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HPLC/ESI-Quadrupole Ion Trap Mass Spectrometry for Characterization and Direct Quantification of Amphoteric and Nonionic Surfactants in Aqueous Samples

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An amphoteric (cocamidopropylbetaine, CAPB) and a nonionic (alcohol polyethoxylate, AE) surfactant were characterized by electrospray ionization quadrupole ion trap mass spectrometry (ESI-MS) as to their homologue distribution and ionization/fragmentation chemistry. Quantitative methods involving reversed-phase gradient HPLC and (+)ESI-MS^n were developed to directly determine these surfactants in hydroponic plant growth medium that received simulated graywater. The predominant homologues, 12 C alkyl CAPB and 9 EO AE, were monitored to represent the total amount of the respective surfactants. The methods demonstrated dynamic linear ranges of 0.5−250 ng (r^2 > 0.996) for CAPB and 8−560 ng (r^2 > 0.998) for AE homologue mixture, corresponding to minimum quantification limits of 25 ppb CAPB and 0.4 ppm AE with 20-μL injections. This translated into an even lower limit for individual components due to the polydispersive nature of the surfactants. The procedure was successfully employed for the assessment of CAPB and AE biodegradation in a hydroponic plant growth system used as a graywater bioreactor.

Just as in Earth-based homes, graywater (nontoilet wastewater) is projected to be the largest waste stream (by mass) generated in long-term crewed space habitats. The estimate of graywater production within space systems is 27 L person^{-1} day^{-1}, which comprises 80% of the total waste output. It is imperative that this wastewater is regenerated in spacecrafts where the water resource is limited and resupply is costly. The National Aeronautics and Space Administration's (NASA) Advanced Life Support (ALS) program is examining the feasibility of developing regenerative life support systems including physicochemical and biological processes, for extended space missions. Our group has been evaluating an approach for recycling graywater through a hydroponic plant production system that utilizes (1) microbial biodegradation of surfactants in the plant rhizosphere and nutrient delivery system and (2) plant transpiration to purify water. The key to the success of such a system is the effective removal of potentially phytotoxic surfactants in the graywater. This requires the close monitoring of the surfactant concentration in the system.

However, the selective determination of nonchromophoric and polydispersive surfactants in environmental samples has always been a challenge. This challenge is mainly due to the difficulty in the separation of structurally similar homologues or oligomers and the detection of nonvolatile and non-light-absorbing molecules. A complex sample matrix such as hydroponic growth medium containing high levels of inorganic and organic substances further compounds the difficulty. There have been many parallel efforts in the separation,[1−5] detection,[1,6−14] and sample enrichment[8,15−17] techniques for determination of surfactants in a variety of matrixes. Until the recent advances in commercial HPLC/MS instruments, the baseline separation of both alkyl homologues and their polyethoxylate oligomers and their sensitive detection without labor-intensive and solvent-laden derivatization[2,17] has remained a problem.

The successful hybridization of HPLC and mass spectrometry reduced the requirement for the chromatographic baseline

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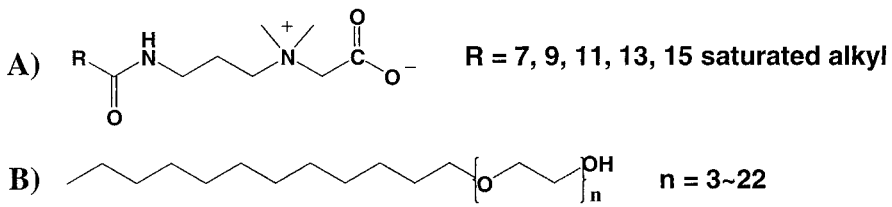

Figure 1. Chemical structures of cocamidopropylbetaines (A) and polyoxyethylene 10 lauryl ether (B).

separation of analytes due to the additional postcolumn separation based upon the analyte ions' mass-to-charge ratio (m/z). In addition, structural information could be obtained through the use of tandem mass spectrometry (MS/MS). The number of applications of HPLC/MS to the determination of surfactants and their degradation intermediates in environmental samples has increased steadily.[9−14] To date, all the reported work has been limited to the analysis of extracted samples and mostly for the determination of nonionic surfactants with a single, linear quadrupole mass spectrometer. To the authors' knowledge, there has only been a single report on the determination of amphoteric surfactants via HPLC/ESI-MS, which was performed by using a single, linear quadrupole mass spectrometer in the negative ESI mode.[18] Our study is the first to explore the use of quadrupole ion trap mass spectrometry and tandem mass spectrometry to characterize and quantify intact amphoteric and polyethoxylated nonionic surfactants. Furthermore, the objective of this study is to establish a direct quantification protocol with no sample enrichment or precleanup prior to instrument analysis, thus to reduce the generation of solvent waste.

EXPERIMENTAL SECTION

Chemicals. Cocamidopropylbetaine (CAPB), an amphoteric surfactant, is the main active ingredient (30.66%) in Miratraine Bet C-30 from Rhodia Inc. (Cranbury, NJ). Polyoxylethylene 10 lauryl ether, an alcohol polyethoxylate (AE) nonionic surfactant, was purchased from Sigma-Aldrich (St. Louis, MO). The chemical structures of the surfactants are shown in Figure 1.

Organic solvents and other reagents were either HPLC grade or analytical reagent grade. Deionized water was further purified by a Barnstead (Dubuque, IA) 4 module E-pure system, which renders organic-free water with resistivity of greater than 18 mΩ.

