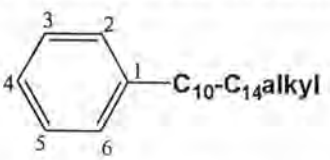
CHAPTER IV
RESULTS AND DISCUSSION

4.1 Synthesis of *p*-nitro linear alkylbenzene and *p*-linear alkylaniline

From the literature search, it was obviously revealed that azo dyes could be used as markers, which were simply detected by extraction with acidic or basic reagents. In recent years, there has been reported on various raw materials used for synthesizing this class of markers. In this research, two important steps to synthesize raw material, *p*-linear alkylaniline, were employed. Step 1 was the nitration of linear alkylbenzene to give *p*-nitro linear alkylbenzene. The other step involved the reduction of *p*-nitro linear alkylbenzene by tin powder in hydrochloric to give *p*-linear alkylaniline.

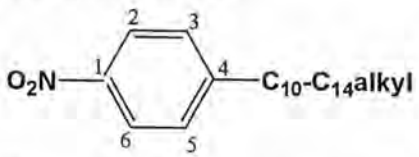
Linear alkylbenzene is a colorless liquid, insoluble in water but freely soluble in organic solvents and alcohols. Its structure is shown below:



The IR spectrum of linear alkylbenzene showed absorption peaks of =C-H stretching of aromatic at 3028 cm^{-1} (w), C-H stretching of aliphatic at 2925 cm^{-1} (s), C=C ring stretching of aromatic at 1604 cm^{-1} (m) and 1495 cm^{-1} (m), C-H stretching of methylene group at 1456 cm^{-1} (m), C-H stretching of methyl group at 1377 cm^{-1} (m), and =C-H out of plane bending of aromatic at 900-690 cm^{-1}(w). In addition, the ^{1}H-NMR spectrum of alkyl and aromatic protons exhibited in the range of 0.75-2.80 ppm and 7.15-7.40 ppm, respectively as the multiplet signals. The ^{13}C-NMR spectrum of linear alkylbenzene revealed the presence of alkyl carbons at 14.0-48.0 ppm, C-1 at 146.5 ppm, C-4 at 125.7 ppm, C-2, C-3, C-5, and C-6 at 128.3 ppm. The mass spectrum of linear alkylbenzene exhibited at 218, 232, 246, 260, and 274 Da/e, which were molecular weights of

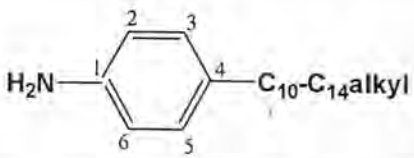
decylbenzene, undecylbenzene, dodecylbenzene, tridecylbenzene, and tetradecylbenzene, respectively.

p-Nitro linear alkylbenzene is a yellow liquid, insoluble in water, freely soluble in organic solvents. Its structure was shown below.



The IR spectrum of *p*-nitro linear alkylbenzene revealed absorption peaks of =C-H stretching of aromatic at 3084 cm^{-1} (w), C-H stretching of aliphatic at 2927 cm^{-1} (s), C=C ring stretching of aromatic at 1604 cm^{-1} (m), and 1466 cm^{-1} (m), N=O asymmetric stretching of nitro group at 1522 cm^{-1} (s), and N=O symmetric stretching of nitro group at 1346 cm^{-1} (s). Moreover, the ^{1}H-NMR spectrum of *p*-nitro linear alkylbenzene showed the aromatic protons at positions 3 and 5 at 7.25 ppm as a doublet with a coupling constant of 8.77 Hz, the aromatic protons at positions 2 and 6 at 8.10 ppm as a doublet with a coupling constant of 8.77 Hz. The ^{13}C-NMR spectrum of *p*-nitro linear alkylbenzene revealed the presence of C-NO_2 at 154.6 ppm, C-4 at 146.5 ppm, C-3 and C-5 at 128.4 ppm, C-2 and C-6 at 123.5 ppm.

p-Linear alkylaniline is a colorless liquid, insoluble in water, freely soluble in organic solvents and alcohols. Since *p*-linear alkylaniline could be oxidized at room temperature, it should be kept in a refrigerator to prevent the oxidation of aniline. Its structure is shown below:



The IR spectrum of *p*-linear alkylaniline revealed absorption peaks of N-H stretching of primary amine at 3467 cm^{-1} (w) and 3384 cm^{-1} (w), =C-H stretching of aromatic at 3026 cm^{-1} (w), C-H stretching of aliphatic at 2925 cm^{-1} (s), N-H bending of primary amine at 1622 cm^{-1} (m), C=C ring stretching of aromatic at 1606 cm^{-1} (w), 1516 cm^{-1} (w) and 1466 cm^{-1} (w), C-H stretching of methylene group at 1458 cm^{-1} (m), C-H stretching of methyl

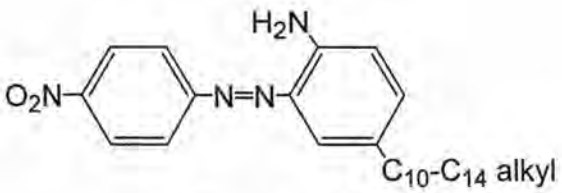
group at 1375 cm^{-1} (w), and C-N stretching of primary aromatic amine at 1273 cm^{-1}(w). The ^{1}H-NMR spectrum of *p*-linear alkylaniline showed the aromatic protons at positions 2 and 6 at 6.62 ppm as a doublet with a coupling constant of 8.32 Hz, the aromatic protons at positions 3 and 5 at 6.94 ppm as a doublet with a coupling constant of 8.32 Hz. The ^{13}C-NMR spectrum of *p*-nitro linear alkylbenzene revealed the presence of C-NH_2 at 144.0 ppm, C-3 and C-5 at 128.4 ppm, C-4 at 127.7 ppm, C-2 and C-6 at 115.3 ppm.

4.2 Syntheses of azo dyes

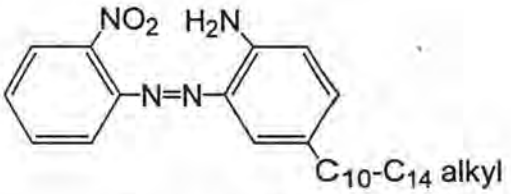
Twenty azo dyes were synthesized by two methods in chapter III and could be categorized into three groups. There were seven phenyl azo anilines (compounds **1a-7a**), which were synthesized by coupling between diazonium salt of aniline derivatives with *p*-linear alkylaniline, seven phenyl azo anilines (compounds **1b-7b**), which were synthesized by coupling between aniline derivatives with diazonium salt of *p*-linear alkylaniline, and six phenyl azo phenols (compounds **1c-6c**), which were synthesized by coupling between phenol derivatives with diazonium salt of *p*-linear alkylaniline. All compounds were fully characterized by spectroscopic evidences including IR, ^{1}H-NMR, and ^{13}C-NMR. Moreover, seven synthesized azo dyes, which gave intense coloration, were characterized to obtain the molecular weight by mass spectroscopic method. The physical properties of the synthesized azo dyes are shown in Table 4.1 and their structures are shown in Figures 4.1 to 4.3.

Table 4.1 The physical properties and %yields of synthesized compounds

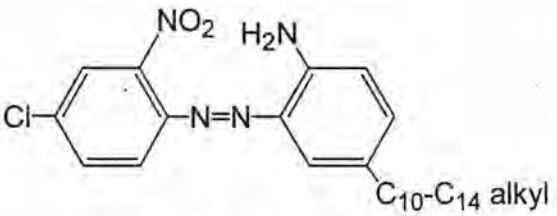
Group of compounds	Compounds	Physical properties (Appearance)	%Yield
Phenyl azo anilines (Group a)	1a	Yellowish brown oil	61
	2a	Yellowish brown oil	69
	3a	Yellowish brown oil	75
	4a	Reddish brown oil	78
	5a	Yellowish brown oil	58
	6a	Yellowish brown oil	73
	7a	Yellowish brown oil	75
Phenyl azo anilines (Group b)	1b	Yellowish brown oil	59
	2b	Yellowish brown oil	67
	3b	Yellowish brown oil	68
	4b	Reddish brown oil	70
	5b	Yellowish brown oil	55
	6b	Yellowish brown oil	73
	7b	Yellowish brown oil	70
Phenyl azo phenols (Group c)	1c	Yellowish brown oil	79
	2c	Reddish brown oil	78
	3c	Reddish brown oil	81
	4c	Reddish brown oil	65
	5c	Reddish brown oil	73
	6c	Reddish brown oil	69



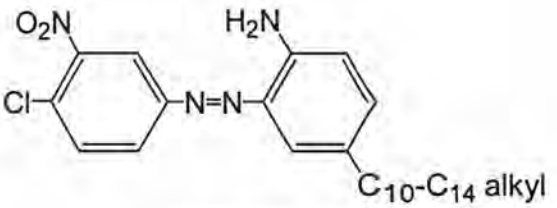
Compound 1a



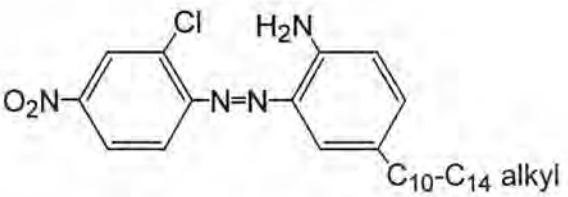
Compound 2a



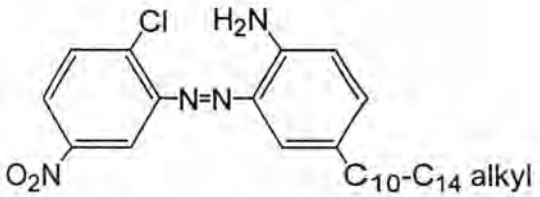
Compound 3a



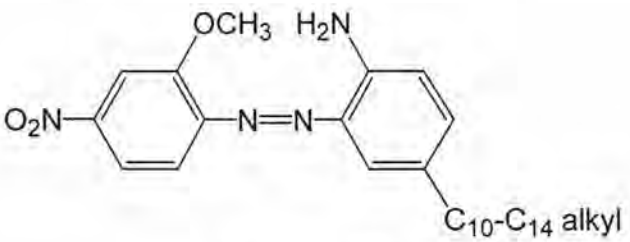
Compound 4a



Compound 5a

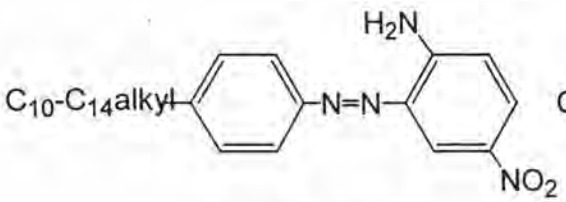


Compound 6a

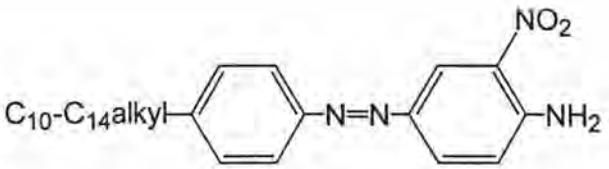


Compound 7a

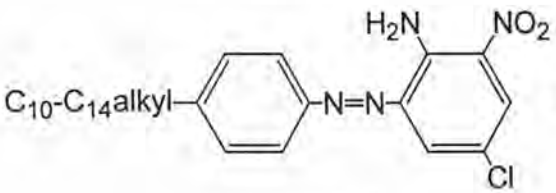
Figure 4.1 The structures of phenyl azo anilines (group a)



