Mitoxantrone Stability in Syringes and Glass Vials and Evaluation of Chemical Contamination

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ABSTRACT

The stability of mitoxantrone hydrochloride solutions was evaluated over 42 days. Mitoxantrone solutions of 2 mg/mL were stored in multidose glass vials, and Monoject syringes at 4°C and 23°C. On days 0, 2, 5, 7, 9, 14, 20, 22, 27, 32 and 42, the mitoxantrone concentration was measured using a stability-indicating liquid chromatographic method. Colour pH, and physical inspection were also completed. On each study day, the concentrations of two chemicals (MBT and HMBT) reported to be leached from the rubber gaskets of some syringes and DEHP, reported to be leached from PVC containers, were also measured.

During the 42-day study period all solutions stored at either 4°C or 23°C retained more than 90% of the initial mitoxantrone hydrochloride concentration. All solutions remained clear and had no change in UV-VIS spectrum absorbance or pH. On day 42, mass balance indicated that less than 3% of the mitoxantrone had degraded. The MBT, HMBT and DEHP concentrations were below detectable limits (100, 100 and 200 ng/mL respectively) in all solutions.

We conclude that mitoxantrone solutions (2 mg/mL) stored at 4°C or 23°C in the original glass vial are stable for 42 days. Since investigations have failed to demonstrate contamination of these solutions stored in Monoject® syringes, we conclude that mitoxantrone solutions are also stable when stored at 4°C or 23°C in these syringes, and maintain acceptable product purity.

Key Words: *mitoxantrone, stability, leached plasticizers*

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RÉSUMÉ

La stabilité des solutions de chlorhydrate de mitoxantrone a été évaluée sur une période de 42 jours. Les solutions de mitoxantrone (2 mg/mL) ont été entreposées dans des flacons de verre multi-dose et dans des seringues Monject à 4°C et 23°C. Les concentrations de mitoxantrone ont été mesurées pendant onze jours d'études différents en utilisant une méthode de chromatographie liquide approuvée pour déterminer la stabilité. La couleur, le pH et l'analyse physique ont aussi été vérifiés. À chaque jour de l'étude les concentrations de deux produits chimiques (MBT et HMBT) qui filtraient des joints de caoutchouc de quelques seringues et du DEHP (qui filtrait des contentants de chlorure de polyvinyle) ont été mesurées.

Pendant la période d'étude de 42 jours, toutes les solutions entreposées soit à 4°C soit à 23°C ont retenu plus de 90% de la concentration initiale de chlorhydrate de mitoxantrone. Elles sont demeurées claires et sans aucun changement d'absorbance de spectre UV-VIS ou de pH. Au jour 42, la balance de masse indiquait qu'environ 3% de mitoxantrone s'était dégradé. Dans toutes les solutions les concentrations de MBT, HMBT et DEHP étaient en dessous des limites décelables.

Nous concluons donc que les solutions de mitoxantrone (2 mg/mL) entreposées à 4°C ou 23°C en flacons de verre originaux sont stables pendant 42 jours. Puisque les analyses n'ont pas réussies à démontrer que les solutions entreposées dans des seringues Monoject étaient contaminées, nous concluons que les solutions de mitoxantrone entreposées dans ces seringues à 4°C ou 23°C sont également stables et maintiennent une pureté acceptable du produit.

Mots clés: *mitoxantrone, stabilité, filtre plastifiant*

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INTRODUCTION

Mitoxantrone (mitozantrone; 1, 4-dihydroxy - 5, 8 - bis [[2 - [(2 - hydroxyethyl) amino] - ethyl] amino] - 9, 10 - anthracenedione; ACS #65271-80-9) is a synthetic anthracenedione used in the treatment of neoplastic diseases. No reports have been published which describe the stability of mitoxantrone in intravenous solutions using stability-indicating assay methodology. The manufacturer provides mitoxantrone as a sterile aqueous solution which is stable for two years.[1] However, once in use the manufacturer recommends that a vial be discarded within 24 hours.[2]

