THE SYNTHESIS OF SOME BIS-SUBSTITUTED FORMAZANS AND THE INVESTIGATION OF THE EFFECT OF SUBSTITUENT UPON THEIR UV-VIS. ABSORPTION λ_{max} VALUES

H. TEZCAN AND N. ÖZBEK

Gazi University, Faculty of Gazi Education, Department of Chemistry,
Teknikokullar, 06500, Ankara-Turkey
Email: habibe@gazi.edu.tr

ABSTRACT

In this study 1,4-bis–[3,3'-phenyl-5,5'-(2-carboxyphenyl)-formaz-1-yl]-benzene-2-sulphonic acid (2,CSPF) and its derivatives with OH groups at the *o-*, *m-* and *p-* positions of 3-phenyl ring were synthesized. The structures of the formazans thus obtained were elucidated by elemental analysis, MASS spectroscopy, IR, ^{1}H-NMR and UV-Vis spectra. Also UV-Visible λ_{max} values were investigated according to substituent effect. The λ_{max} values were observed to shift towards shorter wave lengths by 130 nm in CSPF relation to 1,3,5-triphenylformazan (TPF) due to the fact that the structure of CSPF contained electron withdrawing –COOH and –SO$_3$H groups (hypsochromic effect). The addition of –OH group to 3-phenyl ring of CSPF, was observed a small bathochromic effect in accordance to the electron donating effect of –OH group.

1. INTRODUCTION

There are numerous studies related to formazans such as their synthesis, structure evaluation, photochromic transitions, tautomer formation, redox potentials [1-6] and synthesis of crown formazans [7,8]. There were various metal-formazan complexes synthesized and their thermogravimetric analysis [9] and dissociation and stability constants were investigated [10]. The Lanthanide complexes of some formazans were also synthesized and their stability and formation constants [11] and their electrochemical behaviors were examined [12].

Formazan/tetrazolium system is described as the marker of vitality [13]. These compounds are used in Brucella-ring test in milk [14]. Blue tetrazolium/formazan systems are used to demonstrate enzymes activity in normal and neoplastic tissues

but it was found to be ten times more toxic in vivo (mice) than mono tetrazolium/formazan system itself [15].

Formazan/tetrazolium system is quite useful in the determination of the effect and the selection of anti-cancer drugs [16, 17]. However Kabler states that they can not act as a marker every time, since the age of the cell and medium are also important [18].

The IR and Raman spectra of the formazans were investigated [5, 19] and the effect substituents upon complex forming properties [20], pKa values formazan complexes [21] and the dependence of their absorption properties upon pH were determined [22].

Formazans are easily oxidized to give tetrazolium salts. When given to a living organism these salts are reduced back to formazan depending upon the viability of the organism. These enables the viability of the organism be tested by monitoring formazan formation with spectroscopy. That is why the spectroscopic investigation of formazans is of utmost importance.   .

In our previous work we synthesized various formazans with electron donating and withdrawing groups attached to 1-phenyl ring and investigated the effects of substituents [23]. There are also related studies to the substitution of electron donating [24] and electron withdrawing groups [25] to 3-phenyl ring.

The purpose of this study is to synthesize water soluble macromolecular bis-formazans with various structures and to investigate their spectral properties. The investigation of λ_{max} values which is the measure of coloring properties of formazans is important in regard to their major uses in dye industry and vitality marker in medicine.

2.MATERIAL AND METHODS

This study was carried out in three stages. The first step was the synthesis of 1,3,5–triphenylformazan (TPF, Fig. 1a, as the comparison compound) and new substituted bis- formazans **2-5** (Fig. 1b). The second step was the determination of the spectral properties of these formazans. The third stage was the evaluation of the spectral data and determination of their λ_{max} values. The shifts in λ_{max} values of **2-5** were evaluated in comparison to TPF (**1**) and the shifts in **3-5** were evaluated in comparison to CSPF (**2**).

