The basic ring of thiazolidinedione was synthesized by 1, 3 dipolar cycloaddition of thiourea and monochloro acetic acid. Knoevenagel condensation with substituted aromatic aldehyde was carried out to yield various substituted benzylidene thiazolidine-2, 4-dione. These compounds were subjected to further esterification and substitution reaction to yield our target compounds as described in scheme 1. The synthesised molecules were screened for blood glucose lowering effect (*in vivo*). Characterisation was done by using various physicochemical and spectral techniques. **IVa** and **IV b** showed blood glucose lowering effect in dexamethasone induced insulin resistance model.

Keywords: Antidiabetic; Peroxisome proliferator-activated receptors; Thiazolidinedione; Diabetes mellitus; Hepatotoxicity.

INTRODUCTION

Diabetes mellitus is a heterogeneous group of disorder, characterized by a state of chronic hyperglycemia, resulting from a diversity of etiologies either environmental and genetics, acting jointly.¹ Characteristically, diabetes is a long term metabolic disorder with a number of complication including cardiovascular, renal, neurological, ocular and other such inter-related problems¹. Diabetes mellitus (DM) is one of the major health problems in the world today. The incidence of the disease currently is estimated to reach 210 million by the year 2010 and 300 million by the year 2025. Most cases will be of Type 2 diabetes mellitus, which strongly associated with a sedentary life style and obesity². During the fast decades substituted thiazolidinediones received attention in the field of diabetes. Many thiazolidinedione and their derivatives serve as basic pharmacophore for various biological profiles i.e. antidiabetic, anticancer³, antimalarial⁴, aldose reductase⁵ and anti-inflammatory⁶ and so on. These observations promoted us to synthesis a new series of thiazolidinedione with higher biological activity. In the present paper, the synthesis and screening of novel thiazolidinedione for antidiabetic activity was performed. The structures of the various synthesized compounds were assigned on the basis of elemental analysis, IR and ¹H NMR spectral data. These compounds were also screened for their antidiabetic activity by using dexamethasone induced insulin resistance in rats.

MATERIALS AND METHODS

The chemicals used in the present project work were purchased from Rankem, Merck and Spectrochem. The melting point of the synthesised compound was determined by open capillary with Thiel's melting point tube (capillary tube method) or Thermanik melting point apparatus and was uncorrected. TLC plates were prepared by using Merck Silica Gel 60 GF 254. Visualization was done in UV light chamber at 254 nm, iodine chamber and visualizing agent (Ninhydrin, 2, 4 DNP reagent). The IR spectra of the synthesized compounds were recorded on a Fourier Transform Infra Red spectrometer (model Shimadzu 8400 S) in the range of 400-4000 cm⁻¹ as KBr pellets. (¹H NMR) data of the compound was carried out in Bruker 200 spectrospin NMR at Astra Zeneca Pharma India Limited, Bangalore and Bruker 400 spectrospin NMR at Indian Institute of Science, Bangalore. The solvent used for NMR was CDCl₃.

Scheme 1: Reagents and conditions

(i) p-hydroxybenzaldehyde, benzoic acid, piperidine, toluene, stirred at 80 °C, 16 h. (ii) 4-chlorobenzaldehyde, benzoic acid, piperidine, toluene, stirred at 80 °C, 16 - 20 h. (iii) ethyl chloro formate, anhydrous K₂CO₃, dry acetone stirred over night. (iv) ethanol, refluxed 4-8h.

General method of synthesis of thiazolidine-2, 4-dione (I)

By 1, 3 dipolar cyclo addition as reported procedure.⁷

Preparation of 5-(4-derivative of benzylidene) thiazolidine-2, 4-dione (IIa-b)

Knoevenagel condensation with substituted aromatic aldehydes using weakly basic amine as a catalyst (piperidine) and toluene as a solvent.⁷

Preparation of [4-(2,4-Dioxo-thiazolidin-5-ylidene)methyl]-phenyl-ester ethyl ester (III)