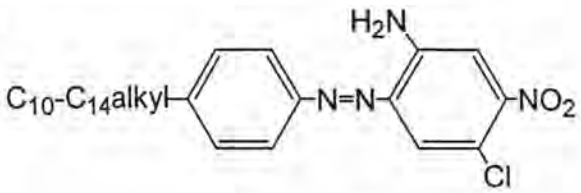
Compound 1b



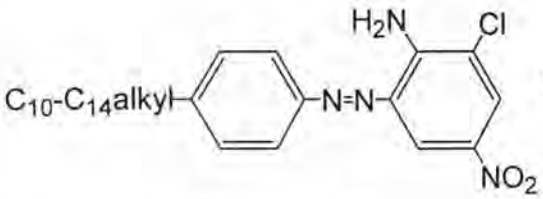
Compound 2b



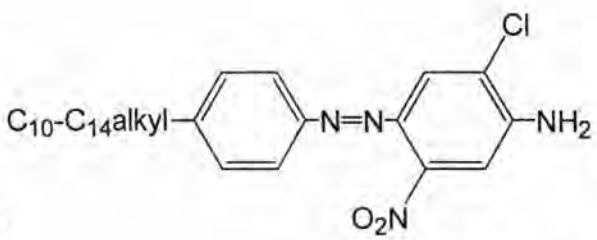
Compound 3b



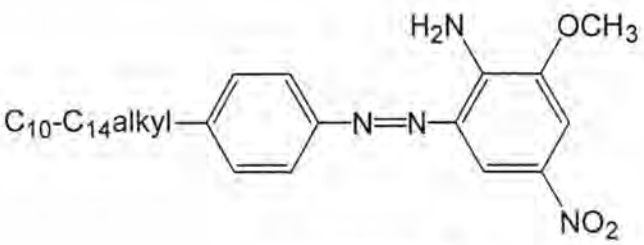
Compound 4b



Compound 5b

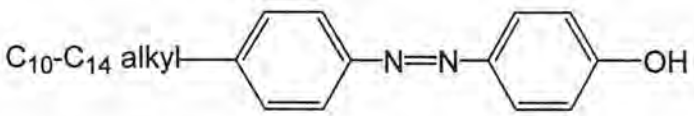


Compound 6b

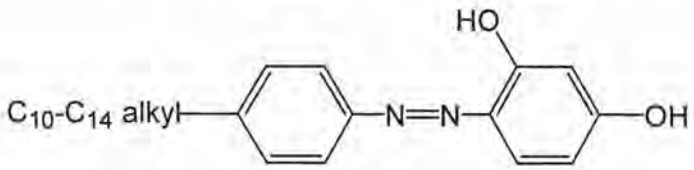


Compound 7b

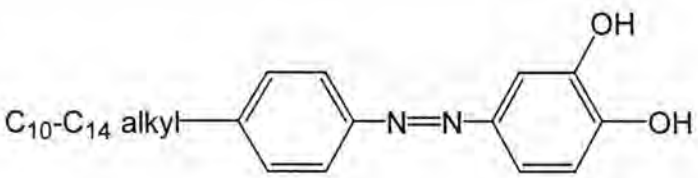
Figure 4.2 The structures of phenyl azo anilines (group b)



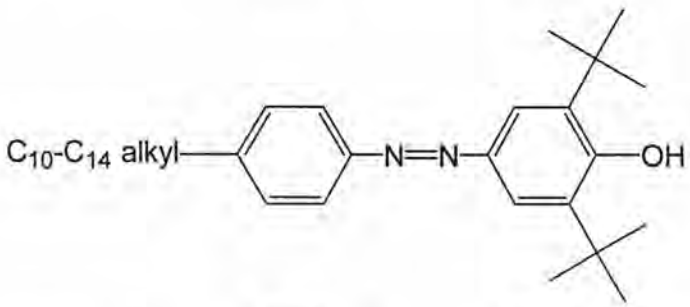
Compound 1c



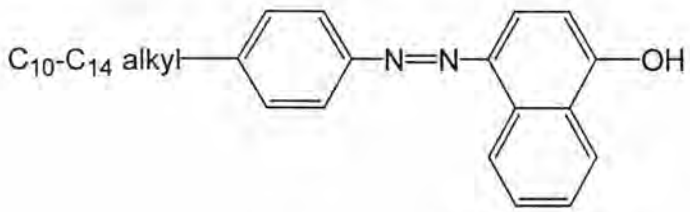
Compound 2c



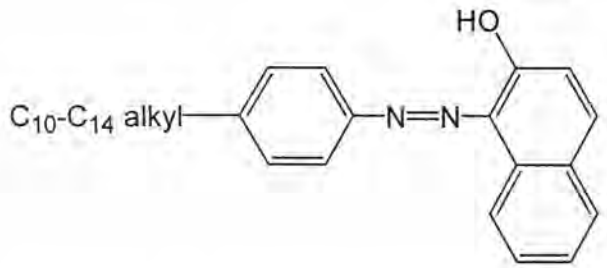
Compound 3c



Compound 4c



Compound 5c



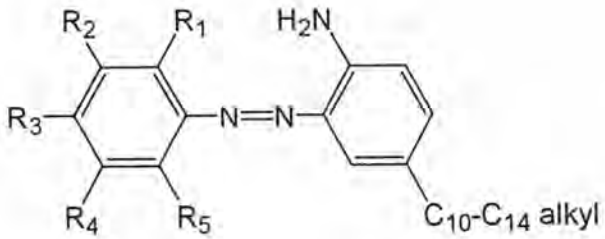
Compound 6c

Figure 4.3 The structures of phenyl azo phenols (group c)

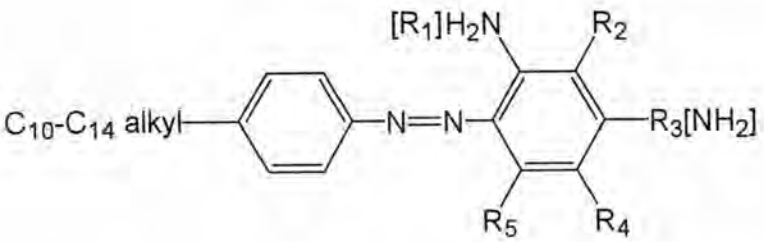
4.3 Spectroscopy of synthesized markers

4.3.1 Infrared Spectroscopy (IR)

Phenyl azo anilines (group a and group b)



Group a



Group b

The FT-IR spectra of phenyl azo anilines (group a and group b) generally revealed the absorption band of N-H stretching of primary amine at 3500-3300 cm^{-1}, C-H stretching of aromatic at 3100-3000 cm^{-1}, C-H stretching of aliphatic at 3000-2900 cm^{-1}, N-H bending of primary amine at 1640-1600 $cm^{-1}$and C=C ring stretching of aromatic at 1605-1460 cm^{-1}. It also revealed N=O asymmetric stretching of nitro group at 1550-1500 cm^{-1}, N=O symmetric stretching of nitro group at 1350-1300 cm^{-1}, C-N stretching of aromatic amine at 1320-1250 cm^{-1}, C-Cl stretching of aryl chloride at 1130-1045 cm^{-1}, C-O asymmetric stretching of aryl alkyl ether at 1248 cm^{-1} and C-O symmetric stretching of aryl alkyl ether at 1038 cm^{-1}. The FT-IR absorption bands assignment of phenyl azo anilines (group a and group b) are tabulated in Tables 4.2 and 4.3, respectively.

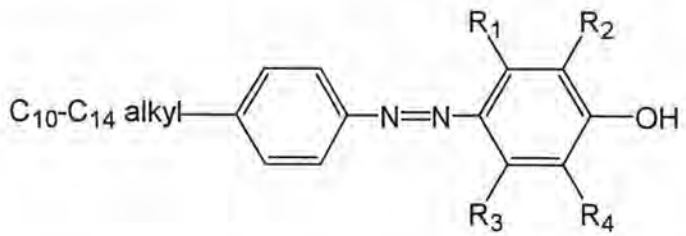
Table 4.2 The FT-IR absorption bands assignment of phenyl azo anilines (group a)

Cpd	Wave number (cm^{-1})										
	N-H		C-H		C=C	N=O		C-N	C-O		C-Cl
	stretching	bending	aromatic	aliphatic	ring	asym.	sym.		asym.	sym.	
1a	3481(w) 3361(w)	1631(m)	3028(w)	2925(s)	1604(m) 1462(m)	1523(s)	1346(s)	1255(w)	-	-	-
2a	3494(m) 3383(m)	1626(m)	3026(w)	2925(s)	1574(m) 1466(m)	1512(m)	1346(m)	1259(m)	-	-	-
3a	3467(w) 3381(w)	1628(m)	3026(w)	2925(s)	1603(m) 1466(m)	1523(m)	1338(m)	1250(m)	-	-	1130(w)
4a	3406(w) 3331(w)	1628(m)	3026(w)	2925(s)	1603(m) 1466(m)	1533(m)	1346(m)	1313(m)	-	-	1130(w)
5a	3492(w) 3377(w)	1624(m)	3026(w)	2932(s)	1590(m) 1466(m)	1525(m)	1336(m)	1279(m)	-	-	1028(w)
6a	3427(w) 3334(w)	1630(m)	3026(w)	2925(s)	1603(m) 1464(m)	1523(m)	1346(m)	1282(m)	-	-	1045(w)
7a	3469(w) 3388(w)	1622(m)	3026(w)	2925(s)	1601(m) 1464(m)	1520(m)	1344(m)	1296(m)	1248(m)	1038(m)	-

Table 4.3 The FT-IR absorption bands assignment of phenyl azo anilines (group b)

Cpd	Wave number (cm^{-1})										
	N-H		C-H		C=C	N=O		C-N	C-O		C-Cl
	stretching	bending	aromatic	aliphatic	ring	asym.	sym.		asym.	sym.	
1b	3458(w) 3356(w)	1637(m)	3028(w)	2925(s)	1600(m) 1464(m)	1531(s)	1348(s)	1250(w)	-	-	-
2b	3499(w) 3386(w)	1627(m)	3029(w)	2920(s)	1603(m) 1466(m)	1514(m)	1344(m)	1259(m)	-	-	-
3b	3492(w) 3383(w)	1628(m)	3026(w)	2922(s)	1603(m) 1466(m)	1520(m)	1338(m)	1250(m)	-	-	1093(w)
4b	3469(w) 3367(w)	1630(m)	3026(w)	2925(s)	1601(m) 1464(m)	1537(m)	1350(m)	1311(m)	-	-	1130(w)
5b	3492(w) 3388(w)	1618(m)	3026(w)	2925(s)	1589(m) 1466(m)	1522(m)	1325(m)	1279(m)	-	-	1022(w)
6b	3473(w) 3354(w)	1628(m)	3026(w)	2925(s)	1603(m) 1464(m)	1527(m)	1346(m)	1250(m)	-	-	1113(w)
7b	3475(w) 3355(w)	1628(m)	3020(w)	2925(s)	1585(m) 1460(m)	1522(m)	1342(m)	1296(m)	1246(m)	1039(m)	-

Phenyl azo phenols (group c)



The FT-IR spectra of phenyl azo phenols (group c) generally revealed the absorption band of O-H stretching of phenol at 3600-3200 cm^{-1}, C-H stretching of aromatic at 3100-3000 cm^{-1}, C-H stretching of aliphatic at 3000-2900 cm^{-1}, C=C ring stretching of aromatic at 1650-1450 cm^{-1}, O-H bending of phenol at 1410-1350 cm^{-1}, and C-O stretching of phenol at 1250-1200 cm^{-1}. The FT-IR absorption bands assignment of phenyl azo phenols (group c) are tabulated in Table 4.4.

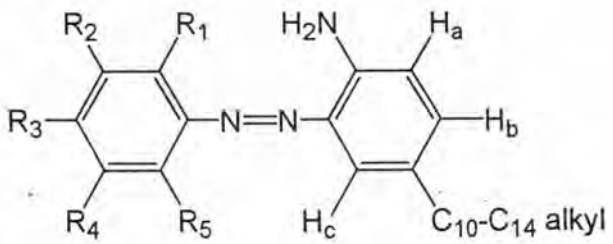
Table 4.4 The FT-IR absorption bands assignment of phenyl azo phenols (group c)

Cpd	Wave number (cm^{-1})					
	O-H stretching	=C-H aromatic	C-H aliphatic	C=C ring	O-H bending	C-O stretching
1c	3600-3200(br)	3026(w)	2924(s)	1589(s), 1466(s)	1377(w)	1248(s)
2c	3600-3200(br)	3041(w)	2925(s)	1604(s), 1469(s)	1406(w)	1248(m)
3c	3600-3200(br)	3028(w)	2925(s)	1604 m), 1454(m)	1389(w)	1250(m)
4c	3643(m) and 3600-3200(br)	3026(w)	2925(s)	1603(m), 1483(w) 1466(m)	1363(w)	1230(w)
5c	3600-3200(br)	3060(w)	2924(s)	1624(m), 1599(m), 1549(m), 1452(m)	1358(w)	1265(w)
6c	3600-3200(br)	3028(w)	2925(s)	1626(s), 1601(s) 1510(s), 1454(s)	1379(w)	1271(m)

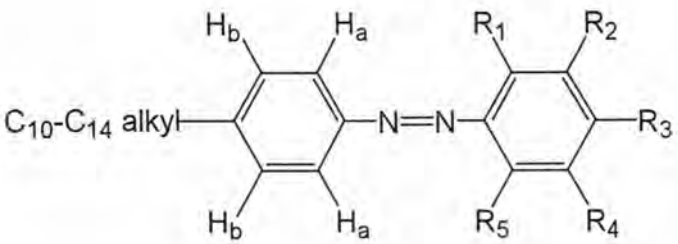
4.3.2 Nuclear Magnetic Resonance Spectroscopy (NMR)

4.3.2.1 ^{1}H-NMR

Phenyl azo anilines (group a and group b)



Group a



Group b

The ^{1}H-NMR spectra of phenyl azo anilines normally revealed 2H of Ar-NH_2 around 4.35 – 7.13 ppm as a broad singlet signal except compound **4b** that showed this signal at 8.64 ppm. The aromatic protons were exhibited as singlet, doublet, and multiplet with various coupling constants around 6.60 - 8.57 ppm. In addition, the chemical shift of alkyl protons adjacent to the aromatic appeared in the range of 0.70-2.90 ppm. The ^{1}H-NMR spectral assignment of phenyl azo anilines (group a and group b) are presented in Tables 4.5 and 4.6, respectively.