It is common for 2 mg/mL of mitoxantrone to be drawn into a syringe and dispensed for administration. Due to the possibility that the dose could be cancelled, knowledge of mitoxantrone stability in a syringe would be important given the cost of mitoxantrone and the relationship reported between expiry dates and wastage.[3] However, many factors in addition to drug stability can affect the expiry date placed on a product. While the financial incentives to extend an expiry date and reduce wastage may be great,[3] patients should not be placed at risk. Rubber gaskets found in some syringes have been reported to release chemicals, primarily 2-mercapto benzothiazole-(MBT) and 2-(2 hydroxyethyl) mercapto benzothiazole-(HMBT)]. Di-2-ethylhexylphthalate-(DEHP) has been leached from PVC containers into the contact solution.[4-9] These compounds produce mild to severe hepatic changes following chronic administration.[10-14] Evaluation of the rate and the amounts of these chemicals which may have leached from the syringe into the solution was thought to be necessary.

It was the aim of this investigation to establish the stability of mitoxantrone hydrochloride solution in an opened original vial and in syringes. To evaluate the possibility of chemicals leaching into the mitoxantrone solutions, the concentrations of MBT, HMBT and DEHP were estimated and the solutions monitored for other contamination during storage in syringes and original glass vials. In order to complete this study, it was necessary to have analytical methods which could separate and quantitate mitoxantrone specifically in the presence of degradation products and leached chemicals, as well as measure the leaching chemicals. This required the development and validation of liquid chromatographic (LC) methods for mitoxantrone, MBT, HMBT and DEHP.

METHOD

Assay Validation

Following set-up of the chromatographic system for mitoxantrone, the suitability of this system for use as a stability-indicating assay was tested by accelerating the degradation of mitoxantrone. Two — 25 mL, 2 mg/mL solutions of mitoxantrone HCl (Lederle, Cyanamid Canada: Novantrone® — lot #9J0440) were used. One solution remained in 0.9% w/v sodium chloride (NS) and the other solution was adjusted to pH 11.5 with 0.5 N NaOH. Each solution was placed in a 30 mL multidose vial with rubber septum closure (SoloPak Laboratories) and incubated in a water bath at 90°C for 240 minutes. Samples were drawn just prior to incubation and at 15, 30, 60, 90, 180 and 240 minutes. This study was stopped at 240 minutes because at this point the mitoxantrone hydrochloride concentration (pH 11.5) had been reduced to approximately 4% of initial, but the mitoxantrone in NS showed no apparent loss in concentration. Chromatograms were inspected for the appearance of additional peaks and the mitoxantrone peak was compared between samples for changes in concentration, retention time and peak shape. To further test if the chromatographic system could separate mitoxantrone from degradation products, the purity of the peak referred to in chromatograms as mitoxantrone was tested by the following methods. First, using a photodiode array detector, complete UV-VIS spectra (200-800 nm, band width 1 nm; resolution 1.4 nm; deuterium lamp: Waters) from the leading edge, middle and tail of the mitoxantrone peak in a degraded sample were compared to UV-VIS spectra observed for the time zero sample. Once spectral purity had been demonstrated mass spectroscopy (MS) was completed on degraded samples, a mitoxantrone reference standard and reference standards of each of the two degradation products. These standards were provided by Lederle Laboratories and included; (i) pure mitoxantrone hydrochloride (98.7% w/w — lot #PC 517), (ii) CL 261-193 (93.3% w/w — lot #12145B-33-1) the major degradation product, and (iii) CL 261-209 (88% w/w — lot #12067B-191-1), a second degradation product. Mass spectrum analysis was completed using a LC-MS-MS system with a MS-MS detector (API III: Sciex) which monitored the LC eluate. Positive and negative orifice voltages of 60V and 90V allowed determination of parent ion mass. Mass spectrum analysis was completed at Mann Testing Laboratories Ltd., Mississauga, Ontario. Using these standards, mass spectroscopy and photodiode array de-

tection, all but one peak in each chromatogram could be identified. Furthermore, the ability of the chromatographic system to separate mitoxantrone from its degradation products could be tested, and loss of mitoxantrone due to degradation could be accounted for by increases in degradation products (mass balance).