1-a

1-b
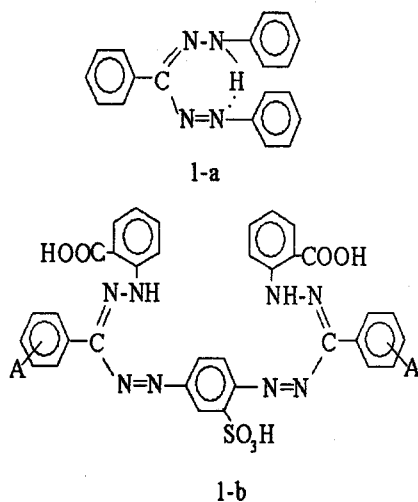
Comp.	A	Abbrev.	Figure
1	--	TPF	1a
2	H	CSPF	1b
3	o-OH	o-HCSPF	1b
4	m-OH	m-HCSPF	1b
5	p-OH	p-HCSPF	1b

**Figure1.** The structure of the formazans synthesized

2.1 Synthesis of Formazans

There are three distinctive routes proposed for the synthesis of formazans in literature. The first one is the condensation of aromatic and aliphatic aldehydes with phenyl hydrazine and the coupling reaction of the resulting hydrazones with diazonium salts [6]. Although this is highly cumbersome and resulting products are difficult to purify, it has the advantage of be able to synthesizing symmetrical and asymmetrical formazans [24, 25]. The second way is the coupling of active methylene compounds with two moles diazonium cations. This is highly practical and easy but only gives symmetrical formazans [19]. The final way is the phase transfer. However it requires special regents such as crown ethers and tetrabuthyl ammonium bromide etc. [6].

The formazans in this study were synthesized by the first way, coupling of hydrazones with diazonium cations in basic media at -5⁰C to 0⁰C. The necessary hydrazones were obtained by the condensation reaction of benzaldehyde (or substituted benzaldehydes) and phenylhydrazine (or substituted phenylhydrazine) at pH 5-6. This is the most difficult and low yielding route in the synthesis of formazans. The purification of the products is cumbersome and requires patience [6]. The reason for the preference of this way in spite of all these setbacks is that the starting material is easily available in every laboratory. We tried to increase the yield in order to provide a cheap way to synthesize formazans for medical, dying and analytic applications.

The reaction mechanism was followed with UV-Visible spectrum in TPF synthesis. The fact that NH proton is more acidic than CH proton, the first diazonium coupling was realized through NH proton, forming orange colored 4-benzylidene-1,3-diphenyltetraz-1-en. This is highly instable in basic media and gives an intramolecular rotation turning into red colored formazans. This is in good accordance with our previous study [25].

The basic buffer solutions employed were 0.1M $HClO_4$ + 0.05 M borax solution (pH 7.60~9.00) and 0.1M NaOH+0.05 M borax solution (pH 9.30~10.80). However, the best yield was obtained with the NaOH+CH_3COONa buffer solution (pH 10~12). The formation mechanism of bis-substituted formazans was given in Scheme 1 and the experimental data were tabulated in Table 1. The structures of the formazans were elucidated by the use of elemental analysis, MASS spectroscopy, IR, ^{1}H-NMR and UV-Vis spectra.
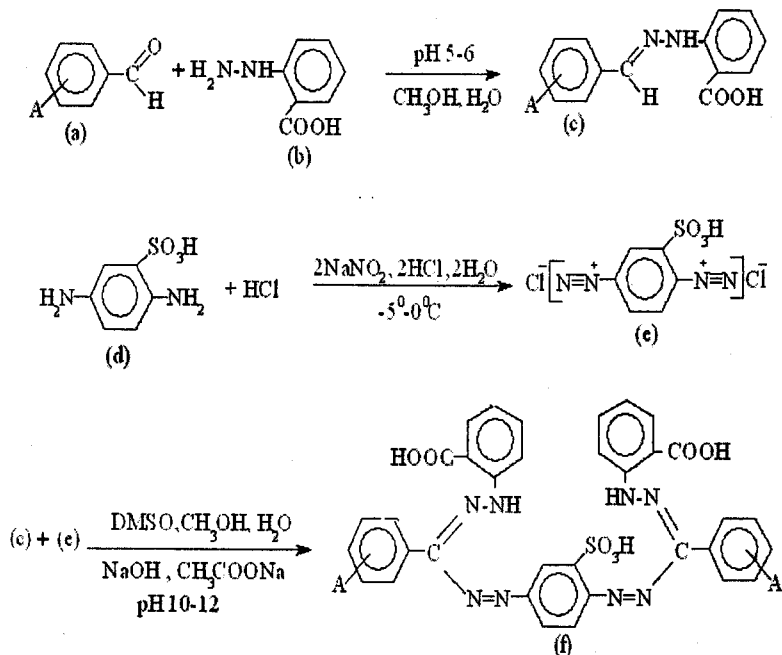
Scheme 1 The formation mechanism of formazans.