Sample Collection and Preparation. Simulated graywater streams (SGW) were prepared by dissolving a predetermined amount of Rhodia Miratraine BET C-30 or Sigma polyoxylethylene 10 lauryl ether in deionized water to reach a desired concentration of CAPB and AE. Wheat (*Triticum aestivum* L. cv. Apogee) plants were grown in recirculating modified half-strength Hoagland's solution (Table 1) via a nutrient film technique (NFT).[19,20] Beginning 4 days after planting (DAP), some of the seedlings were exposed to Hoagland's solution supplemented with SGW containing either CAPB or AE. In the first experiment, SGW containing either AE or CAPB was added to maintain a preset liquid level in nutrient reservoirs, so the SGW input varied (1.8−4.8 g of surfactant m^{-2} of growing area day^{-1}) as plants grew and evapotranspiration increased. In the second experiment, SGW was added at a constant rate of 2 g of surfactant m^{-2} day^{-1}. This input rate would result in a surfactant concentration of ∼33 mg L^{-1} within 24 h if no degradation occurred. The hydroponic nutrient solution amended with SGW was sampled throughout the experiment to examine the persistence of the surfactants and their impact on plant growth. At 20 DAP (in both experiments), a single pulse of SGW was added to nutrient reservoirs to reach a targeted AE or CAPB concentration of 100 mg L^{-1}. One-milliliter samples were withdrawn from the reservoirs at intervals. All samples were filtered through a 0.2-μm nylon Acrodisc HPLC syringe filter (Pall Gelman) into an HPLC autosampler vial and subjected to instrument analysis without further manipulation immediately or stored in a −25 °C freezer until analysis.

Table 1. Elemental Composition of the Half-Strength Modified Hoagland's Solution

macronutrient	NO_3	PO_4	K	Ca	Mg	SO_4
concn (mM)	7.5	0.5	3.0	2.5	1.0	1.0
micronutrient	Fe	Mn	Zn	Cu	BO_4	Mo
concn (μM)	60	7.4	0.96	1.04	7.13	0.01

Liquid Chromatography. Liquid chromatography was carried out with a Thermo Separation Products (San Jose, CA) SpectraSystem HPLC equipped with a quaternary pump (P4000) and an autosampler (AS 3000). Chromatographic separation was performed on an Alltima C18 column (5-μm particle size, 2.1 mm × 250 mm) preceded by a guard column (4 × 4 mm, 5-μm particle) of the same packing from Alltech (Deerfield, IL). Mobile phase A was either water containing 5% methanol and 3% acetic acid for CAPB analysis or water containing 5% methanol and 30 mM ammonia acetate buffer for AE analysis. Mobile phase B was 100% acetonitrile for both CAPB and AE analyses. The HPLC flow rate was 0.2 mL min^{-1}. For separation of CAPB homologues, a mobile-phase gradient was used with the percentage of B varying as follows: 10% for 5 min, linear increase to 70% over 15 min, and then maintenance at 70% for 30 min. For separation of AE homologues, the percentage of B was varied as follows: 10% for the first 5 min, linear increase to 70% min over 5 min, 70% for 35 min, linear increase to 90% over 5 min, and then maintenance at 90% for 5 min. Prior to each analysis, the percentage of B was decreased from the maximum of the previous gradient to the starting percentage of B over 10 min followed by a 7-min column equilibration period.

Mass Spectrometry. The effluent from the HPLC column was introduced to a Thermo Finnigan (San Jose, CA) quadrupole ion trap mass spectrometer (LCQ Deca) through an electrospray ionization interface (ESI) operated in the positive ionization mode. ESI parameters were optimized for each target compound to achieve a stable and high abundance of the parent ions. This was

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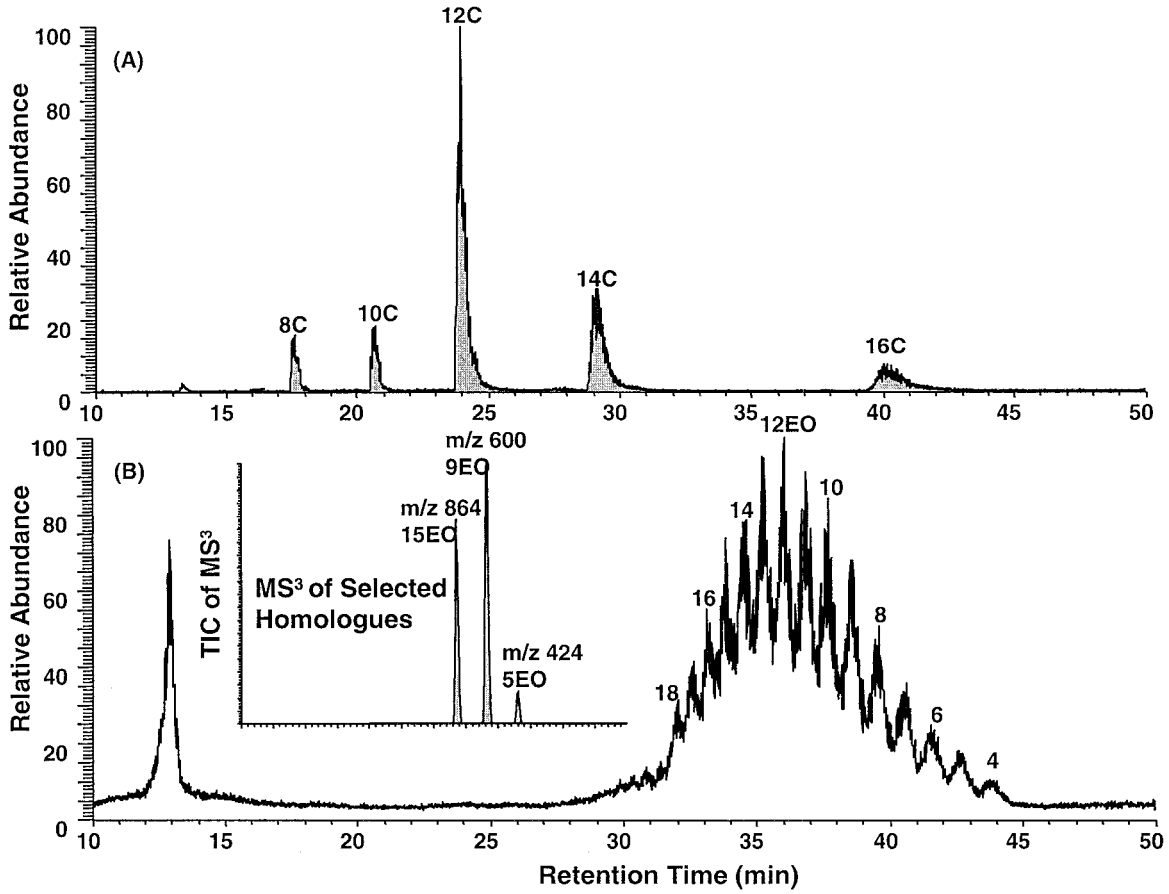

