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The quantitative determination of aspirin and its degradation products in a model solution aerosol

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Abstract: Formulation of pressurized aerosol solutions in propellants for inhalation requires the use of high quantities of surfactants to solubilize the drug. Due to the lipophilic nature of these surfactants, analytical difficulties are created for those wishing to quantify the drug and its degradation products. In order to quantify drug and degradation products by LC it is necessary to separate surfactant and analytes prior to chromatography. To illustrate a typical situation, a method was developed for the analysis of acetylsalicyclic acid ($\approx 2.5 \times 10^{-3}$ M) and its major degradation products (salicylic acid, acetylsalicylsalicylic acid and salicylsalicylic acid) solubilized in trichloromonofluoromethane (CFC-11) containing 10^{-2} M sorbitan trioleate (Span 85®). Surfactant extraction problems were reviewed experimentally. The presentation of all analytes and the surfactant, dissolved in hexane, to silica solid phase extraction columns, followed by elution in a polar solvent, was found to be an efficient way of separating this lipophilic surfactant from the analytes. The final assay employed propellant evaporation, reconstitution of the non-volatiles in hexane, normal phase solid phase extraction (recoveries of 100 ± 10% were observed for all analytes), elution and dilution with mobile phase, and reversed-phase liquid chromatography (Econosphere® C_8 5 μm, 4.6 × 250 mm). The assay utilized a mobile phase of water, methanol, tetrahydrofuran and 1 M phosphoric acid with ultraviolet detection at 275 nm. Using external standards, linear calibration curves of peak height versus concentration were obtained for all analytes in the expected concentration ranges ($r > 0.991$). As it is described, the assay had a relative standard deviation of ≤3.7% for all analytes.

Keywords: *Surfactant; aerosol; solution; solid phase extraction; LC.*

Introduction

At the present time, pressurized metered dose inhalers (MDIs) are formulated almost exclusively as suspensions of micronized drug in blends of chlorofluorocarbon propellants (CFCs). Low concentrations of surfactants (typically 1/10th the drug concentration) are included to prevent particulate aggregation. With the enforcement of the Montreal Protocol, CFCs must be replaced with environmentally friendly propellants. The most likely replacements are the hydrofluorocarbons, HFC-134a (1,1,1,2-tetrafluoroethane) and HFC-227 (1,1,1,2,3,3,3-heptafluoropropane) [1], which have negligible stratospheric ozone depleting potentials [2]. Since the solubilities of many drugs in these propellants are significantly higher than in the conventional CFC propellants [3], physical instability due to crystal growth [4] becomes a problem when suspension aerosols are formulated.

Unfortunately, the solvency of the propellants alone [3, 5] is rarely sufficient to enable a solution aerosol to be formulated in the absence of surfactants and/or cosolvents. Low concentrations of surfactants are favoured because cosolvents like ethanol (required in higher concentrations) reduce lung penetration by conferring low volatility on formulations [6]. However, while physical stability can be a problem with pressurized suspension aerosols (which are usually chemically stable), this is traded for chemical stability problems when drug is dissolved in propellants. Thus there is a need for the aerosol formulator to identify and quantify the drug and its major breakdown products [7] in a prepared analytical sample which often contains relatively large concentrations of lipophilic surfactants. Typically, surfactants like sorbitan trioleate, oleic acid, lecithin and others [3] are employed both to solubilize drug and lubricate the metering valve in final, pressurized, solution formulations [8]. These can cause significant problems such as shortened column life and interfering peaks, requiring extraordinary development and validation of the various analytical

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procedures for the final dosage form. Previous reports in the cosmetic and pharmaceutical industries indicate the difficulty of separating surfactants and excipients from the compound(s) of interest [9–12]. For these reasons, we have chosen to study a model solution aerosol in which chemical breakdown was believed to be inevitable (acetylsalicylic acid, aspirin, solubilized in trichloromonofluoromethane, CFC-11, with the aid of a fairly high concentration of sorbitan trioleate, Span 85®). In this paper, our purpose was to determine firstly, whether we could identify and quantify the aspirin and its degradation products using LC, and secondly, whether our method would stand up to the normal analytical standards expected of assays for a final dosage form [13].