Table 1-a. Experimental data of the formazans synthesised

Comp	m.p / °C (lit)	Yield / % (lit)	Color
	Experimental data		
1	172-173 (172-174)[6]	75 (54)[6],(63)[24]	Cherry red
2	255-256	55	Salmon
3	213	46	Cyclamen
4	>300	53	Yellow-brown
5	207	54	Light brown

Table 1-b. Elemental analysis of the formazans synthesised

	Elemental analysis							
	Calculated				Found			
	C	H	N	S	C	H	N	S
1	76.00	5.33	18.66	-----	75.97	5.29	18.69	-----
2	59.13	3.76	16.23	4.63	59.01	3. 67	16.12	4.68
3	56.51	3.60	15.51	4.43	56.48	3.54	15.47	4.55
4	56.51	3.60	15.51	4.43	56.48	3.54	15.47	4.55
5	56.51	3.60	15.51	4.43	56.48	3.54	15.47	4.55

As seen from **Table 1.a.** that the yield of TPF is higher than that reported in literature [6, 24]. These have been made the results of carefully pH adjustment and showing scrutiny to each step. The lowest yield was obtained at the *o*-position in **2-5** formazans. This can be attributed to the fact that substituents are the closest to the reaction site in this position, which sterically hinders the coupling reaction. The relative increase in yield at both *m*- and *p*- positions verifies this hypothesis [25]. If an electronic effect (resonance and inductive effects) were dominant, the best yield would be obtained at *o*-position.

The biggest difficulty was encountered in the synthesis of *m*-OH bis formazan **4**. The resulting product was resinous and non crystallizable. The synthesis was repeated several times using dilute conditions and various pH values, to prevent this situation. However, it was not possible to obtained perfect crystals.

2.2 Spectral properties
As seen from **Table 2** the ^{1}H-NMR data showes the δ values of CSPF shifted to lower field compared to those of TPF. For instance δ values for Ar-H is 7.55 ppm in TPF (**1**) and showed a 1.00 ppm shift to the lower field and became 8.55 ppm in CSPF. This is perfectly justifiable since there are electron withdrawing groups such as two –COOH and one –SO$_3$H in the structure of CSPF (**2**). In compounds <u>3</u>-<u>5</u> the substitution of –OH groups at *o*-, *m*- and *p*- positions resulted δ values to show a slight shift to higher fields compared with compound <u>2</u>. This is quite expectable from the electron donating feature of –OH groups. However the shift towards the higher fields as a result of substituting OH groups to the ring is lower compared with the shift towards the lower fields as a result of substitution of –COOH and –SO$_3$H. For instance δ value makes a shift of 0.63 ppm higher field from CSPF to *o*-HCSPF (8.55 to 7.92 ppm). This Δδ value is lower in *m*-position compared with *o*-position since there is no resonance effect and only a weak inductive effect at *m*-position. This shift at *p*-position is higher than *m*- and lower than *o*- positions. Because at *p*-position the inductive effect is diminished but there is a resonance effect. That is why the δ values of compounds 3-5 are at highly lower fields compared with that of TPF but a little bit higher than CSPF. These peaks are compatible with the structure

depicted in Figure-1b [26]. The N-H, Ar-OH, -COOH and $-SO_3H$ groups were observed in the expected regions (Table 2).