Following this first phase of evaluation and validation, the accuracy and reproducibility of standard curves was tested over a five-day period. On each day a 2 mg/mL vial of mitoxantrone hydrochloride solution (Lederle, Cyanamid Canada: Novantrone® — lot #9J0440) was used as a secondary reference stock solution. Samples of this stock solution were diluted to 2.0 mL to obtain reference solutions with final concentrations of 1.5, 0.5, and 0.25 mg/mL. These reference solutions served to construct a standard curve. Each sample was chromatographed in duplicate. As well, a 1.0 mg/mL sample of mitoxantrone hydrochloride (Novantrone — Lederle Laboratories — lot #9J0440) prepared on each day was chromatographed and its concentration determined and compared to its known concentration (recovery).

Stability Study

On study day zero, twelve 2 mg/mL vials of mitoxantrone hydrochloride (Lederle, Cyanamid Canada: Novantrone® — lot #9J0440) were used. Six mitoxantrone samples were kept in the original manufacturer's vial and the contents of the other six vials were drawn into and stored in 12 mL Monoject PVC plastic syringes (Division of Sherwood Medical). Three vials and syringes were stored at 4°C and the other three vials and syringes were stored at 23°C for the duration of the study. On each of the 11 study days, these solutions were tested for mitoxantrone, MBT, HMBT and DEHP content, UV-VIS spectra (200-800 nm), pH and changes in physical appearance (colour, clarity and particulate matter).

Mitoxantrone LC Analysis

Solutions containing 2 mg/mL of mitoxantrone hydrochloride were stored at 4°C or 23°C, and sampled on each of the 11 study days (0, 2, 5, 7, 9, 14, 20, 22, 27, 32, and 42). On each study day samples of 0.5 mL, were drawn from each container with a 1 mL 27 gauge tuberculin syringe. One microlite of each of these samples was directly chromatographed in duplicate.

On each study day standards were prepared from mitoxantrone hydrochloride (Lederle, Cyanamid Canada: Novantrone® — lot #9J0440). Aliquots of this solution were diluted to obtain standards with final concentrations of 1.5, 0.5, and 0.25 mg/mL. One microlitre of each of these standards and a blank were directly chromatographed in duplicate, and served to construct a standard curve. An additional standard of 1.0 mg/mL prepared and chromatographed on each day, was used to calculate recovery.

The chromatographic system consisted of an isocratic solvent pump (Spectra Physics: Model 8700) which delivered a mixture of acetonitrile (Fisher: cat. #A998), 0.2% triethylammonium phosphate (TEAP) buffer (2 mL of triethylamine (Sigma: cat. #T-O886) per litre of water adjusted to pH = 2.5 with phosphoric acid) and 0.01 M tetrabutylammonium hydrogen sulfate (Sigma: cat. #T-1134) through a reversed phase (250 mm x 4.6 mm) C-18, 5μm column (Beckman: Ultrasphere ODS #235329) at 1.5 mL/min. The ratio of acetonitrile to TEAP buffer was 40:60 and was held constant during a chromatographic run. On each day the composition of the mobile phase was adjusted to achieve a retention time for mitoxantrone between 114 and 120 seconds. Retention times did not vary by more than three seconds during each analytical day (mean maximal variation: 1.2 seconds).

The column eluate was monitored with a variable wavelength ultra violet detector (Kratos: Spectroflow 783) at 243 nm. A 10 mV signal output from the detector was integrated and recorded on a chromatography integrator (Spectra Physics: SP4270) and archived on computer diskette.

Standard curves were prepared daily, as previously described. The peak area of the mitoxantrone HCl peak was subjected to least squares linear regression and the actual mitoxantrone HCl concentration, from each solution, was interpolated from these curves and recorded. Concentrations were reported to the nearest 0.01 mg/mL.

Measurement of Degradation Products

On days zero and 42, standard curves for each of the degradation products were prepared to allow calculation of the amount of each present. Since degradation products are present in such small concentrations, to increase the sensitivity of the method, an increased volume of sample (100 μL) was injected onto the column. This re-

quired the use of a semi-preparative (250 mm x 10 mm) C-18, 5 μm reverse phase column (Beckman: Ultrasphere ODS #235238) as the loading capacity is exceeded with an injection of 200 μg or more of mitoxantrone.

