Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2012, 4(9):4260-4265

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Synthesis, characterization and antimicrobial activities of 2,2′-dinitrodiphenylmethanes

A. Jepa Malar*[a], M. Christudhas[a] and G. Allen Gnana Raj[b]

[a]Department of Chemistry, N. M. Christian College, Marthandam, Tamilnadu, India- 629165
[b]Department of Chemistry, Scott Christian College (Autonomous), Nagercoil, Tamilnadu, India-629003,

---

ABSTRACT

The present study reports the synthesis of 2,2′- dinitrodiphenylmethanes from 2-bromoaniline and formaldehyde in acidic medium followed by nitration using potassium nitrate in 98% H_2SO_4. Dichloro derivatives of 2,2′-dinitrodiphenylmethanes have been synthesized by Sandmeyer's reaction. The chemical structure of these compounds have been confirmed by UV-visible, FTIR and ^{1}H-NMR spectroscopic analysis. The compounds have been screened for their antibacterial and antifungal activity.

Keywords : 2-Bromoaniline, Formaldehyde, 2,2′- Dinitrodiphenylmethanes, Sandmeyer's reaction, antimicrobial activity.

INTRODUCTION

The chemistry of 2,2′- dinitrodiphenylmethane compounds had been investigated extensively for the last several decades leading to new synthetic routes, variety of structures and their potential biological applications[1-4]. The survey of literature reveals that no attempt has been made to study the behavior of 2,2′-dinitrodiphenylmethanes derived from the condensation of 2-bromoaniline and formaldehyde. The infrared spectra of nitro groups have been the subject of detailed investigation during the past several years [5-8]. It is well known that several 2,2′-dinitrodiphenylmethane compounds have anti-fungal and antibacterial activity. Nitroaromatics are hazardous chemicals that display several manifestations of toxicity, including skin sensitization [9], immunotoxicity [10], germ cell degeneration [11], inhibition of liver enzymes [12] and also a conjectured carcinogenicity [13]. Nitro aromatic compounds are cytotoxic, it has been proposed that their cytotoxicity is due to the generation of reactive oxygen species such as superoxide radical anion, singlet oxygen and hydrogen peroxide [14]. Possible formation of highly mutagenic or carcinogenic nitrocompounds is unquestionsbly of great environmental interest [15]. Nitrated polycyclic aromatic compounds (nitro PAC), including nitrated polycyclic aromatic ketones, have been found to be mutagenic as well as carcinogenic. 3-Nitrobenzathrone (3NB), a powerful bacterial mutagen, was first identified by investigators using bioassay-directed fractionation and chemical analysis of diesel emission particle extracts. 3NB is an effective human cell mutagen, significantly including mutations at the tk and hprt loci in both cell lines. Investigators have reported the formation of DNA adducts by 3NB both *in vitro* and *in vivo* and urinary metabolities of 3NB have been found in workers occupationally exposed to diesel exhaust [16]. The present investigation describes the synthesis, characterization and antimicrobial studies of 2,2′-dinitrodiphenylmethane and its dihalo derivatives.

EXPERIMENTAL SECTION

All the chemicals used were of GR grade obtained from Merck, India and were used without further purification.

The melting points of the compounds were determined by digital Auto Melting point apparatus, Labronies. The purity of the synthesized compounds were checked by TLC using silica gel "G" as adsorbent and visualizing through Heber Scientific Mini UV viewer. IR spectra were recorded on a Perkin-Elmer Precisely Spectrum 100 in the 4000 – 400 cm^{-1} region. UV - visible spectra were obtained on a Perkin – Ellmer Lamda 750 UV – vis spectro meter using dimethyl sulphoxide as solvent in the 200 – 800 nm regions. The ^1H NMR spectral analysis were performed on a JEOL 300 JMTC – 300 / 54 spectrometer using tetramethyl silane as internal standard. Chemical compositions of the synthesized compounds were carried out using Hitachi S-4800 field emission scanning Electron microscope (FE-SEM) equipped with EMAX elemental analyzer.

Synthesis

4,4′-Diamino-3,3′-dibromodiphenylmethane (DPM 1)

4,4′-Diamino-3,3′-dibromodiphenylmethane was prepared by the condensation of 2-bromoaniline with formaldehyde in acidic medium following the procedure reported by Scanlan [17]. 2-Bromoaniline (17.202g), formaldehyde (1.505g) and conc. HCl (50mL) were heated at about 60^oC for three hours in a RB flask (Scheme - 1). The product formed was filtered, washed with water, and dried in vacuum and crystallized in ethanol a colourless shining flakes of 4,4′-diamino-3,3′-dibromodiphenylmethane (m.p.115^oC) was obtained.

