AN ULTRA-VIOLET SPECTROPHOTOMETRIC METHOD FOR THE DETERMINATION OF PICRATE, STYPHNATE AND PICROLONATE IN *S*-ALKYL-*N*-PHENYLTHIURONIUM SALTS

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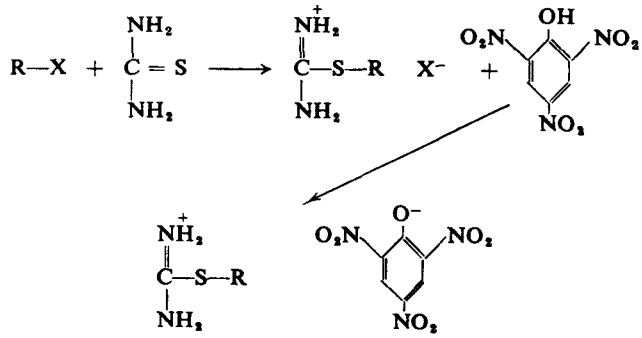
By selecting suitable wavelengths to measure the extinction, it has been found possible to determine the percentage of picrate, styphnate and picrolonate in *S*-alkyl-*N*-phenylthiuronium salts in ethanol solution by an ultra-violet spectrophotometric method. The method is rapid and accurate to ± 1 per cent.

IN a previous communication[1] we reported that *S*-alkyl-*N*-phenylthiuronium picrates, styphnates and picrolonates were useful as derivatives for the characterisation of alkyl halides. It has been shown previously[2] that alkyl halides react with thiourea to form *S*-alkylthiuronium halides which precipitate as *S*-alkylthiuronium picrates with picric acid, Scheme 1.



Scheme 1

Elemental analysis of the *S*-alkyl-*N*-phenylthiuronium picrates, styphnates and picrolonates[1] suggested that these salts were of the same type as the *S*-alkylthiuronium picrates. To check the composition of all the salts which were prepared and to widen the analytical scope of the method, we have developed a spectrophotometric method of determining the picrate, styphnate and picrolonate content of the respective *S*-alkyl-*N*-phenylthiuronium salts.

N-Phenylthiourea does not absorb in the wavelength region 350 to 400 mμ whereas picric acid absorbs strongly in this region. The total absorption of *S*-alkyl-*N*-phenylthiuronium picrate in the region 350 to 400 mμ will therefore be due to the picrate ion only. An application of this principle to the determination of the molecular weight of amine picrates has been reported[3]. Since both styphnic acid and picrolonic acid absorb in the region 350 to 400 mμ, then the principle should be applicable to *S*-alkyl-*N*-phenylthiuronium styphnates and picrolonates.

EXPERIMENTAL

Apparatus

A Hilgar and Watts Uvispek H.700.304 equipped with a quartz prism was used. Matched pairs of silica cuvettes of 1 cm. optical path length

were used for the determination of absorption spectra (for readings below 360 mμ). Matched pairs of glass cuvettes of 1 cm. optical path length were used for the assay readings above 360 mμ.

Melting points were taken on a Kofler block and are corrected.

Material

Solvent. Ethanol (95 per cent) which complied with Appendix IV H of the British Pharmacopoeia 1958 was used. Picric acid, styphnic acid and picrolonic acid were of the purity described previously[1]. *N*-Phenylthiourea was of M.A.S. grade (Hopkin and Williams). *S*-Alkyl-*N*-phenylthiuronium picrates, styphnates and picrolonates were prepared as described previously[1].

Piperidine picrate. Piperidine (0·85 g.) and picric acid (2·3 g.) were dissolved in hot ethanol (95 per cent). On cooling, the product crystallised out. The precipitate was collected and recrystallised twice from

TABLE I

ABSORPTION OF *N*-PHENYLTHIOUREA IN ETHANOL (95 PER CENT) (6·44 MG. IN 100 ML. OF SOLUTION). 1 CM. MATCHED SILICA CUVETTES

Wavelength (mμ)	Extinction
400	0·000
385	0·000
380	0·000
370	0·000
360	0·000
350	0·000
340	0·003
330	0·028
320	0·05
310	0·153
300	0·535
290	1·671

ethanol (95 per cent). Yellow crystals m.p. 151° (Lit.[4] 151°). Found: N, 17·8. Calc. for $C_{11}H_{14}O_7N_4$: N, 17·84. The following compounds were all prepared in a similar manner.

cis-2,6-*Dimethylpiperidine picrate* m.p. 165° (Lit.[5] 162—164°). Found: N, 16·3. Calc. for $C_{13}H_{18}O_7N_4$: N, 16·4.