The mobile phase was identical to that used with the analytical column, but the flow rate was increased in direct proportion to the cross-sectional area of the column to 7.0 mL/min.

pH

On each of the 11 study days, the pH of one solution in the original vial and syringe stored at 4°C or 23°C was measured and recorded to the nearest 0.05 of a pH unit. The pH meter (Fisher: Accumet-model 925) was fitted with a microprobe glass body electrode (Fisher: cat. #13-639-280) and was standardized each day with two commercially available buffer solutions. (pH 7: Fisher: cat. #S108-500 and pH 4: Fisher: cat. #SO-B-98).

The pH of each solution was obtained by withdrawing 1 mL of solution and placing it in a glass test tube. The solution was allowed to come to room temperature before the pH was measured.

Physical Inspection

On each of the 11 study days, each container was inspected visually for colour and clarity. Visual inspection for particulate matter was carried out against a black and white background.

Colour

On each study day a complete UV-VIS spectrum (200 nm - 800 nm) was recorded using a photodiode array detector (Waters: 990 - resolution 1.4 nm, Band width 1 nm). One microlite of each solution was injected into a LC system, without a column, and the UV-VIS spectrum 200 nm - 800 nm) determined using a photodiode array detector. The mobile phase in this system was 50% acetonitrile and 50% water and this was pumped at 0.5 mL/min. To eliminate any carry over from one sample to the next, between mitoxantrone samples, 10 μL of acetonitrile was injected into the LC solvent path. This is a more extensive test than measuring the absorbance at one discrete wavelength, since the entire spectrum is evaluated. Whole spectra from each sample were then compared for maxima, minima and overall similarity using the PDA 990 (Waters) software. Identical spectra have a match of 100% ± 1%.

MBT HMBT and DEHP LC Analysis

2-(2-hydroxyethyl) mercapto benzothiazole (HMBT) was synthesized according to the method described by Sexton[5] from 2-mercapto benzothiazole (Sigma) and 2-chloroethanol (Aldrich). The product, recrystallized twice from benzene had a melting point of 56-57°C (literature 56-58°C[15]; 56-57°C[16]) and chromatographed as a single major peak (estimated purity 98%) with a mass spectrum identical to that reported for HMBT.[16] On each study day, 40 mg of 2-mercapto benzothiazole (MBT — Sigma: M-2274), 40 mg of HMBT and 25μL of dioctylphthalate (di-2-ethylhexyl phthalate — DEHP: Aldrich D20, 115-4) were dissolved in separate 100 mL quantities of methanol. Aliquots of each were further diluted with methanol to 10 mL in a volumetric flask to achieve final standard concentrations of 250, 500, 1000 and 2000 ng/mL. Samples of 50 μL were chromatographed in duplicate. The coefficient of variation for duplicates, for each method, routinely averaged less than 2% on each day. The functional limit of sensitivity of these measurements was 100 ng/mL for both MBT and HMBT and 200 ng/mL for DEHP.

Samples of 0.5 mL drawn for mitoxantrone analysis were also subjected to analysis for MBT, HMBT and DEHP. The mobile phase consisted of a linear gradient that ranged between 20% acetonitrile and 80% TEAP (pH = 2.5) and 100% acetonitrile over 60 minutes and was pumped at 1.5 mL/min through a 5 μm reverse phase C-18 column (Beckman: Ultrasphere ODS #235329). The UV-VIS spectra of the eluate was monitored between 200 nm and 800 nm with a photodiode array detector (990: Waters).

Data Reduction and Statistical Analysis

Means (+/-standard deviation) were calculated for analyses completed in duplicate or triplicate. These means are reported in summary tables. Reproducibility was assessed by the coefficient of variation (CV — standard deviation divided by the mean). Mean results from different days of an identical test were compared statistically by least squares linear regression to determine if there was an association between the observed result and time. Log-linear and linear-linear fits for the data from the accelerated degradation study (90°C) were compared for goodness of fit by the Maximum likelihood method of Box and Cox.[17,18] Analysis of variance and the least significant difference multiple range test or Student's t test (where

appropriate) were used to compare differences between temperatures solutions for similar analytical tests. The five percent level was used as the *a priori* cut-off for significance.

