Asian Journal of Chemistry; Vol. 23, No. 6 (2011), 2801-2805

ASIAN JOURNAL OF CHEMISTRY

www.asianjournalofchemistry.co.in

Antioxidant Potential of New Methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide Derivatives

MUHAMMAD NADEEM ARSHAD[1], ISLAM ULLAH KHAN[1,*], MUHAMMAD ZIA-UR-REHMAN[2] and MUHAMMAD SHAFIQ[1]

[1]Material Chemistry Laboratory, Department of Chemistry, Faculty of Science, Government College University, Lahore-54000, Pakistan
[2]Applied Chemistry Research Centre, PCSIR laboratories Complex, Lahore-54600, Pakistan

*Corresponding author: Tel: +92 3334046740; E-mail: iukhan@gcu.edu.pk

(*Received*: 13 November 2010; *Accepted*: 1 March 2011) AJC-9668

An easy synthesis of a series of N-alkyl-1,2-benzothiazine-1,1-dioxide derivatives from commercially available saccharine is reported in order to explore their antioxidant potential. The newly synthesized compounds were characterized by spectroscopic techniques (FT-IR, NMR, MS) and single crystal X-ray diffraction analyses. All the synthesized compounds were screened for their DPPH and FRAP analyses to determine their antioxidant activity.

Key Words: 1,2-Benzothiazine-1,1-dioxide, N-Alkylation, Antioxidant activity, Crystal structure.

INTRODUCTION

Several biochemical reactions generate reactive oxygen species (ROS) in human body and these are capable of damaging crucial bio-molecules. These undesired species may lead to a number of diseases like cerebrovascular disease[1], cancer[2], arteriosclerosis[3,4], atherosclerosis, heart disease, senility, aging, behcet's disease, crohn's disease, cataracts,sunburn, ulcers, osteoporosis, rheumatoid arthritis, diabetes mellitus, emphysema, stroke[5,6]. Considerable attention is being given these days to the exploration of possible therapeutic antioxidant molecules in controlling degenerative diseases associated with marked oxidative damage[7-9].

In continuation of our work on the synthesis of antioxidant benzothiazines[10,11], we here report the synthesis of methyl-4-hydroxy-2*H*-2 alkyl-1,2-benzothiazine-3-carboxylate-1,1-dioxides and their derivatives. Cyclic sulfonamides (sultams) are considered important and useful heterocycles due to their enormous potential as pharmaceutical and agricultural agents[12], chiral auxiliaries[13] and therapeutic compounds[14]. Among these, 1,2-benzothiazine-1,1-dioxide derivatives got enough significance[15-17] due to their applications as antiinflammatory[18], anticancer[19], analgesic[20], antilukemic[21], antimicrobial[22] and antipyretic[23] agents. Besides, some of their derivatives have also been reported as metal chelators[24]. According to the literature, most of the work on methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide nucleus has been carried out at its 3-position by reacting the ester group with amines to get corresponding amides *e.g.*, antiinflammatory drugs like piroxicam[25] and meloxicam[26] (Fig. 1) or its hydrazinolysis followed by intramolecular cyclization to pyrazoles[11]. It is interesting to note that not much attention has been given on the synthesis of a series of N-alkyl derivatives of methyl 4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide.

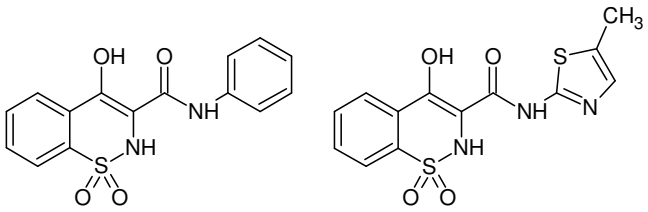
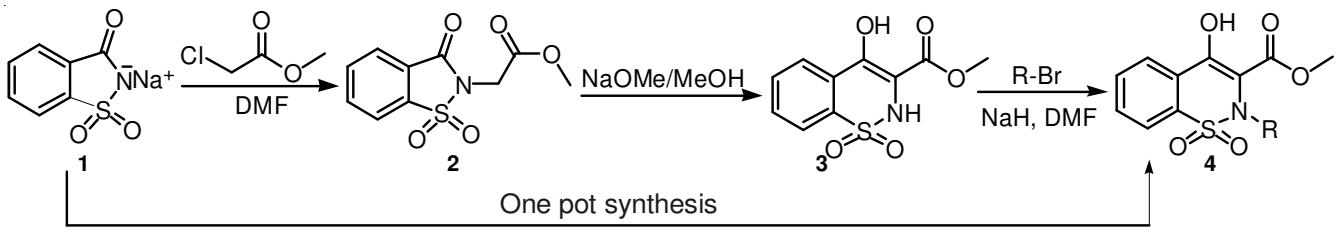