Trimethylamine picrate m.p. 226° (Lit.[4] 225°). Found: N, 19·5. Calc. for $C_9H_{12}O_7N_4$: N, 19·45.

Tropane picrate m.p. 281° (Lit.[6] 281°). Found: C, 47·31; H, 5·101. Calc. for $C_{14}H_{17}O_7N_4$: C, 47·45; H, 5·08.

Piperidine styphnate. Recrystallised twice from ethanol (50 per cent) m.p. 204–206° (decomp.). Found: N, 17·1. Calc. for $C_{11}H_{14}O_8N_4$: N, 17·0.

cis-2,6-*Dimethylpiperidine styphnate.* Recrystallised twice from ethanol (50 per cent) m.p. 208—210° (decomp.). $C_{13}H_{18}O_8N_4$ requires N, 15·6. Found: N, 15·45.

Tropane styphnate. Recrystallised from ethanol (50 per cent) m.p. 266°. Found: N, 15·1. Calc. for $C_{14}H_{18}O_8N_4$: N, 15·14.

Quinine styphnate. Recrystallised from ethanol (95 per cent) m.p. 155—156° (Lit.[6] 154°). Found: N, 12·3. Calc. for $C_{26}H_{27}O_{10}N_5$: N, 12·3.

Tropane picrolonate. Tropane (2 ml.) was poured into a saturated solution of picrolonic acid in ethanol (95 per cent). After thirty minutes the crystals were filtered off and recrystallised from ethanol (95 per cent) m.p. 207°. Found: N, 18·1: Calc. for $C_{18}H_{22}O_5N_5$: N, 18·0.

The following picrolonate salts were similarly prepared.

Diethylamine picrolonate m.p. 256—260° (decomp.). Found: N, 20·5. Calc. for $C_{14}H_{19}O_5N_5$: N, 20·77.

cis-2,6-*Dimethylpiperidine picrolonate* m.p. 262–265° (decomp.). $C_{17}H_{23}O_5N_5$ requires N, 18·57. Found: N, 18·6.

Piperidine picrolonate m.p. 270—273° (decomp.). Found: N, 19·7. Calc. for $C_{15}H_{19}O_5N_5$: N, 19·6.

Measurements

N-*Phenylthiourea.* The extinction of a solution of *N*-phenylthiourea (6·44 mg. in 100 ml. of solution) was determined between the wavelengths 290 to 400 mμ. The results are given in Table I.

Picrate ion. The absorption of a solution of piperidine picrate (1·36 mg. in 100 ml. of solution) was determined between the wavelengths 340 to

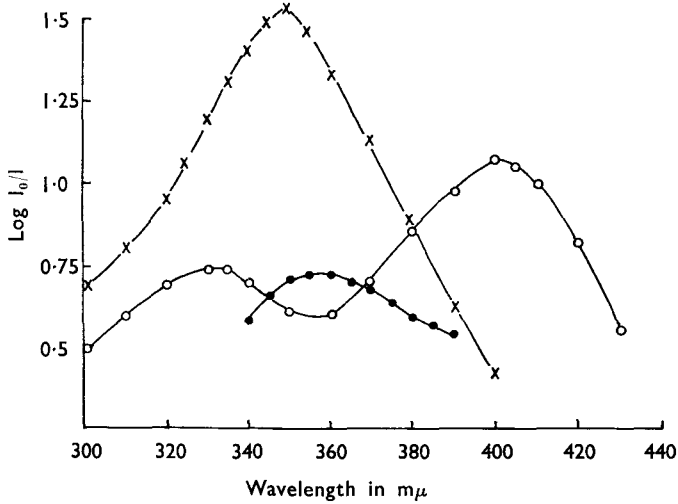


FIG. 1. Absorption spectra of picrate, styphnate and picrolonate in ethanol, 95 per cent. 1 cm. silica cuvettes.
Piperidine picrate, 1·36 mg. in 100 ml. ●—●
Piperidine styphnate, 2·05 mg. in 100 ml. ○—○
Diethylamine picrolonate, 2·124 mg. in 100 ml. ×—×

390 mμ (Fig. 1). The wavelength 385 mμ was selected as suitable for the determination of picrate.

Validity of Beer-Lambert Law for picrate in ethanol. Solutions of different concentrations of an authentic sample of *S-n*-butyl-*N*-phenylthiuronium picrate in ethanol were prepared and the extinction determined at 385 mμ in 1 cm. glass cuvettes (Fig. 2).