The paper describes an application of disposable Extract-Clean® columns to quantitatively analyze acetylsalicylic acid (ASA, aspirin), which was found to degrade to salicylic acid (SA), acetylsalicylsalicylic acid (ASSA), and salicylsalicylic acid (SSA), in this non-aqueous solution (Fig. 1). The bulk of the lipophilic surfactant and these compounds could be retained on a sample pre-treatment silica column. The analytes alone could then be eluted using a polar mobile phase. This enabled the eluant to be diluted and analysed for ASA, SA, ASSA, and SSA by reversed-phase LC. While the final assay was adequate for subsequent chemical kinetic studies, the need to separate the analytes from a matrix containing large amounts of surfactant prior to their quantification precluded the achievement of the normal standards for accuracy and precision expected in final dosage form analysis [13].

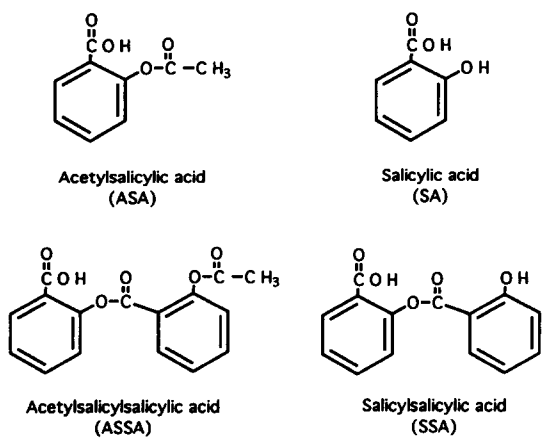

Figure 1
Structures of analytes in non-aqueous solution.

Experimental

Chemicals, reagents and extraction materials

ASA (the Sigma Chemical Co., St Louis, MO, USA), SA (City Chemical Corp., New York, NY, USA), SSA (Eastern Chemical, Hauppauge, NY, USA), sodium hydroxide (JT Baker, Phillipsburg, NJ, USA), acetic anhydride (Aldrich, Milwaukee, WI, USA), sorbitan trioleate (Fluka, Ronkonkoma, NY, USA), trichloromonofluoromethane (CFC-11; DuPont, Wilmington, DE, USA) and absolute ethanol (Pharmco, Bayonne, NJ, USA) were used as received. ASSA was synthesized from SSA via a well known acetylation procedure [14]. Methanol (Fisher Scientific, Raleigh, NC, USA), tetrahydrofuran (Fisher Scientific, Raleigh, NC, USA), and *o*-phosphoric acid (Fisher Scientific, Raleigh, NC, USA) were LC grade and the water was reverse-osmosis purified. The silica sorbent columns (Extract Clean®, Stock no. 209250) were purchased from Alltech (Deerfield, IL, USA).

Instruments and conditions

The LC system consisted of an Isocratic LC Pump (Model 250, Perkin–Elmer, Norwalk, CT, USA) equipped with an auto-sampler (Model ISS 100, Perkin–Elmer, Norwalk, CT, USA). UV detection (Shimadzu UV Spectrophotometric Detector Model SPD-6A; Kyoto, Japan) of ASA, SA, ASSA, and SSA was employed at 275 nm. Peak heights were recorded with an integrator (Model HP 3396A, Hewlett–Packard, Wilmington, DE, USA). Samples were chromatographed on C_8 5 μm columns (Econosphere®, 4.6 × 250 mm; Alltech, Deerfield, IL, USA) and the guard column (Uptight Precolumn 2 mm × 2 cm; Thomson Instrument Co., Springfield, VA, USA) filled with C_8 pellicular matter (Upchurch Perisorb RP-8 30–40 μm Pellicular; Thomson Instrument Co., Springfield, VA, USA) was attached directly to the analytical column. The mobile phase of methanol–tetrahydrofuran–phosphoric acid (1 M)–water (44:5:5:qs to 100 by volume) was filtered through a 1.0 μm glass fibre filter (Gelman Sciences, Ann Arbor, MI, USA) and degassed under vacuum with sonication (Model 5200, Branson Ultrasonic Corp., Danbury, CT, USA) prior to use. Mobile phase was delivered at a flow rate of 1 ml min^{-1}. Sample injection volume was 20 μl. The analyses were carried out at ambient temperature.