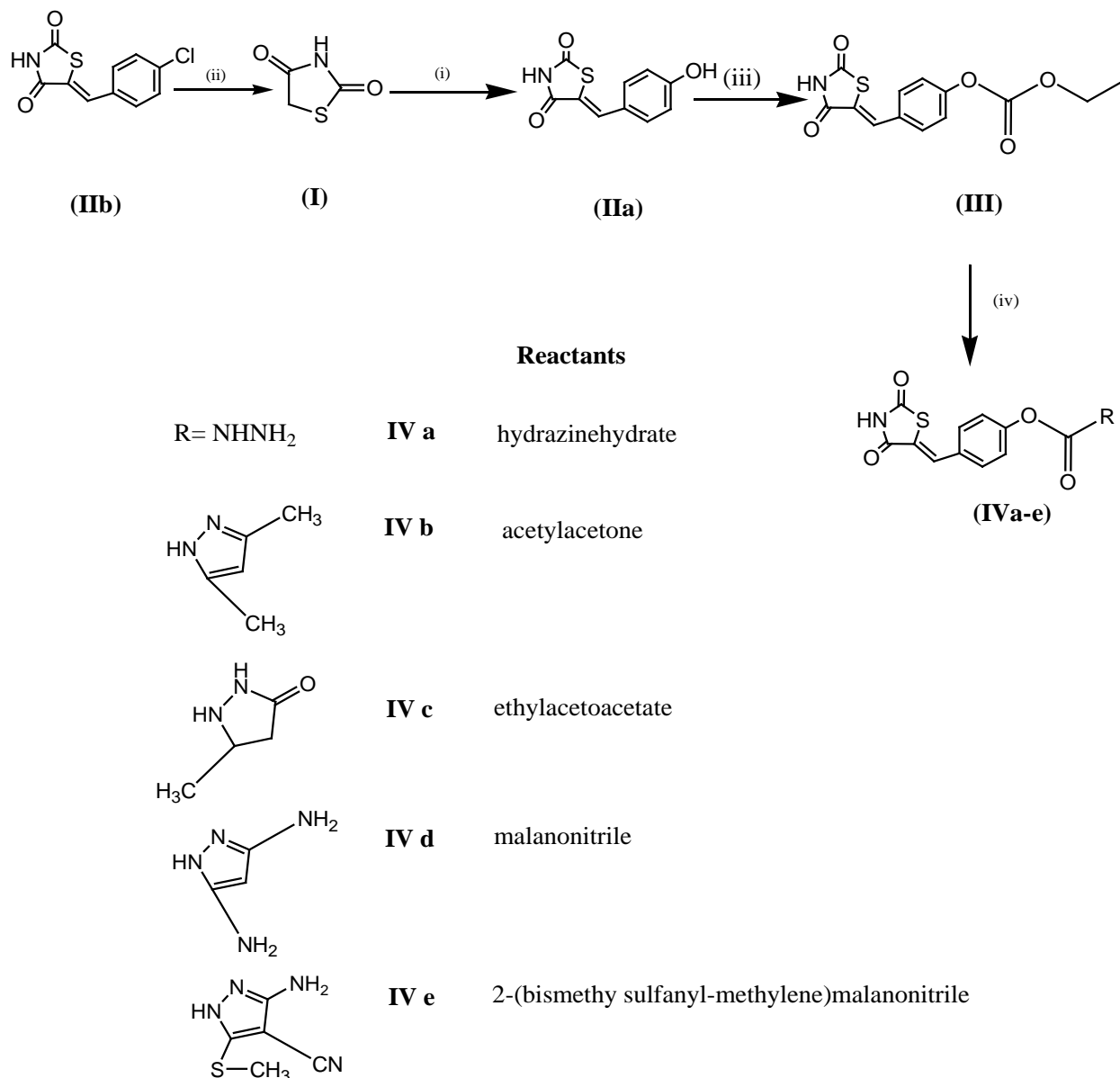
Was prepared by overnight stirring of III with ethyl chloro formate, anhydrous K₂CO₃ and dry acetone as per reported procedure.⁸

Preparation of Hydrazinecarboxylic acid [4-(2, 4-Dioxo-thiazolidin-5-ylidene)methyl]-phenyl-ester (IVa-e)

To a suspension of compound (III) (0.01mol) in 40 ml ethanol 0.015 mol of hydrazine hydrate was added and the reaction mixture was refluxed for 2h. The resulting mixture was allowed to cool and filtered. The solid obtained dried and recrystallised with hot water (IVa).⁸

General procedure for the preparation of compounds (IVb-e)

A series of novel pyrazole derivatives were synthesized by the cycloaddition of diketones with the respective hydrazine derivatives. Compound (IV a) (0.2 mol) and acetyl acetone (0.2 mol), ethylacetoacetate, malanonitrile, 2-(bismethyl sulfanyl-methylene) malanonitrile, in ethanol (10ml) was refluxed with continuous stirring for 6-10 h to obtain compound (IVb-e). The reaction was monitored by TLC after the completion of reaction mixture was cooled at room temperature and stirred for 10-15 mins, the resulting solid mass was filtered, washed with small amount of ethanol and dried. Recrystallisation by ethanol.⁹⁻¹¹



Scheme 1

Antidiabetic Activity

Dexamethasone induced insulin resistance in rats

Exogenous administration of glucocorticoids (Ex: dexamethasone) in rats causes hyperglycemia, hyperinsulinaemia, associated with insulin resistance.¹² Institution of Animals Ethics Committee has approved the experimental protocol (IAEC/NCP/44/10).

