Analytical Method for a Cationic Fabric Softener in Waters and Wastes

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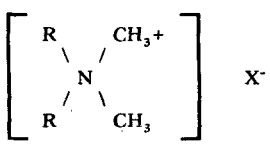
ABSTRACT

The Waters and Kupfer disulfine blue method for cationic surfactants has been extensively modified in order to extend its applicability to the broad range of samples necessary for elucidating the removal and biodegradability of the cationic fabric softener distearyl dimethyl ammonium chloride (DSDMAC) during sewage treatment. In addition, these modifications have overcome the need for preconditioning anion-exchange resin columns and the instability of the disulfine blue-quaternary ion association compound, both recognized by the authors as problems associated with the Waters and Kupfer method. The addition of a thin layer chromatographic procedure has made possible the semiquantitative estimation of the $DSDMA^+$ cation contribution to the measured level of disulfine blue active substances.

INTRODUCTION

Cationic surfactants, although representing only a small percentage of total surfactant production, are broadly used in consumer products and industrial applications for their bacteriostatic, antistatic and textile softening properties. Compounds that have been used extensively as fabric softeners have the quaternary ammonium (I) structure, with R being a hydrophobic, long alkyl chain derived from fatty acid or petrochemical sources.



(I)

Since these materials have the potential for widespread introduction into waste treatment and disposal systems, analytical methodology is needed for determining such surfactants at very low levels in media ranging from sewage treatment plant sludges to potable waters. Although Jungermann (1) has compiled a variety of analytical procedures for the determination of cationic surfactants, the published work of Waters and Kupfer (2) presented the first significant approach to the determination of material such as (I) in wastewater samples in the presence of anionic surfactants. In their procedure, the cationic surfactant is colorimetrically determined as its disulfine blue-ion association compound following removal of interfering anionics by means of an anion-exchange step.

The method for measurement of disulfine blue active substances (DBAS) is nonspecific since many quaternary ammonium compounds and imidazoline-type surfactants, as well as certain protonated alkyl amines, amine oxides and possibly other nitrogen containing cationics give a positive response to the test. The work of Michelson (3) suggested that thin layer chromatography (TLC) offered a viable approach to adding specificity to the colorimetric measurement.

This paper presents significant modifications of the Waters and Kupfer method for the determination of distearyldimethyl ammonium chloride (DSDMAC), a fabric softener widely used in consumer products, in a broad range of waste and water samples. Further, it presents sample collection and preservation conditions that minimize losses of DSDMAC due to adsorption on sample container surfaces, a problem that has apparently not been recognized by other investigators. Specificity for the $DSDMA^+$ cation, on a semiquantitative basis, is achieved using TLC separation of the disulfine blue active substances.

EXPERIMENTAL—ANALYTICAL METHOD (Fig. 1)

Principle

A selected volume, or weight, of the sample was evaporated to dryness. The residue was subjected to extraction and ion-exchange steps that remove insoluble matter, inorganic chlorides and interfering anionic surfactants such as linear alkylbenzene sulfonate (LAS). The fraction containing the cationics was then reacted with disulfine blue, an anionic dye, to form a blue, chloroform extractable cationic-disulfine blue ion association compound that was measured spectrophotometrically, with DSDMAC being used for calibration.

The disulfine blue responsive cationics, measured as DBAS, were adsorbed on a cation exchange resin, from