4,4′-Diamino-5,5′-dibromo-2,2′-dinitrodiphenylmethane (DPM 2)

4,4′-Diamino-3,3′-dibromodiphenylmethane was nitrated using a mixture of anhydrous potassium nitrate and 98 % sulphuric acid at 0^0 C [18]. A solution of potassium nitrate (0.05 mol) in conc. H_2SO_4 (18N) at 0^0 C was added to an ice-cold solution of 4,4′-diamino-3,3′-dibromodiphenylmethane (0.025 mol) in conc. H_2SO_4 (18N) drop wise over a period of half an hour with stirring. The stirring was continued for another three hours, keeping the reaction mixture at 0^0 C. The reaction mixture was diluted by pouring it over crushed ice and was then neutralized with ice-cold ammonia solution (1:1). The orange yellow solid was filtered, washed thoroughly with water and dried (Scheme - 2). This product was recrystallised from ethyl alcohol-ethyl acetate mixture (90:10) and that furnished orange red flakes of 4,4′-diamino-5,5′-dibromo-2,2′-dinitrodiphenylmethane [19] (m.p.215^o C) was formed.

5,5′-Dibromo-4,4′-dichloro-2,2′-dinitrodiphenylmethane (DPM 3)

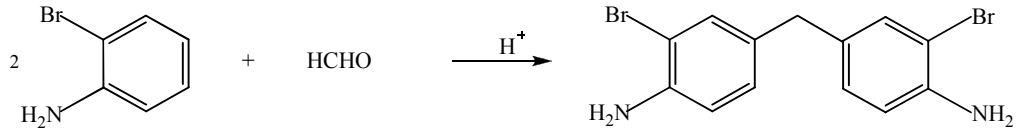
4,4′-Diamino-5,5′-dibromo-2,2′-dinitrodiphenylmethane (8.14 g) dissolved in 1:1 hydrochloric acid (30 mL) was diazotized at 0^0 C with sodium nitrite (16.9 g). The reaction mixture was then added to a cold solution of cuprous chloride (11.6 g) in conc.HCl (30 mL) (Scheme - 3). The crude product formed was chromatographed on a column of neutral alumina using petroleum ether – benzene (5:1 v/v) as eluant [20]. The product thus obtained was crystallized from benzene – petroleum ether (1:10 v/v) as pure pale yellow flakes (m.p.245^o C).

Antimicrobial screening

The *in vitro* antimicrobial activity of the synthesized 2,2′-dinitrodiphenyl methane derivatives on selected bacteria *Pseudomonas aeruginosa (NCIM 2026), Proteus vulgaris (NCIM 2027), Staphylococcus aureus(NCIM 2127), Klebsiella pneumonia (NCIM 5082)* and *Escherichia Coli (NCIM 2563)* and two fungi *Penicillium notatum(NCIM 745)* and *Aspergillus niger(NCIM 616)* was carried out. The antimicrobial action was studied by sterile disc method using concentration of 1mg/ml. Amikacin was used as a standard for antibacterial screening and Flucanazole was used as a standard for antifungal screening. The growth of the microbes was measured by recording the diameter of the inhibition zone.