Fig. 1. Structures of some well known oxicam drugs

In the present study, antioxidant activities (DPPH and FRAP) of a number of N-alkyl derivatives of methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide have been discussed. For the reason, a series of such compounds has been synthesized; first through multi-step procedure and secondly by one pot synthetic methodology with improved overall yields and lesser reaction times (Table-1, **Scheme-I**).

EXPERIMENTAL

All the chemicals were purchased from E. Merck, BDH or Fluka and used without purification. However, solvents were purified through distillation. ^{1}H and ^{13}C NMR spectra were recorded on a varian AM400 instrument. Chemical shifts are



Scheme-I: Lay out for the synthesis of N-substituted derivatives of methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide

TABLE-1
COMPARATIVE YIELDS OF SYNTHESIZED DERIVATIVES IN TWO METHODS

Entry	Comp. No.	R	Yield (%)	
			Multi-step synthesis (overall)	One pot synthesis
1	**4a**	*iso*-Propyl	64	83
2	**4b**	*n*-Butyl	67	89
3	**4c**	1-Butenyl	65	86
4	**4d**	*n*-Pentyl	59	84
5	**4e**	1-Pentenyl	66	89
6	**4f**	Benzyl	62	87
7	**4g**	2-Phenylethyl	65	85
8	**4h**	3-Phenylpropyl	58	88
9	**4i**	2-Methoxy-2-oxoethyl	67	86
10	**4j**	2-Ethoxy-2-oxoethyl	62	92

reported in ppm referenced to the residual solvent signal. FT-IR spectra were recorded on a Perkin-Elmer 1600-FT spectrometer. Mass spectra were recorded on Agilent 5973N instrument using EI mode. Melting points were recorded on a electrothermal (Griffin 1090) melting point apparatus and are uncorrected. X-Ray diffraction data were collected at 100 (2) K on a Siemens SMART three-circle X-ray diffractometer equipped with an APEX II CCD detector (Bruker-AXS) and an Oxford cryosystems 700 cryostream, using MoK_α radition (0.71073 Å) source and a graphite monochromator and the data were corrected for Lorentz and polarization effects and for absorption using multi-scan method[27,28].

Methyl-2-(1,1,3-trioxo-2,3-dihydro-1,2-benzothiazol-2-yl)acetate (2): A mixture of methyl chloroacetate (3.95 g, 36.56 mmol) and sodium saccharin (**1**) (7.49 g, 36.56 mmol) in dimethyl formamide (40 mL) was heated at 60 ºC for 0.5 h. The contents of the reaction mixture was cooled to room temperature and poured into ice cold water (250 mL), white solid produced was filtered and washed with water. The solid was dried to produce (**2**) as a white powder; yield: 89 %; m.p. 116-117 ºC. IR (KBr, ν_{max}, cm^{-1}): 1752, 1677, 1342, 1182, ^1H NMR: (400 MHz) ($CDCl_3$) δ: 7.76 -7.82 (4H, aromatic), 4.43 (2H, s, CH_2), 3.79 (3H, s, OCH_3); MS (E/I) m/z: 255 [M^+].

Methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (3): Methyl-2-(1,1,3-trioxo-2,3-dihydro-1,2-benzothiazol-2-yl)acetate ester (**2**) (2.30 g, 100 mmol) was added to the solution of sodium methoxide, prepared by refluxing the sodium metal (0.92 g, 40.0 mmol) in methanol (15 mL) under N_2 and heated at 55 ºC until the appearance of precipitates. The contents were poured on ice cold water mixture and pH adjusted to about 3 using HCl (15 %). The orange slurry produced was poured into conc. HCl (20 mL). The precipitates obtained were filtered off, washed with water, dried and recrystallized from methanol by slow evaporation to get ester (**3**). Yield: 78 %; m.p. 172-173 ºC. (Lit m.p. 172 ºC)[29]. IR (KBr, α_{max}, cm^{-1}): 3192, 1665, 1349, 1163; ^1H NMR: (400 MHz) ($CDCl_3$) δ: 12.31 (1H, s, OH_{enolic}), 8.14 (1H, br s, NH) 7.75 -7.82 (4H, aromatic), 4.11 (3H, s, OCH_3); MS (E/I) m/z: 255 [M^+].

General procedure for the synthesis of N-alkyl derivative of methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide: A solution of methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (**3**) (100 mmol) in DMF (5 mL) was added to *n*-hexane washed suspension of sodium hydride (200 mmol of 64 %). The reaction mixture was stirred for 0.5 h followed by the addition of alkyl bromide (120 mmol) and the resulting mixture was allowed to stir at room temperature for 2-3 h for **a-j**. Contents were poured over crushed ice and pH was adjusted to 2 using 1 N HCl. Precipitates obtained were filtered, washed with water and dried to get the corresponding product.

One pot synthesis of N-alkyl derivatives of methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide: To a three neck round bottom flask, fitted with a mechanical stirrer, nitrogen inlet and a vent, a mixture of methyl chloroacetate (6.32 g, 5.84 mmol), sodium saccharin (**I**) (8.00 g, 3.74 mmol) and dimethyl sulfoxide (20 mL) was heated on a water bath. After 20 min, reaction mixture was cooled to 25 ºC and sodium methoxide (2.4 g; 4.32 mmol) was added to it with constant supply of nitrogen (10 mL min^{-1}). The contents were kept stirred for 10 min and the temperature was maintained at 0 ºC followed by addition of sodium hydride (3.76 g, 7.84 mmol). After 10 min, temperature of the reaction mixture was again maintained at 20 ºC followed by drop wise addition of alkyl bromides (6.72 mmol). Contents were poured over a mixture of crushed ice (100 g) and concentrated hydrochloric acid (10 mL) after stirring for a period of 15 min. The precipitates obtained were filtered and dried.

Methyl-4-hydroxy-2-(propan-2-yl)-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4a): White powder; yield: 83 %; m.p. 100-101 ºC. IR (KBr, ν_{max}, cm^{-1}): 3486, 1659, 1375, 1150; ^1H NMR: (400 MHz) ($CDCl_3$) δ: 12.12 (1H, s, OH_{enolic}), 8.06 (1H, d, aromatic), 7.87 (1H, d, aromatic), 7.68 (2H, t, aromatic), 3.93 (3H, s, CH_3), 3.03 (1H, m, CH), 1.09 (6H, d, CH_2); MS (E/I) m/z: 297.09 [M^+].

Methyl-2-butyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4-b): White powder; yield: 89 %; m.p. 78 ºC. IR (KBr, ν_{max}, cm^{-1}): 3442, 1654, 1344, 1176; ^1H NMR: (400 MHz) ($CDCl_3$) δ: 12.10 (1H, s, OH_{enolic}), 8.04 (1H, d, aromatic), 7.85 (1H, d, aromatic), 7.70 (2H, m, aromatic), 3.95 (3H, s, CH_3), 3.48 (2H, q, $=CH_2$), 1.19 (2H, m, $=CH_2$), 1.08 (2H, m, CH_2), 0.74 (3H, t, CH_3); MS (E/I) m/z: 311.07 [M^+].

Methyl-2-(but-3-en-1-yl)-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4-c): White powder; yield: 86 %; m.p. 66-68 ºC. IR (KBr, ν_{max}, cm^{-1}): 3472, 1658, 1346, 1176; ^{1}H NMR: (400 MHz) ($CDCl_3$) δ: 12.12 (1H, s, OH_{enolic}), 8.03 (1H, d, aromatic), 7.86 (1H, d, aromatic), 7.72 (2H, m, aromatic), 5.49 (1H, m, =CH), 4.79 (2H, m, $=CH_2$), 3.95 (3H, s, OCH_3), 1.98 (2H, m, CH_2); MS (E/I) m/z: 309.08 [M^+].

