G Ital Med Lav Erg 2020; 42:2, 109-120
ISSN 1592-7830

MEDICINA DEL LAVORO

Fabio Sciubba[1], Mariangela Spagnoli[2], Sergio Iavicoli[2], Gaia Asaro[3], Alessio De Luca[4], Gaspare Guglelmi[5], Maurizio Delfini[1]

Efficacy of sodium hypochlorite in the degradation antineoplastic drugs by NMR spectroscopy

[1] University of Rome La Sapienza - Department of Chemistry (S. Cannizzaro Building), Piazzale Aldo Moro 5, 00185 Rome, Italy
[2] INAIL - Italian Workers Compensation Authority - Department of Occupational and Environmental Medicine, Epidemiology and Hygiene, Via di Fontana Candida 1, 00078 Monte Porzio Catone, Rome, Italy
[3] A.R.N.A.S. Ospedali Civico Di Cristina Benfratelli - Farmacia Ospedaliera, Piazza Nicola Leotta 4, 90127 Palermo, Italy
[4] Fondazione Policlinico Universitario Agostino Gemelli - Farmacia Interna, Largo Agostino Gemelli 8, 00168 Rome, Italy
[5] A.O.R.N. Antonio Cardarelli - U.O.C. Farmacia Ospedaliera, Via Antonio Cardarelli 9, 80131 Napoli, Italy
[1] Maurizio Delfini, University of Rome La Sapienza, Department of Chemistry (S. Cannizzaro Building), P.le Aldo Moro 5, 00185 Rome, Italy

ABSTRACT. Antineoplastic drugs are used to treat cancer, having their therapeutic effect by inhibiting the cell division process. Although cancer cells, due to their rapid growth, are more sensitive to the toxic effects of chemotherapeutic agents, healthy cells and tissues may also be damaged. Many studies show acute and chronic toxicity both in patients treated with chemotherapy and in exposed workers. In fact, exposure to these substances can also be linked to the formation of different types of secondary tumors. The International Agency on Research on Cancer (IARC) included some antineplastic drugs in Group 1 (carcinogenic to humans), in Group 2A (probable carcinogens for In recent years, many studies have evidenced the presence of antineoplastic drug contamination on work surfaces, materials and floors and based on these observations, international and national guidelines have been published to limit occupational exposure, with particular attention to procedures post-preparation of chemotherapy to limit as much as possible the accumulation of contaminated residues.
The aim of the following study is to determine the effectiveness of the degradation of four antineoplastic drugs: 5-fluorouracil, azacitidine, cytarabine and irinotecan using a low concentration of sodium hypochlorite solution (0.115%). The analytical platform used to monitor the degradation course of the substances under examination was hydrogen nuclear magnetic spectroscopy (^{1}H NMR). In the same experimental conditions the effectiveness of the degradation of the same antineoplastic drugs with a 99.9% ethanol solution was also evaluated. The study showed that the best degradation efficiency (> 90%) is obtained with the hypochlorite solution after 15 minutes.

Key words: *antineoplastics, degradation, NMR, sodium hypochlorite.*

RIASSUNTO. Efficacia dell'ipoclorito di sodio nella degradazione di farmaci antiblastici mediante risonanza magnetica nucleare. I farmaci antiblastici sono farmaci utilizzati nelle terapie oncologiche che svolgono il loro effetto terapeutico inibendo il processo di divisione cellulare. Sebbene le cellule tumorali, a causa del loro rapido accrescimento, siano maggiormente sensibili agli effetti tossici degli agenti chemioterapici anche le cellule e i tessuti sani possono subire un danno. L'esposizione a tali sostanze, infatti, può essere collegata anche alla formazione di diversi tipi di tumori secondari.
L'International Agency on Research on Cancer (IARC) ha inserito alcuni farmaci antiblastici classificandoli come cancerogeni per l'uomo (Gruppo 1), probabili cancerogeni per l'uomo (Gruppo 2A) e possibili cancerogeni per l'uomo (Gruppo 2B).
Negli ultimi anni molti studi hanno evidenziato la presenza di contaminazione da farmaci antiblastici su superfici di lavoro,

Introduction

Antineoplastic drugs (ADs) carry out their therapeutic effect by inhibiting the process of cell division. They damage the DNA and initiate apoptosis, preventing the development and spread of neoplastic cells. They also affect rapidly dividing normal cells, therefore are likely to suppress the bone marrow, suppress growth, impair healing, cause sterility and cause hair loss.