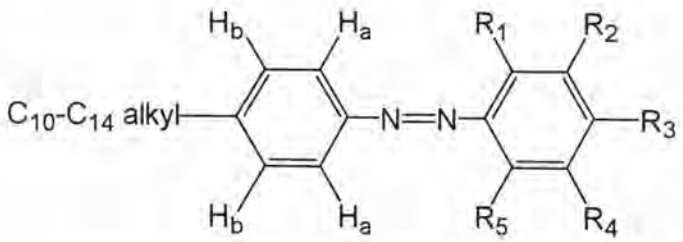
Table 4.5 The ^{1}H-NMR spectral assignment of phenyl azo anilines (group a)

Cpd	Substituents					Chemical shift, δ(ppm)								
							H of R					H_a	H_b	H_c
	R_1	R_2	R_3	R_4	R_5	-N<u>H</u>$_2$	R_1	R_2	R_3	R_4	R_5			
1a	H	H	NO_2	H	H	5.27 (*s,br*)	8.06(*d*, J=8.19 Hz	8.29(*d*, J=8.19 Hz)	-	8.29(*d*, J=8.19 Hz)	8.06(*d*, J=8.19 Hz)	6.66(*d*, J=8.29 Hz)	7.09(*d*, J=8.29 Hz)	7.58 (*s*)
2a	NO_2	H	H	H	H	6.10 (*s, br*)	-	8.09 (*td*, J=1.50 8.45 Hz)	7.88 (*td*, J=1.15 8.45 Hz)	7.38 (*td*, J=1.15 8.45 Hz)	8.09 (*td*, J=1.50 8.45 Hz)	6.62(*d*, J=8.70 Hz)	6.79 (*dd*, J=1.44 8.70 Hz)	7.69(*d*, J=1.44 Hz)
3a	NO_2	H	Cl	H	H	5.25 (*s, br*)	-	8.07 (*m*)	-	8.07 (*m*)	8.07 (*m*)	6.82(*d*, J=8.50 Hz)	7.02(*d*, J=8.50 Hz)	7.69 (*s*)
4a	H	NO_2	Cl	H	H	5.46 (*s, br*)	8.42 (*s*)	-	-	7.87(*d*, J=8.54 Hz)	8.12(*d*, J=8.54 Hz)	6.85(*d*, J=8.40 Hz)	7.09(*d*, J=8.40 Hz)	7.61 (*s*)
5a	Cl	H	NO_2	H	H	5.00 (*s, br*)	-	8.25 (*s*)	-	8.18(*d*, J=8.27 Hz)	8.26(*d*, J=8.27 Hz)	6.74(*d*, J=8.40 Hz)	7.19(*d*, J=8.40 Hz)	7.64(*d*, J=1.99 Hz)
6a	Cl	H	H	NO_2	H	5.48 (*s, br*)	-	7.54(*d*, J=8.74 Hz)	8.38(*d*, J=8.74 Hz)	-	8.57 (*s*)	6.84(*d*, J=8.38 Hz)	7.16(*d*, J=8.38 Hz)	7.68(*d*, J=1.82 Hz)
7a	OCH_3	H	NO_2	H	H	5.01 (*s, br*)	3.89(*s*)	7.64(*d*, J=2.26 Hz)	-	7.25 (*m*)	7.75 (*m*)	6.60(*d*, J=8.30 Hz)	6.71(*d*, J=8.30 Hz)	7.59 (*s*)

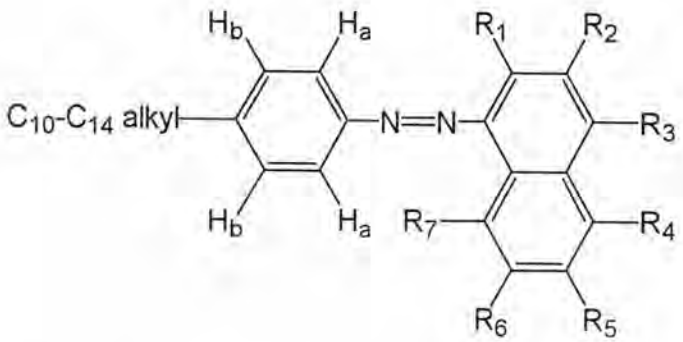
Table 4.6 The ^{1}H-NMR spectral assignment of phenyl azo anilines (group b)

Cpd	Substituents					Chemical shift, δ(ppm)						
						H of R					H_a	H_b
	R_1	R_2	R_3	R_4	R_5	R_1	R_2	R_3	R_4	R_5		
1b	NH_2	H	H	NO_2	H	5.44 (*s, br*)	7.14(*d*, J=9.00 Hz)	8.29(*d*, J=9.00 Hz)	-	8.62(*s*)	7.55(*d*, J=8.02 Hz)	6.60(*d*, J=8.02 Hz)
2b	H	NO_2	NH_2	H	H	8.37(*d*, J=1.61 Hz)	-	-	6.74(*d*, J=8.44 Hz)	7.79(*dd*, J=1.05, 8.50 Hz)	7.61(*dd*, J=1.39, 7.59 Hz)	6.86(*dd*, J=1.39, 7.59 Hz)
3b	NH_2	NO_2	H	Cl	H	-	-	8.39 (*s*)	-	8.12 (*s*)	7.69(*d*, J=7.56 Hz)	6.90(*d*, J=7.56 Hz)
4b	NH_2	H	NO_2	Cl	H	8.64 (*s, br*)	7.95 (*s*)	-	-	8.11	7.65(*d*, J=7.60 Hz)	6.78(*d*, J=7.60 Hz)
5b	NH_2	Cl	H	NO_2	H	7.13 (*s, br*)	-	8.45 (*s*)	-	8.61 (*s*)	7.68 (*d*, J=7.16 Hz)	6.64 (*d*, J=7.16 Hz)
6b	H	Cl	NH_2	H	NO_2	8.10 (*s*)	-	7.03 (*s, br*)	7.89 (*s*)	-	7.51(*d*, J=7.46 Hz)	6.89(*d*, J=7.46 Hz)
7b	NH_2	OCH_3	H	NO_2	H	4.35 (*s, br*)	3.77 (*s*)	7.65(*d*, J=2.36 Hz)	-	8.31(*d*, J=2.36 Hz)	7.78(*dd*, J=8.67, 2.35 Hz)	6.61(*d*, J=8.67 Hz)

Phenyl azo phenols (group c)



Compounds **1c-4c**



Compounds **5c-6c**

The ^{1}H-NMR spectrum of Ar-OH generally showed a broad singlet signal around 5.12 – 6.70 ppm. The aromatic protons exhibited as singlet, doublet, and multiplet with various coupling constants around 6.47 - 8.18 ppm. The other signals, alkyl protons, were detected around 0.70-2.90 ppm. The ^{1}H-NMR spectral assignment of compounds **1c-4c** and compounds **5c-6c** are shown in Tables 4.7 and 4.8, respectively.

Table 4.7 The ^{1}H-NMR spectral assignment of phenyl azo phenols (group c); compounds **1c – 4c**

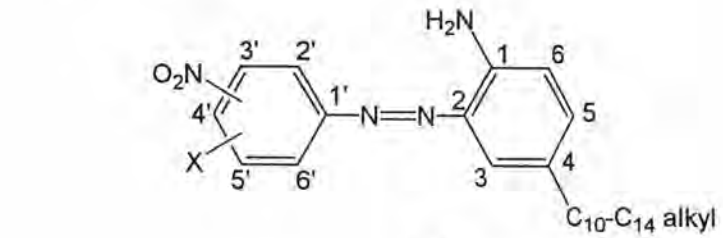
Cpd	Substituents					Chemical shift, δ(ppm)						
						H of R					H_a	H_b
	R_1	R_2	R_3	R_4	R_5	R_1	R_2	R_3	R_4	R_5		
1c	H	H	OH	H	H	7.84 (*dd*, J=8.43, 2.16 Hz)	7.10 (*dd*, J=2.16, 8.43 Hz)	5.27 (*s, br*)	7.10 (*dd*, J=2.16, 8.40 Hz)	7.84 (*dd*, J=2.16, 8.43 Hz)	7.66 (*d*, J=8.06 Hz)	6.92 (*d*, J=8.06 Hz)
2c	OH	H	OH	H	H	6.30 (*s, br*)	6.47 (*d*, J=2.40 Hz)	6.30 (*s, br*)	7.65 (*dd*, J=2.40, 8.72 Hz)	6.56 (*d*, J=8.72 Hz)	7.68 (*d*, J=8.36 Hz)	7.23 (*d*, J=8.36 Hz)
3c	H	OH	OH	H	H	7.12 (*s*)	5.12 (*s, br*)	5.12 (*s, br*)	7.02 (*d*, J=8.35 Hz)	7.83 (*d*, J=8.35 Hz)	7.71 (*dd*, J=1.38, 7.38 Hz)	6.89 (*dd*, J=1.38, 7.38 Hz)
4c	H	*t*-Bu	OH	*t*-Bu	H	7.82 (*s*)	1.57 (*s*)	5.31 (*s, br*)	1.57 (*s*)	7.82 (*s*)	7.70 (*d*, J=7.33 Hz)	6.94 (*d*, J=7.33 Hz)

Table 4.8 The ^{1}H-NMR spectral assignment of phenyl azo phenols (group c); compounds **5c – 6c**

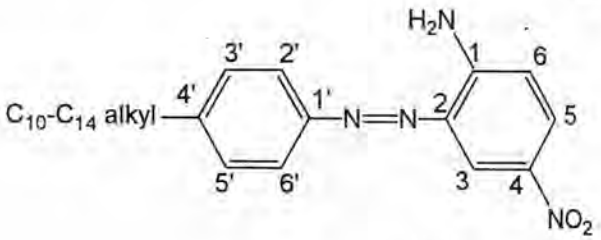
Cpd	Substituents							Chemical shift, δ(ppm)								
								H of R							H_a	H_b
	R_1	R_2	R_3	R_4	R_5	R_6	R_7	R_1	R_2	R_3	R_4	R_5	R_6	R_7		
5c	H	H	OH	H	H	H	H	8.18(*d*, J=7.80 Hz)	7.19 (*d*, J=7.80 Hz)	6.70 (*s, br*)	7.73 (*d*,J=7.58 Hz)	7.25 (*t*, J=7.43 Hz)	7.59 (*t*, J=7.43 Hz)	7.44 (*dd*, J=1.55, 7.30 Hz)	7.39(*d*, J=7.50 Hz)	6.83(*d*, J=7.50 Hz)
6c	OH	H	H	H	H	H	H	3.81 (*s, br*)	7.22(*d*, J=8.83 Hz)	7.71 (*m*)	7.71 (*m*)	7.51 (*t*, J=7.01 Hz)	7.71 (*m*)	7.58(*d*, J=8.21 Hz)	7.68(*d*, J=7.88 Hz)	6.92(*d*, J=7.88 Hz)

4.3.3.2 ^{13}C-NMR

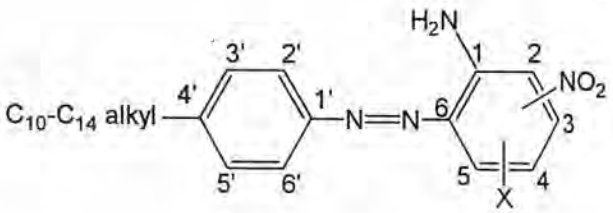
Phenyl azo anilines



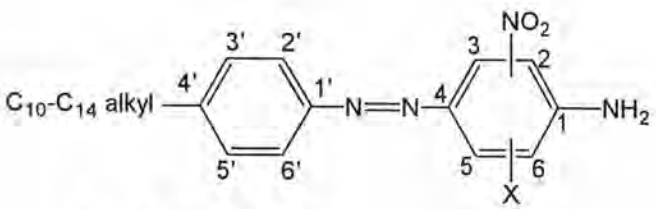
Group a



Compound **1b**



Compounds **3b, 4b, 5b** and **7b**



Compounds **2b,** and **6b**

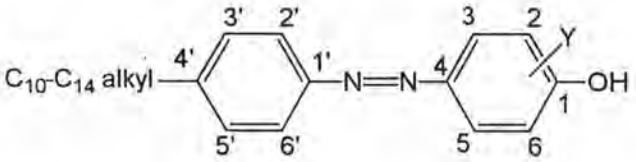
X = H, Cl, and OCH_3

The ^{13}C-NMR spectra of phenyl azo anilines (group a) revealed the signals of aromatic carbons around 111.1 - 158.3 ppm, a C-NH_2 showed around 138.1 – 150.8 ppm. The chemical shifts of alkyl and methoxy groups were observed around 11.0 - 47.9 ppm and at 55.8 ppm, respectively. The ^{13}C-NMR spectral assignment of phenyl azo anilines (group a) are tabulated in Table 4.9.