Procedures

(1) *Synthesis of acetylsalicylsalicylic acid.* Sodium hydroxide (4.66 g) was dissolved in 50 ml of water. SSA (20 g) was dissolved in the basic solution at room temperature. Approximately 40 g of crushed ice was added and the mixture stirred vigorously. Acetic anhydride (9.09 ml) was added and the solution stirred until crystal formation ceased. The yield was filtered, dried under vacuum and purified by recrystallization from an aqueous ethanolic solution held at 4°C overnight. The product (ASSA) was filtered and dried under vacuum.

(2) *Preparation of standard solutions and dosage form samples.* Matrix-free standards (Table 1) were manufactured by adding the analytes to a 100-ml volumetric flask and diluting to volume with mobile phase. These standards were analysed directly by LC.

Five separate analyte solutions containing 10^{-2} M sorbitan trioleate in trichloromonofluoromethane (CFC-11) were prepared by weight and analysed directly following manufacture. These were prepared by manufacturing a dosage form sample (Table 1) by mixing the surfactant and all analytes in a screw cap compatibility container (Aerosol Laboratory Equipment, Walton, NY, USA). The container was sealed, weighed and shaken (Wrist action shaker, Model 75, Burrell Corp., Pittsburgh, PA, USA) until all components were dissolved. Volumes (5, 4, 3, 2, and 1 ml) of the dosage form sample were removed from the compatibility container using grade A glass pipettes and diluted to 5 ml with 10^{-2} M sorbitan trioleate in CFC-11. The solutions were then allowed to evaporate in a fume hood. The residues were reconstituted in 5 ml of hexane and analysed. They were considered to be typical of differently formulated dosage forms stored for different lengths of time in stability tests.

(3) *Solid phase extraction.* A vacuum manifold (Vac-Elut Sorbent Phase Extraction Vacuum Box, Varian Analytical Instruments, San Fernando, CA, USA) and columns packed with 500 mg silica (Extract Clean®, Stock no. 209250; Alltech, Deerfield, IL, USA) were used. One silica column for each sample was inserted into the vacuum manifold. After each column was conditioned with 2 ml hexane at −2 in. Hg, 1 ml of sample solution was added to the column using a calibrated pipette and applied to the column at the same pressure. Waste materials were collected in a vacuum flask placed between the pump and the vacuum manifold. A 5-ml volumetric flask was then placed under the column. Mobile phase, 2 × 2 ml, was used to elute the analytes at −10 in. Hg. The vacuum was maintained to complete solvent elution in all cases. The eluant was collected, diluted to volume with mobile phase, mixed and analysed via LC.

(4) *Chromatographic procedure.* Once a stable baseline had been established (no visible drift during a baseline plot of at least 20 min), samples (prepared in triplicate) were injected in duplicate. A matrix-free standard solution was injected in duplicate after every two samples analyses (every four injections). For example, a series of 14 injections consisted of the following:

injection no. 1 — standard solution, replicate no. 1;

injection no. 2 — standard solution, replicate no. 2;

injection no. 3 — sample no. 1 from unknown no. 1, replicate no. 1;

Table 1
Formulation of samples and standards for analysis

	Matrix-free standards		Dosage form samples	
Component	Target (g)	Actual (g)	Target (g)	Actual (g)
CFC-11	—	—	75.00	75.11
Sorbitan trioleate	—	—	0.5138	0.5137
ASA	0.00400	0.00398	0.02250	0.02218
SA	0.00300	0.00313	0.01125	0.01061
ASSA	0.00100	0.00080	0.00187	0.00174
SSA	0.00100	0.00087	0.00187	0.00136
Mobile phase	100 ml	100 ml	—	—