Methyl-4-hydroxy-2-pentyl-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4d): White powder; yield: 84 %; m.p. 73-74 ºC. IR (KBr, ν_{max}, cm^{-1}): 3465, 1656, 1354, 1178, ^{1}H NMR: (400 MHz) ($CDCl_3$) δ: 12.18 (1H, s, OH_{enolic}), 8.05 (1H, t, aromatic), 7.83 (1H, t, aromatic), 7.69 (2H, m, aromatic), 3.94 (3H, s, OCH_3), 3.27 (2H, t, CH_2), 1.22 (2H, m, CH_2), 1.02 (2H, m, CH_2), 0.70 (3H, t, CH_3); MS (E/I) m/z: 325.11 [M^+].

Methyl-4-hydroxy-2-pentenyl-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4e): White powder; yield: 89 %; m.p. 70-71 ºC. IR (KBr, ν_{max}, cm^{-1}): 3452, 1665, 1349, 1176; ^{1}H NMR: (400 MHz) ($CDCl_3$) δ: 12.18 (1H, s, OH_{enolic}), 8.08 (1H, t, aromatic), 7.85 (3H, m, aromatic), 5.58 (1H, m, =CH), 4.87 (1H, d, $=CH_2$), 4.83 (1H, d, $=CH_2$), 3.92 (3H, s, OCH_3), 3.46 (2H, t, CH_2), 1.81 (2H, m, CH_2), 1.33 (2H, m, CH_2); MS (E/I) m/z: 323.06 [M^+].

Methyl-2-benzyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4f): Colourless crystals. Yield 87 %; m.p. 148-150 ºC. IR (KBr, ν_{max}, cm^{-1}): 3439, 1648, 1362, 1177; ^{1}H NMR: (400 MHz) ($CDCl_3$) δ: 12.04 (1H, s, OH_{enolic}), 7.74 (1H, d, aromatic), 7.65 (1H, d, aromatic), 7.56 (2H, t, aromatic), 6.98 (3H, m, aromatic), 6.88 (2H, d, aromatic), 4.65 (2H, s, CH_2), 3.94 (3H, s, OCH_3); MS (E/I) m/z: 345.08 [M^+].

Methyl-4-hydroxy-2-(2-phenylethyl)-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4g): Colourless crystalline; yield: 85 %, m.p. 107-108 ºC. IR (KBr, ν_{max}, cm^{-1}): 3452, 1653, 1368, 1179; ^{1}H NMR: (400 MHz) ($CDCl_3$) δ: 12.08 (1H, s, OH_{enolic}), 8.04 (1H, d, aromatic), 7.87 (3H, m, aromatic), 7.10 (3H, m, aromatic), 6.96 (2H, d, aromatic), 3.97 (3H, s, OCH_3), 3.80 (2H, t, CH_2), 2.58 (2H, t, CH_2); MS (E/I) m/z: 359.07 [M^+].

Methyl-4-hydroxy-2-(3-phenylpropyl)-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4h): Colourless crystalline; yield: 88 %; m.p. 76-77 ºC. IR (KBr, ν_{max}, cm^{-1}): 3441, 1651, 1358, 1181; ^{1}H NMR: (400 MHz) ($CDCl_3$) δ: 12.78 (1H, s, OH_{enolic}), 7.98 (1H, d, aromatic), 7.87 (3H, m, aromatic), 7.01 (3H, m, aromatic), 6.87 (2H, d, aromatic), 3.93 (3H, s, OCH_3), 3.46 (2H, t, CH_2), 2.61 (2H, t, CH_2), 1.51 (2H, m, CH_2); MS (E/I) m/z: 373.10 [M^+].

Methyl-4-hydroxy-2-(2-methoxy-2-oxoethyl)-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4i): Colourless crystalline; yield: 86 %; m.p. 108-110 ºC. IR (KBr, ν_{max}, cm^{-1}): 3485, 1661, 1364, 1176; ^{1}H NMR: (400 MHz) ($CDCl_3$) δ: 12.15 (1H, s, OH_{enolic}), 8.08 (1H, d, aromatic), 7.82 (3H, m, aromatic), 4.45 (2H, s, CH_2), 3.96 (3H, s, OCH_3), 3.35 (3H, s, OCH_3); MS (E/I) m/z: 327.04 [M^+].