Figure 2. HPLC/(+)ESI-normal scan total ion chromatograms (TIC) of the CAPB homologues in mirataine BET C-30 (A) and of the AE homologues in polyoxylethylene 10 lauryl ether (B). The inset of (B) is a MS^3 TIC of the selected AE homologues

Table 2. ESI and MS/MS Parameters for Determination of 12 C Alkyl CAPB and AE with 9 EO Units

ESI interface parameters	12 C alkyl CAPB	AE with 9 EO
capillary temperature (°C)	260	200
capillary voltage (V)	9.0	8.0
ion spray voltage (kV)	5	5
tube lense offset (V)	25	10
shealth gas flow (arbitrary unit)	51	53
auxiliary gas flow (arbitrary unit)	17	3
MS^n parameters		
parent ion (m/z)	343	600
isolation width	2	2
activation energy[a] for MS^2	52%/0.25q/30 ms	30%/0.25q/30 ms
activation energy[a] for MS^3	n/a[b]	30%/0.25q/30 ms

[a] Activation energy is % CID/CID-q/CID time. [b] Not available.

accomplished by introducing analytes into mass spectrometer through direct infusion via the LCQ's syringe pump along with an HPLC flow (0.2 mL min^{-1}) at the solvent composition that the target compound was expected to elute from the column. Specifically, the optimized conditions for the determination of the predominant homologues of CAPB and AE are listed in Table 2. For the tandem mass spectrometric experiments, the parameters for parent ion isolation and collision-induced dissociation (CID) energy were also optimized to achieve the highest efficiency for the formation of desired product ions (Table 2). The selected product ion $[(M + H) - 103]^+$ from the protonated CAPB was monitored, while all the product ions between m/z 165 and 610 generated from MS^3 of the AE ammonia adducts $[M + NH_4]^+$ were monitored. The chromatographic retention time and MS^n spectra were used for positive identification of target components.

Quantitation. A series of standard solutions were prepared from Rhodia's Mirataine BET C-30 and Sigma's C12E10 AE in half-strength Hoagland's solution, and analyzed by HPLC/(+)-ESI-MS^n to establish an external calibration curve. A five-level calibration was carried out at the beginning and the end of each set of samples for each batch of mobile phase. A QC standard was also run after every 10−15 sample injections to ensure the stability of the system and validity of the calibration curve. A blank was routinely run between samples with low surfactant concentrations. Since pure individual homologues are not readily available, standard curves were generated from the responses of predominant homologues, 12 C alkyl CAPB and 9 EO unit AE, and the total amount of the respective surfactants. Qualitative and quantitative data were processed with Xcalibur software (version 1.1).

RESULTS AND DISCUSSION

Characterization and Determination of an Amphoteric Surfactant, CAPB. The chromatographic behavior of the zwitterionic CAPB homologues at neutral pH was similar to that of nonionic compounds; the CAPB homologues eluted in order of the increasing length of their hydrophobic alkyl chains (Figure 2A). The addition of acetic acid (a weak acid) to the mobile phase to facilitate ionization for MS detection did not affect the