injection no. 4 — sample no. 1 from unknown no. 1, replicate no. 2;
injection no. 5 — sample no. 2 from unknown no. 1, replicate no. 1;
injection no. 6 — sample no. 2 from unknown no. 1, replicate no. 2;
injection no. 7 — standard solution, replicate no. 3;
injection no. 8 — standard solution, replicate no. 4;
injection no. 9 — sample no. 3 from unknown no. 1, replicate no. 1;
injection no. 10 — sample no. 3 from unknown no. 1, replicate no. 2;
injection no. 11 — sample no. 1 from unknown no. 2, replicate no. 1;
injection no. 12 — sample no. 1 from unknown no. 2, replicate no. 2;
injection no. 13 — standard solution, replicate no. 5;
injection no. 14 — standard solution, replicate no. 6 . . .
Data analysis is described in what follows.

A series of experiments were performed to document (a) percentage recovery of analytes from dosage form samples; (b) relative standard deviation (RSD) of the assay for a typical lot of Extract Clean® silica columns; (c) assay variability due to inter-lot variation in Extract Clean® silica columns; and (d) the linearity of calibration curves for each of the analytes in dosage form samples spiked with known analyte concentrations. For purposes of clarity, these are described more fully in Results and Discussion.

The guard column was consistently repacked after ≤160 sample injections.

(5) *Data analysis.* Each average analyte (ASA, SA, ASSA and SSA) concentration in each unknown sample (e.g. sample no. 1, above, injections nos 3 and 4) was calculated from

$$(Conc)_A = \frac{PH_A \times (Conc)_{A,Std}}{PH_{A,Std}}$$

where *PH* represents average peak height and the subscripts A and Std refer to analyte and standard solution, respectively. $(Conc)_{A,Std}$ was held constant throughout (Table 1). PH_A was calculated as the mean of the duplicate injections (e.g. injections no. 3 and 4). $PH_{A,Std}$ was calculated as the mean of the four replicates which bracketed the sample injections in question (e.g. injections no. 1, 2, 7 and 8, bracket injections no. 3 and 4). Since each sample was prepared in triplicate and injected in duplicate, this procedure generated three values for $(Conc)_A$ for each analyte in each unknown. The final concentration for each analyte in each unknown was given by the average of these values.

Analyte recovery from silica Extract Clean® columns was calculated from: percentage recovery = (amount recovered × 100)/(amount added to column) where the amount recovered was given by: amount recovered = (average $(Conc)_A$) × (reconstitution volume).

A single blinded analysis of dosage form samples (Table 2) was also performed and coefficients of variation were calculated from: RSD = (sample standard deviation × 100)/mean.

Results and Discussion

(1) *Synthesis of acetylsalicylsalicylic acid.* The product's (ASSA) structure and purity were verified using IR and NMR spectroscopy and melting point determinations. Purity was further verified using LC. This procedure ensured the absence of ASA, SA, and SSA.

(2) *Preparation of standard solutions and dosage form samples.* Typical chromatograms of a matrix-free standard and dosage form

Table 2
Mean values and RSDs for the assay of ASA, SA, ASSA and SSA determined from blind dosage form samples analysed using a single lot of silica columns (Extract Clean®, 500 mg silica, Alltech, Deerfield, IL, USA)

Analyte	Amount added to formulation ($\mu g\ g^{-1}$)	Amount determined by assay ($\mu g\ g^{-1}$)	% of actual amount added	RSD (%)* (n 5)
ASA	278.85	267.97	96.09	3.46
SA	152.71	158.02	103.48	3.67
ASSA	17.40	17.40	100.00	3.30
SSA	32.27	32.27	100.00	3.14

* RSD = (sample standard deviation/mean) × 100.

sample after solid phase extraction are shown in Figs 2 and 3. They both show some non-interfering peaks. In Fig. 2, the small peaks at 2.5 and between 3 and 4 min were associated with the matrix-free standard and were not analyte (or reference standard) associated. These peaks (Figs 2 and 3) were dosage form (probably propellant CFC-11 and surfactant) associated and remained effectively constant and independent of analyte concentration in samples.