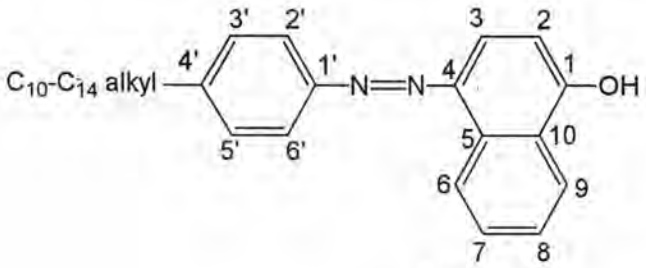
Table 4.9 The ^{13}C-NMR spectral assignment of phenyl azo anilines

Cpd	Chemical shift, δ(ppm)												
	C-1	C-2	C-3	C-4	C-5	C-6	C-1′	C-2′	C-3′	C-4′	C-5′	C-6′	OCH_3
1a	138.1	139.2	124.8	130.9	132.4	118.4	158.3	125.3	125.7	151.8	125.7	125.3	-
2a	138.0	138.8	122.6	128.8	130.5	116.9	147.8	143.0	123.0	131.0	135.7	122.8	-
3a	138.1	138.9	123.0	129.0	131.1	115.0	147.5	143.8	123.6	131.4	135.8	123.3	
4a	137.5	138.0	122.8	129.3	132.1	115.1	151.2	120.0	148.3	132.0	130.2	130.4	
5a	138.4	138.4	125.7	128.1	129.0	117.5	158.0	127.7	125.7	149.2	124.4	125.7	-
6a	138.3	139.7	125.1	129.8	131.2	113.0	154.1	135.9	130.2	127.8	147.2	121.0	-
7a	137.1	140.8	119.2	128.3	131.5	117.0	143.5	151.4	111.7	145.4	118.1	122.0	55.8
1b	147.1	141.0	119.0	138.4	126.9	116.6	149.5	123.5	128.5	142.5	128.5	123.5	-
2b	145.2	135.5	119.0	144.8	129.4	116.5	146.5	125.7	128.3	142.1	128.3	125.7	-
3b	134.0	135.7	127.0	125.0	131.3	144.5	154.3	121.0	127.7	144.8	127.7	121.0	-
4b	137.9	111.1	151.6	119.7	125.6	146.4	148.0	122.7	128.2	144.9	128.2	122.7	-
5b	146.4	122.5	127.0	138.0	117.6	138.6	149.0	124.3	128.4	138.8	128.4	124.3	-
6b	150.8	126.2	124.3	139.0	142.1	113.5	149.1	120.6	128.2	143.5	128.2	120.6	-
7b	131.7	148.9	111.5	137.2	110.6	140.3	148.9	122.0	128.3	140.8	128.3	122.0	55.8

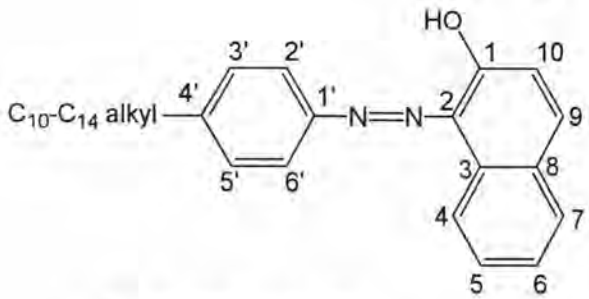
Phenyl azo phenols (group c)



Compounds **1c-4c**



Compounds **5c**



Compounds **6c**

Y= H, OH, and *t*-Bu

The ^{13}C-NMR spectra of phenyl azo phenols (group a) revealed the signals of aromatic carbons around 103.9 – 162.4 ppm, a C-OH showed around 147.0 – 162.4 ppm. The chemical shift of alkyl group was observed around 11.0 - 47.9 ppm. The ^{13}C-NMR spectral assignment of phenyl azo phenols are exhibited in Table 4.10 and Table 4.11.

Table 4.10 The ^{13}C-NMR spectral assignment of phenyl azo phenols (group c); compounds 1c-4c

Cpd	Chemical shift, δ(ppm)											
	C-1	C-2	C-3	C-4	C-5	C-6	C-1′	C-2′	C-3′	C-4′	C-5′	C-6′
1c	158.7	116.0	124.9	149.6	124.9	116.0	151.0	122.6	128.1	146.9	128.1	122.6
2c	162.4	103.9	158.4	132.8	126.0	109.6	149.7	121.1	128.9	147.3	128.9	121.1
3c	147.3	145.4	109.8	147.0	116.1	116.9	150.7	121.1	128.8	139.8	128.8	121.1
4c	154.0	135.7	119.8	114.0	119.8	135.7	150.6	124.7	127.3	139.6	128.3	124.7

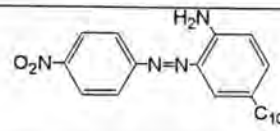
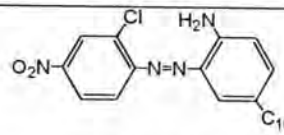
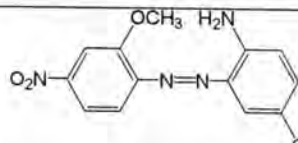
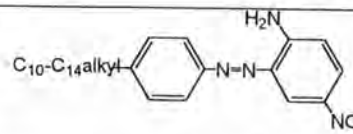
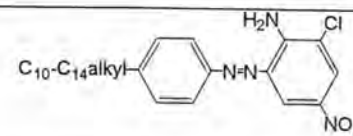
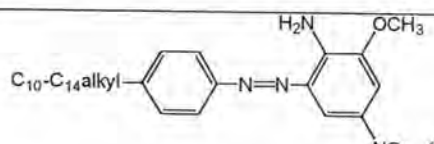
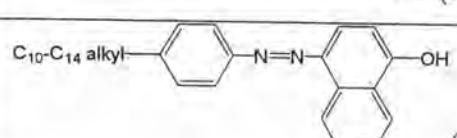
Table 4.11 The ^{13}C-NMR spectral assignment of phenyl azo phenols (group c); compounds 5c-6c

Cpd	Chemical shift, δ(ppm)											
	C-1	C-2	C-3	C-4	C-5	C-6	C-1′	C-2′	C-3′	C-4′	C-5′	C-6′
5c	154.8	109.7	121.7	143.2	129.6	128.0	147.0	124.2	128.7	139.5	128.7	124.2
	C-7	C-8	C-9	C-10								
	126.3	126.3	123.1	125.5								
6c	C-1	C-2	C-3	C-4	C-5	C-6	C-1′	C-2′	C-3′	C-4′	C-5′	C-6′
	147.0	135.0	128.0	126.5	126.3	124.3	150.5	123.1	127.8	143.1	127.8	123.1
	C-7	C-8	C-9	C-10								
	127.8	128.0	133.6	118.5								

4.3.3 Mass spectroscopy

Seven synthesized azo dyes, which gave intense color in extracted phase, were characterized by mass spectroscopy. The mass spectrum of each compound exhibited five molecular weights of mixture from C_{10}-alkyl to C_{14}-alkyl as shown in Table 4.12 and Figures A.12, A.25, A.32, A.36, A.49, A.56 and A.72, respectively.

Table 4.12 The mass spectrums of azo dye compounds

Compounds	Mass spectrum (Da/e)				
	C_{10}-alkyl	C_{11}-alkyl	C_{12}-alkyl	C_{13}-alkyl	C_{14}-alkyl
 **(1a)**	382	396	410	424	438
 **(5a)**	417	431	445	459	473
 **(7a)**	412	426	440	454	468
 **(1b)**	382	396	410	424	438
 **(5b)**	417	431	445	459	473
 **(7b)**	412	426	440	454	468
 **(5c)**	388	402	416	430	444

4.4 Detection of markers

In this research, the detection of marked HSD was the extraction with fuel immiscible extractant, which caused the marker to react or complex, producing a clearly defined color.

All marked HSD were shaken with 2% potassium hydroxide in ethylene glycol in order to observe their basically developed colors, which were summarized in Table 4.13

Table 4.13 The developed color of markers extracting with 2% potassium hydroxide in ethylene glycol

Markers	Developed colors
1a	**Red**
2a	Yellow
3a	Yellow
4a	Yellow
5a	**Red**
6a	Yellow
7a	**Pale green**
1b	**Red**
2b	Yellow
3b	Yellow
4b	Yellow
5b	**Purple**
6b	Yellow
7b	**Pale green**
1c	Yellow
2c	Yellow
3c	Yellow
4c	Yellow
5c	Yellowish orange
6c	Yellow

From Table 4.13, there were 4 markers, which produced intense red coloration in extracted phase. These markers were phenyl azo aniline, which had similar structures. Compound **1a**, which gave red coloration, was therefore chosen to study suitable extractant system for the detection of marker in HSD.

4.4.1 Suitable extractant system for the detection of marker in HSD

Aliquot of 10 ppm of compound **1a** in HSD was extracted with acid and basic extractant systems. The acid system included 10% hydrochloric acid, 10% formic and 10% acetic acid in cosolvent comprising 40% propylene glycol (PG) and 60% methanol (MeOH). The basic system included 1% - 5% potassium hydroxide (KOH), 10% -50% diethylamine (DEA), and 10% - 50% ethylenediamine (EDA) dissolved in 40% propylene glycol (PG) and 60% methanol (MeOH). The volume ratio of marked HSD to each extractant was 6: 1. In selecting the appropriate extractant system, the color developed in the extracted phase, the intensity and stability of the developed color, and time to separate from an oil phase were considered. The UV/VIS absorption at maximum wavelength of each extracted phase was measured and illustrated in Tables 4.14-4.17 and Figures 4.3-4.5.