Mitoxantrone concentrations were considered "acceptable", or "within acceptable limits" if the concentration on any day of analysis was not less than 90% of the initial (Day zero) concentration. Measurable concentrations (>100 ng/mL) of MBT, HMBT and 200 ng/mL DEHP were considered unacceptable.

RESULTS

Accelerated Degradation and Assay Validation

At 90°C, mitoxantrone 2 mg/mL, adjusted to pH 11.5 with sodium hydroxide degraded in an apparent first order fashion. This part of the study was stopped at 120 minutes when less than 5% of the initial mitoxantrone concentration remained in the solution (Figure 1). When the commercially available solution was incubated at 90°C for up to 2750 minutes, no reduction in mitoxantrone concentration could be observed. (Figure 1) The peak corresponding to mitoxantrone in each chromatogram was evaluated for UV-VIS spectrum similarity. UV-VIS spectra (200 nm - 800 nm) obtained from the leading edge, middle, and tail of mitoxantrone peaks from each sample in the accelerated study and a reference standard, were all identical with respect to maxima, minima and overall shape. Maximum deviation between UV-VIS spectrum (200 nm - 800 nm) fits was 1% (Table I). The mass spectrum of the LC eluate fraction corresponding to the mitoxantrone peak was also identical to the mitoxantrone reference standard.

During the accelerated study

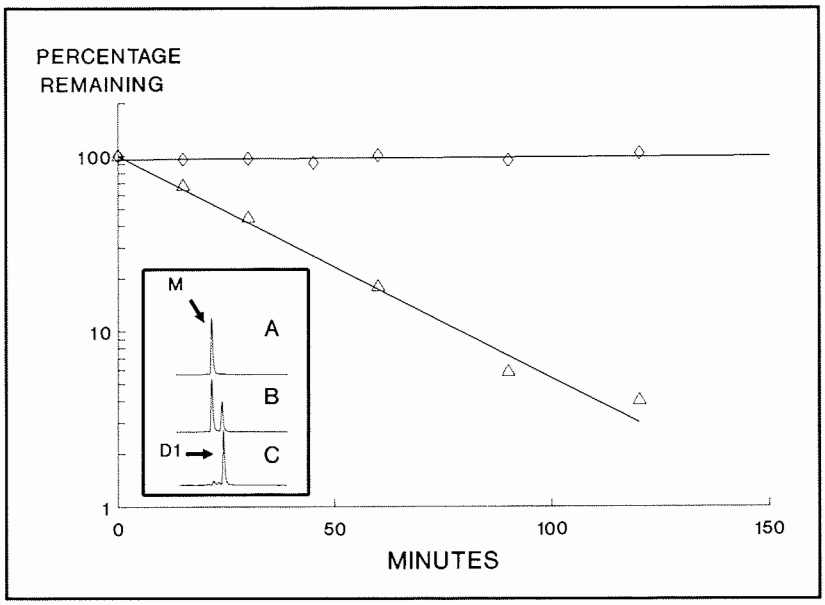

Figure 1: Rate of loss of mitoxantrone during the accelerated study at 90°C in the commercially available solution at pH 3.5 (open diamonds) and in a solution adjusted to pH 11.5 with 0.5N sodium hydroxide (open triangles). During the study, as the mitoxantrone concentration declined, degradation products could be observed in chromatograms (Insert panels B and C, after 15 and 90 minutes, respectively, compared to time zero — Panel A). The final concentration of mitoxantrone is 100% of initial in the commercial solution and 5% in the sample containing NaOH (pH 11.5). D1 represents the major degradation product, CL 261-193 and M represents mitoxantrone.