(3) *Solid phase extraction.* In order to prepare samples for liquid–liquid extraction, the residues were successfully dissolved in organic solvents, to form a cocktail. However, liquid–liquid extraction of the analytes proved impractical due to emulsion formation upon the addition of any amount of water (sorbitan trioleate was completely insoluble in water or the mobile phase used in the assay). Furthermore, repetitive injection of any of these emulsions (formed, for example, by sonication of the cocktail with mobile phase alone) resulted in highly variable chromatograms (nonreproducible peak areas, nonreproducible peak heights and rapid increase in back pressure). These problems, which were

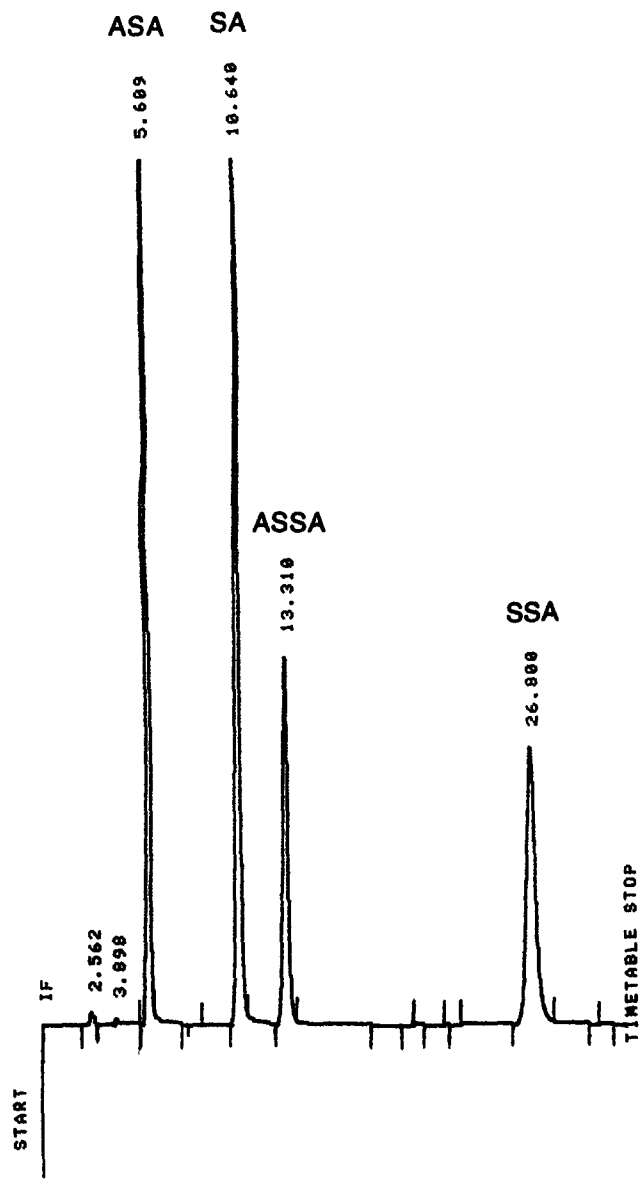

Figure 2
Typical chromatogram of a matrix-free standard solution prepared in mobile phase containing ASA (48 μg ml^{-1}), SA (40 μg ml^{-1}), ASSA (14 μg ml^{-1}), and SSA (17 μg ml^{-1}).