Table 2. The ^{1}H-NMR data of formazans 1-5, (400 MHz, in $CDCl_3$)

Comp.	Ar-H	N-H	Ar-OH	COOH	SO₃H
1	7.55-6.70(m,15H)	1.14 (s,1H)	---	---	---
2	8.55-6.90(m,21H)	2.50-2.25(s,2H)	---	10.90(s,2H)	11.28(s,1 H)
3	7.92-6.85(m,19H)	2.75-2.60(s,2H)	3.10(s,2H)	10.80(s,2H)	11.27(s, 1H)
4	8.50-6.65(m,19H)	2.55-2.20(s,2H)	3.40(s,2H)	10.85(s,2H)	10.85(s,1H)
5	7.98-6.65(m,19H)	2.05-1.70(s,2H)	3.20(s,2H)	10.85(s,2H)	10.85(s,1H)

The investigation of the **IR** data in **Table 3** reveals that -C=N stretching band is located at 1500 cm^{-1} in TPF. This shows the presence of chelation in TPF. NH hydrogen forms a hydrogen bond upon the electron pair on azo-nitrogen $(-N = \overset{..}{N} -)$ and causes chelation and therefore tautomerism [1, 5, 25]. There is a symmetry element in the molecule (Scheme 2)
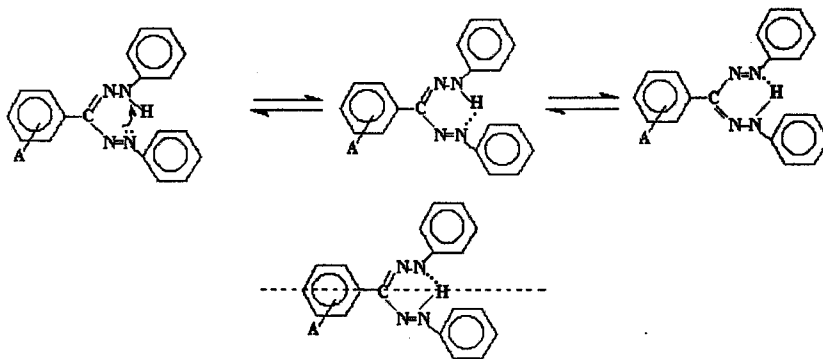
Scheme 2 Molecular chelation and symmetry

Table 3. The IR spectral data of formazans 1-5, (in KBr, cm^{-1})

Comp.	C=N stretching	N=N stretching	Aromatic C=C stretching	CNNC Skeletal vibration	Ar-OH stretching	-COOH stretching	-SO$_3$H stretching
TPF 1	1500	1358	1600	800~600	3050-300	------	----
CSPF 2	1540-1580	1455-1335	1629	930~600	3600	3400-3300	1420-1330
o-HCSPF 3	1590-1520	1418-1330	1620	850~620	3480-3300	3390-3300	1385-1220
m-HCSPF 4	1635-1580	1450-1335	1630	920~610	3600-3200	3420-3210	1410-1340
p-HCSPF 5	1600-1530	1420-1335	1622	850~620	3400-3220	3395-3300	1380-1220

The C=N stretching peak is observed above 1635-1520 cm^{-1} in compounds 2-5. This indicates that the fact that bis-formazans are contain –SO$_3$H and two –COOH groups increases the tension and inhibit the chelation to great extent. However it is observed that although in a very small extend there is the presence of chelate form in the equilibrium. Since it is known that if C=N stretching band is located in 1565-1551 cm^{-1} or higher. There is no chelation and the molecule is in the excited state. On the other hand the location of the C=N stretching band between 1510 -1500 cm^{-1} is the indication of chelation [6, 27]. Our upper limit for 2-5 formazan at 1640 cm^{-1} is the indication of the absence of chelation while the lower limit of 1520 cm^{-1} verifies the presence of a very small chelation. The fact that the peaks at 1529 cm^{-1} are very weak is also another indication of the very small extent of the chelation ratio.