Group I: Normal control - Received 0.25% CMC p.o and sterile water for injection i.m.

Group II: Dexamethasone control - Received 0.25% CMC p.o and Dexamethasone 0.7 mg/Kg i.m.

Group III: Rosiglitazone treated - Received Rosiglitazone 0.72 mg/Kg in 0.25% CMC p.o and Dexamethasone 0.7 mg/Kg i.m.

Group IV: IVa treated - Received IVa, 0.72 mg/Kg in 0.25% CMC p.o and dexamethasone 0.7 mg/Kg i.m.

Group V: IVb treated - Received IVb, 0.72 mg/Kg in 0.25% CMC p.o and dexamethasone 0.7 mg/Kg i.m.

Treatment was continued for 10 days. On day 10, after overnight fasting, blood samples were collected from all the animals by puncturing the retro orbital plexus under mild ether anaesthesia.¹³

RESULTS AND DISCUSSION

The synthesis of parent ring was done by 1, 3 cycloaddition of thiourea and monochloroacetic acid using H₂O and Conc. HCl as a solvent. It was proved by comparing observed m.p with literature m.p. ¹H NMR spectra which clearly show two singlets at 4.31 and 11.98 indicating the presence of -CH₂ and -NH. Further Knoevenagel condensation reaction with substituted aromatic aldehydes leads to structure II a and II b. The formation of compounds were confirmed by visualizing agents (2, 4 DNP, Phosphomolybdic acid) and IR. The phenolic benzylidene intermediate was subjected to reaction with ethylchloroformate to form ester i.e., III. This was then made to react with hydrazine hydrate using ethanol as solvent to get respective hydrazino derivatives IV. These compounds were treated with different diketones (acetylacetone, ethylacetoacetate), malanonitrile, 2-(bismethy sulfanyl-methylene)malanonitrile), ethanol used as solvent to obtain compounds IVa-e as described in scheme 1. Physical and spectroscopical data described in Table 2-3 and Figure 1-2.

Table 1: List of synthesised compounds

1	IV	Hydrazinecarboxylic acid[4-(2,4-Dioxo-thiazolidin-5-ylidenemethyl)-phenyl-ester	
2	IVa	3,5-Dimethyl-pyrazole-1-carboxylic acid 4-(2,4-dioxo-thiazolidin-5-ylidenemethyl)-phenyl ester	
3	IVb	5-Methyl-3-oxo-pyrazolidine-1-carboxylic acid 4-(2,4-dioxo-thiazolidin-5-ylidenemethyl)-phenyl ester	
4	IVc	Pyrazole-1-carboxylic acid 4-(2,4-dioxo-thiazolidin-5-ylidenemethyl)-phenyl ester	
5	IVd	3-Amino-4-cyano-5-methylsulfanyl -Pyrazole-1-carboxylic acid 4-(2,4-dioxo-thiazolidin-5-ylidenemethyl)-phenyl ester	

Table 2: Physical properties of synthesised compounds

Sl. no	C.C.*	Molecular formula	M.Wt.	% yield	State	R _f	Mobile Phase
1.	IV	C ₁₁ H ₉ N ₃ O ₄ S	279.27	78.5%	semisolid	0.60	n-Hex :EA 2 : 1
2.	IVa	C ₁₆ H ₁₃ N ₃ O ₄ S	343.36	65.7%	semisolid	0.62	n-Hex : EA 2 : 1
3.	IVb	C ₁₅ H ₁₃ N ₃ O ₅ S	347.35	76.5%	semisolid	0.60	n-Hex : EA 2 : 1
4.	IVc	C ₁₄ H ₉ N ₃ O ₄ S	315.30	66.7 %	semisolid	0.57	n-Hex : EA 2 : 1
5.	IVd	C ₁₅ H ₁₂ N ₄ O ₄ S	355.33	63.2%	semisolid	0.69	n-Hex : EA 2: 1

Table 3: Elemental analysis and spectral data of synthesised compounds

Fig 1	Elemental analysis	I.R. values (cm ⁻¹)
a	C=30.76; H= 2.58; N= 11.96; O= 27.32; S= 27.38	3470.06, (-N-H), 2915.19, 2974.61 (C-H str.) 1776.50,1737.99 (keto C=O str.), 1522(C=C str.),
b	C=54.29; H=3.19; N= 6.33; O=21.70; S=14.49	3429.29, (-OH str.), 3236.66 (-NH str.), 1718,1703(C=O), 1680, 1664.31 (C=C str.), 1275(C-C)
c	C= 50.11; H= 2.52; Cl= 14.79; N= 5.84; O= 13.35; S= 13.38	3236.66 (-NH str.), 1718,1703(C=O), 1680, 1664.31 (C=C str.), 1275(C-C), C-Cl 752

Sl.no	Comp.code	NMR In (DMSO-d ₆) (δ) values in ppm from TMS
Fig 2	I	4.130 (s, 2H,-S-CH ₂ -C=O) 11.98(1s, 1H, -NH-).