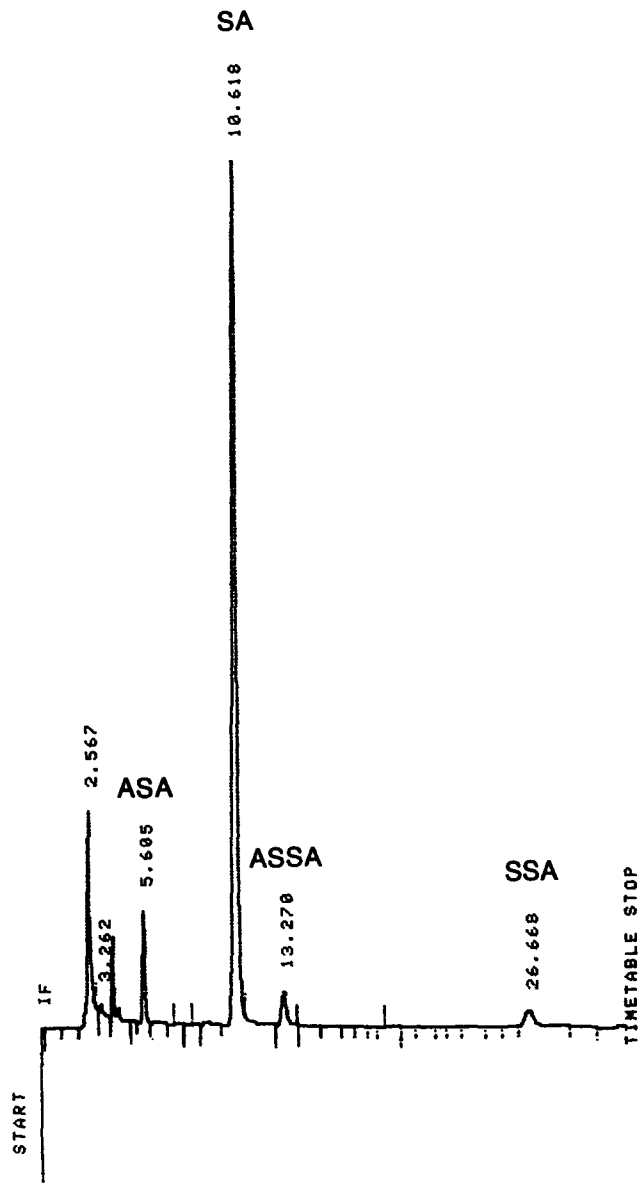

Figure 3
Typical chromatogram of a dosage form sample after solid phase extraction containing ASA (3.1 μg ml^{-1}), SA (37.5 μg ml^{-1}), ASSA (1.3 μg ml^{-1}), and SSA (1.1 μg ml^{-1}).

emulsion and thus surfactant induced, could not be resolved by regularly changing the guard column. Thus, it was necessary to remove as much of the surfactant as possible prior to LC. This was eventually accomplished using solid phase extraction.

Two different amounts of silica sorbent were investigated (200 and 500 mg) and various solvents were tested as eluants. The larger columns were chosen for the final assay since the 200 mg bed of silica was unable to retain the analytes completely at the expected concentrations, as determined by percentage studies (percentage recovery <50%, as determined in preliminary studies). Of the various eluting solvents attempted (acetonitrile, ethanol, hexane, methanol, mobile phase and tetrahydrofuran), only the mobile phase allowed separation of all analytes from the surfactant after loading the cocktail onto the column in a solution of hexane. The uniqueness of the final method resides in the fact that silica can effectively retain the lipophilic surfactant and the relatively polar analytes simultaneously. Furthermore, the use of the polar mobile phase allows the elution of the analytes

while the surfactant is retained on the column. The presentation of this lipophilic surfactant in hexane enables relatively strong binding to occur between the polar headgroups of the surfactant and polar silica. In effect, the lipophilic nature of this particular surfactant is confounded by its presentation to a polar sorbent in a non-polar solvent. This leads to the ability of the eluting solvent, the mobile phase, to remove the analytes more effectively than the surfactant from the silica. In an independent series of experiments to test the ability of the silica columns to retain the surfactant, various concentrations of sorbitan trioleate were dissolved in hexane. Hexane samples (1 ml) were applied to the silica columns, the hexane was collected in clean and previously weighed collection tubes after passing through the silica columns, and subsequently evaporated. The collection tubes were then reweighed and it was determined that >100 mg of surfactant (typical amounts in this assay) could be retained without exceeding column capacity.

The mean recovery of the analytes from "dosage form samples" (Table 1) is shown in Table 3. After analyte adsorption from hexane solutions containing all analytes in a typical "working range" of concentrations (see Table 3), column elution in mobile phase was performed to produce mean recoveries for ASA, SA, ASSA and SSA of 101.1, 101.3, 101.9 and 103.6%, respectively, indicating complete retention and elution in all cases. These experiments showed that the silica sorbent efficiently retained both analytes and surfactant prior to elution with mobile phase.