Table I: Concentrations of mitoxantrone and degradation compounds on day zero and 42

Retention Time (Min.)	Compound Identification	Mass	UV-VIS Spectra Match*	Percent of mass present			
				Day 0		Day 42	
				4°C	23°C	4°C	23°C
1.65	Impurity	[444]	940	2.23	2.27	2.27	2.17
1.95	Mitoxantrone	444	989	97.09	97.46	94.22	94.05
2.45	D-2:CL 261-209	442	872	.12	.20	.86	1.22
2.70	D-1:CL 261-193	357	830	.49	.07	2.64	2.56
	Percent Degradation Products**(%)			2.91	2.54	5.78	5.95

* UV-VIS spectra taken from the middle of the peak of interest. Match is against a 2.0 mg/mL mitoxantrone standard; A value of 1000 indicates a perfect match.

** Percent of degradation products calculation requires corrections for changes in mass to allow mass balance. Mass determined using mass spectroscopy. Numbers in parenthesis are assumed molecular weights that were used when confident mass spectra could not be obtained.

two degradation products were observed. Both were well separated from the mitoxantrone peak and did not interfere with its quatitation (Figures 1 and 2, Panels A, B and C, products labelled D1 and D2). One other peak (indicated as 'I' in Figure 2) and well separated from mitoxantrone was observed in chromatograms. This compound was present in all ungraded stock solutions and did not interfere with mitoxantrone quantitation (Figure 2, Panel B). Its presence appeared unrelated to mitoxantrone degradation since its peak area did not change during the accelerated studies. It was, therefore, considered a contaminant.

These results (predictable degradation, observed separation of degradation products and mitoxantrone, mitoxantrone UV-VIS spectrum purity in each peak, mass spectrum identification of the mitoxantrone peak and separation of mitoxantrone from known and other degradation products) indicated that this analytical method was stability-indicating.

Accuracy, and reproducibility were then evaluated. Mitoxantrone, 10 mg, was added to 10 mL of water and the concentration of this sample estimated. The results of this investigation indicated that the mitoxantrone concentration was measured accurately (recovery for all samples was within 97%-103%) and reproducibility (CV for all replicates averaged 1%). The reproducibility of samples run on different days averaged 3.5%. This indicated that differences of 8% or more could be confidently detected with acceptable error rates.[19]

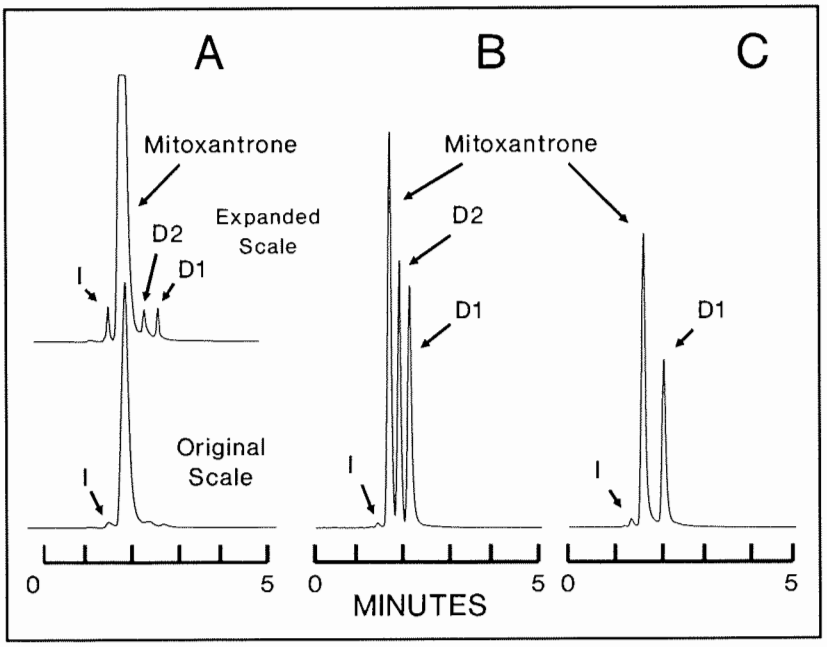

Figure 2: Chromatograms observed during the study period. Panel A (lower tracing) is typical of chromatograms observed during the stability study — shown in its original scale on day 42. The upper tracing has been magnified 10 times to show degradation products/contamination. From the original scale chromatogram it is apparent that even after 42 days degradation products are not prominent. Panel B is a chromatogram observed when water is spiked with mitoxantrone and two known degradation products in concentrations of approximately equal magnitude. Panel C is the chromatogram observed at 15 minutes in the accelerated degration study in a solution adjusted to pH 11.5 with 0.5N sodium hydroxide. In these chromatograms D1 is the major degradation product, CL 261-193; D2 is a minor degradation product, CL 261-209; and I represents a probable contaminant. Mitoxantrone is appropriately marked.