Methyl-2-(2-ethoxy-2-oxoethyl)-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4j): Colourless crystalline; yield: 92 %; m.p. 95-96 ºC. IR (KBr, ν_{max}, cm^{-1}): 3479, 1665, 1372, 1187; ^{1}H NMR: (400 MHz) ($CDCl_3$) δ: 12.09 (1H, s, OH_{enolic}), 8.11 (1H, d, aromatic), 7.84 (3H, m, aromatic), 4.43 (2H, s, CH_2), 4.16 (2H, q, CH_2), 3.92 (3H, s, OCH_3), 1.18 (3H, t, CH_3); MS (E/I) m/z: 341.04 [M^+].

Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay: DPPH solution (3 mL, 25 mg L^{-1}) in methanol was mixed with appropriate volumes of neat or diluted sample solutions. The reaction progress of the mixture was monitored at 515 nm over a time period till TEC_{50} was obtained. Upon reduction, the colour of the solution faded. The percentage of the DPPH remaining was calculated as[29]:

$$DPPH_{rem}\ (\%) = \frac{[DPPH]_{rem}}{[DPPH]_{t=0}} \times 100$$

A kinetic curve showing the scavenging of DPPH radical in terms of decrease in absorbance at 593 nm as a function of time (min) was plotted for each fraction of the samples. EC_{50}, the concentration that causes a decrease in the initial DPPH concentration by 50 % and TEC_{50}, the time needed to reach the steady state with EC_{50} concentration were calculated from the kinetic curve.

Ferric reducing antioxidant power (FRAP) analysis: The reducing capacity of synthetic compound was measured according to the method of Benzie and Strain[30]. Freshly prepared FRAP solution contained 25 mL of 300 mM acetate buffer (pH 3.6), 2.5 mL of 10 mM TPTZ solution in 40 mM HCl solution and 2.5 mL of 20 mM ferric chloride solution. The mixture was incubated at 37 ºC throughout the monitoring period. 3 mL of FRAP reagent was mixed with 100 µL of sample and 300 µL of distilled water. Absorbance readings were taken at 593 nm after every minute for 6 min. Results were compared with standard curve of ferrous sulfate.

RESULTS AND DISCUSSION

The synthesis of the target molecules was accomplished starting from commercially available sodium saccharin as a sweetening agent. Synthesis of this nucleus involves a base catalyzed isomerization of methyl-1,2-benzothiazoline-3(2*H*)-one-2-acetate-1,1-dioxide to the 1,2-benzothiazine nucleus analogous to Gabriel-Colman rearrangement[31] as reported by Abe[32] and later improved by Lomberdino *et al.*[33]; there are other approaches for its synthesis including a patent filed by Unverferth[34] transforming the methyl-2-{[(2-methoxy-2-oxoethyl)amino]sulfonyl}benzoate to the corresponding 1,2-benzothiazine. On the other hand, Pátek[35] reported a detailed study of the Dieckmann condensation of the former to the later using different bases. In the current studies, the whole reactions were tried to be carried out in one pot fashion under continuous nitrogen flow achieving an overall yield of 83-92 % and are compared with multi-step synthesis carried out by literature procedures[36]. Spectroscopic studies like FT-IR, ^{1}H NMR and MS were used to identify the intermediates and final products. X-Ray crystallographic analysis has also been employed to study the various interactions and their three dimensional arrangement in the molecules.