Some types of ADs have been linked with different kinds of second cancers. The cancers most often linked to chemotherapy are myelodysplastic syndrome and acute myelogenous leukaemia. Acute lymphocytic leukaemia has also been linked to chemotherapy. Some solid tumour cancers have also been linked to chemotherapy for certain cancers, such as testicular cancer. The classification of secondary carcinogenic effects of ADs is updated by The International Agency on research on Cancer (IARC) (1).

The main routes of exposure are the inalatory exposure to aerosols, dusts and vapours, the skin contact with chemicals, polluted surfaces, biological contaminated material and the working clothing.

The secondary carcinogenic potential for ADs is critically evaluated in terms of therapy risk assessment for cancer patients with a life-threatening disease, but it must be not an issue for physicians and personnel involved in the therapy. For the health care workers who are exposed to antineoplastic agents as part of their work practice, precautions should be taken to eliminate or reduce exposure as much as possible. Pharmacists who prepare these drugs or nurses who may prepare and/or administer them are the two occupational groups who have the highest potential exposure to antineoplastic agents. Additionally, physicians and operating room personnel may also be exposed through the treatment of patients. Hospital staff, such as shipping and receiving personnel, custodial workers, laundry workers and waste handlers, all have potential exposure to these drugs during the course of their work.

Particular attention must be paid to the preparation, transport, administration and cleaning of the areas of the manipulation of antineoplastic drugs, and during wasting operations or accidental spills.

materiali e pavimenti e sulla base di queste osservazioni sono state pubblicate delle linee guida internazionali e nazionali per limitare l'esposizione professionale, ponendo anche particolare attenzione alle procedure di pulizia post preparazione dei farmaci, per limitare quanto più possibile l'accumulo di residui contaminati.
Lo scopo del seguente studio è determinare l'efficacia della degradazione di quattro farmaci antiblastici: 5-fluorouracile, azacitidina, citarabina e irinotecano mediante una soluzione di ipoclorito di sodio a bassa concentrazione (0,115%).
La piattaforma analitica impiegata per monitorare il decorso della degradazione delle sostanze in esame è stata la spettroscopia magnetica nucleare di idrogeno (^{1}H-NMR). Nelle stesse condizioni sperimentali è stata anche valutata l'efficacia della degradazione degli stessi farmaci antiblastici con una soluzione di etanolo al 99,9%. Lo studio ha evidenziato che la migliore efficienza nella degradazione (>90%) si ottiene con la soluzione di ipoclorito dopo soli 15 minuti.

Parole chiave: antiblastici, degradazione, RMN, ipoclorito di sodio.

Cleaning of the areas, woods and not disposable individual protective equipment, such as masks or glasses, must be addressed to a complete degradation of antineoplastic agents by using a proper inactivation solution. This operation must be repeated during waste disposal, after inactivation of containers used for cleaning and their residues must be degradated by means of the inactivating solution.

The exposure of hospital personnel handling ADs since seventies was reported (2) followed by a series of studies reporting the exposure risk for hospital personnel (3-4). The consequence was the introduction of procedures dedicated to the ADs handling (5-6). Considering the epidemiologic importance of cancer, Guidelines on safe handling of ADs are expected to be continuously updated and more and more comprehensive (7).

In Italy, the safe and health of hospital workers who handle hazardous drugs is regulated by the D.L. 81/2008 in which all the preventive and corrective actions for the safe and health of workers are described.

In order to contain the workplace contamination, the preparation, compounding and administration procedures of Ads are typically carried out in the "Unità Farmaci Antiblastici" (UFA), a dedicated and restricted/controlled area. Within this area, specific handling of Ads is usually carried out using Class II Biological Safety Cabinets (BSCs). Some Class II BSCs recirculate airflow within the cabinet or exhaust contaminated air back into the work environment through HEPA filters.

However, the Class II BSCs do not prevent the generation of contamination within the cabinet and the effectiveness of such cabinets in containing hazardous drug contamination depends on operators' use of proper technique. A recent review on the exposure to cytostatic drugs verified that the presence of contamination by cytostatic drugs was confirmed in many hospitals across all five continents. In all cases, contamination was found in the cabinet, on the floor in front of the cabinet, and in other places of the Hospital Pharmacy (8).