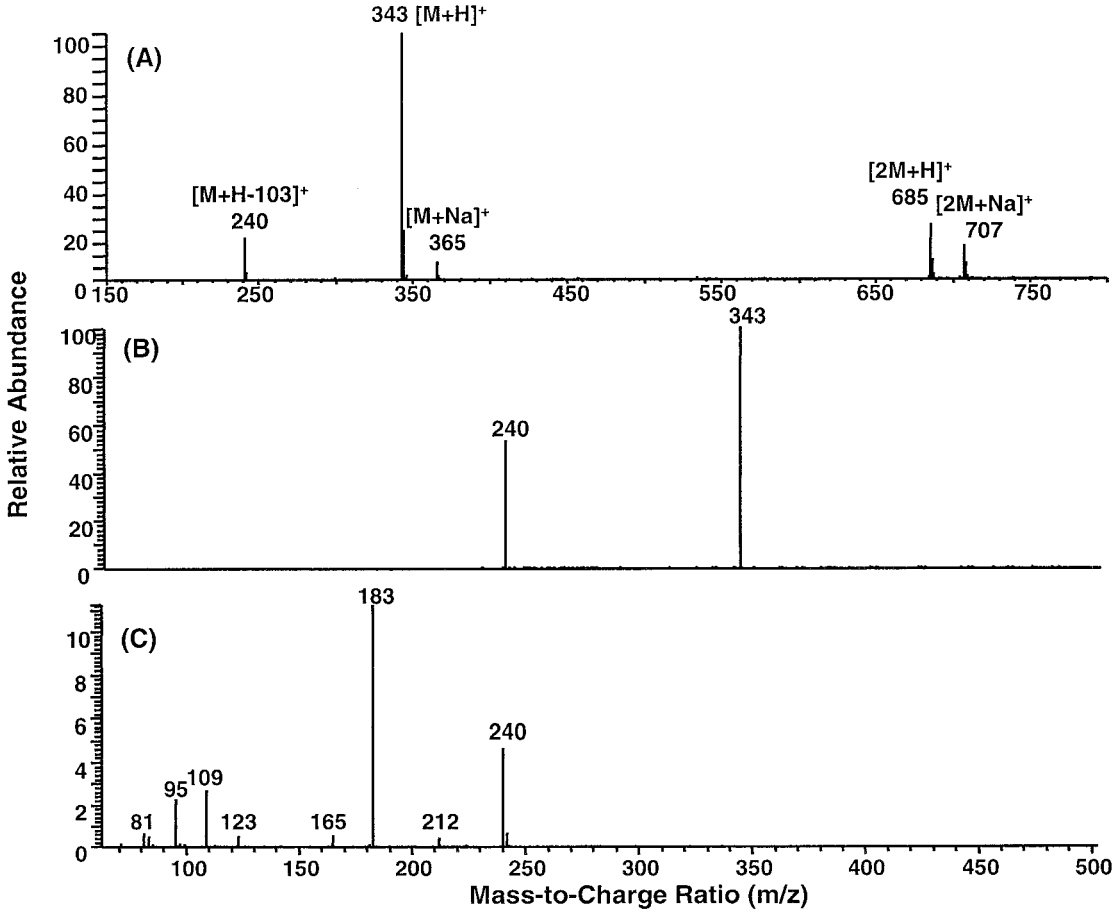

Figure 3. The (+)ESI normal mass spectrum of 12 C CAPB homologue (A), the MS/MS spectrum of its *m*/*z* 343 protonated molecule (B), and the MS^3 spectrum of the *m*/*z* 240 product ion generated from *m*/*z* 343 (C).

Table 3. CAPB Homologues and Their Major Ions Formed in (+)ESI Mode[a]

	8 C CAPB		10 C CAPB		12 C CAPB		14 C CAPB	
ions	*m*/*z*	RA[b]	*m*/*z*	RA[b]	*m*/*z*	RA[b]	*m*/*z*	RA[b]
$[M + H - 103]^+$	184	14.8	212	11.7	240	16.7	268	14.4
$[M + H]^+$	287	100.0	315	100.0	343	100.0	371	100.0
$[M + Na]^+$	309	23.4	337	25.7	365	8.4	393	19.2
$[M + H + M]^+$	573	5.6	629	3.7	685	21.2	741	10.2
$[M + Na + M]^+$	595	28.3	651	27.4	707	13.7	764	23.2
$[3M + H]^+$	859	0.7	943	0.4	1027	4.6	1111	1.9
$[3M + Na]^+$	881	1.9	965	0.7	1049	6.4	1133	3.0

[a] Values were the average of three analyses of 47.92 mg L^{-1} CAPB and 14 spectra of each analysis around peak apex (±0.1 min). [b] RA, percentage relative abundance.

characteristics of their interaction with column packing. Although separation of these homologues can be achieved by 70% acetonitrile under isocratic condition, a low percentage of organic solvent at the initial stage was necessary for samples containing inorganic ions to prevent precipitation of salt on the column. With the gradient program specified in the Experimental Section, inorganic salts contained in samples were eluted at the void volume. The early-eluting inorganic salts and nontarget compounds eluting during the first 10 min were diverted to waste via a the LCQ's six-position Rheodyne valve to prevent clogging of the heated capillary and buildup on the skimmer lenses of the electrospray interface.

The amphoteric surfactant homologues were protonated readily in the acidic mobile phase, producing predominantly their $[M + H]^+$ ions under the optimized (+)ESI conditions. In contrast, the corresponding signals in the negative ESI mode were much weaker in this mobile phase, which is true in a basic (pH 8) mobile phase reported by Eichhorn and Knepper.[18] Therefore, we focused our investigation on the positive ESI. Accompanying the protonated molecules, the sodiuated molecule, $[M + Na]^+$, protonated and sodiuated dimers, and respective trimers were formed to varying degrees (Figure 3A). Fragmentation of the $[M + H]^+$ ions also occurred during ESI or ion injection and resulted in $[(M + H) - 103]^+$ fragment ions due to the loss of dimethyl glycine (103 u). (+)ESI mass spectra of CAPB homologues were very similar with differences only in the relative abundance and masses (28 u apart) of these ions (Table 3). The difference in the relative intensity of various ions among homologues indicated that the