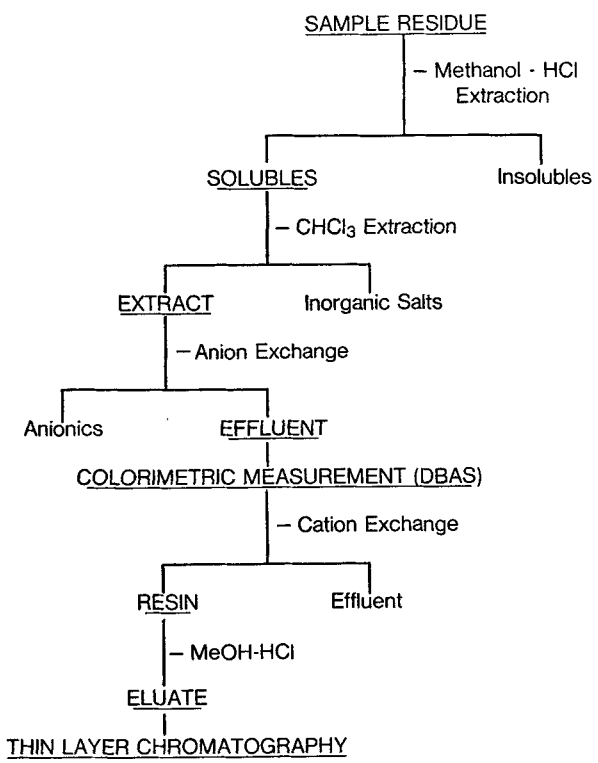


FIG. 1. Analytical schematic.

which they were eluted with acidified methanol. This separation was necessary to effect the removal of substances that would interfere with the TLC separation. An aliquot of the eluted material was spotted and developed, along with known μg quantities of DSDMAC, using standard TLC techniques. Estimation of the $DSDMA^+$ contribution to total DBAS, on a percentage basis, was based on visual comparison of the TLC-separated $DSDMA^+$ in the DBAS sample with the known DSDMAC standards.

Apparatus

Ion-exchange columns of conventional design were used that accommodate a resin bed of ca. 11 x 180 mm. The columns should be fitted with joints to accommodate a Teflon stopcock joint assembly and a 250-mL solvent reservoir.

For TLC apparatus, any suitable equipment to handle 20 x 20 cm glass plates can be used, including micropipettes, spotting template, desiccating storage cabinet and developing tank.

Reagents

Disulfine Blue VN-150 (Acid Blue 1, C.I. 42045) or Patent Blue VF Hi Conc were obtained from American Hoechst Corp., Dyes and Pigments Division. The anion exchange resin was Bio-Rex 9, 50-100 mesh, chloride form, obtained from Bio-Rad Laboratories. For cation exchange resin, we used Amberlyst 15 from Rohm and Haas Co. Neodol 45-7, an ethoxylated fatty alcohol, was purchased from the Industrial Chemicals Division of Shell Chemical Co. Distearyl dimethyl ammonium chloride was of high purity and of known percent active. Solvents and acids were reagent grade.

Solutions

Neodol 45-7 (1,000 mg L^{-1}) (1.00 g, 100% active basis) was dissolved in 1 L distilled water. Distearyl dimethyl ammonium chloride (DSDMAC) was accurately weighed (0.1000 g, 100% active basis), dissolved in chloroform and brought to a volume of 100 mL with chloroform. This was stock solution A and contained 1,000 μg mL^{-1} of DSDMAC. Stock solution A (10 mL) was pipetted into a 100-mL volumetric flask and brought to volume with chloroform. This was diluted stock solution B containing 100 μg mL^{-1} DSDMAC. We pipetted 10 mL of diluted stock solution B into a 100-mL volumetric flask and brought to volume with chloroform. This was the working standard solution containing 10 μg mL^{-1} DSDMAC.

Acidic (HCl) methanol was prepared by adding 6 vol of concentrated hydrochloric acid to 150 vol of methanol; it was mixed well and stored in a stoppered glass container.

Disulfine blue stock solution was prepared from a mixture of 30 mL ethyl alcohol and 270 mL distilled water. Disulfine blue powder (0.250 g) was dissolved in the aqueous ethanol and brought to a volume of 250 mL with the alcohol-water mixture.

Sulfuric acid (10% by vol) was made by cautiously adding 50 mL concentrated sulfuric acid to ca. 400 mL distilled water in a 500-mL volumetric flask. The acid was allowed to cool to room temperature and was brought to volume with distilled water.

For 25% sulfuric acid, we slowly added one part concentrated sulfuric acid to 3 parts chilled, distilled water.

Disulfine blue working solution was prepared by pipetting 20 mL of the disulfine blue stock solution into a 500-mL volumetric flask containing ca. 250 mL distilled water. We added 50 mL of the 10% (by vol) sulfuric acid solution and brought to volume with distilled water. After transferring this solution into a 1-L separatory funnel, 250-mL distilled water was added. Five mL of the DSDMAC diluted stock solution B was added by pipet. Chloroform (50 mL) was added and the funnel was shaken vigorously for 1 min. The layers were allowed to separate and the lower chloroform layer was discarded. The chloroform extraction was repeated three additional times, and each time the extracts were discarded. Disulfine blue working solution was stored in a glass container.