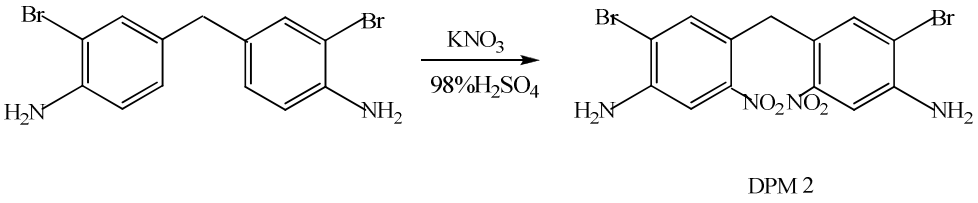
RESULTS AND DISCUSSION

Mechanism



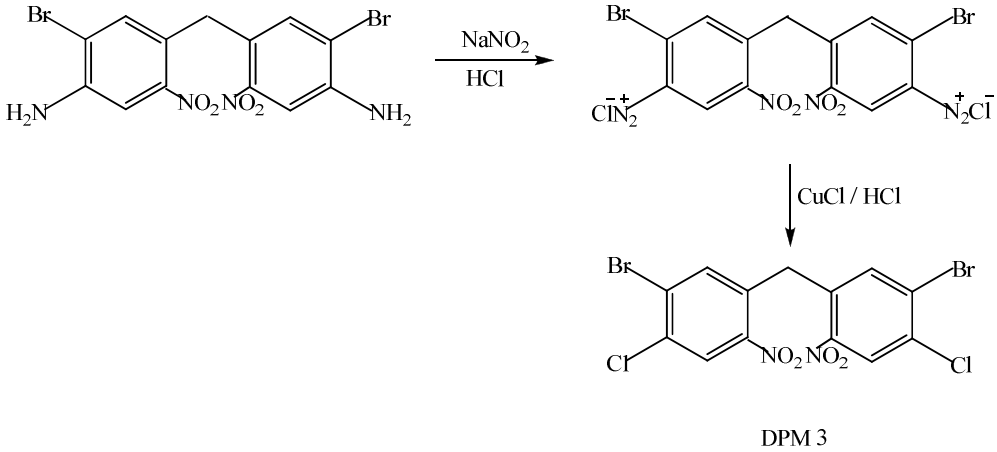
DPM 1

Scheme -1



DPM 2

Scheme -2



DPM 3

Scheme -3

The molecular formula is obtained from elemental analysis. The elemental analysis, molecular formula and melting points are given in Table 1.

Table - 1 : Analytical data of the compounds

Compound	Elemental Analysis (%)						Molecular Formula	Melting point (˚C)
	C	H	N	O	Br	Cl		
DPM 1	43.83	3.42	7.88	-	44.87	-	$C_{13}H_{12}N_2Br_2$	115
DPM 2	34.98	2.25	12.59	14.33	35.85	-	$C_{13}H_{10}N_4O_4Br_2$	215
DPM 3	32.18	1.25	5.75	13.23	32.93	14.66	$C_{13}H_6N_2O_4Cl_2Br_2$	245

Spectral Analysis

UV – visible spectra

The DPM 1 shows λ_{max} value of 330 nm. The absorption band at 330 nm is due to an n→π* transition of the amino group of the compound. The DPM 2 shows λ_{max} value of 380 nm. This absorption band is due to n→π* transition of amino and nitro group. The absorption bands due to the nitro group of aromatic nitro compounds are generally hidden under the intense bands arising from the π→π* transitions of the aromatic ring. The nitro group being very highly electron withdrawing in nature causes marked bathochromic shifts of the aromatic absorption bands. The DPM 3 shows λ_{max} value of 332. This is due to n→π* transition of the nitro group. The difference in λ_{max} value of all these compounds indicates the formation of new compound in each step. The UV spectral data of the synthesised compounds are presented in Table 2.

FTIR spectra

IR spectrum of the DPM 1 and 2 show a band centered around 1620 cm^{-1} which has been assigned to the presence of N-H bending vibration [21]. The absorption band at 3350 and 3440 cm^{-1} in DPM 1 and 3350 and 3500 cm^{-1} DPM 2 corresponds to the N-H asymmetric and symmetric stretching vibrations respectively [22]. The band at 1523 and 1347 cm^{-1} in DPM 2 and 1550 and 1350 cm^{-1} in DPM 3 is due to asymmetric and symmetric N-O stretching vibrations. The band at 1465 cm^{-1} in DPM 1 and 2 and 1450 cm^{-1} is due to C-H bend methylene group. C-H out of plane bending occurs at frequency 750, 770 and 720 cm^{-1} in DPM 1, 2 and 3 respectively [23]. The band at 3072, 3200 and 3075 cm^{-1} in DPM 1,2, and 3 is due to aromatic C-H group. The band at 650, 650and 625 cm^{-1} in DPM 1, 2 and 3 is due to C-Br stretching vibration. The band at 850 cm^{-1} in DPM 3 is due to C-Cl stretching vibration. The IR spectral data of the synthesised compounds are presented in Table 2.

^{1}H NMR spectra

The signal at δ 3.84, 3.43 and 3.39 in DPM 1, 2 and 4 is due to two methylene proton (s, 2H, Ph-CH_2-Ph). The signal at δ 4.74 and 4.20 in DPM 1 and 2 is due to four amine proton (s, 4H, NH_2). The signal for aromatic proton exist as a multiplet.