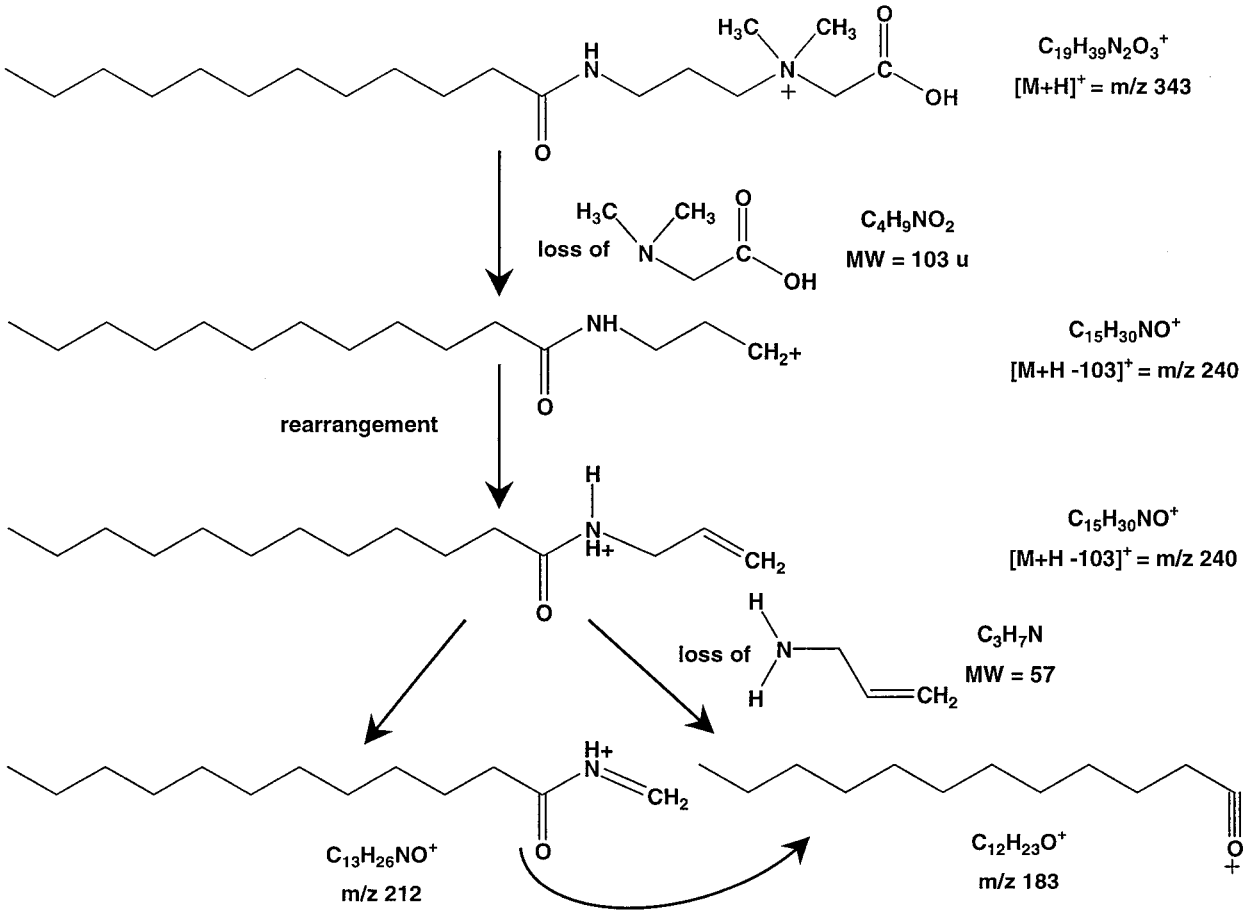

Figure 4. Probable mechanism of 12 C CAPB fragmentation in an ESI quadrupole ion trap mass spectrometer.

Table 4. Effect of CAPB Concentration on the Percent Relative Abundance (RA) of Various Ions Formed during (+)ESI[a]

ions	*m*/*z* of 12 C CAPB	RA for different CAPB concns (mg L^{-1}) 4.79	23.96	47.92
$[(M + H) - 103]^+$	240	11.1	13.0	16.7
$[M + H]^+$	343	100.0	100.0	100.0
$[M + Na]^+$	365	38.0	13.2	8.4
$[M + H + M]^+$	685	3.0	12.4	21.2
$[M + Na + M]^+$	707	40.4	21.2	13.7
$[3M + H]^+$	1027	<1.0	2.6	4.6
$[3M + Na]^+$	1049	1.2	5.5	6.4

[a] Values were the average of three analyses of each concentration and 14 spectra per analysis around the peak apex (±0.1 min).

alky chain length had a significant influence on the ionization process. This prohibited the estimation of homologue composition in the surfactant by directly using area percentage of their corresponding protonated molecules. The relative abundances of protonated dimers and trimers increased with surfactant concentration (Table 4). On the other hand, the sodiated ions were reduced, which may be due to the limited sodium ions in the mobile phase.

The protonated molecule, $[M + H]^+$, may be monitored for quantification in a selected ion monitoring (SIM) mode but lacked the structurally significant information needed for target compound identification in a complex mixture. Greater specificity and structural information were obtained through MS/MS. The MS/MS CID of the $[M + H]^+$ ions produced predominantly the $[(M + H) - 103]^+$ ions (Figure 3B), which is characteristic of glycinebetaine-type amphoteric surfactants. An increase in the CID energy did not enrich the spectrum. Consequently, the $[(M + H) - 103]^+$ ions were monitored in the SRM mode for quantification. Further CID of the product ion resulted in a MS^n spectrum (Figure 3C) that allowed us to propose a fragmentation pathway as shown in Figure 4.

Hydroponic plant growth medium representing a complex sample matrix contained not only various organic substances of both plant and microbial origins but also high levels of inorganic ions used as plant nutrients (Table 1). To determine the effect of inorganic ions within sample matrix on (+)ESI SRM response, three different concentrations of CAPB were prepared both in deionized water and in fresh half-strength Hoagland's solution. There were two preparations for each concentration, and two analyses of each preparation were performed by the HPLC/(+)-ESI-MS/MS in the SRM mode. The response obtained with Hoagland's solution was generally higher than that obtained with deionized water (Table 5). The enhanced response was greater for samples containing a lower concentration of the surfactant, but the change in signal due to the presence of inorganic ions was mostly less than 10% (Table 5). This minor matrix effect was corrected by preparing the calibration standards in the Hoagland's solution.