Table 4.14 The visual color of compound 1a (10 ppm) in HSD extracting with acid reagents

Extractant systems	Visual color	Time to separate (min)
10% HCl in 40% PG and 60% MeOH	Dark yellow	7
10% HCOOH in 40% PG and 60% MeOH	Dark yellow	7
10% CH_3COOH in 40% PG and 60% MeOH	Dark yellow	7

Table 4.15 The absorbance of the developed color of compound 1a (10 ppm) in HSD extracting with 1 – 5 % potassium hydroxide solution

Extractant system	λ_{max}	Absorbance	Visual color	Time to separate (min)
1% KOH in 40%PG and 60%MeOH	522.0	0.8733	Red	10
2% KOH in 40%PG and 60%MeOH	522.9	0.8397	Red	10
3% KOH in 40%PG and 60%MeOH	522.5	0.8041	Red	10
4% KOH in 40%PG and 60%MeOH	523.3	0.8596	Red	8
5% KOH in 40%PG and 60%MeOH	525.1	0.8681	Red	8

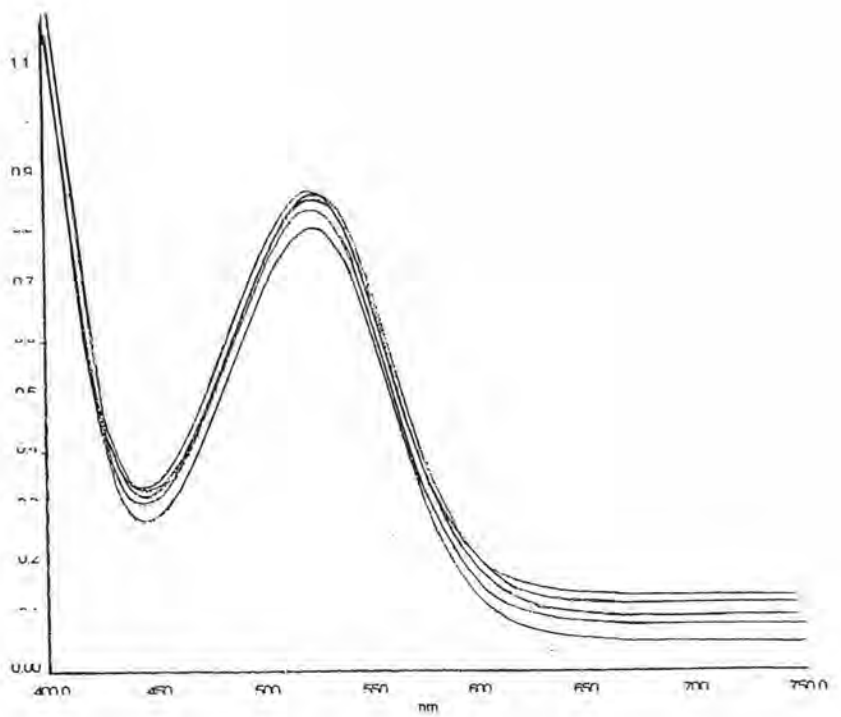


Figure 4.4 The absorbance of the developed color of compound 1a (10 ppm) in HSD extracting with 1 – 5 % potassium hydroxide solution

Table 4.16 The absorbance of the developed color of compound 1a (10 ppm) in HSD extracting with 10 – 50 % diethylamine solution

Extractant system	λ_{max}	Absorbance	Visual color	Time to separate (min)
10% DEA in 40% PG and 60% MeOH	523.1	0.6626	Red	6
20% DEA in 40% PG and 60% MeOH	524.7	0.7428	Red	6
30% DEA in 40% PG and 60% MeOH	525.3	0.8336	Red	6
40% DEA in 40% PG and 60% MeOH	526.7	1.0522	Red	6
50% DEA in 40% PG and 60% MeOH	527.0	1.1741	Red	6

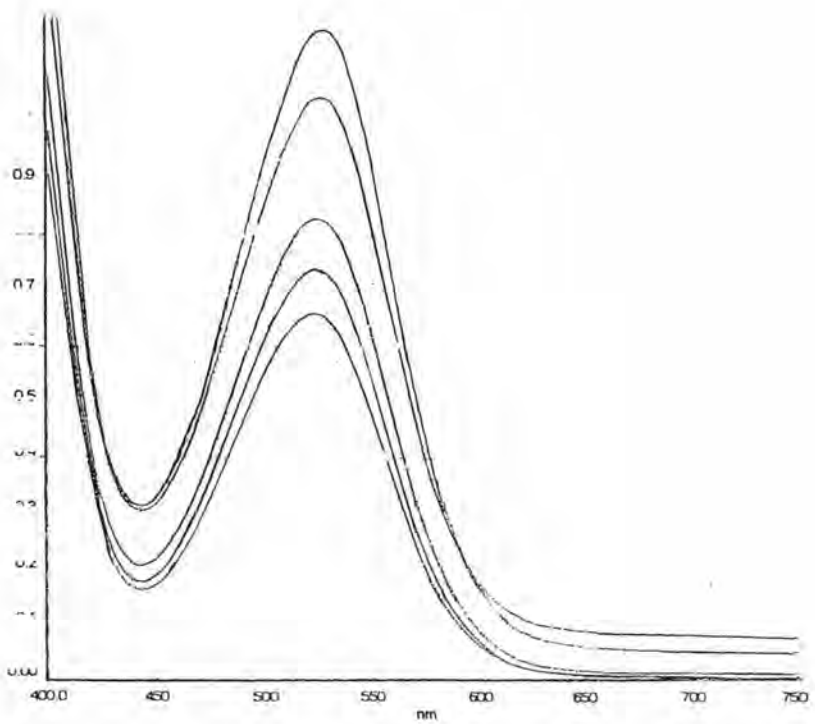


Figure 4.5 The absorbance of the developed color of compound 1a (10 ppm) in HSD extracting with 10 – 50 % diethylamine solution

Table 4.17 The absorbance of the developed color of compound 1a (10 ppm) in HSD extracting with 10 – 50 % ethylenediamine solution

Extractant system	λ_{max}	Absorbance	Visual color	Time to separate (min)
10% EDA in 40% PG and 60% MeOH	527.1	0.7236	Red	7
20% EDA in 40% PG and 60% MeOH	533.4	0.7432	Red	7
30% EDA in 40% PG and 60% MeOH	538.4	0.7790	Reddish purple	6
40% EDA in 40% PG and 60% MeOH	545.4	0.8030	Purple	6
50% EDA in 40% PG and 60% MeOH	551.8	0.8402	Purple	6

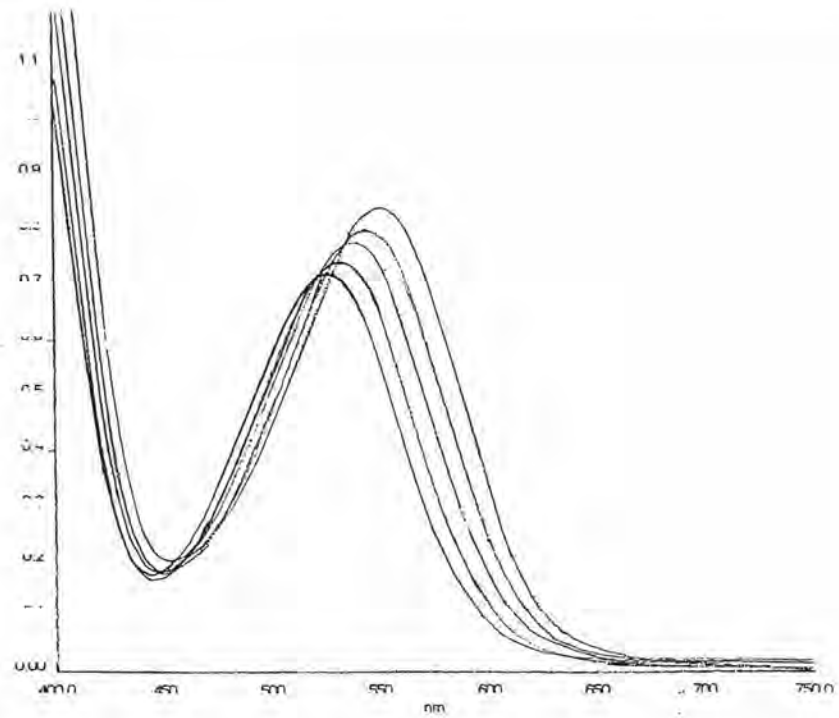


Figure 4.6 The absorbance of the developed color of compound 1a (10 ppm) in HSD extracting with 10 – 50 % ethylenediamine solution

From the extractant systems in Tables 4.14-4.17, it was found that extraction using acid reagents produced a dark yellow color in extracted phase, while extraction using basic reagents produced from red to purple coloration. Thus, the basic reagent could produce a color, which was more clearly distinguishable from HSD color, than the acid reagent. As a result, the acid reagent did not gain considerable attention as the extractant for the detection of markers in this research.

Pertaining to extraction using basic reagents, it was shown that the least time to separate the extracted phase from the oil phase was six minutes. This was due to the fact that each solution contained a methanol as cosolvent, which was difficult to separate from the diesel. However, it was preferred that the extractant contain the cosolvent because it helped to solvate both ionic and non-ionic species. As a consequence, the cosolvent had significantly advantageous effects with respect to shade and intensity of the developed color. From Tables 4.15-4.17, potassium hydroxide system used longer time to separate than the diethylamine and ethylenediamine systems. This might be because potassium hydroxide solution must contain more volume of methanol than the amine solution. In addition, it was difficult to prepare the precise concentration of potassium hydroxide solution because potassium hydroxide is typically quite moist. Hence the absorbance of the extracted phase using potassium hydroxide solution was not related to its concentration, which is shown in Table 4.15 and Figure 4.3. Furthermore, the color developed with potassium hydroxide solution was unstable, which reduced the ability to obtain quantitative determinations. For these reasons, the potassium hydroxide system was not suitable for the extraction of marker in this research.

For the amine-extractant system, it was found that diethylamine solution gave the maximum absorbance of extracted phase and it was higher than ethylenediamine at the same concentration. Nevertheless, diethylamine solution could be oxidized very easily within 24 hours at room temperature. Consequently, the coloring of the extracted phase was changed, which led to the errors during the quantitative determination of markers. In addition, the color developed from compound 1a with diethylamine solution was red,

which was not as intense as might be desired. Because of its odor, diethylamine was less preferred to be the extractant in the detection of marker.

The ethylenediamine system had advantage in its fairly stable and the developed color in the extracted phase was unchanged after 3 days at room temperature. Furthermore, in the detection reaction, it gave more distinguishable coloration at longer maximum wavelength. It could be concluded that the most suitable system for the extraction of marker from HSD in this research was the ethylenediamine system.

From Table 4.17 and Figure 4.5, it was observed that the maximum wavelength of developed color would shift to the longer wavelength when the percentage of ethylenediamine was increased. This effect is the "Bathochromic shift" or "Red shift". One explanation for this effect is that the energy level of the excited state was reduced by dipole-dipole interaction and hydrogen bonding between ethylenediamine and the marker. Hence 50% ethylenediamine in cosolvent comprising 40% propylene glycol and 60 % methanol was the most suitable system for the detection of marked commercial in this research.

4.4.2 Optimum shaking time

Shaking was a method that led to react or complex the marker in HSD with the extractant. Therefore shaking should be restricted at an optimum time, which could substantially remove all of the markers from HSD. In this study, 10 ppm of compound **1a** in HSD was shaken with suitable extractant from 4.2.1 comprising 50% ethylenediamine, 20% propylene glycol and 30% methanol. The volume ratio of marked HSD to extractant was 6: 1. The various shaking times were varied from 10 seconds to 50 seconds. The extracted phase of each shaking time was measured by UV/VIS absorption at its λ_{max} as shown in Table 4.18. In addition, this absorbance was plotted with its shaking time invidually for the comparison as a graph in Figure 4.7.

Table 4.18 The absorbance of the developed color with various shaking times (sec)

Time (sec)	λ_{max} (nm)	Absorbance
10	551.7	0.6120
20	551.9	0.7493
30	551.7	0.8398
40	551.8	0.8393
50	552.0	0.8397

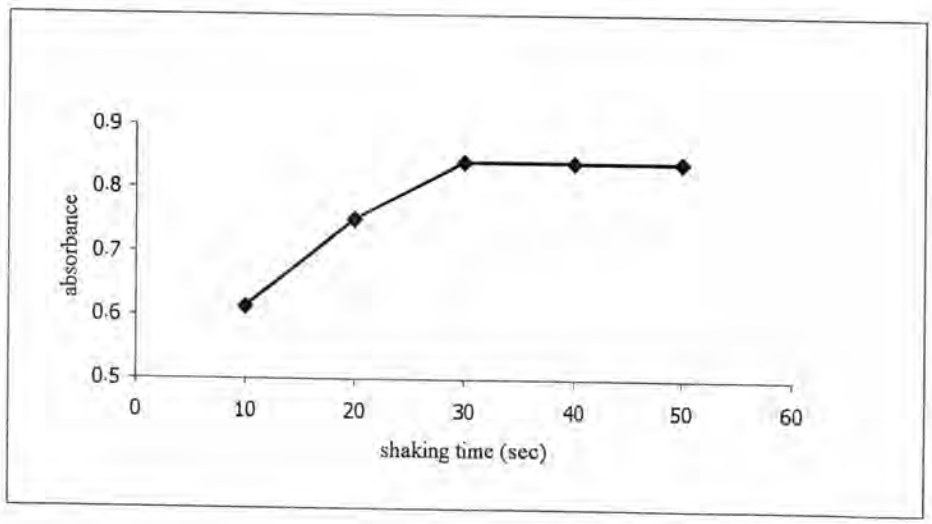


Figure 4.7 Relation of shaking time (sec) and absorbance

From Table 4.18 and Figure 4.7, the absorbance was increased according to shaking time from 10 seconds to 30 seconds, and it was constant from 30 seconds to 50 seconds. This meant that 30 seconds of shaking time was the optimum shaking time, which could extract the marker from HSD to extracted phase completely. Consequently, 30 seconds of shaking time was used throughout this study.