Single crystal X-ray crystallography of methyl-2-benzyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide (4f): All the non-hydrogen atoms were refined with anisotropic displacement parameters. The aromatic and aliphatic hydrogen atoms were positioned geometrically and

TABLE-2
CRYSTALLOGRAPHIC DATA AND STRUCTURE REFINEMENT DETAILS OF **4f**

Empirical Formula	$C_{17}H_{15}NO_5S$	Formula Weight	345.36
Temperature (K)	296(2)	Wavelength (Å)	0.71073
Crystal system	Monoclinic	Space group	P2(1)/c
a (Å)	9.492 (15)	α (°)	90.00
b (Å)	10.961 (17)	β (°)	99.758(2)
c (Å)	15.050 (23)	γ (°)	90.00
Volume (Å^3)	1543.12 (7)	Z, Calculated density (g cm^{-3})	4, 1.49
F(000)	719.9	Absorption coefficient (mm^{-1})	0.238
Crystal size (mm)	0.43 × 0.25 × 0.19	Range for data collection (°)	2.2 to 28.4
R(int)	0.0331	Reflection collected/unique	13430/3719
Completeness to θ	99.7 % (26)	Absorption correction	Multi-scan
Goodness-of-fit on F^2	1.063	Data/restraints/parameters	3719/0/222

treated as riding atom over their parent carbon atoms. Crystallographic data and refinement details are given in Table-2.

With the interest to study geometrical behaviour of the molecules of the series, compound **4f** was crystallized and studied by single crystal X-ray crystallography (Fig. 2).

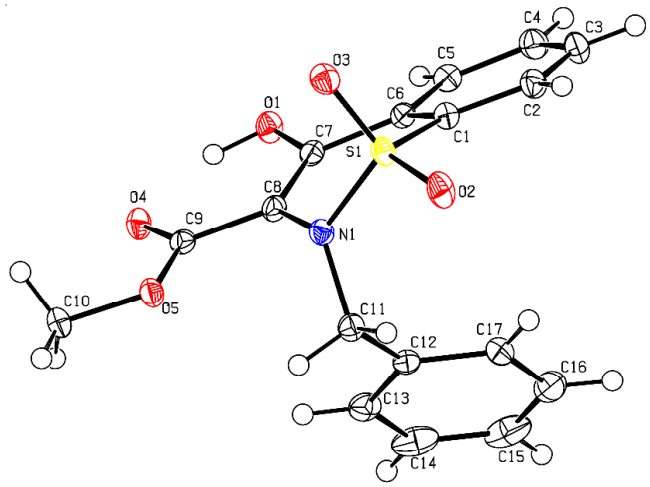


Fig. 2. ORTEP diagram for **4f** with the thermal ellipsoids drawn at 50 % probability level

Its crystal structure showed that the benzyl ring is attached to the thiazine ring system in such a way that the planer phenyl ring C11/C12/C13/C14/C15/C16 is oriented at dihedral angles of 44.52 (0.03)º and 51.43 (0.03)º with respect to the planar benzene ring C1/C2/C3/C4/C5/C6 and envelope shaped thiazine nucleus C1/C6/C7/C8/N1/S1, respectively. The dihedral angle between the two fused rings C1/C2/C3/C4/C5/C6 and C1/C6/C7/C8/N1/S1 is 11.67 (0.06)º. The O---H···O intramolecular hydrogen bonding interaction in the molecule is found which explain the enolic behaviour of the molecule.

Antioxidant assay: The results of antioxidant activity and the list of tested compounds using DPPH and FRAP method is presented in Figs. 3 and 4, respectively in comparison with that of reference compounds trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) and BHA (butylated hydroxyanisole). The findings of this study have revealed that most of the derivatives have shown antioxidant activities. According to the FRAP method, compound **4i** has shown maximum antioxidant activity while **4h** has shown the minimum activity. Compounds **4i** and **4j** exhibited almost same activities; both of these derivatives have lone pair containing oxygen

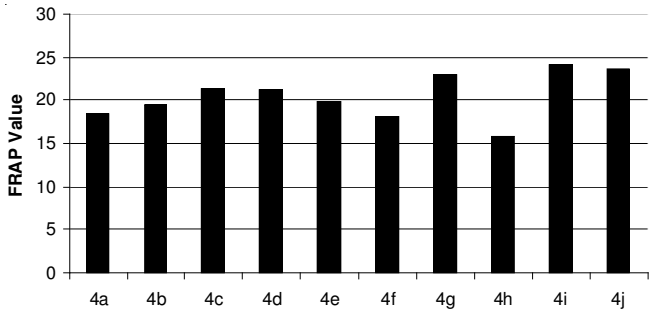


Fig. 3. A plot showing the antioxidant activity of synthetic compounds (**4a-j**) measured by FRAP method