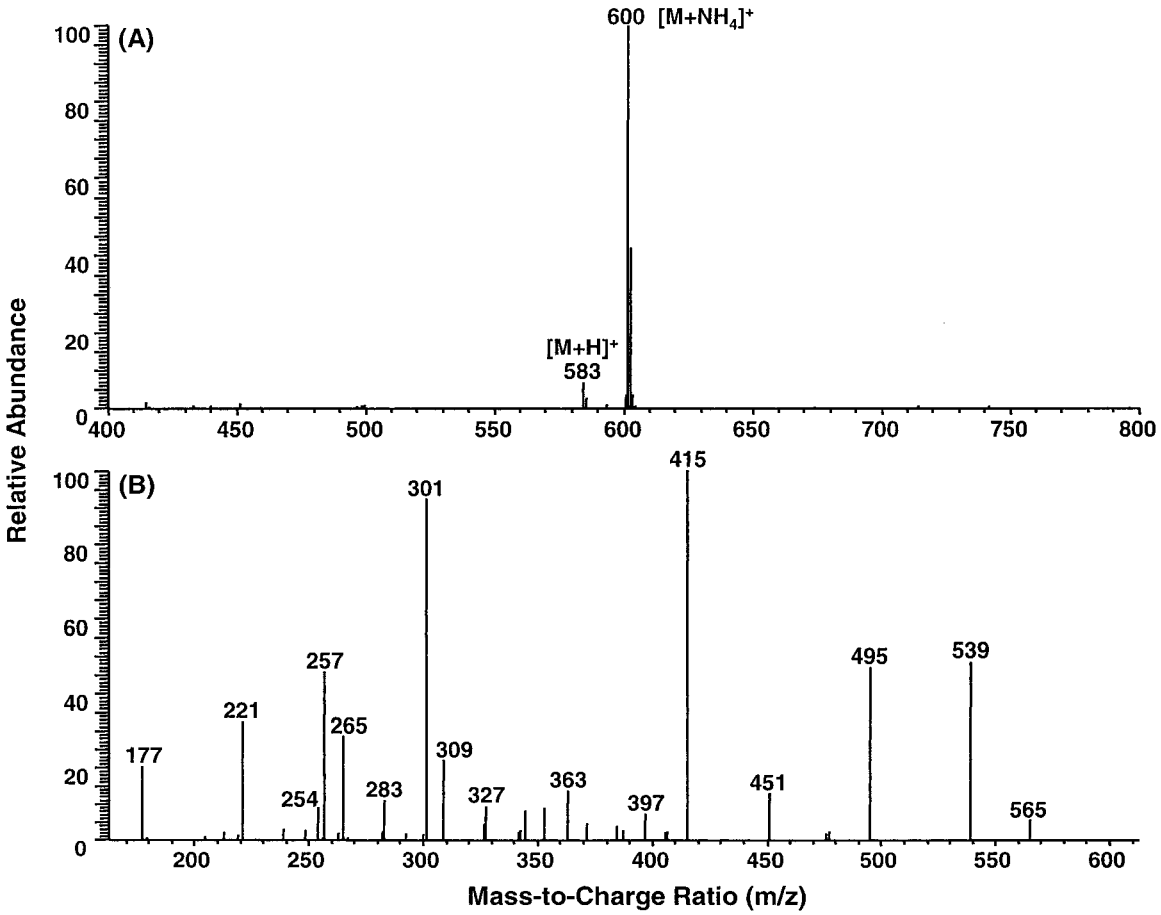

Figure 5. The (+)ESI normal mass spectrum of 9 EO AE (A) dominated by the $[M + NH_4]^+$ ion that undergoes CID to form the *m*/*z* 583 $[M + H]^+$ ion (spectrum not shown). The protonated molecule further undergoes CID to produce the MS^3 spectrum (B).

Table 5. Effect of Inorganic Ions on the Response of (+)ESI MS/MS SRM

bulk CAPB concn (mg L^{-1})	% change[a] observed by (+)ESI MS/MS SRM			
	8 C	10 C	12 C	14 C
5.03	9.3	7.9	10.6	2.9
25.13	4.9	2.3	4.4	0.6
50.25	2.5	0.8	−0.2	−0.4

[a] Percent change was calculated by the following equation: [peak area(Hoagland) − peak area(H_2O)] ÷ [peak area(H_2O)] × 100.

Table 6. Effect of Surfactant Concentration and Hydrophobicity of the Molecule on Partition between Water and the Liquid−Air Interface

bulk CAPB concn (mg L^{-1})	% reduction in aqueous phase after aspiration ($n = 2$)			
	8 C	10 C	12 C	14 C
5.03	31.2	26.3	27.6	46.4
25.13	4.4	8.5	25.1	57.9
50.25	−5.0	−0.3	14.0	61.1

Unlike other samples, extensive mixing of surfactant samples should be avoided because of their tendency to foam, and different homologues may foam to different extents. To investigate the effect of foaming, different concentrations of CAPB in half-strength Hoagland's solution were analyzed before and immediately after a 30-s aspiration with helium. Aspiration of the samples resulted in the partial transfer of surfactant molecules from the aqueous phase to the liquid−air interface (i.e., the foam). The lower the initial aqueous surfactant concentration, the greater was the percentage loss (Table 6). Furthermore, the longer alkyl chain homologues had a higher tendency to foam, resulting in greater losses. This behavior was directly related to the longer alkyl chains being more hydrophobic. Therefore, it is important to allow the foam to subside before sampling and analysis.

Due to the lack of individual homologue standards, calibration of the quantitative method was based on the response of the most abundant homologue (12 C alkyl component) in (+)ESI MS/MS SRM versus the total amount of the CAPB homologue mixture. The method provided a dynamic linear range between 0.5 and 250 ng with an R^2 greater than 0.996. The minimum quantification limit was ~0.5 ng of analyte injected on-column, which translated to a concentration of 25 ppb in a 20-μL sample. It was highly reproducible. Multiple injections of the same sample resulted in less than 1.5% RSD, and 1 week of continuous analyses exhibited less than 10% RSD.