Molecular extinction coefficient of picrate at 385 *mμ in ethanol.* Solutions in ethanol were prepared of a number of salts of picric acid. The extinctions of these solutions were determined at 385 mμ using 1 cm. matched glass cuvettes. The molecular extinction coefficient was calculated for each compound and the mean taken for assay purposes (Table II).

Determination of picric content of S-*alkyl*-N-*phenylthiuronium picrates.* A weighed amount of each picrate was dissolved in ethanol (1 to 4 mg.

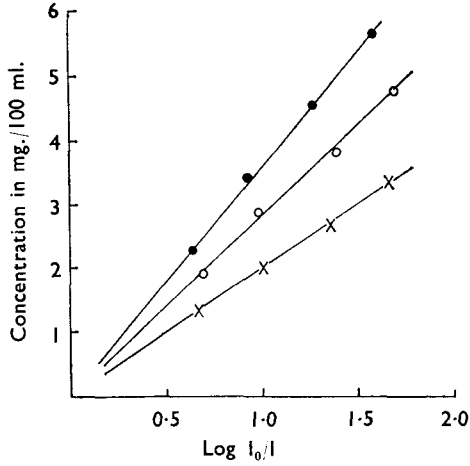


FIG. 2. Verification of Beer-Lambert law for picrate, styphnate and picrolonate in 95 per cent ethanol.

S-n-Butyl-*N*-phenylthiuronium picrate, 385mμ ●—●.
S-n-Pentyl-*N*-phenylthiuronium styphnate, 400 mμ ○—○.
S-n-Pentyl-*N*-phenylthiuronium picrolonate, 350 mμ ×—×.

in 100 ml. of solution). The extinction was determined using 1 cm. glass matched cuvettes at 385 mμ. The precentage picrate was calculated using the following formula.

$$\text{Per cent picrate} = \frac{E \times 228{\cdot}1 \times 100}{\epsilon \times C}$$

where 228·1 = ionic weight of picrate ion
E = Extinction of solution
ϵ = Molecular extinction coefficient of picrate
C = Concentration of picrate in g./l.

A list of values obtained is given in Table III.

Styphnate ion. The absorption of a solution of piperidine styphnate (2·05 mg. in 100 ml. of solution) was determined between 300 to 440 mμ (Fig. 1). The wavelength 400 mμ was selected as the most suitable for the determination of styphnate.

Validity of Beer-Lambert Law for styphnate ion in ethanol. Solutions of different concentrations of an authentic sample of *S-n*-pentyl-*N*-phenylthiuronium styphnate in ethanol (95 per cent) were prepared and the extinction determined at 400 mμ in a 1 cm. glass cuvette (Fig. 2).

Molecular extinction coefficient of styphnate at 400 *mμ in ethanol.* Solutions in ethanol were prepared of a number of known styphnate salts. The extinctions of these solutions were determined at 400 mμ using 1 cm. matched glass cuvettes. The molecular extinction coefficient was calculated for each compound and the mean taken for assay purposes (Table II).

Determination of styphnate content of S-*alkyl*-N-*phenylthiuronium styphnates.* A weighed amount of each styphnate was dissolved in ethanol (1 to 4 mg. in 100 ml. of solution). The extinction was deter-

TABLE II

MOLECULAR EXTINCTION COEFFICIENTS OF PICRATE, STYPHNATE AND PICROLONATE IN ETHANOL (95 PER CENT)

Compound	Molecular extinction coefficient
Picrate—385 *mμ*	
Picric acid	12,310
Piperidine picrate	12,280
2,6-Dimethylpiperidine picrate	12,340
Tropane picrate	12,230
Trimethylamine picrate	12,310
Mean	12,300
Styphnate—400 *mμ*	
Styphnic acid	16,990
Piperidine styphnate	17,000
Tropane styphnate	16,900
2,6-Dimethylpiperidine styphnate	16,950
Quinine styphnate	16,930
Mean	16,950
Picrolonate—350 *mμ*	
Picrolonic acid	24,190
Tropane picrolonate	24,140
Diethylamine picrolonate	24,160
Piperidine picrolonate	24,200
2,6-Dimethylpiperidine picrolonate	24,190
Mean	24,170

mined using 1 cm. glass matched cuvettes at 400 mμ. The percentage styphnate was calculated using the formula given for picrate, but the molecular weight of the styphnate ion (244) was substituted for that of the picrate ion. A list of results is given in Table III.