Assay variation was assessed using a dosage form sample containing CFC-11, sorbitan trioleate (10^{-2} M), and analytes in concentrations described in the second column of Table 2. Intra and inter lot variations are summarized in Table 4. Typically, when the assay was performed on the dosage form samples as described, recovery of all analytes was excellent and reproducibility was good (RSD ≤3.7% in all cases, Table 2). Coefficients of variation for the use of silica columns from three different batches were less than or equal to those associated with the assay itself (≤3.7%, Table 2) in all cases with the exception of aspirin when assayed using column lot no. 1 (RSD = 11.73%, Table 4). This high assay variability for ASA in the case of lot no. 1 of silica sorbent made it essential to screen the assay performance of each batch of solid phase extraction columns to preclude the use of columns showing large intra lot variation. Prior to use of a particular lot, assay variation is now assessed using a dosage form sample containing all analytes (Table 2). In the event that RSD >3.7% for any analyte, that silica sorbent lot is rejected. None of the results reported elsewhere in this paper include values determined using lot no. 1 columns; either lot no. 2 or lot no. 3 was used. Moreover, the method, as described in our operating procedures, requires that intra lot coefficients of variation of ≤3.7% are documented for all analytes prior to acceptance of a given batch of Extract Clean® silica columns.

Table 3
Average recovery of the analytes at various concentration

Analyte	Concentration range ($\mu g\ g^{-1}$)	Mean % recovery
ASA	58.66–293.29	101.1
SA	28.06–140.30	101.3
ASSA	4.60–23.01	101.9
SSA	3.60–17.98	103.6

Table 4
Inter lot variability due to use of silica columns (Extract Clean®, 500 mg silica, Alltech, Deerfield, IL, USA) from different batches

Analyte	Lot no. 1 RSD (%)	Lot no. 2 RSD (%)	Lot no. 3 RSD (%)	Lot 1-2-3 RSD (%)
ASA	11.73	2.63	1.09	12.76
SA	2.44	2.21	1.46	2.39
ASSA	3.07	2.20	1.74	2.50
SSA	2.60	2.30	1.44	2.58

* RSD = (Sample standard deviation/mean) × 100.
n = 4 for lots 1, 2 and 3.
n = 12 in the case of inter lot variability (1-2-3).

(4) *Chromatographic procedure.* The LC analysis of aspirin and its degradation products is not a new concept [15, 16]. In the presence of a large excess of a lipophilic surfactant, however, direct sample injection onto the analytical column becomes almost impossible. This obstacle is recognized by those in the field as a major deterrent in the development of pressurized solution aerosols for inhalation [17]. Even though aspirin is not used in aerosol preparations, it is known to degrade by a variety of well characterized mechanisms, and both it and its degradation products have molecular sizes and lipophilicities typical of compounds which are included in metered dose inhalers (albuterol, isoproterenol etc.) [18].

In the absence of sorbitan trioleate, the mobile phase and column characteristics were optimized to achieve adequate separation with a reasonable analysis time. In preliminary studies, retention times of all analytes were found to be exceedingly long, or peak separation inadequate, when a C_{18} column was used with a variety of acidified methanolic mobile phases (methanol–phosphoric acid (1 M) in water (0–60:5%)) similar to those previously reported in the literature [15, 16]. These mobile phases were also investigated with a C_8 column. These failed for a variety of reasons: inadequate peak separation, excessive analysis time, and/or immiscibility with dosage form samples. The mobile phase reported in this paper (methanol–tetrahydrofuran–phosphoric acid (1 M)–water; 44:5:5:qs to 100 by volume) allowed for adequate separation with maximum total retention times consistently less than 35 min.