-N=N- **stretching** bands in TPF was observed 1358 cm^{-1}. This value shows chelation in the molecule since this band is expected at 1400-1450 cm^{-1}. This decrease in wave number stems from the resonance stabilization due to the formation of chelation in the molecule. In substituted formazans 2-5, -N=N-stretching bands was observed between 1455-1330 cm^{-1}. These values reveal that the excited state of the molecule, distortion of the chelation and probable the presence of trans-formation (1455 cm^{-1}). The values of 1420 cm^{-1} or higher is the indication of –N=N- bands of the formazans without chelation and the values of 1442 cm^{-1} or higher is the verification of the trans conformation of the molecule [6,27]. Also the very weak bands observed between 1360-1335 cm^{-1} is the proof of the presence of chelate form in equilibrium with a very small extend. CNNC skeleton vibration peaks were observed in the fingerprint region as expected from the structure of formazans. N-H and Ar-OH stretching peaks appeared between 3600-3000 cm^{-1}. Other peaks were also in accordance with structure of formazans.

There are three distinctive peaks in the **UV-Visible** spectra of formazans. The typical broad peak (λ_1) observed at 350~ 600 nm is specific to formazans. The peak (λ_2) at ~300nm corresponds to n→π* transition of –N=N – and the peak (λ_3) ~272 nm is due to π→π* transition of –C=N- groups [3, 19, 24]. This study is concerned with λ_{max_1} values of formazans, since it is specific to formazans. The investigation of UV-Vis spectral data tabulated in **Table 4** shows that the λ_{max_1} values specific to formazans, TPF and bis-formazans (CSPF) are observed at 483 nm and 352 nm respectively. There is a shift of $\Delta\lambda_{max_1}$ =131 nm towards to shorter wavelengths (hypsochromic effect). This is in accordance with the fact that CSPF contains compared with TPF electron withdrawing groups such as –COOH and –SO_3H in its structure (Figure 1b). In compounds **3-5** there are OH groups attached to 3,3-phenyl rings in addition to electron withdrawing –COOH and –SO_3H groups in compound **2**. The λ_{max_1} values were observed at 356, 353 and 354 nm as a result of the attachment of OH groups to *o-*, *m-* and *p-* position of 3,3'-phenyl rings (compound **3-5**). This shows that the $\Delta\lambda_{max}$ values shifted by 4, 1 and 2 nm towards the longer wavelengths compared with CSPF (bathochromic effect). This is explained by the electron donating nature of OH groups. The most distinctive shift was observed at *o-* position ($\Delta\lambda_{max_1}$ = 4 nm). That is because OH group attached at *o-*position is the closest to the reaction site. The inductive effect is the most intensive in this position and there is also resonance effect, which are in the same direction making a cumulative influence. Therefore the effect is expected to be maximum. There is no resonance effect at *m-* position and the inductive effect is highly diminished. That was why there was a small shift such as $\Delta\lambda_{max_1}$ =1 nm in this position. In *p*-position the inductive effect vanishes and -OH acts as an only electron donating group with resonance effect. Since it is far away from the reaction site compared with *o*-position the shift is lower than *o*-position but higher than *m*-position where there is no resonance effect, ($\Delta\lambda_{max_1}$ =2 nm).

Table 4. UV-Visible absorption λ_{max} values of formazans **1-5** (CH$_3$OH, 10^{-4} molL^{-1})

Comp.	λ_{max1}(nm) (Abs)	λ_{max2}(nm) (Abs)	$\Delta\lambda_{max1}$ according to TPF (nm)	$\Delta\lambda_{max1}$ according to CSPF (nm
1	483 (0.370)	335 (0.614)		
2	352 (0.912)	233 (0.565)	131	
3	356 (1.590)	218 (1.714)	127	4
4	353 (0.912)	218 (1.714)	130	1
5	354 (1.222)	219 (1.282)	129	2

Column 4: $\Delta\lambda_{max1}= \lambda_{max_1}$ (TPF) - λ_{max_1} (substituted formazans)

Column 5: $\Delta\lambda_{max1}= \lambda_{max_1}$ (CSPF) - λ_{max_1} (HCSPF)