Preparation of Ion-Exchange Columns

The Bio-Rex 9 and Amberlyst 15 ion-exchange resins were conditioned by covering with methanol and allowing to stand 30 min with occasional stirring. A plug of glass wool was placed in the bottom of an ion-exchange column, ca. two-thirds of the column depth was filled with methanol and then the preconditioned resin was added until a resin bed depth of 160-180 mm was attained. The prepared column was rinsed with 100 mL methanol.

Sample Preservation/Stabilization

Five mL of the 1,000 mg L^{-1} Neodol 45-7 solution and 10 mL of formaldehyde (37% solution) were added to the sample container for each 1 L of sample at the time of collection. This procedure applies to tap water, river water, sewage influent and effluent samples. Sewage sludge and river bottom sediment samples were preserved by addition of 50 mL formaldehyde solution to each 1 L of sample.

DBAS Procedure

The first step in the procedure was to evaporate an appropriate volume or weight of the sample to dryness on a steam bath under a stream of nitrogen or clean, dry air. The sample should contain 5-500 μg of DBAS. The following schedule could be used as a guide.

Type of sample	Volume or weight	Beaker size for final evaporation (mL)
Sewage influent	100 mL	200
Sewage effluent	200 mL	200
River water	1000 mL	200
Tap water	2000 mL	200
Sewage sludge (wet)	1.0-1.5 g	50
River sediment (dry)	1.0-1.5 g	50

Effluent samples were added in increments as evaporation proceeded. River and tap water samples were evaporated in increments to a volume of 75-100 mL in a 500-mL Erlenmeyer flask by gently boiling on an electric hot plate with two boiling stones and 5-mL concentrated hydrochloric acid being added prior to the start of the concentration. Following concentration, the sample was transferred into a 200-mL beaker and the flask was rinsed with two 20-mL portions of methanol into the beaker. Alternatively, river and tap water samples could have been concentrated to 75-100 mL using a rotating vacuum evaporator at 45 C. The concentrated sample was then cautiously acidified with 5 mL concentrated hydrochloric acid, transferred to a 200-mL beaker, and the evaporator flask was rinsed with methanol into the beaker.

Following evaporation of the sample to dryness, 20 mL of the acidified methanol solution was added and all of the residue from the sides and bottom of the beaker was thoroughly dislodged with a spatula. The solution was placed over live steam on the steam bath until the volume was reduced to ca. 10 mL; it was then swirled and the contents of the beaker were immediately poured into a

centrifuge tube. A second 20-mL portion of the acidified methanol solution was added to the beaker which was replaced on the steam bath, but not over live steam. The first extract was centrifuged at maximal speed for several minutes and then the supernatent liquid was transferred into a 150-mL extraction flask with a disposable pipet.

Approximately 2 mL of the second extract was pipetted into the centrifuge tube with the same pipet and the residue in the bottom of the tube was dislodged by drawing liquid in and out of the pipet several times. The pipet was rinsed with the second extract, which was added to the centrifuge tube. The remainder of extract was transferred into the tube, centrifuged and transfer operations were performed as described for the first extract. This operation was repeated a third time.

The combined acidic methanol extracts were evaporated to dryness on the steam bath under a stream of nitrogen or air. The residue in the flask was transferred into a 250-mL separatory funnel using 100 mL distilled water in ca. 25-mL portions, adding 5 mL concentrated hydrochloric acid to the final water rinse. The flask was rinsed with 50 mL chloroform, which was added to the contents of the funnel. The funnel was shaken vigorously for 1 min, layers were allowed to separate, and the lower chloroform layer was drained into a clean, dry 150-mL extraction flask. The chloroform rinse of the original flask and extraction of the aqueous solution were repeated with two additional 50-mL portions of chloroform. The combined chloroform extracts were evaporated to dryness.