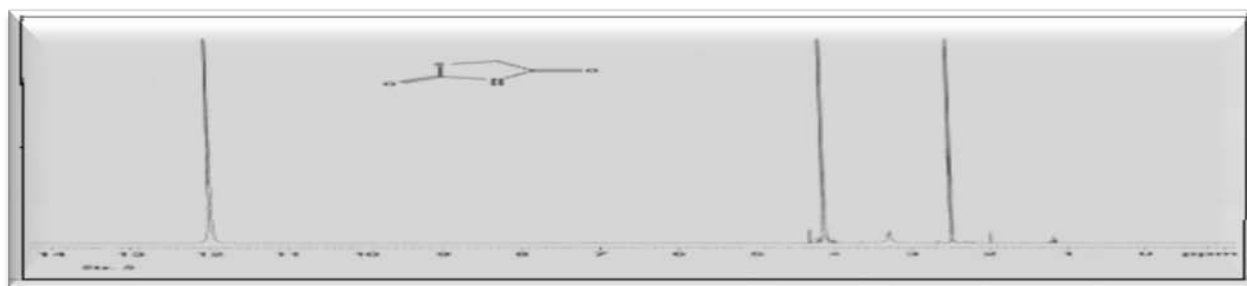


Fig. 2: ¹H NMR Spectra of Thiazolidine-2, 4-dione (I)

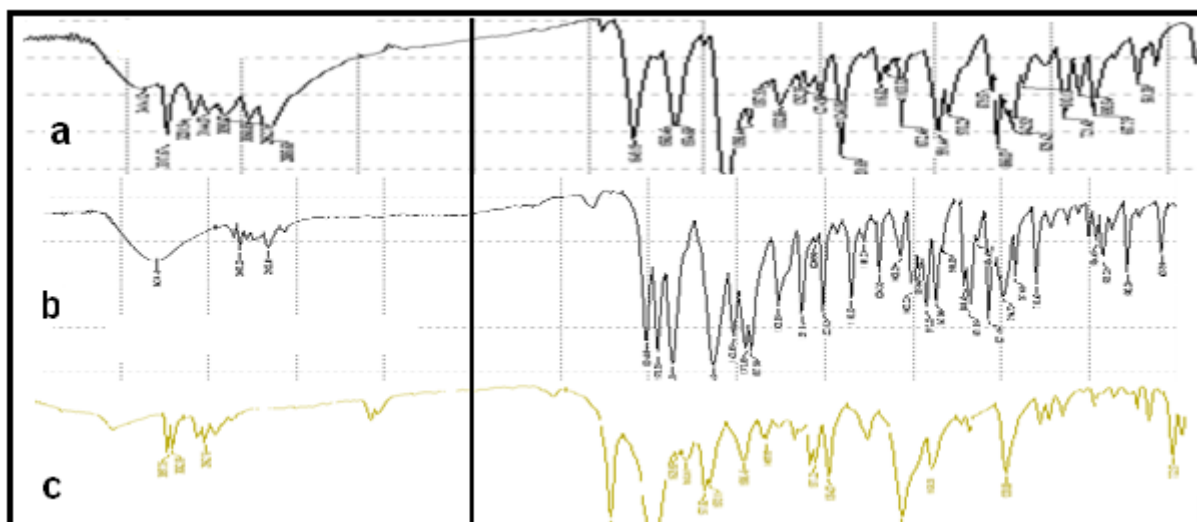


Fig. 1:

- a. FTIR spectra of Hydrazinecarboxylic acid[4-(2,4-Dioxo-thiazolidin-5-ylidenemethyl)-phenyl-ester (**IV**)
 b. FTIR spectra of 3,5-Dimethyl-pyrazole-1-carboxylic acid 4-(2,4-dioxo-thiazolidin-5-ylidenemethyl)-phenyl ester (**IVa**)
 c. FTIR spectra of 5-Methyl-3-oxo-pyrazolidine-1-carboxylic acid 4-(2,4-dioxo-thiazolidin-5-ylidenemethyl)-phenyl ester (**IVb**)

Table 4: Effect of IVa and IVb on blood glucose levels (mg/dl) in dexamethasone induced insulin resistance model in rats.