Picrolonate ion. The absorption of a solution of diethylamine picrolonate (2·124 mg. in 100 ml. of solution) was determined between 300 to 420 mμ (Fig. 1). The wavelength 350 mμ was selected as the most suitable for the determination of picrolonate.

Validity of Beer-Lambert Law for the picrolonate ion in ethanol. Solutions of different concentrations of an authentic sample of *S-n*-pentyl-*N*-phenylthiuronium picrolonate in ethanol were prepared and the extinctions determined at 350 mμ in a 1 cm. silica cuvette (Fig. 1).

Molecular extinction coefficient of picrolonate at 350 *mμ in ethanol.* Solutions in ethanol were prepared of a number of salts of picrolonic acid. The extinctions of these solutions were determined at 350 mμ using 1 cm. matched silica cuvettes. The molecular extinction coefficient was calculated for each compound and the mean taken for assay purposes (Table II).

Determination of picrolonate content of S-*alkyl*-N-*phenylthiuronium picrolonates.* A weighed amount of each picrolonate was dissolved in ethanol (1 to 2·5 mg. in 100 ml. of solution). The extinction was determined using 1 cm. silica matched cuvettes at 350 mμ. The percentage picrolonate was calculated and is given in Table III, using the formula given above but substituting the molecular weight of the picrolonate ion (263) for that of the picrate ion.

TABLE III

DETERMINATION OF PICRATE, STYPHNATE AND PICROLONATE IN *S*-ALKYL-*N*-PHENYLTHIURONIUM PICRATES, STYPHNATES AND PICROLONATES*

Compound	Picrate		Styphnate		Picrolonate	
	Calc.	Found	Calc.	Found	Calc.	Found
Methyl	57·71	57·23	59·34	59·55	61·15	60·84
Ethyl	55·73	55·50	57·42	57·46	59·21	59·33
n-Propyl	53·89	54·36	55·55	55·12	57·41	57·45
n-Butyl	52·14	51·88	53·84	54·06	55·75	55·44
n-Pentyl	50·53	50·27	52·22	52·20	54·12	53·96
n-Hexyl	49·00	50·07	50·70	50·74	52·59	53·01
n-Heptyl	47·57	48·03	49·26	48·71	51·16	51·05
n-Octyl	46·22	46·78	47·92	48·76	49·80	49·72
n-Nonyl	44·95	45·68	46·63	46·49	48·52	48·67
n-Decyl	43·75	43·22	45·51	45·32	47·30	47·11
Allyl	54·14	54·01	55·81	55·98	57·66	57·51
1-But-3-enyl	52·40	52·38	54·08	53·51	55·95	56·03
1-Pent-4-enyl	50·76	50·71	52·43	52·47	54·39	54·40
Benzyl	48·28	48·83	49·97	50·02	51·87	51·40
p-Nitrobenzyl	44·08	44·84	45·76	45·93	47·64	47·66
Cetyl	37·66	37·31	39·27	39·01	41·09	41·26
s-Propyl	53·89	53·18	55·55	55·81	57·41	57·69
s-Butyl	52·14	52·19	53·84	54·26	55·75	55·50
Isobutyl	52·14	52·89	53·84	53·46	55·75	55·68
2-Pentyl	50·53	49·99	52·22	51·78	54·12	53·96
3-Pentyl	50·53	50·51	52·22	51·50	54·12	54·46
Isopentyl	50·53	50·89	52·22	51·99	54·12	54·46
Isohexyl	49·00	48·63	50·70	51·25	52·59	52·67

* Picrate λ = 385 mμ, 95 per cent ethanol, 1 cm. glass cuvettes.
Styphnate λ = 400 mμ, 95 per cent ethanol, 1 cm. glass cuvettes.
Picrolonate λ = 350 mμ, 95 per cent ethanol, 1 cm. silica cuvettes.

DISCUSSION

The ultra-violet spectrophotometric determination of picrates, styphnates and picrolonates is a quick and accurate method. The results (Table III) indicate that the *S*-alkyl-*N*-phenylthiuronium salts consisted of one mole of *S*-alkyl-*N*-phenylthiuronium ion and one mole of picrate, styphnate or picrolonate. The method is not limited to the determinations of these ions only, but could be used for the determination of the molecular weight of a cation of a picrate, styphnate or picrolonate provided that the molar relation between the cation and the acid was known and that the cation did not absorb at the wavelengths at which the determinations were made. Since milligram quantities of the salts are used for the spectrophotometric determination, then it is possible to use this method for the determination of molecular weights on a micro scale.

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