Chloroform extractable material was dissolved in ca. 2 mL chloroform and 10 mL of methanol and then poured into the ion-exchange column containing the Bio-Rex 9 anion exchange resin. The stopcock was opened to collect the column effluent in a clean, dry 150-mL extraction flask at a rate of ca. one drop/sec. The flask was rinsed with several additional portions of methanol, with the rinses added to the column each time the liquid level was ca. 1 cm above the top of the resin bed. The solvent reservoir was attached to the top of the column and we continued passing methanol through the column until ca. 130 mL of effluent had been collected. This was placed on a steam bath and evaporated to dryness.

The column effluent residue was dissolved in 10 mL chloroform, then added to a 250-mL separatory funnel containing 70 mL of the disulfine blue working solution. The mixture was shaken vigorously for 1 min and the layers were allowed to separate. The lower chloroform layer was drained into a 20-mL vial which was placed on the steam bath under a stream of nitrogen to evaporate. Chloroform extraction was repeated two more times, using each 10-mL portion of chloroform to rinse the sample flask prior to addition to the separatory funnel. Each of the chloroform extracts was added to the vial followed by evaporation to dryness. Ten mL of methanol was pipetted into the vial, which was capped, and then the contents were mixed by shaking. Absorbance of the methanol solution was measured in a 10-mm cell at 625 nm using a methanol blank to balance the spectrophotometer at zero absorbance. Micrograms of DBAS were read in terms of the calibration standard from the *calibration curve* prepared as described in the following section. Increased sensitivity for samples containing less than 10 μg of DBAS may be achieved by dissolving the residue in the 20-mL vial in 5 mL of methanol; samples containing more than 100 μg of DBAS must be diluted as described under "Preparation of Calibration Curves."

Preparation of Calibration Curves

Since the DBAS level in the various types of samples may vary considerably, calibration curves covering the 0-100 μg and 100-500 μg DSDMAC ranges were required. We prepared the 0-100 μg curve by adding 0, 20, 50 and 100 μg of DSDMAC (0, 2, 5 and 10 mL of the working standard solution) to four 250-mL separatory funnels, each containing 70 mL of the disulfine blue working solution, with sufficient fresh chloroform added to make 10 mL chloroform volume; we proceeded as described in the final paragraph under "Procedure." A calibration curve was constructed by plotting absorbance vs μg of DSDMAC.

The 100-500 μg DSDMAC calibration curve was prepared by using 100, 300 and 500 μg DSDMAC (1, 3 and 5 mL of diluted stock solution B) in the manner described. The residue was dissolved in the 20-mL vial in 10 mL of methanol and then 5 mL of the methanol solution was pipetted into a 25-mL volumetric flask. The flask was brought to volume with methanol and absorbance of the diluted solution was measured. All samples that fell within this range likewise had to be diluted five-fold prior to the absorbance measurement.

Calculations

$$\mu g \text{ DBAS/L} = \frac{\mu g \text{ from calibration curve} \times 1000}{\text{sample volume (mL)}}$$

$$\text{mg DBAS/L} = \frac{\mu g \text{ from calibration curve}}{\text{sample volume (mL)}}$$

$$\mu g \text{ DBAS/g} = \frac{\mu g \text{ from calibration curve}}{\text{sample weight (g)}}$$

TLC Procedure

Methanol solution containing the DBAS was transferred into the chromatographic column containing Amberlyst 15 cation exchange resin. For samples containing less than 100 μg of DBAS, the solution used for the absorbance measurement had to be returned to the 20-mL vial and the absorbance cell rinsed with methanol into the vial. For samples containing 100-500 μg of DBAS, the 5 mL of methanol solution remaining in the 20-mL vial was used for this chromatographic separation, with the μg of DBAS used in "Calculations" being one-half of the determined μg of DBAS. Using standard techniques, we rinsed the 20-mL vial with several portions of methanol and continued rinsing the resin column with methanol until at least 125 mL of effluent had been collected. The effluent was discarded and a clean, 150-mL extraction flask was placed under the column. The column was eluted with 100 mL of acidified methanol followed by 20 mL methanol at a rate of ca. one drop/sec. The eluate was evaporated to dryness on a steam bath under a stream of nitrogen or air. The column should be rinsed with 100-125 mL methanol before reusing. The eluate residue was transferred with several small portions of chloroform into a 2-dram vial and the chloroform was removed by evaporation.