GROUPS →	GROUP 1 NORMAL CONTROL	GROUP 2 DEXA CONTROL	GROUP 3 DEXA + Rosiglitazone	GROUP4 DEXA + IVa	GROUP5 DEXA + IVb
Serum glucose (mg/dl)	85.08	262.1	112.5	99.57	101.7
MEAN	±	±	±	±	±
±	3.064	6.398 ^{a***}	3.023 ^{b***}	0.995 ^{b***}	2.430 ^{b***}
SEM →					

Values are expressed as mean ± S.E.M., n = 6, DEXA = dexamethasone 0.7 mg/kg, i.m.

Rosiglitazone = Rosiglitazone 0.36 mg/kg, p.o., twice a day,

IVa = IVa 0.36 mg/kg, p.o., twice a day,

IVb = IVb 0.36 mg/kg, p.o., twice a day,

^a when compared with normal control; ^b when compared with dexamethasone control.

***p<0.001 highly significant;

CONCLUSION

The purpose of the present work was to synthesize, characterize and evaluate the biological activity of substituted thiazolidinedione derivatives. The compounds were studied for antidiabetic activity. The activity was found to be favourable when compared to standard.

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REFERENCES

- Shashikant PR, Prajact K, Dighe NS, Bhawar SB, Ana N, Ashwini P *et al*. Synthesis and evaluation of some new thiazolidinedione derivatives for their antidiabetic activities. A J Res Chem. 2009; 2(2):123-26.
- Jithendra CH, Muralidharan P. Anti- hyperglycemic and anti oxidant activities of the ayurvedic drug Nisha Kathakathadhi churam in alloxan-induced hyperglycemic rats. Int J Green Pharm. 2009; 66-69.
- Kim S, Lee JJ, Heo DS. PPARγ ligands induce growth inhibition and apoptosis through p63 and p73 in human ovarian cancer cells. Biochem Biophys Res Commun 2011.
- Sunduru N, Srivastava K, Rajakumar S, Puri SK, Saxena JK, Synth MS *et al*. Synthesis of novel thiourea, thiazolidinedione and thioparabanic acid derivatives of 4-aminoquinoline as potent antimalarials. Bioorg Med Chem Lett. 2009; 19:2570-2573.
- Soni LM, Gupta AK, Kaskhedikar SG. QSAR study of 5-arylidene-2, 4-thiazolidinedione as aldose reductase inhibitor. 2008; 17:258-266.
- Amla A, Youssef M, White S, Erika B, Ibrahim M. Synthesis and biological evaluation of novel pyrazolyl-2,4- TZD as anti-inflammatory and neuro protective agents. Bioorg Med chem. 2010; 18:2019-2028.
- Murugan R, Anbazhagan S, Narayanan SS. Synthesis and in vivo antidiabetic activity of novel dispiropyrrrolidines through [3 + 2] cycloaddition reactions with thiazolidinedione and rhodanine derivatives. Eur J Med Chem. 2009; 44:3272-3279.
- Nargund LVG, Reddy GRN and Prasad VH. Anti-inflammatory activity of substituted 1, 3, 4 -oxadiazoles. J Ph Sci. 1994; 83(2):246-47.
- Gopal KAR, Son AJ, Kalirajan K. Anti-epileptic and anti-microbial activity of novel 1-(unsubstituted/substituted)-3,5 dimethyl-1H pyrazole derivative. Int J Chem Tech Res. 2010; 2(1):45-49.
- Patel CK, Rami CS, Panigraha, Patel CN. Synthesis and biological evaluation of (4-substituted benzylidene)-3-methyl-1-

- (substituted phenyl sulfonyl and substituted benzoyl)-1Hpyrazol-5(4H)-one as anti-inflammatory agent. *J Chem Pharm. Res.* 2010; 2(1):75-78.
11. Shah NK, Patel MP, Patel RC. Synthesis of pyrazole by condensation of hydrazino with malanonitrile. *Ind J chem.* 2009; 48:1170-1173.
 12. Andrew RC, Walker BR. Glucocorticoids and insulin resistance: old hormones, new targets. *Clin Sci (Lond).* 1999; 96(5):513-523.
 13. Jeong IK, Oh SH, Kim BJ, Chung JH, Min YK, Lee MS *et al.* The effects of dexamethasone on insulin release and biosynthesis are dependent on the dose and the duration of treatment. *Diabetes Res Clin Pract.* 2001; 51(3):163-171.