Mitoxantrone Stability Study

Over the 42-day study period there was no significant trend for the mitoxantrone concentration to change in any solution stored at 4°C or 23°C. All concentrations on all study days remained greater than 90% of the time zero concentration. For all solutions, at both temperatures, in vials and syringes, the peak identified as mitoxantrone maintained UV-VIS purity across its elution profile, (UV-VIS spectra obtained on the leading edge, middle, and tailing edge of the peaks were identical) and was also identical to authentic mitoxantrone, observed in standards and day zero samples.

The concentration of the two degradation products, noted during the accelerated degradation studies, were observed and measured on days zero and 42. As indicated in Table I, the total amount of mitoxantrone mass found as degradation products or impurities on day zero was 2.54% and 2.91% for solutions stored at 23°C and 4°C, respectively. On day 42, this mass had increased to 5.95% and 5.78%, respectively (Table I), indicating that approximately 3% of the mitoxantrone present on day zero had degraded by day 42. However, approximately 2.25% of the mitoxantrone mass found on day zero, existed as the impurity, and degradation of mitoxantrone during the 42-day study period can be primarily attributed to degradation of mitoxantrone to D-1. No significant difference in degradation rate was apparent between solutions stored in syringes compared to glass vials (since no significant degradation in any solution was observed). Although solutions

stored at room temperature had a greater increase in the amount of degradation product formed, these differences could not be detected by measurement of total concentration.

During the 42-day study period, pH remained virtually unchanged (pH 3.43-3.65) in solutions stored at 4°C or 23°C. UV-VIS absorbance spectra determined for solutions stored at 4°C and 23°C on each day did not demonstrate any shift in maxima or minima throughout the study period; spectra determined on day zero were superimposable on day 42 and were not significantly different (less than 1% total deviation from 200 nm - 800 nm).

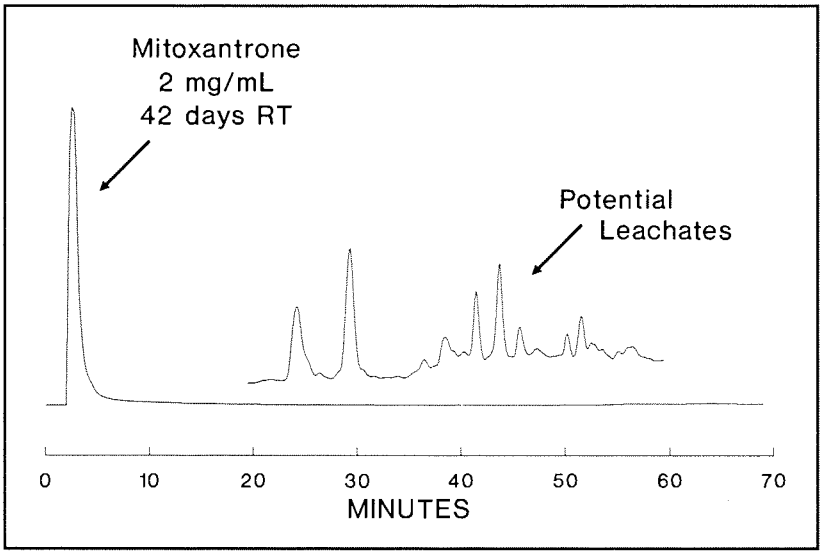

Figure 3: Tracing observed during MBT, HMBT and DEHP analysis on day 42. In this gradient system MBT elutes at 10 minutes, HMBT at 12 and DEHP at 51. The trace marked 'Potential Leachates' represents leachates contaminating a methanol solution stored in a Monoject syringe. It is apparent that none of these chemicals contaminates the mitoxantrone solution even after 42 days storage at room temperature (RT).