4.4.3 Efficiency of extractant system

From 4.4.1 and 4.4.2, the suitable extractant system was an extractant comprising 50% ethylenediamine, 20% propylene glycol and 30% methanol, when the shaking time was 30 seconds. Efficiency of this extractant system was studied to determine the extracted percentage of this system. The calibration curve of compound **1a** dissolving in the extractant was produced for comparing between the amount of marker, which was removed into extracted phase, with the amount of marker originally added to HSD. This calibration curve was exhibited in Figure 4.8, where the calibration equation was

$$Y = -1.800121e\text{-}03 + 1.370011e\text{-}02 \times X$$

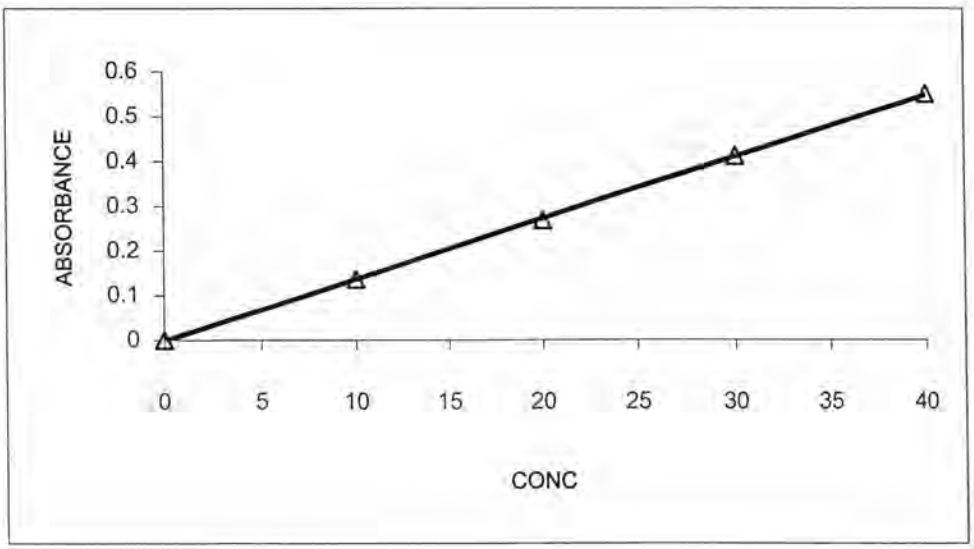


Figure 4.8 The calibration curve of compound **1a** dissolving in extractant

Subsequently, 30 ml of HSD containing 4 ppm of compound **1a** (0.1200 mg of compound 1a) was extracted with 5 ml of extractant. The UV-VIS absorption of the extracted phase was **0.3201**. It was compared with the above calibration curve and indicated the concentration as 23.50 ppm. This 5 ml of extracted phase containing 23.50 ppm of compound **1a** meant there was 0.1175 mg of compound **1a** in extracted phase. As a result, the weight percentage of compound **1a** in extracted phase to compound **1a** in the

original HSD was 97.92 %. This procedure was repeated, and gave the extracted percentage of this extractant system as 97.92 ± 0.2 % by weight.

This extracted percentage showed that the extraction with extractant comprising 50% ethylenediamine, 20% propylene glycol, and 30% methanol for 30 seconds was a high efficiency system. The ethylenediamine solution was therefore used for the extraction of all of the markers from HSD in this research.

The synthesized markers were marked in HSD at 15 ppm, and they were subsequently shaken with an extractant comprising 50% ethylenediamine, 20% propylene glycol, and 30% methanol for 30 seconds, except for compound **6c** that provided substantial red color in HSD. The volume ratio of marked HSD to the extractant was 6: 1, each visual color of marker in extracted phase was observed as shown in Table 4.19 and Figure 4.9.

Table 4.19 Visual color of markers in extracted phase

Markers	Visual color
1a	Purple
2a	Yellow
3a	Yellow
4a	Yellow
5a	Red
6a	Yellow
7a	Green
1b	Purple
2b	Yellow
3b	Yellow
4b	Yellow
5b	Violet

Table 4.19 Visual color of markers in extracted phase (cont.)

Marker	Visual color
6b	Yellow
7b	Green
1c	Dark yellow
2c	Dark yellow
3c	Dark yellow
4c	Dark yellow
5c	Reddish orange

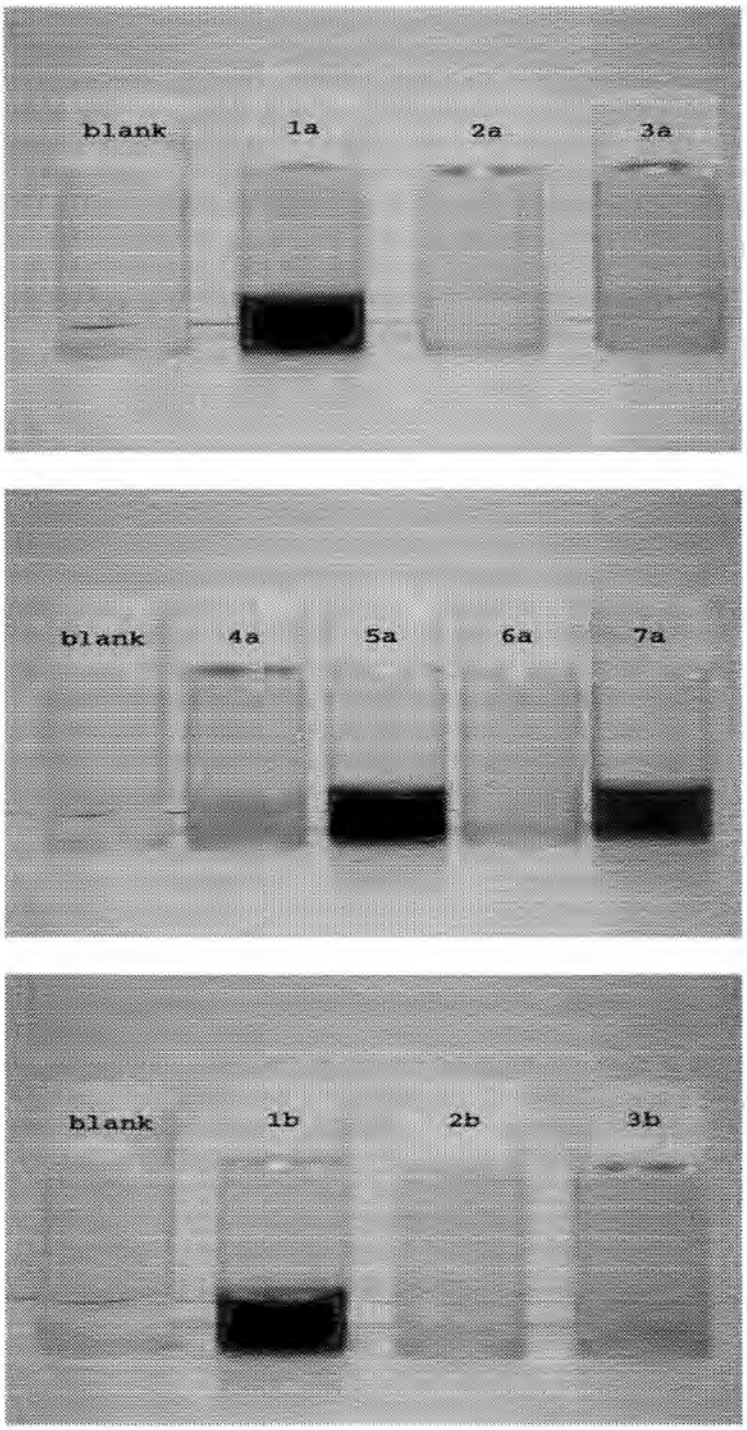


Figure 4.9 Unmarked HSD and marked HSD (15 ppm) when they were extracted with 50% ethylenediamine extractant, using 6:1 volume ratio of HSD to extractant

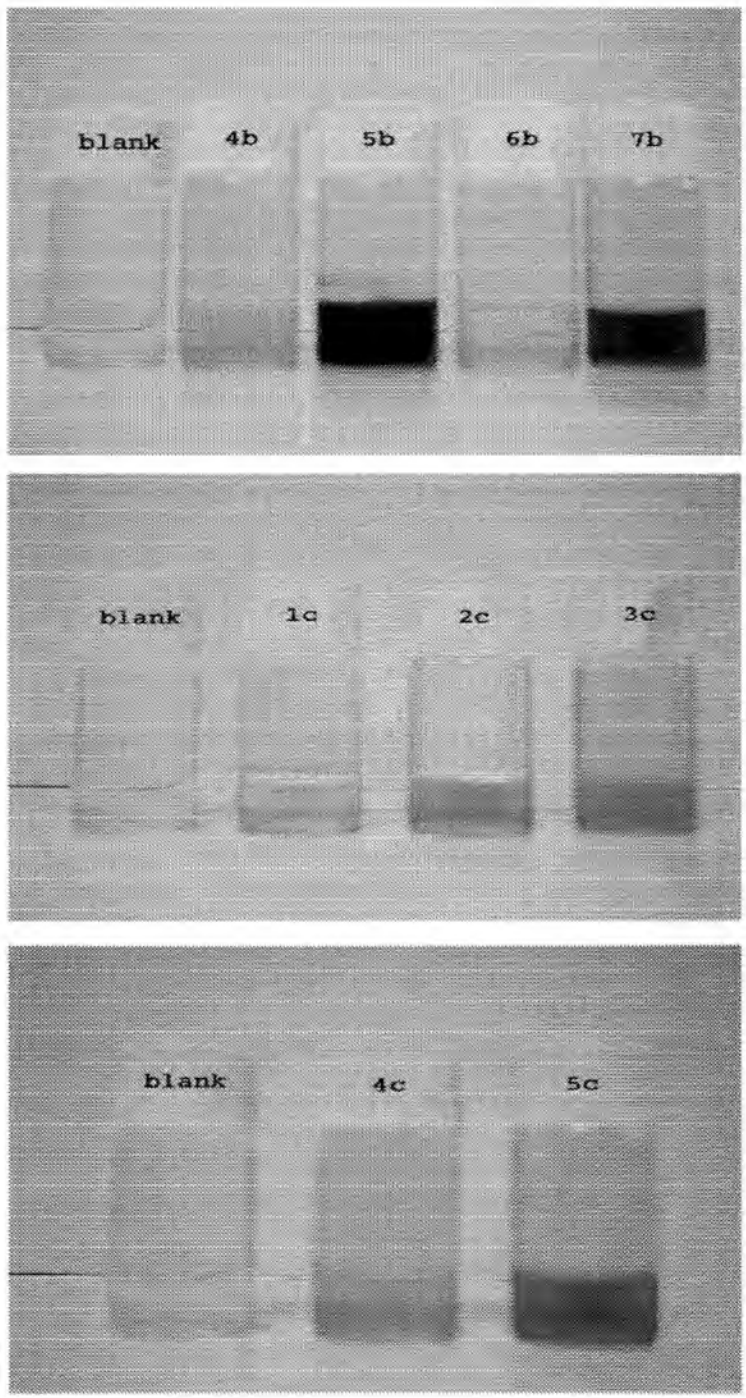
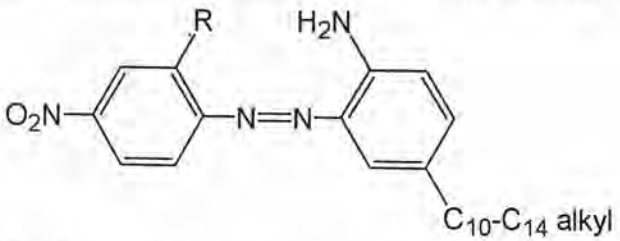


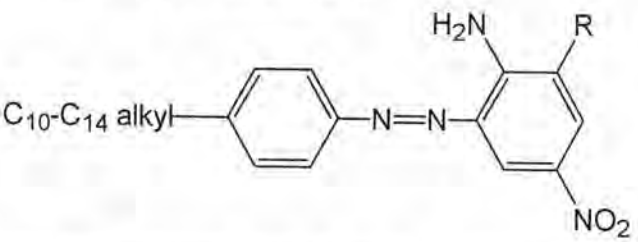
Figure 4.9 Unmarked HSD and marked HSD (15 ppm) when they were extracted with 50% ethylenediamine extractant, using 6:1 volume ratio of HSD to extractant (cont.)