Characterization and Determination of a Nonionic Surfactant, Alcohol Polyethoxylate. Studies have demonstrated that separation of AEs based on the number of ethylene oxide (EO) groups is better achieved under normal-phase chromatographic conditions.[1,3] However, normal-phase chromatography is not compatible with direct injection of aqueous samples. In this study, the combination of a reversed-phase C18 column and water/methanol/acetonitrile gradient provided adequate resolution be-

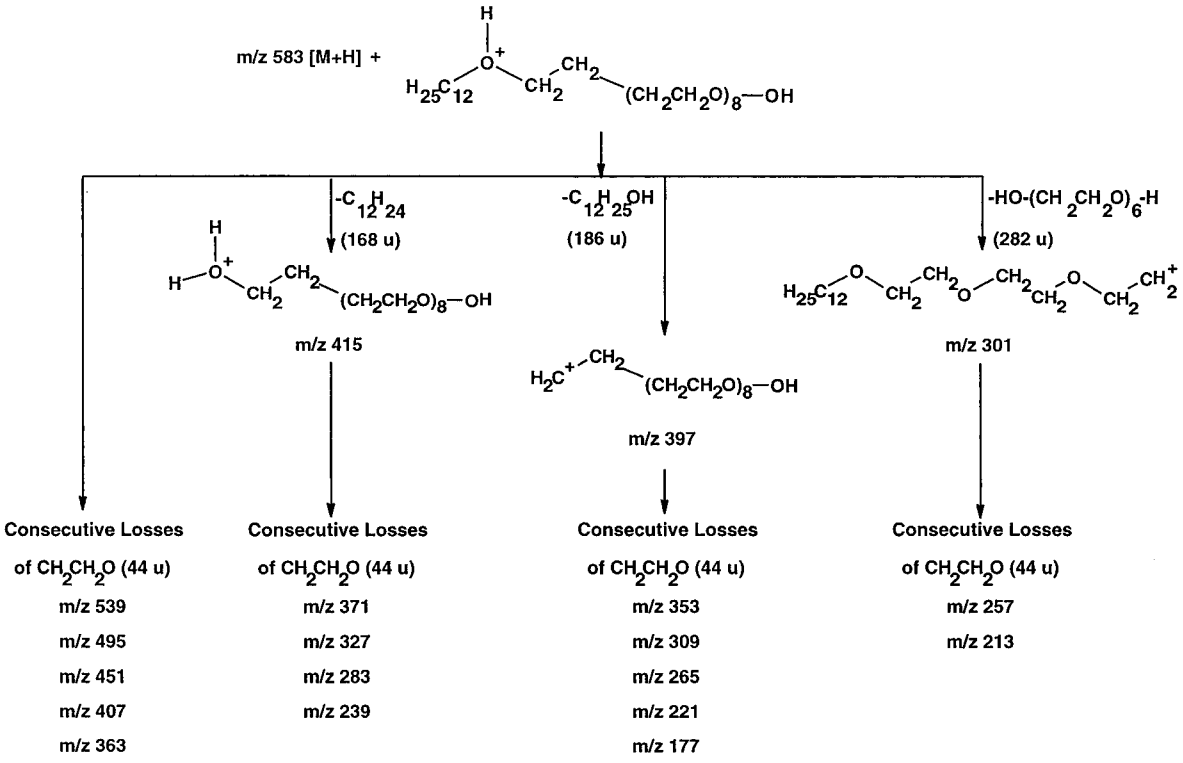

Figure 6. Probable fragmentation pathway for the (+)ESI-MS^3 of the *m/z* 583 $[M + H]^+$ ion generated from the MS/MS of the *m/z* 600 $[M + NH_4]^+$ ion of the 9 EO AE.

tween the AE homologues, which eluted from the column in the order of decreasing EO number (Figure 2B). On-line HPLC/MS and direct infusion analyses of the standard material also revealed that the polyoxyethylene 10 lauryl ether consisted of homologues with ethoxylate units ranging from 3 to 22.

The AE had a greater tendency to form the $[M + NH_4]^+$ adduct ion than its protonated molecule $[M + H]^+$. The AE adducts NH_4^+ so efficiently that the $[M + NH_4]^+$ ions were formed even when no NH_4^+ was intentionally added to the mobile phase, although there may have been trace residual NH_4^+ ions from previous analyses. Therefore, the mobile phase was modified with ammonium acetate to provide a consistent source of NH_4^+ ions and enhance the formation of the $[M + NH_4]^+$ ions and thus the sensitivity. The addition of 30 mM ammonium acetate to the aqueous mobile phase A increased the MS responses to different degrees for homologues with different EO units. The signal increased 26%, 11%, and 140% for the AE (bulk concentration of 30.5 mg L^{-1}) with 15, 9, and 4 EO units, respectively. The relative intensity of $[M + NH_4]^+$ to $[M + H]^+$ ions increased as the number of EO units increased. This could be attributable to the fact that the AE with a higher number of EO units adducts NH_4^+ more readily than those with a lower number of EO units. As a result, when compared to the system with limited ammonium ions, the increase in sensitivity with added ammonium ions is greater for the AEs with the lower number of EO units.

The $[M + NH_4]^+$ ions readily undergoes CID, producing the $[M + H]^+$ ions with high efficiency due to a neutral loss of NH_3 (17 amu). Further tandem mass spectrometry (i.e., MS^3 of the $[M + NH_4]^+$ ions) resulted in structure-specific fragment ions such as $[M + H - C_2H_4O]^+$, $[M + H - C_{12}H_{24}]^+$, $[M + H - C_{12}H_{25}OH]^+$ and other ions following consecutive losses of CH_2CH_2O from these (Figures 5 and 6). The (+)ESI-MS^3 spectrum rendered great selectivity or specificity for target analytes; thus, the spectrum and total ion count were used for qualitative and quantitative assessment, respectively, of the AE.