MBT, HMBT and DEHP Concentrations

MBT, HMBT and DEHP concentrations in mitoxantrone solutions stored at 4°C or 23°C in the manufacturer's original glass vials and syringes (Monoject) could not be measured because the concentrations of these three chemicals were below detectable limits. This was also the case in a previous investigation, where a methanolic solution was used to extract these three chemicals from Monoject syringes.[29] In fact, it was determined that even with a methanolic solution, MBT, HMBT and DEHP could not be extracted from a Monoject syringes.[20] It was concluded that these chemicals are not found in Monoject syringes. However, although contamination of a methanol solution stored in plastic PVC syringes with unidentified chemicals was observed in a previous investigation,[29] none of these unknown peaks were present in the chromatograms obtained for any mitoxantrone solution, even after prolonged storage (Figure 3).

DISCUSSION

Statistical analysis of the mitoxantrone HCl concentration time data in this study was limited to least squares linear regression because demonstration of a trend for the concentration to decrease was considered more important than demonstrating a statistical difference in concentration between any two days. In fact, the random fluctuations in concentration around a line of 'best fit' are not of practical importance and should be considered 'noise' or experimental error. All conclusions and recommendations made from the data generated by this study have been based on results obtained by least squares linear regression analysis. Least squares log-linear regression indicates that less than a three percent change in concentration occurred over the 42-day study period, as calculated from the line of best fit through the data. In studies such as this, where no change in concentration of the drug can be detected, assurance that the analytical method is specific for the compound of interest is important. This was demonstrated as part of the assay validation procedure. During the accelerated degradation study, we were able to degrade mitoxantrone, separating the degradation products from the parent compound chromatographically. The identity of these compounds was confirmed by mass spectroscopy and comparison with reference standards. These methods are considered important and necessary to assure the validity of the analytical method used in a stability study.[21-23] Failure to carry out these steps could result in the validity of the results being questioned.

No other studies have been published concerning the stability of mitoxantrone in intravenous solutions using stability-indicating assay methodologies. Ostroy and Gams[24] report that a 40 mg/L solution of mitoxantrone is not

stable for extended periods. Although these investigators demonstrate that mitoxantrone degrades to at least two degradation products, the solution used and the degree of degradation observed were not reported. Dilute solutions of doxorubicin, an anthracycline, have also been reported to have short expiry dates,[25,26] yet more concentrated solutions are stable.[27-29] This may be explained by the dependance of photo-degradation on concentration.[30] Although not specifically reported for mitoxantrone, a small zero-order rate of photodegradation could explain the results of Ostroy and Gams,[24] since both this investigation and the manufacturer's data[1] indicate that a 2 mg/mL solution of the drug is stable.

During this stability study, solutions of mitoxantrone were monitored for the presence of plasticizers (MBT, HMBT and DEHP) leached from rubber closures and syringe tips. The analytical methods failed to demonstrate the presence of any of these or any other compounds. Although rubber gaskets have been reported to release both MBT and HMBT into contact solutions,[12,13] many syringe manufacturers have reformulated their rubber gaskets so that MBT is not used.[31,32] While a parallel investigation demonstrated that methanol solutions stored in Monoject or Terumo syringes can become contaminated with a variety of chemicals other than MBT, HMBT or DEHP,[20] the saline solutions used in this and the previous study were free of contamination. It is apparent that a number of factors such as the type of solvent, storage temperature and exposure time also affect the amount of contamination.

CONCLUSION

We conclude that mitoxantrone solutions (2 mg/mL) retain more than 90% of the initial mitoxantrone HCl content for 42 days, when stored at 23°C or 4°C. During this evaluation of stability, no contamination by MBT, HMBT or DEHP was observed. Maximum recommended expiry date is 42 days when stored at 4°C or 23°C. However, this expiry date should only be used after due consideration of sterility and the contamination rate of IV admixtures in an IV additive program.

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