From Table 4.19 and Figure 4.9, each marker provided the different developed color in the extracted phase, so the extraction could be used as the qualitative determination in field test. Markers, which gave distinguishable developed color, included compounds **1a**, **5a**, **7a**, **1b**, **5b**, **7b**, and **5c**. When considering the structure of these markers, it was found that three markers in group a substituted with the nitro group in the para-position to the azo group, would give intense coloration when extracted with ethylenediamine solution. The structures of these markers are shown below.



Where R = H in compound 1a
R = Cl in compound 5a
R = OCH_3 in compound 7a

On the other hand, three markers in group b substituted with the nitro group in the para-position to the amine group would give intense coloration. The structures of these markers are exhibited below.



Where R = H in compound 1b
R = Cl in compound 5b
R = OCH_3 in compound 7b

The last structure, which gave intense coloration was phenyl azo naphthol having a hydroxy group in the para-position to the azo group (compound 5c). The coloration of this structure was stronger than phenyl azo phenol, which was substituted with the hydroxy group in the same position. The structures of markers in group c are shown in Figure 4.3.

However, the reaction or complexation of ethylenediamine and cosolvent with the marker was not fully understood. They were known to perform two functions. First, the reaction or complexation results in the solvate-soluble marker being rendered soluble in aqueous medium, and thereby extractable in aqueous medium. Secondly, the reaction or complexation developed the color.

4.5 Quantitative determination of markers in marked HSD

Quantitative determinations were particularly important in the case where dilution is suspected, e.g. dilution of a higher-taxed fuel with a lower-taxed fuel. In this research, the quantitative determination of markers in marked HSD was divided into 2 systems. First, the volume ratio of HSD to extractant was fixed at 6:1. The calibration curve of marker in extracted phase was produced for the quantitative determination to obtain the minimum and suitable concentration. Secondly, the concentration of markers in HSD was fixed at 5 ppm except for compound **5b** that gave a very strongly visual color in the extracted phase, which was fixed at 3 ppm. The marked HSD was extracted at various volume ratios of HSD to extractant for the determination of suitable volume ratio. Each suitable volume ratio was used for the extraction of marker in HSD to produce the calibration curve of marker for the quantitative determination.

4.5.1 Quantitative determination of markers at 6:1 volume ratio of marked HSD to extractant

The UV/VIS absorbance in extracted phase of markers was measured by UV/VIS spectrophotometer at its maximum wavelength (λ_{max}), which is shown in Table 4.20.

Table 4.20 The UV/VIS absorbance at its maximum wavelength (λ_{max}) and visual color in the extracted phase of markers

Markers	Conc. (ppm)	Visual colors	λ_{max} (nm)	Absorbances
1a	4	**Purple**	552.8	0.3201
2a	15	Yellow	418.1	0.8570
3a	15	Yellow	423.6	0.3150
4a	15	Yellow	381.9	0.2021
5a	4	**Red**	529.5	1.0469
6a	15	Yellow	394.6	0.4175
7a	5	**Green**	589.5	0.1412

Table 4.20 The UV/VIS absorbance at its maximum wavelength (λ_{max}) and visual color in the extracted phase of markers (cont.)

Markers	Conc. (ppm)	Visual colors	λ_{max} (nm)	Absorbances
1b	2	**Purple**	553.0	0.3263
2b	15	Yellow	418.1	0.7956
3b	15	Yellow	424.0	0.4358
4b	20	Yellow	398.7	0.4090
5b	0.5	**Violet**	560.2	0.5318
6b	15	Yellow	400.9	0.8068
7b	15	**Green**	587.9	0.3756
1c	5	Dark yellow	440.0	1.2059
2c	5	Dark yellow	440.6	1.1548
3c	15	Dark yellow	466.3	0.3439
4c	15	Dark yellow	453.9	0.2793
5c	5	**Reddish orange**	512.0	0.8008

The absorbance of each marker was employed to estimate the range of concentration in its calibration curve, which was a plot between absorbance and the concentration of the marker in HSD. Since the maximum wavelength of each marker in this extractant was specific, the absorbance of each marker in each concentration had to be measured at its maximum wavelength. Each marker provided different development colors at different maximum wavelengths because substituents in each structure were different. In group a, the chlorine group, which resulted in development color of compound 5a indicated maximum wavelength, which was shorter than compound 1a where R was hydrogen. While the methoxy group provided the longest maximum wavelength of development color of compound 7a. On the contrary, the chlorine group, which resulted in the development of compound 5b indicated maximum wavelength longer than compound 1b and methoxy group provided the longest maximum wavelength

of development color of compound 7b. As the discussion in 4.4.3, the mechanism of coloration between ethylenediamine and the marker was not fully understood.

The concentration, which gave absorbance close to 0.39, was the suitable concentration of marker in HSD because this absorbance provided the lowest error [27]. The minimum concentration of marker in HSD was the least concentration, which could be detected by the naked eye. The calibration equations, the suitable concentrations, and the minimum concentrations of markers in HSD are listed in Table 4.21.

Table 4.21 The calibration equations for the quantitative determinations of markers

Cpd	λ_{max}	Calibration equation	Correlation coefficient	Min. conc. (ppm)	Suit. conc. (ppm)
1a	552.8	Y=-5.002632e-04+8.060011e-02*X	0.999895	2	5
2a	418.1	Y=-2.689459e-03+4.954169e-02*X	0.999755	6	8
3a	423.6	Y=-3.663784e-03+1.554227e-02*X	0.999658	15	25
5a	529.5	Y=1.593017e-03+2.680884e-01*X	0.999889	0.5	2
6a	394.6	Y=-2.23071e-04+2.69563e-02*X	0.999881	10	15
7a	589.5	Y=-1.20038e-03+2.9941719e-02*X	0.999885	9	13
1b	552.6	Y=2.670000e-03+1.514620e-01*X	0.999905	1	3
2b	418.1	Y=2.209730e-03+5.685291e-02*X	0.999621	5	7
3b	424.0	Y=-5.818000e-03+2.535600e-02*X	0.999724	10	15
5b	560.2	Y=3.526393e-03+8.745992e-01*X	0.999884	0.1	0.5
6b	400.9	Y=1.433962e-05+3.343745e-02*X	0.999994	8	12
7b	587.9	Y=-5.925135e-03+2.726408e-02*X	0.999676	10	15
1c	440.0	Y=-8.132000e-03+2.320400e-01*X	0.999831	1	2
2c	440.6	Y=-2.238000e-03+1.68007e-01*X	0.999815	1	2
4c	453.9	Y=-6.081081e-05+1.820149e-02*X	0.999978	15	20
5c	512.0	Y=1.410270e-03+1.221242e-01*X	0.999950	2	3

From Table 4.21, the correlation coefficients in the calibration equations of markers were close to 1, this meant that these calibration curves were the precise quantitative determinations. As a consequence, each calibration curve was used for the quantitative determination. HSD, which were marked with these markers at suitable concentration and then released to the petrol station, were detected or monitored by shaking with 50% ethylenediamine in 20% propylene glycol and 30% methanol. The extracted phase could be compared by color with a solution of known concentration and the marker content was determined by comparing its absorbance with the calibration equation.

Some compounds were considerably used as markers since they could be detected at the low concentration including compounds **1a**, **5a**, **1b**, **5b**, **1c**, **2c**, and **5c**. However, compounds **1c** and **2c**, which provided yellow color in the extracted phase, should not be used as markers because the yellow was not significantly different from the HSD color. Owing to the quantity of the marker, it would be decreased with fuel release to consumers; diluting the next batch of fuel might bring about the loss of the marker. To prevent this problem, the concentration of the marker in general was used from 3 to 5 ppm. As a consequence, compounds **5a**, and **1b** would be added into HSD at 5 ppm. Compound **5b** could be added into HSD at only 3 ppm because it gave very intense violet in the extracted phase. The suitable volume ratio of marked HSD to extractant was then studied by shaking with extractant at various volume ratios. After that, the calibration curves at suitable volume ratio were produced for the quantitative determination. Furthermore, compounds **7a** and **7b**, which could be detected at 12 ppm and 15 ppm, respectively, would be studied for suitable volume ratio since they developed intense green color in the extracted phase. They would be added into the HSD at 5 ppm and were studied using a similar manner to that described for the study of compound **5a**.

4.5.2 Quantitative determination of markers at suitable volume ratio of marked HSD to extractant

Five compounds including compounds **5a**, **7a**, **1b**, **5b**, and **7b** were studied for the suitable volume ratio of marked HSD to extractant by shaking with extractant at various volume ratios. The UV/VIS absorbance in extracted phase at various volume ratios of marked HSD to extractant of each compound are tabulated in Table 4.22.

Table 4.22 The UV/VIS absorbance with various volume ratios of marked HSD to extractant at its maximum wavelength (λ_{max})

Compounds	Conc. (ppm)	Ratio	λ_{max}	Absorbance
5a	5	1:1	532.9	0.2839
		2:1	532.6	0.4956
7a	5	15:1	584.2	0.4356
		16:1	584.0	0.4720
1b	5	3:1	552.0	0.4316
		4:1	551.3	0.5781
5b	3	0.8:1	559.7	0.3622
		1:1	559.8	0.4099
7b	5	16:1	584.7	0.3718
		18:1	584.2	0.4185

The volume ratios of marked HSD to extractant, which gave an absorbance close to 0.39 [27], were chosen for the quantitative determination. Each suitable volume ratio was used for the extraction of various concentrations of marker in HSD with the extractant. The calibration of marker in HSD was a plot between the absorbance of the extracted phase and the concentration of the marker in HSD. The calibration equations of markers in HSD are listed in Table 4.23.

Table 4.23 The calibration equations for the quantitative determination of marker at suitable volume ratio of marked HSD to extractant

Compounds	Ratio	λ_{max}	Calibration equation	Correlation coefficient
5a	2:1	532.6	Y=5.611321e-04+9.963302e-02*X	0.999962
7a	15:1	584.2	Y=-4.432623e-03+7.240721e-02*X	0.999914
1b	3:1	552.0	Y=1.893019e-04+8.667472e-02*X	0.999911
5b	1:1	559.8	Y=-1.24727e-03+1.372511e-01*X	0.999728
7b	18:1	584.2	Y=-1.460943e-03+8.461415e-02*X	0.999898

From the whole study of the quantitative determination, there were seven compounds, which could be used as markers in HSD, including compound **1a**, 4-linear alkyl-2-(4-nitro)phenyl azo aniline; compound **5a**, 4-linear alkyl-2-(2-chloro-4-nitro) phenyl azo aniline; compound **7a**, 4-linear alkyl-2-(2-methoxy-4-nitro)phenyl azo aniline; compound **1b**, 4-nitro-2-(4-linear alkyl)phenyl azo aniline; compound **5b**, 2-chloro-4-nitro-6-(4-linear alkyl)phenyl azo aniline; compound **7b**, 2-methoxy-4-nitro-6-(4-linear alkyl)phenyl azo aniline; and compound **5c**, 4-(4-linear alkyl) phenyl azo naphthol. These markers could be added into HSD with 2 patterns. First these markers were added at individual concentration using the volume ratio of marked HSD to extractant as 6:1 for the detection. Second the markers were added at 5 ppm except for compound **5b** that was added at 3 ppm using individual volume ratio of each marked HSD to extractant. This made it more confusion to imitate the markers by others. The visual colors of markers in 2 patterns of the detection are shown in Figures 4.10-4.17.