The residue was dissolved in the 2-dram vial in twice as many μL of chloroform as the determined μg of DBAS, except as noted above. A 10μL aliquot of the chloroform solution was spotted along with 1, 2, 3, 4 and 5 μL of the DSDMAC stock solution A, 2 cm from the bottom and 1.5 cm apart on a 20 x 20 cm Silica Gel G plate. The plate was developed to a distance 1 in. from the top of the plate in a solvent system of chloroform, methanol and water (75:23:3). The R_f of $DSDMA^+$ is 0.48. Visualization was by spraying and charring using 25% sulfuric acid. The intensity and size of the sample spot were compared with those of the standards and the μg quantity of DSDMAC in the sample was estimated.

Calculations

$$\% \text{ DSDMAC in total DBAS} = \frac{\mu g \text{ DSDMAC found by TLC} \times 100}{\mu g \text{ DBAS}}$$

DISCUSSION

Sample Preservation/Stabilization

Early in this investigation, ^{14}C methyl-labeled DSDMAC was added at the 5 $\mu g\ L^{-1}$ level to tap and stream waters and at the 25 $\mu g\ L^{-1}$ level to sewage treatment plant primary influent and final effluent samples at the time of collection at four different sites. Liquid-scintillation counting of the samples, carried through the anion exchange step of the procedure, showed erratic recoveries of the radiolabeled DSDMAC, with most recoveries being less than 50%.

Most of the "missing" tagged material was found in methanol rinses of the empty sample containers, giving evidence that the low and erratic recoveries were attributable to adsorption of the labeled DSDMAC on the surfaces of the sample container. Further testing showed that surface adsorption occurred with both glass and plastic containers, the adsorption progressing with time.

A finding that an ethoxylated alcohol, such as Neodol 45-7 (Shell Chemical Co. Industrial Chemicals Division) was effective as an additive promoting the "solubility" of DSDMAC in aqueous solutions suggested that addition of this material to a sample at the time of collection might minimize losses due to adsorption on sample container surfaces. Samples were collected at five locations. Five mg of Neodol 45-7 and 10 mL formaldehyde solution/L of sample were added to the container prior to the addition of the sample and radiolabeled DSDMAC at levels described above. Liquid-scintillation counting of these samples, carried through the anion exchange step of the DBAS procedure, showed recoveries to range from 70-100%, with the average recovery for 61 determinations being 90%. No significant difference in average recovery was found for the four different sample types. Further, no difference was found between samples analyzed in the first week after collection and replicates analyzed 2 months after collection. These findings, supported by the finding of insignificant levels of labeled DSDMAC in methanol rinses of the empty sample containers, confirmed that the addition of Neodol 45-7 provided an effective means of minimizing, if not eliminating, losses of DSDMAC due to adsorption on container surfaces.

Acidified Methanol Extraction

The initial methanol extraction step of the Waters and Kupfer method has been replaced with an acidified methanol extraction. This procedural modification was made to effect removal of DSDMAC bound to clays and clay-like materials such as would be found in stream and stream sediment samples. Clays contain significant levels of polymeric silicates that carry negative charges and act in much the same manner as cation exchange resins. Acidified methanol was found to effect complete removal of DSDMAC bound to sediments. Incomplete recovery of added DSDMAC resulted when hot, unacidified methanol was used as the extractant.

Chloroform Extraction

The acidified methanol extractables from most samples include appreciable inorganic chloride that, if not removed, would (a) pass through the anion exchange resin and (b) result in partial elution of anionic surfactants from previous samples for which the resin column had been used. The effect of (a) could be the possible introduction of chlorides into the colorimetric step of the DBAS method and, as shown in Table I, cause a negative interference with the colorimetric measurement. A chloroform extraction step has been added to the original method to eliminate inorganic chlorides.

Further, a test designed to demonstrate the effect of (b) was conducted by carrying duplicate samples of DSDMAC in distilled water and saline solution through the DBAS procedure. In this test, the chloroform extraction step was omitted from one of each of the duplicate samples and the anion exchange resin, for each sample, was pretreated with LAS (linear alkylbenzenesulfonate). The conditions and results, given in Table II, demonstrate the additional necessity for removal of inorganic chlorides.