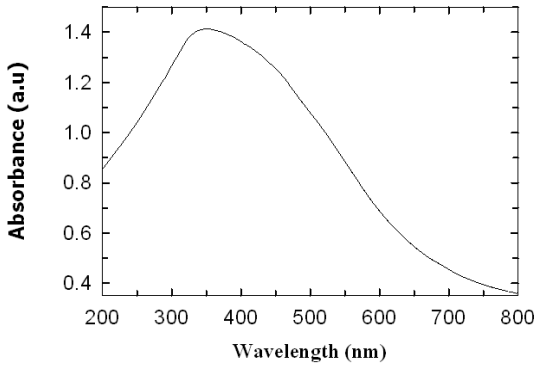


Fig 1 UV – visible spectrum of 5,5′-Dibromo-4,4′-dichloro-2,2′-dinitrodiphenylmethane

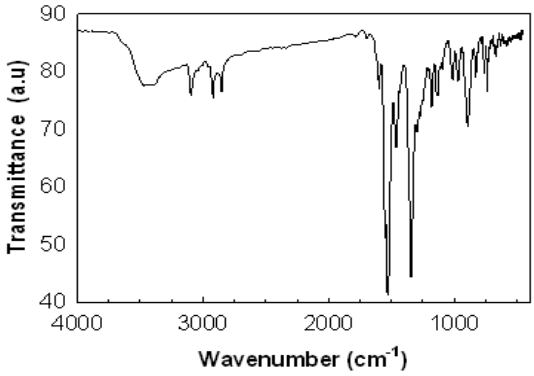


Fig 2 IR spectrum of 5, 5′-Dibromo-4,4′-dichloro-2,2′-dinitrodiphenylmethane

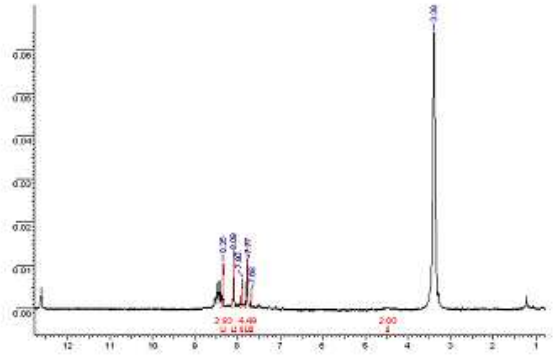


Fig 3 ^{1}H NMR spectrum of 5,5′-Dibromo-4,4′-dichloro-2,2′-dinitrodiphenylmethane

Table - 2 : Infrared and UV-visible Spectra of 2.2′-dinitrodiphenylmethane compounds

Compound	Frequency of the peak (cm^{-1})										UV- visible spectra λ_{max} (nm)
	υ(N-H) (Bend)	υ(N-H) (Asym)	υ(N-H) (Sym)	υ(C-H) (Methylene)	υ(C-H) (Aromatic)	υ(C-N)	υ(N-O) (Asym)	υ(N-O) (Sym)	υ(C-Br)	υ(C-Cl)	
DPM 1	1620	3350	3440	1465	3072	1175	-	-	650	-	330
DPM 2	1620	3350	3500	1465	3200	1175	1523	1347	650	-	380
DPM 3	-	-	-	1450	3075	-	1550	1350	625	850	332

Antimicrobial activity studies

The results of the antibacterial and antifungal screening of the 2,2′-dinitrodiphenylmethane derivatives with *Pesudomonas aeruginosa, Proteus vulgaris, Staphylococcus aureus, Klbsiella pneumonias, Escherichia Coli, Penicillium notatum and Aspergillus niger* by sterile disc method are given in Table 3. The synthesized compounds

exhibited varying degree of antibacterial and antifungal activity against the test organisms. The substituent which attached to the benzene ring, affected the antimicrobial and antifungal activity. The zone of inhibition differs with respect to the different positions of the substituents of 2,2′-dinitrodiphenylmethane derivatives. With same substituent but different position in the benzene ring, the activity are affected differently. From the Table 3 it is seen that the DPM 1 shows the highest antibacterial activity with the zone of inhibition 20mm against *Staphylococcus aureus* and antifungal activity with the zone of inhibition 17mm against *Penicillium notatum and Aspergillus niger*. The DPM 2 shows the highest antibacterial activity with the zone of inhibition 16mm against *Escherichia Coli* and antifungal activity with the zone of inhibition 12mm against *Penicillium notatum.* The DPM 3 shows the highest antibacterial activity with the zone of inhibition 17mm against *Staphylococcus aureus* and antifungal activity with the zone of inhibition 10mm against *Penicillium notatum and Aspergillus niger*. Apparantly chloro substituent on the benzene ring is the most active substituent. From the Table 3 it is clear that all the newly synthesized compounds are highly active against the bacteria *Staphylococcus aureus and the fungi Penicillium notatum*. According to Overtons concept of cell permeability, the lipid membranes that surround the cell favours the passage of only the lipid soluble material due to which lipid solubility is an important factor, which control antimicrobial activity[24].