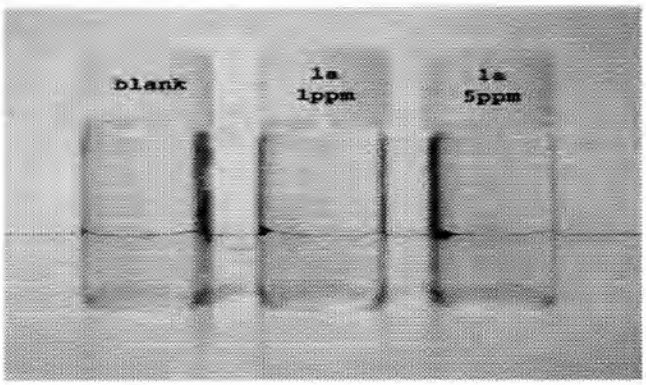


Figure 4.10 Visual color of HSD containing 1 ppm and 5 ppm of compound **1a** comparing with unmarked HSD (blank)

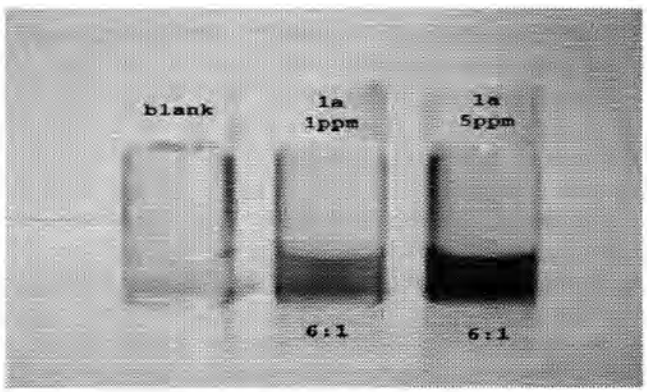


Figure 4.11 Visual color of compound **1a** in HSD when it was extracted with 50% ethylenediamine extractant, using 6:1 volume ratio of marked HSD to extractant

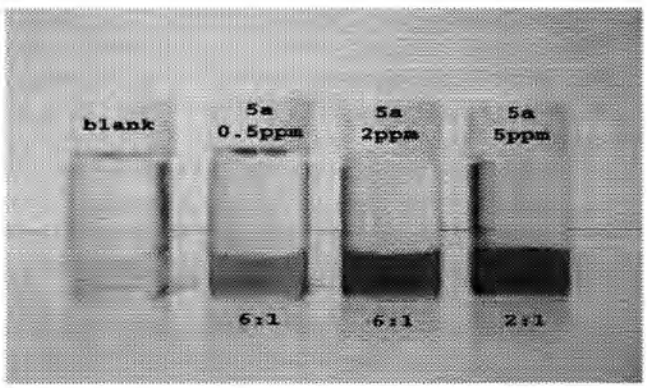


Figure 4.12 Visual color of compound **5a** in HSD when it was extracted using 6:1 and 2:1 volume ratios of marked HSD to extractant

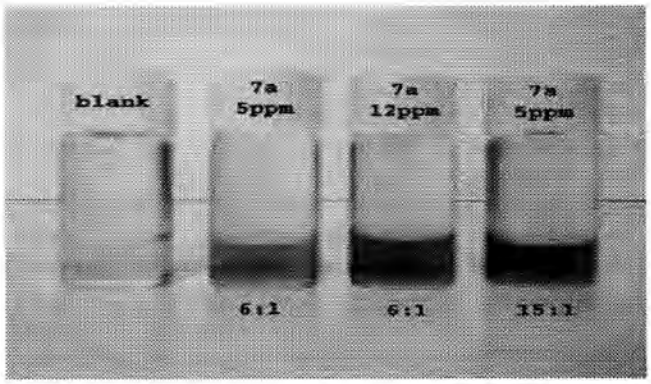


Figure 4.13 Visual color of compound **7a** in HSD when it was extracted using 6:1 and 15:1 volume ratios of marked HSD to extractant

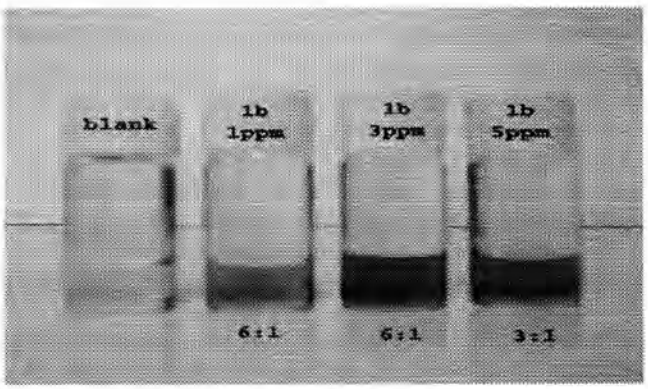


Figure 4.14 Visual color of compound **1b** in HSD when it was extracted using 6:1 and 3:1 volume ratios of marked HSD to extractant

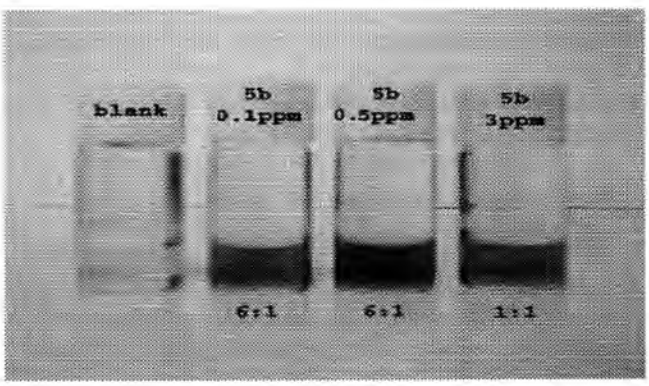


Figure 4.15 Visual color of compound **5b** in HSD when it was extracted using 6:1 and 1:1 volume ratios of marked HSD to extractant

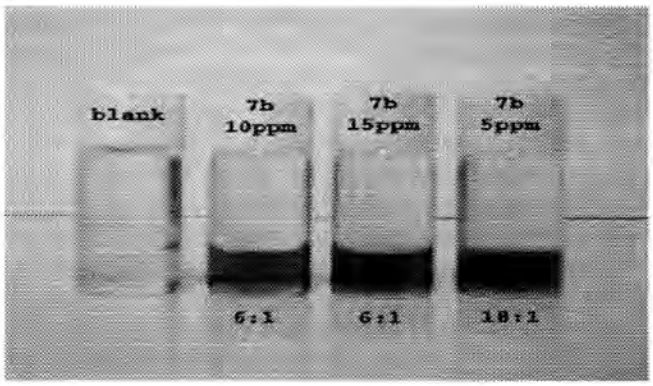


Figure 4.16 Visual color of compound **7b** in HSD when it was extracted using 6:1 and 18:1 volume ratios of marked HSD to extractant

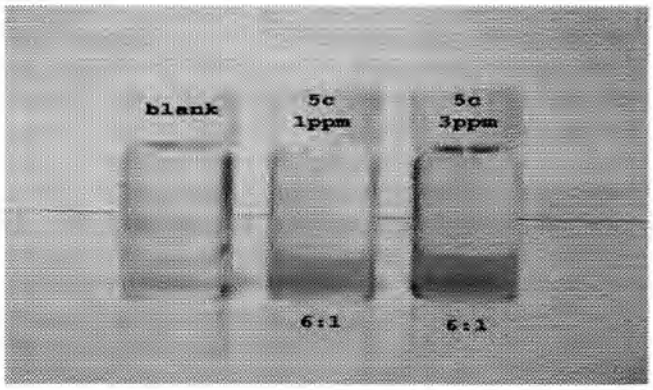


Figure 4.17 Visual color of compound **5c** in HSD when it was extracted using 6:1 volume ratio of marked HSD to extractant

In field test, the difficulty of quantitative determination of markers by UV-VIS spectrophotometer might be encountered. A developed color's strip might be used as a rough and ready method to determine the quantity of the marker in the specimen of HSD using the principle similar to the indicator paper. The developed color's strip of compound **5b** using volume ratio of marked HSD to extractant as 1:1, is shown in Figure 4.18

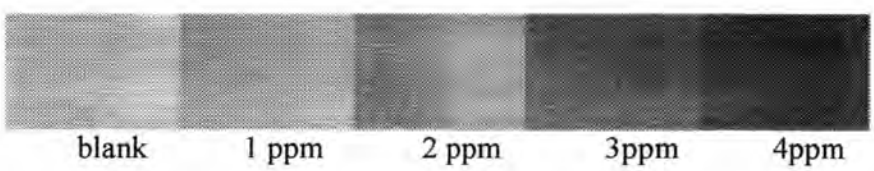


Figure 4.18 The developed color's strip of compound **5b** using 1:1 volume ratio of marked HSD to extractant

From Figure 4.18, HSD containing compound **5b** could be detected the quantity in a simple field test by comparing the developed color in the extracted phase of the specimen with this color strip. However, the absorbance of extracted phase should be measured by UV-VIS spectrophotometer if the precisely quantity of marker in HSD was desired.

4.6 Effect of marker on the physical properties of marked HSD

HSD containing 3 ppm of compound **5b** was employed for physical properties testing, using the ASTM methods. The results are shown in Table 4.24.

Table 4.24 The effect of marker on the physical properties of marked HSD

Test items	ASTM	Limit	Result	
			Unmarked HSD	Marked HSD
API gravity@60°F	D1298	Report	38.7	38.7
Specific gravity@15.6/15.6°C	D1298	Report	0.8314	0.8314
Calculated cetane index	D976	47 min	56.0	56.0
Kinematic viscosity@40°C, cSt	D445	1.8-4.1	3.183	3.148
Pour point, °C	D97	10	-8	-8
Flash point, °C	D93	52	67	64
Sulfur content, %wt	D5453	0.05	0.0429	0.0432
Copper strip corrosion (3 hrs, 50 °C)	D130	No.1	No.1	No.1

Table 4.24 The effect of marker on the physical properties of marked HSD (cont.)

Test items	ASTM	Limit	Result	
			Unmarked HSD	Marked HSD
Distillation: (correct. temp)	D86			
IBP, °C		Report	177.0	173.0
10% rec, °C		Report	211.2	209.6
50% rec, °C		Report	279.0	279.0
90% rec, °C		357max	351.9	351.6
Total acid number, mgKOH/g	D664		0.02	0.01
Color	D1500 Visual	2.0max	L<0.5 Yellow	L<0.5 Yellow

From Table 4.24 the physical properties of marked HSD were not significantly different from unmarked HSD. Both marked and unmarked HSD provided similar specific gravity, calculated cetane index, kinematic viscosity, pour point, flash point, sulfur content, copper strip corrosion, distillation properties, total acid number and color. This means that this compound did not have any effect on the physical properties of the marked HSD. Consequently, these azo compounds can possibly be used as the marker in HSD.

4.7 Stability of markers in HSD

Seven compounds, which gave visual color in extracted phase including compounds **1a**, **5a**, **7a**, **1b**, **5b**, **7b**, and **5c** were chosen for the stability studying of the marker in HSD. In this study, the quantities of markers added into commercial HSD were monitored for 3 months, using the UV/VIS spectroscopic technique. The concentrations of marked HSD in the period of 3 months are shown in Table 4.25.

Table 4.25 The concentration of marked HSD in the period of 3 months

Markers	Month	Concentration in HSD (ppm)		
		1st	2nd	Average
1a	1	5.07	5.05	5.06
	2	5.08	5.08	5.08
	3	5.10	5.08	5.09
5a	1	4.01	4.01	4.01
	2	4.03	4.01	4.02
	3	4.03	4.04	4.04
7a	1	5.05	5.00	5.03
	2	5.07	5.08	5.08
	3	5.10	5.11	5.11
1b	1	5.03	5.01	5.02
	2	5.05	5.07	5.06
	3	5.07	5.10	5.09
5b	1	3.05	3.03	3.04
	2	3.05	3.06	3.06
	3	3.09	3.08	3.09
7b	1	5.03	5.05	5.04
	2	5.09	5.08	5.09
	3	5.15	5.11	5.13
5c	1	3.01	3.01	3.01
	2	3.04	3.05	3.05
	3	3.09	3.07	3.08

From Table 4.25, the concentrations of these compounds in HSD were unchanged from the initial concentrations that meant these compounds were stable after at least 3 months storage in HSD fuel. As a result, these compounds could be used as markers in HSD, which in general are consumed within 3 months after being released to the market.