TABLE I

Effect of Chloride on Disulfine Blue Colorimetric Response

Test surfactant	Chloride (Cl^-) added[a]	% Response
100 µg DSDMAC	0 mg	100
↓	6	72
	30	40
	60	25
	120	17

[a]Chloride added to disulfine blue working solution + DSDMAC.

TABLE II

Elimination of Negative Chloride Effect by Chloroform Extraction

DSDMAC	Matrix 100 mL	$CHCl_3$ extraction	LAS on Bio-Red 9 resin	DSDMAC recovery
100 µg	Distilled H_2O	Yes	50 µg	112%
↓	Saline solution[a]	Yes	↓	97
	Distilled H_2O	No		93
	Saline solution[a]	No		47

[a]Saline solution contained 0.175 g Cl^-.

TABLE III

LAS Removal by Unconditioned Bio-Rex 9 Anion Exchange Resin

Resin condition	DSDMAC (µg)	LAS (µg)	Mol /LAS/ mol DSDMAC	DSDMAC found (µg)
Used column	100	200	3.4	102
↓	50	4000	136	49
	20	3000	255	21
	10	5000	850	11
Fresh column	100	0		101
↓	100	200	3.4	100
	100	500	8.5	101
	100	800	13.6	97
	50	200	6.8	50
	20	200	17.0	20

Anion Exchange Resin

Bio-Rex 9 resin was found to be as effective for removing anionic surfactants from methanol solution as is Bio-Rad

TABLE IV

Typical DBAS Reproducibility Data

Sample type	mg L^{-1} or $\mu g\ g^{-1}$*
Influent	2.3; 2.4
↓	1.8; 1.9
	1.2; 1.2
	0.85; 0.84
↓	0.71; 0.75
Final effluent	0.06; 0.05
↓	0.23; 0.26
	0.22; 0.27
↓	0.04; 0.05
River water	0.03; 0.04
↓	0.04; 0;04
	0.04; 0.03
Digester supernatent liquor	4.2; 4.5
Primary sludge*,[a]	11.4; 10.8
	143; 156
	220; 235
Digester sludge*,[a]	470; 490
	420; 410
River sediment*,[b]	20; 21
↓	34; 38
↓	44; 45

[a]Wet weight basis.
[b]Dry weight basis.

TABLE V

Typical Recoveries of Added DSDMAC

Sample type	Added	Found	% Recovery
Influent	50 μg	45 μg	90
↓	50	40	80
	50	48	96
Final effluent	40	48	120
River water	50 μg	41 μg	82
River sediment	40 μg/g	44 μg/g	110
	40	30	75
Primary sludge	68	83	122
	82	98	119

AGl-X2, the anion-exchange resin used in the Waters and Kupfer procedure. However, the Bio-Rex 9 resin does not need preconditioning with a cationic surfactant as Waters and Kupfer found necessary with Bio-Rad AGl-X2. The results in Table III show excellent removal of LAS and recovery of DSDMAC with Bio-Rex 9 resin.

Stability of Disulfine Blue Extracts

The problems of color instability and adsorption of the disulfine blue-cationic compound on optical cells from chloroform solution, experienced by Waters and Kupfer, have been overcome by the use of methanol as described in the procedure. Although the combined chloroform extracts of both standards and samples may turn green, yellow or colorless during evaporation, the blue color is regenerated in methanol. The color of the methanol solution of the disulfine blue-cationic compound does not adsorb on optical cells and has been found to maintain its stability over a period of at least 8 days.

TABLE VI

Typical TLC Reproducibility Data

Sample type	% $DSDMA^+$
Influent	50; 50
↓	50; 60
	40; 40
Final effluent	60; 40
	60; 80
Primary sludge	40; 40
Digester sludge	30; 30
Digester supernatent liquor	40; 50
	30; 40
River sediment	30; 30
↓	20; 20
	30; 50
River water	20; 40

Precision and Accuracy

The precision of the DBAS method is shown by the reproducibility data for different sample types presented in Table IV. The accuracy of the method, based on recoveries of DSDMAC added to various sample types, is indicated by the results given in Table V. The duplicate values given in Table VI are an indication of the reproducibility of the TLC procedure. These values were obtained on duplicate samples and represent a semiquantitative estimate of the percentage of DBAS that is attributable to the $DSDMA^+$ cation.

ACKNOWLEDGMENT

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