Another important point was the λ_{max1} values of bis-formazans **2-5** relative to TPF. λ_{max1} values of compounds **2-5** were observed at 352, 356, 353, and 354 nm respectively. There was a shift towards a relatively short wavelength (hypsochromic effect) due to the fact that bis-formazans contain highly electron withdrawing groups. The fact that there were shifts of 131,127, 130, 129 nm for substituted formazans **2-5** relative to TPF is in good accordance with the structure of these compounds (Figure 1). These shifts changed slightly to the opposite direction upon attachment of –OH groups to the molecule. The UV spectra of the compounds are given in Figure 2 comparative manner.
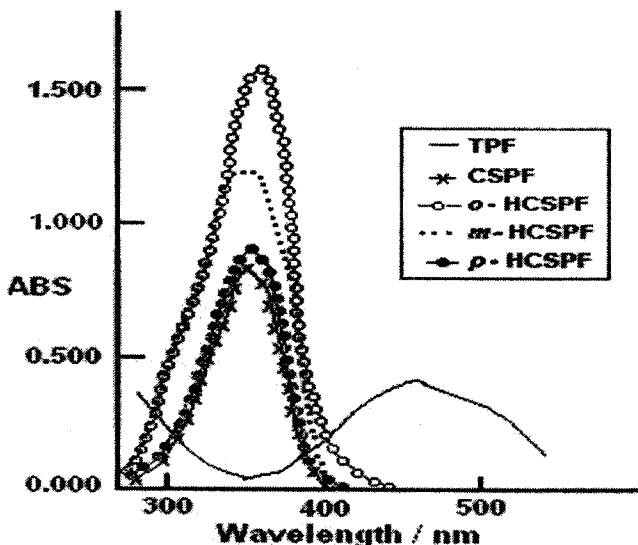
Figure 2. UV-Vis spectra of formazans (CH_3OH, 10^{-4} mol/l)

3.CONCLUSION

Formazans are colored compound ranging from red to orange or blue depending upon their structures. However they did not have their deserved place in dying industry due to their relatively high cost. They are used in medicine such in testing the vitality, determination of the cancerous tissue and selection of cancer drugs due to their specific color features. The most widely used formazans in medicine for this purpose is BTT [15, 16]. However this compound is ten times more toxic than the normal formazans. It is hoped that the formazans synthesized in this study are less toxic and more suitable for medical applications. However this is a subject of another medical study. As seen the application of these compounds is based upon their color features. That is why this study is mainly focused on λ_{max} values of various formazans and the substituents which effect the λ_{max} values.

Formazans are polydentate ligands with donor atoms and they are used for analytical purposes for forming complexes with trace metals. It also hoped that the formazans synthesized in this study are suitable for these applications. Contrary to other formazans our compounds are soluble in water.

4. EXPERIMENTAL

4.1 General

The H-NMR spectra of the formazans synthesized were taken with BRUKER 400 MHz ^{1}H-NMR spectrometer in 10^{-4} M CDCl$_3$. The IR spectra were recorded with the use of MATTSON 100 FT-IR spectrometer, using KBr discs in the range of 4000-400 cm^{-1}. The UV-Vis spectra were obtained with UNICAM UV2-100 UV-visible spectrometer in 1 cm quartz cell, in 10^{-4} M CH$_3$OH using 325 nm UV lamb. The elemental analysis were carried out with CHNS-932 LECO and the MASS spectra were taken micro mass UK Platform-II device at 70 eV electron impact mode.

4.2 General Synthesis

Synthesis of the formazans were carried out starting from benzaldehyde (or substituted benzaldehydes), hydrazine (or substituted hydrazines) and aniline (or substituted anilines). The condensation reaction of benzadehyde (or substituted benzaldehyde) and related substituted phenyl hydrazine was carried out with the usual way to obtain hydrazones. The hydrazones (substituted hydrazones) thus formed were coupled with the substituted diazonium cation obtained from substituted aniline derivatives to yield formazans.