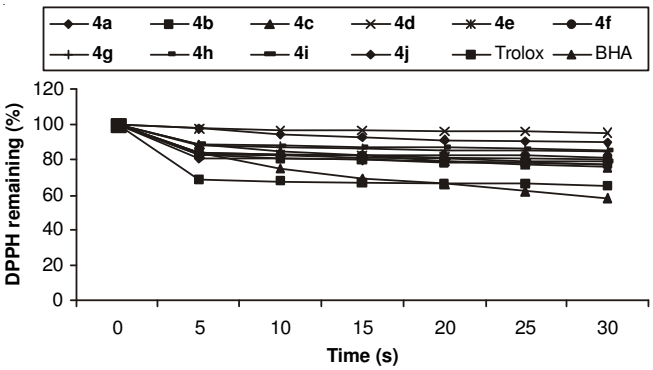


Fig. 4. Percentage DPPH remaining values of the compounds (**4a-j**) as a function of time (s)

atom in the *n*-alkyl groups. Compounds **4a-g** have intermediate activity as all these derivatives have been alkylated with alkyl groups having no lone pair containing atom in their structure.

According to the results of the DPPH assay, compound **4c** exhibited the best activity than the standard antioxidants (BHA and trolox); compound **4e** exhibited good activity. Compounds **4i** and **4j** has also shown good activity while other derivatives have moderate activity. From these observations, it can be concluded that the derivatives which have lone pair containing atoms and π bonds in their *n*-alkyl groups have good antioxidant activity while aromatic ring and simple aliphatic alkyl group containing no hetero atoms has no prominent affect on overall activity of the compounds.

Conclusion

Synthesis of N-alkyl derivatives of methyl-4-hydroxy-2*H*-1,2-benzothiazine-3-carboxylate-1,1-dioxide has been reported in a straight forward and convenient one step strategy

with over all good yields of the target N-substituted molecules. These molecules have been synthesized in order to find out their antioxidant potential. The results of antioxidant activity have revealed that groups with heteroatoms and π bonds have shown good antioxidant activities while aromatic ring has no prominent affect on it.

ACKNOWLEDGEMENTS

One of the authors, Muhammad Nadeem Arshad gratefully acknowledges the Higher Education Commission (HEC), Islamabad, Pakistan, for providing a scholarship under the Indigenous Ph.D. Program batch II (PIN 042-120607-PS2-183). He is also grateful to Dr. K. Travis Holman for the provision of necessary facilities to carry out a part of this work in his laboratories at the Department of Chemistry, Georgetown University, Washington D.C., USA.