Due to the harsh nature of the mobile phase (pH $\approx$ 2, to maintain analytes in their nonionized form [19]) the retention times of the analytes decreases progressively with increasing numbers of injections. While a new column showed an SSA retention time of 35 min, this value was reduced to 15 min after 400 injections. Repeated use of this method has shown, however, that the frequent use of external standards negated any complicating effects, with respect to data analysis (similar results of those described in Tables 2–4 were recorded for "new" or "old" C_8 analytical columns). The calibration curve for the matrix-free standard solutions in typically encountered concentrations remains linear throughout the life of the column ($r > 0.998$ with a new column and $r > 0.991$ with a used column; $n = 9$). Columns were routinely replaced when chromatograms showed incomplete resolution of the analytes from either the early eluting peaks which were dosage form related (ASA retention time less than 4.25 min) or each other (peak height separations less than 1.3 min).

(5) *Data analysis.* The LC assay variation for dosage forms usually involves a proof that the RSD of the assay is $\leq$2% [13]. Assays which utilize single pass extraction procedures have often been noted to have difficulty meeting this criterion [20, 21]. The variance of the analysis procedure is shown for individual peak heights for each analyte in the final column of Table 3. Clearly, the RSDs of individual results are too large to validate the assay based upon single injection results for either USP validation purposes (the most stringent USP standard requires RSD $\leq$ 2%) or for our own laboratory's kinetic analysis purposes ($\leq$ 5%). During routine dosage form analysis, LC often employs the mean from two injections. In this assay, concentrations are determined by comparison of the results from the mean of duplicate injections of three samples of unknown (each of which has passed through a separate solid phase extraction column) to the mean from duplicate injections of two standards of the same concentration, neither of which has passed through the adsorbent (unknown, $n = 6$; known, $n = 4$, see injection sequence, procedure no. 4). In this way, the final assay has RSD for analytes $\leq$3.7% (Table 2). This final variance is sufficient for our laboratory's chemical kinetic purposes. In the case of final dosage form analysis, however, the difficulties involved with the analysis of solution dosage form aerosols can best be illustrated by observing that even with this assay in its present form (RSD $\leq$ 3.7%) some further steps still need to be taken to validate at the 2% level. These may include the use of an internal standard which was rejected in our investiation due to the difficulty of selecting a material which behaved similarly to all of the analytes. However, for the purposes of our kinetic analysis of factors affecting breakdown and reaction mechanisms, the assay has good linearity for all analytes (Table 5). Furthermore, the error associated with the samples was equivalent to the error associated with the analysis of the external matrix-free standard solutions, indicating that the bulk of this error was also inherent in the procedures which did not require surfactant removal. Thus, the presence of trace quantities of surfactant following solid phase extraction did not account for the variance in the assay

Table 5
Linarity of six point calibration curves for analytes in dosage form samples

Analyte	Linear range ($\mu g\ ml^{-1}$)	r	RSD (%)* ($n = 6$)
ASA	17.42–87.10	0.9981	0.35–4.70
SA	8.33–41.67	0.9983	0.72–4.38
ASSA	1.37–6.83	0.9978	1.03–7.81
SSA	1.07–5.34	0.9979	0.87–4.05

* RSD = (sample standard deviation/mean) × 100.

and, at least a portion of this variance appeared to be instrumental in origin.

Conclusion

This method allows the separation and determination of aspirin and its major degradation products in a solution aerosol having a high surfactant concentration. The use of normal phase (silica) adsorption from hexane and subsequent analyte elution in a polar mobile phase, successfully removed a large excess of an oily surfactant (sorbitan trioleate). The analytes, when reconstituted in mobile phase, could be quantified with reference to non-matrix derived external standards prepared in mobile phase alone. Due to errors (reconstitution, extraction, elution, dilution, the use of external standards, and integration) the assay failed to meet the most stringent USP requirements for dosage form analysis (RSD $\leq$ 2%). This confirms the conventional wisdom which recognizes that analytical precision problems often defeat pressurized solution aerosol products during their development. Nevertheless, the RSDs were sufficiently small ($\leq$3.7%) to enable the continued study of the kinetics of drug breakdown in reversed-micellar non-aqueous solutions. With solution aerosol development becoming increasingly probable in the future with the use of alternative propellants, methods similar to those described in this paper may prove useful during dosage form analysis.

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