4.2.1 Synthesis of 1,3,5 –Triphenylformazan, _1_

The condensation reaction was carried out by 40 ml methanolic solutions of benzaldehyde (2.12 g, 0.02mol) and phenylhydrazine (2.16g, 0.02 mol), adjusting the pH at 5-6 to give light yellow colored benzaldehyde phenlylhydrazone. The product was recrystallized from methanol. It was dissolved in 75 ml methanol under reflux. A basic buffer solution prepared with NaOH (2.50 g), CH$_3$COOH (3.50 g) and 200 ml methanol was added to the above solution. At the other side benzendiazonium chloride solution was prepared by the usual way with aniline (1.86 g, 0.02 mol), concentrated HCl (5 ml), NaNO$_2$ (1.40 g), at -5 to 0^{0}C. The benzendiazonium chloride solution was added to the benzaldehyde phenlylhydrazone solution which was cooled down to -5 to 0^{0}C, in drop wise manner with constant stirring. Stirring was continued for two hours at the same temperature resulting the formation of a bright red colored 1,3,5-triphenylfromazan. The solution was kept in the fridge for two days and the resulting product was recrystallized from methanol+dioxan.

4.2.2. Synthesis of 1,4-Bis-[3,3'-phenyl-5,5'-(2-carboxyphenyl) –formaz-1-yl]-benzen-2-sulphonic acid (CSPF, 2)

Benzaldehyde (2.12 g, 2.04 ml, 0.02 mol) was diluted with methanol (40ml). 2-Hydrazynobenzoic acid (3.76 g, 0.02mol) was dissolved in a mixture of methanol (80 ml) and water (20 ml). 2-Hydrazynobenzoic acid solution was added to the benzaldehyde solution with constant stirring in drop wise fashion adjusting the pH value to 5-6. A light yellow colored benzaldehyde-2-carboxyphenyl hydrazone immediately precipitated out. The product is recrystallized from methanol water-mixture. Hydrazone was dissolved in methanol (75 ml) water (25 ml) mixture under reflux. The basic buffer solution was prepared with by dissolving NaOH (2.50 g) and sodium acetate (3.50 g) in 200 ml methanol under reflux and added to hydrazone solution as prepared above. The mixture was kept until it cooled down to room temperature. This basic buffer hydrazone mixture was cooled down to 0^0C for coupling reaction (stock solution).

At the other side a 2-sulphobenzen-1,4-di- diazonium chloride solution was prepared with 2,5-diaminobenzen sulphonic acid (1.88 g , 0.01mol) concentrated HCl (5ml) NaNO$_2$ (1.40 g ,0.02 mol) in usual way at -5 to 0^0C . This diazonium solution was added in drop wise manner to basic buffer hydrazone solution as prepared above and cooled down to 0^0C, in ice bath at constant stirring for the coupling reaction. Care was taken for the temperature not exceeds -5 to 0^0C. The reaction was completed in 110 minutes. The pH 6 was adjusted to approximately by the addition of HCl for the precipitation of the product 2. The mixture was stirred for two more hours at the same temperature. The mixture was kept in the fridge at the same temperature for five days. The salmon colored formazan **2** precipitated out. The product was recrystallized from methanol+water+dioxan mixture.
Elemental Analysis for C$_{34}$H$_{26}$O$_7$N$_8$S, M: 690.30. Mass m/z (eV): 690.12 (M+, 0.51%), 658(2.21%), 599(5.27 %), 520(11.48 %), 300(47.25 %), 255(78.89 %), 240(67.33 %), 188(22.46 %).

4.2.3 Synthesis of 1,4-Bis-[3,3'-(o-hydroxyphenyl)-5,5'-(2-carboxyphenyl) –formaz-1-yl]-benzen-2-sulphonic acid (o-HCSPF, 3)

o-Hydroxybenzaldehyde (2.44g, 0.02 mol) was diluted with methanol (45 ml). 2 Hydrazynobenzoic acid (3.76 g, 0.02 mol) was dissolved in a mixture of methanol (80 ml) water (20 ml) under reflux . Then o-hydroxybenzaldehye solution was added to 2 -hydrazynobenzoic acid in dropwise manner with constant stirring keeping the PH at 5-6 and the temperature at 25^0C. The color changed from light yellow to dark yellow and the product of 2-hydroxybenzaldehye-2-carboxyphenylhydrazone was precipitated out. The product was kept at room temperature for two days, recrystallized from methano+water-mixture. It was then dissolved in 85 ml methanol and 35 ml water mixture under reflux and constant stirring. It was cooled down to room temperature. The basic buffer solution was prepared NaOH (2.50 g) and sodium acetate (3.50 g) in 200 ml methanol under

reflux and added the hydrazone solution as prepared above. The mixture was cooled down to 0^0C and kept ready for the coupling reaction (stock solution)