Table 3: Invitro antimicrobial activity of compounds and their inhibition zone in mm

Micro organism	Zone of Inhibition (mm)				
	DPM 1	DPM 2	DPM 3	Amikacin	Flucanazole
Bacteria *Pesudomonas aeruginosa*	12	10	12	25	-
Proteus vulgaris	18	10	13	25	-
Staphylococcus aureus	20	13	17	28	-
Klbsiella pneumonia	13	11	12	20	-
Escherichia Coli	10	16	9	28	-
Fungi *Penicillium notatum*	17	12	10	-	20
Aspergillus niger	17	10	10	-	18

CONCLUSION

In the present study, some new 2,2′-dinitrodiphenylmethane derivatives have been synthesized and characterized by UV-visible, IR and ^{1}HNMR spectral analysis. The antimicrobial data show that all these compounds are active against pathogenic species. Moreover, the studies show the significant antimicrobial activity to *Staphylococcus aureus* and *Penicillium notatum*. The compounds also inhibit the growth of fungi and bacteria to a greater extent as the concentration is increased. The compounds can be used as potent antibacterial and antifungal agents. Further study is needed for the identification of active site. It is essential to predict the leading molecule and drug like property at the onset of drug design which will helps in drug development.

REFERENCES

[1] M.B. Ferrari, S. Capacchi, F. Bisaglic and G.P. Pelosi, *Inorg. Chim. Acta*., **2001**, 81, 312.
[2] Vandim Yu. Kukushkin and Armando J.L. Pombeiro, *Coord Chem.Revewies*., **1999**, 181, 147.
[3] Vandim Yu. Kukushkin and Armando J.L. Pombeiro, *Coord Chem.Revewies*., **1996**, 156,333.
[4] R. Shakru, N.J.P. Subhashini, K. Sathish Kumar, Shivaraj, *J. Chem. Pharm. Res*., **2010**, 2(1): 38-46.
[5] C.N.R. Rao, *Chemical Applications of Infrared Spectroscopy*, Academic Press, New York., **1963**.
[6] N. Kornblum, H.E. Ungnade and R.A. Smiley, *J. Org. Chem*., **1956**, 21, 377.
[7] J.F. Brown, *J. Am. Chem. Soc*., **1955**, 77, 6341.
[8] J. Mason and J. Dunderdale, *J. Chem. Soc*., **1956**, 759.
[9] Cronin MTD, B.W. Gregory, T.W. Schultz. *Chem. Res. Toxicol*.**1998**, 11, 902–908.
[10] Q Li, M Minami, T Hanaoka, Y Yamamura. *Toxicol*. **1999**; 137:35–45.
[11] K Shinoda, K Mitsumori, K Yasuhara, C Uneyama, H Onodera, K Takegawa, M Takahashi, T Umemura. *Arch. Toxicol*. **1998**; 72:296–302.
[12] Sajan M, Reddy G, Kulkarni AP. *Int. J. Toxicol*. **2000**; 19:285–292.
[13] Ohkuma Y, Kawanishi S. *Biochem. Biophys. Res. Commun*. **1999**;257: 555–560.
[14] H. Sies, H. Degroot. *Toxicol. Lett*. 64-65(**1992**) 547.
[15] N. Matykiewiczova et al / *Journal of photochemistry and Photobiology A: Chemistry* 187 (**2007**) 24-32.
[16] P.T. Phousongphouang, *J. Arcy / Atmospheric Environment* 37 (**2003**) 3189-3199.
[17] J.T. Scanlan, *J. Am. Chem. Soc*., **1935**, 57, 860.
[18] P. Ehrlichand, H. Bauer, *Ber. Dtsch. Chem. Ges*., **1915**, 48, 502.
[19] K. Sudheesh Kumar, *Orient. J. Chem*., **2010**, 26 (4), 1393-1399.
[20] L. Mascarelli, B. Toschi, and , T. Zambonini, *Atti. Acad. Naz. Lincei*., **1910**, (II) 19, 338.
[21] P.S. Kalsi, *Spectroscopy of Organic Compounds*, 6th Edition., **2004**, 110.

[22] G. Socrates, *Infrared Characteristic Group Frequencies*, John Wiley and Sons, New York., **2004**.
[23] S. Gunasekaran, and S.R. Varadhan, *Orient. J. Chem*., **1995**, 11, 27.
[24]Shivaraj et al, *J. Chem.Pharm. Res.*, **2010,** 2(1), 38-46.