In theory, all homologues may be quantified in a single analysis. However, due to the time required for each MS^3 scan and the great number of AE homologues, this would have resulted in poorer sensitivity and inadequate sampling. To augment the sensitivity, only three representative homologues with 15, 9, and 5 EO units were monitored routinely (Figure 2B inset). These three homologues exhibited linear correlation between their MS^3 responses and the mixture concentration with R^2 values greater than 0.997 for all three homologues in a five-level calibration with two analyses per level. Since the correlation was established between MS responses of the individual homologues and the total amount of homologue mixture, the dynamic linear range was dependent on the relative abundance of the homologue in the mixture and the efficiency of its signal generation by (+)ESI-MS^3. The AE with 9 EO units had the highest response under the experimental conditions and was used for estimation of the concentration of homologue mixture in samples. The lower and upper quantification limits were approximately 8 and 560 ng of the AE mixture injected (corresponding to concentrations of 0.4 and 28 mg L^{-1} with 20-μL injection).

As opposed to analysis of the amphoteric surfactant, inorganic ions present in sample matrix had negligible influence on the MS signal (data not shown).

Application of the Method To Monitor the Biodegradation of CAPB and AE in a Hydroponic Plant Growth System. Biodegradation of CAPB and AE in the hydroponic plant growth system was studied by the determination of intact surfactant molecules using the HPLC/(+)ESI-MS^n methods described above. Wheat (*T. aestivum* L.) was grown in a half-strength Hoagland's

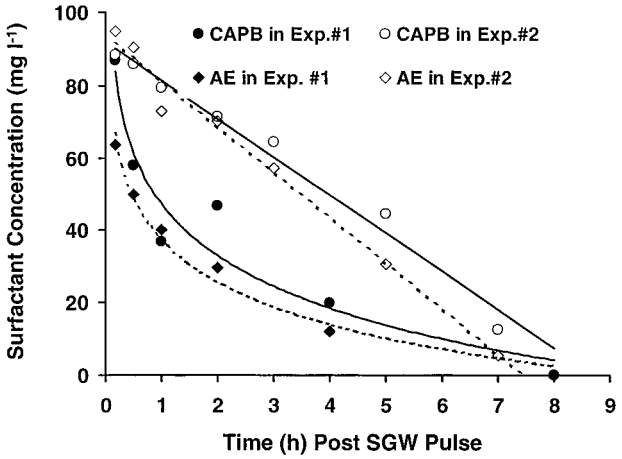

Figure 7. Change in concentrations of CAPB and AE following a pulse of SGW to a hydroponic plant growth system that had been previously acclimated to either a varied amount of surfactant up to 4.8 g m^{-2} day^{-1} (experiment 1) or to a constant 2 g m^{-2} day^{-1} dose of surfactant (experiment 2).

solution (Table 1) with addition of SGW containing either CAPB or AE in the two different feeding modes described in the Experimental Section from 4 to 20 DAP, and the surfactant concentration in hydroponic growth medium was monitored daily. The degradation rate of both surfactants was slower than the input rate for the first 2 days, resulting in a slight accumulation of the surfactants. The CAPB and AE concentrations in the growth medium reached to levels of 20 and 15 mg L^{-1} (average of three trials), respectively, within first 24 h. This corresponded a degradation of 30% and 50% of the added CAPB and AE, respectively.

On DAP 20, a single pulse of simulated graywater containing CAPB or AE was added, and the degradation of these surfactants was examined over next 8 h. Figure 7 shows that both CAPB and AE had logarithmic decay in the first experiment and a linear decay in the second experiment. Recall from the Experimental Section that the simulated graywater input for the first experiment was ramped up to 4.8 g m^{-2} day^{-1}, more than twice the 2 g of surfactant m^{-2} day^{-1} input for the second experiment, Thus, the results suggest that a system acclimated to a high load of surfactant has a higher capacity for degradation. The results of both the daily monitoring and the time course study also suggest that AE has a more rapid acclimation and slightly higher degradation rate than CAPB.

CONCLUSIONS

This study elucidated the fragmentation pathway of both CAPB and AE during positive ESI and established that on-line HPLC/ion trap mass spectrometry is sensitive and reproducible for direct determination of these surfactants in a complex aqueous sample matrix. The quantification method had comparable sensitivity for direct determination of CAPB (25 ppb vs 30 ppb obtained with single, linear quadrupole instrument in the negative mode[18]). Although the sensitivity for determination of AE was lower compared with reported values obtained with extracted samples, this method is more advantageous because it eliminates the need for sample extraction and provides greater specificity by being able to perform multiple stages of MS/MS (MS^n). Application of the method to the analysis of hydroponic plant growth medium spiked with surfactants also demonstrated that the method is suitable for samples containing a significant amount of inorganic ions (~200 μS electrical conductance, Table 1) and organic materials (up to 50 mg L^{-1} dissolved organic carbon and 2.5 mg L^{-1} tannic acid equivalent). The minimum detection limit is near or below the chronic toxicity level (ranging from 0.1 to 20 mg L^{-1}) of nonionic surfactants to algae, fish and (in)vertebrates.[21,22] Our previous studies have shown that higher plants such as wheat, soybean, and lettuce can tolerant much higher surfactant concentrations without reduction in the yield.[23] Hence, the analytical method can provide feedback information for controlling graywater input based on the surfactant concentration in the nutrient solution.

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