At the other side a 2-sulphobenzen-1,4-di- diazonium chloride solution was prepared with 2,5-diaminobenzen sulphonic acid (1.88 g, 0.01mol) concentrated HCl (5ml), $NaNO_2$ (1.40 g, 0.02 mol) in usual way at -5 to 0^0C. This diazonium solution was added in dropwise manner to basic buffer hydrazone solution, prepared as above and cooled down to 0^0C, in ice bath at constant stirring for the coupling reaction. The reaction was carried out in salt-ice bath. Ice cubes were added to the medium in order to ensure that the temperature does not exceed 0^0C. The reaction was completed in 1.5 hours. Cyclamen colored formazan **3** formed. The pH 5-6 was adjusted to approximately by the addition of HCl for the precipitation of the product **3**. The mixture was kept in the fridge for 5 days and the resulting product was re crystallized from methanol+dioxin+water mixture. Elemental Analysis for, $C_{34}H_{26}O_9N_8S$, M: 722.15. Mass: m/z(eV): 722.01 (M+, 3.27%), 391.22 (4.27%), 258.42 (0.12%), 256.11 (7.62%), 238.13 (9.06%), 182.17 (23.85%), 181.11 (96.94%), 165.13 (21.18%), 154.19 (13.27 %), 153.19 (24.74 %), 152.12(22.45%), 134.05 (7.11%).

4.2.4.Synthesis of 1,4-Bis-[3,3'-(m-hydroxyphenyl)-5,5'-(2-carboxyphenyl)–formaz-1-yl]- benzen-2-sulphonic acid (m-HCSPF, 4)

The reaction was the same as in the synthesis of *o-HCSPF, 3* by the use of *m*-hydroxybenzaldehyde (2.44g, 0.02mol). However the resulting product was gum-like resinous and oily polymeric mass .The experiment was repeated several times in very dilute media and varied pH without any success. There was not a definite melting point (mp >300^0C)

4.2.5 Synthesis of 1,4-Bis-[3,3'-(p-hydroxyphenyl)-5,5'-(2-carboxyphenyl) – formaz-1-yl]-benzen-2-sulphonic acid (p-HCSPF, 5)

The reaction was the same as in the synthesis of *o-HCSPF 3,* by the use of p-hydroxybenzaldehyde (2.44g, 0.02 mol) resulting in red-brown colored formazan.

ÖZET

Bu çalışmada 1,4-bis–[3,3'-fenil-5,5'-(2-karboksifenil)-formaz-1-il]-benzen-2-sulfonik asit (2,CSPF) ve onun 3-fenil halkasının *o-*, *m-* *ve* *p-* köşelerine –OH grubunun bağlı olduğu türevleri sentezlendi. Elde edilen formazanların yapıları, elementel analiz, MASS spektroskopisi, IR, ^{1}H-NMR and UV-Vis spekrumları ile tayin edildi. Ayrıca UV-Vis spektrumları üzerine substituent etkileri incelendi. λ_{max} değerlerinin incelenmesi ile TPF ye göre CSPF'nin λ_{max} değerlerinde –COOH ve –SO$_3$H gibi electron çekici grupların bağlı olmasına uygun olarak 130 nm kadar hipsokromik etki görüldü. CSPF'nin 3-fenil halkasına electron salıcı grup olan –OH grubunun, *o-*, *m-* *ve* *p-* köşelerine bağlanması ile de –OH'ın electron salıcı olmasına uygun olarak küçük bir batokromik etki gözlendi.

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