REFERENCES

1. C. Leeuwenburgh and J.W. Heinecke, *Curr. Med. Chem*., **8**, 829 (2001).
2. I.M. Lee and R.S. Paffenbarger, *Am. J. Epidemiol*., **151**, 293 (2000).
3. I.M. Lee, R.S. Paffenbarger Jr. and C.H. Hennekens, *Aging (Milano)*, **9**, 2 (1997).
4. C. Sen, L. Packer, O. Hanninen, Amsterdam and R.J.M. McCarter, In Studies in Handbook of Oxidants and Antioxidants in Exercise, p. 797 (2000).
5. F. Dreher, N. Denig, B. Gabard, D.A. Schwindt and H.I. Maibach, *Dermatology*, **198**, 52 (1999).
6. D. Darr and I. Fridovich, *J. Invest. Dermatol*., **102**, 671 (1994).
7. J. Fuchs, M.E. Huflejt, L.M. Rothfuss, D.S. Wilson, G. Carcamo and L. Packer, *J. Invest. Dermatol*., **93**, 769 (1989).
8. Y. Shindo, E. Witt and L. Packer, *J. Invest. Dermatol*., **100**, 260 (1993).
9. K. Briviba and H. Sies, San Diego, CA: Academic Press, p. 107 (1994).
10. M. Zia-ur-Rehman, J.A. Choudary, M.R.J. Elsegood, H.L. Siddiqui and K.M. Khan, *Eur. J. Med. Chem*., **44**, 1311 (2009).
11. M. Ahmad, H.L. Siddiqui, M. Zia-ur-Rehman and M. Parvez, *Eur. J. Med. Chem*., **44**, 694 (2010).
12. W.R. Rough, S.L. Gwaltney, J. Cheng, K.A. Scheidt, J.H. McKerrow and E. Hansell, *J. Am. Chem. Soc*., **120**, 10994 (1998).
13. K.H. Ahn, C. Ham, S.-K. Kim and C.-W. Cho, *J. Org. Chem*., **62**, 7047 (1997).
14. Z. Xiao and J.W. Timberlake, *J. Heterocycl. Chem*., **37**, 773 (2000).
15. W.J. Layman Jr., T.D. Greenwood, A.L. Downey and J.F. Wolfe, *J. Org. Chem*., **70**, 9147 (2005).
16. D.K. Barange, T.C. Nishad, N.K. Swamy, V. Bandameedi, D. Kumar, B.R. Sreekanth, K. Vyas and M. Pal, *J. Org. Chem*., **72**, 8547 (2007).
17. D.K. Rayabarapu, A. Zhou, K.O. Jeon, T. Samarakoon, A. Rolfe, H. Siddiqui and P.R. Hanson, *Tetrahedron*, **65**, 3180 (2009).
18. G. Trapani, A. Reho, F. Morlacchi, A. Latrofa, P. Marchini, F. Venturi and F. Cantalamessa, *Farmaco. Ed. Sci*., **40**, 369 (1985).
19. A. Scozzafava, T. Owa, A. Mastrolorenzo and C.T. Supuran, *Curr. Med. Chem*., **10**, 925 (2003).
20. R. Wammack, M. Remzi, C. Seitz, B. Djavan and M. Marberger, *Eur. Urol*., **41**, 596 (2002).
21. R. Silvestri, G. Marfe, M. Artico, G.L. Regina, A. Lavecchia, E. Novellino, M. Morgante, C.D. Stefano, G. Catalano, G. Filomeni, E. Abruzzese, M.R. Ciriolo, M.A. Russo, S. Amadori, R. Cirilli, F. La Torre and P.S. Salimei, *J. Med. Chem*., **49**, 5840 (2006).
22. M. Zia-ur-Rehman, J.A. Choudary, S. Ahmad and H.L. Siddiqui, *Chem. Pharm. Bull*., **54**, 1175 (2006).
23. J. Chiani and E.H. Wiseman, *Biochem. Pharm*., **21**, 2323 (1972).
24. D.L. Daniela, B. Francesco and C. Renso, *J. Chem. Soc. Dalton Trans*., 1993 (1998).
25. H. Li-Ren, H. Yaw-Bin, W. Pao-Chu and T. Yi-Hung, *Int. J. Pharmaceut*., **106**, 1 (1994).
26. D. Fowler, K. Isakow, N. Caulkett and C. Waldner, *Cand. Vat. J*., **44**, 603 (2003).
27. Z. Otwinowski and W. Minor, *Methods Enzymol*., **276**, 307 (1997).
28. R.W.W. Hooft, COLLECT, Nonius BV, Delft, The Netherlands (1998).
29. K. Shimada, K. Fujikawa, K. Yahara and T. Nakamura, *J. Agric. Food Chem*., **40**, 945 (1992).
30. I.E.F. Benzie and J.J. Strain, *Anal. Biochem*., **239**, 70 (1996).
31. J.G. Lombardino, E.H. Wiseman and W. Mclamore, *J. Med. Chem*., **14**, 1171 (1971).
32. K. Abe, S. Yammamoto and K. Matsui, *Yakagaku Zasshi*, **76**, 1058 (1956).
33. S. Gabriel and J. Colman, Ber. **33**, 980, 996, 2630 (1900); **35**, 2421 (1902).
34. K. Unverferth, G. Laben, W. Gunther, D. Lohmann, E. Kretzschmar, H. Kassebaum, L. Lucke, E. Jassmann, H. Hilger and H. Runge, J. Ger. (G.D.R), Patent 247672 (1983); *Chem. Abstr*., **108**, 150061 (1988).
35. M. Pátek and F. Hampl, *Coll. Czech. Chem. Commun*., **54**, 3267 (1989).
36. M. Zia-ur-Rehman, S. Ahmad and J.A. Choudary, *Bull. Korean Chem. Soc*., **26**, 1771 (2005).