Organic solvents, typically ethanol and isopropanol, are usually employed in the clean-up procedures of BSCs on the basis of their ability to solve organic substances as Ads are. The clean-up effectiveness is depending on the solubility of the organic contaminant, but also the type of wipe and the operative procedure play an important role. Ethanol and isopropanol are also used for the work areas sterilization but some limitation of ethanol in sanitization of work surfaces of BSCs as the inefficacy against spores and non lipid viruses, has pointed out (9). Other concerns in the use of organic solvents are linked to their flammability, and incompatibility to some material (*i.e.* rubber hardening and glue dissolution).

The chemical deactivation of a hazardous substance is generally preferred to the clean-up, but no single process has been found to deactivate all currently available hazardous drugs (10).

In Italy, the first recommendations on the decontamination procedures of the workplace are reported in a specific guideline by I.S.P.E.S.L. (11) containing, among others, indication on the use of sodium hypochlorite solution for the work surfaces clean-up.

The efficacy of sodium hypochlorite in the cleaning of contaminated surfaces by ADs was previously reported (12). The decontamination effectiveness is higher than the typical cleaning procedure with organic solvents or cleansers, as a complete and fast chemical degradation (oxidation) due to the high oxidative reductive potential (ORP) of these solutions is to be associated to the solvent effect and high pH. It is usually also recommended for biological decontamination especially in case of accidental blood and biological fluids spillage (13).

The effectiveness of chemical oxidation by sodium hypochlorite strongly depends on concentration, as ORP is function of the hypochlorite ion concentration according to Nernst equation. A fast and complete degradation is usually obtained at high concentration sodium hypochlorite solutions (bleach). However, bleach is very corrosive and will cause damage of stainless-steel, over time, if used to clean the BSC bench. A corroded bench is more difficult to efficiently be cleaned due to the physical and chemical modification of the surface.

Diluted solutions of sodium hypochlorite can be used to clean up the BSCs bench and preserve the bench surface form corrosion. However, the efficacy in the decontamination should be evaluated, the oxidative degradation being dependent on the ADs chemical structure.

The evaluation of the efficacy of sodium hypochlorite in the decomposition of organic substances is carried out by determining the degradation time and residual concentration at a fixed end time.

Currently, chromatographic techniques are primarily employed for the determination of residual concentration of organic contaminant, due to specificity and sensitivity. Besides the chromatographic techniques, the use of NMR for quantitative purposes is growing due to recent advances in technology and software (14). Nuclear magnetic resonance (NMR) spectroscopy is a primary analytical methodology, because a direct proportionality exists between the signal integral and the number of protons (^{1}H) giving rise to it. In particular, quantitative ^{1}H-NMR has certain advantages: (i) structural and quantitative data can

be obtained simultaneously; (ii) the time for sample preparation is relatively short; (iii) NMR is non-destructive; and (iv) simultaneous determination of several classes of analyte in a mixture is possible, (iv) no compound-specific recalibration is required. The analytical performances in terms of accuracy, precision and linearity are very high as internal standards are usually used and they can be fully validated (15). Last but not least, a series of measures can be performed at different times on the same sample. These characteristics make NMR very suitable for the determination of the reaction kinetics (16, 17).

The aim of this study was the determination of the efficacy of 0,115 % solution of sodium hypochlorite in the decomposition of ADs by means of ^{1}H-NMR spectroscopy. In order to evaluate the effect of chemical structure of ADs on the extent of the decomposition four different ADs were selected: 5-fluorouracil, azacitidine, cytarabine and irinotecan. End point for the test was > 90% degradation within 15 minutes reaction time. The analytical methodology was validated and linearity, precision accuracy and sensitivity (limit of quantization) were evaluated. The efficacy of the 0,115% sodium hypochlorite solution was compared to a 99,9% ethanol solution in the same experimental conditions.

Materials and methods

Materials

5-Fluorouracil, Azacitidine, Cytarabine, Irinotecan, deuterated water, deuterated ethanol, 3-(trimethylsilyl)-propionic-2,2,3,3-d4 acid sodium salt (TSP) and hexamethyldisiloxane (HMDSO) were purchased from Sigma-Aldrich (St. Louis, MO, USA). Amuchina® (sodium hypochlorite 1,15 % (w/v), Batch 0001, Cod. 419601) was provided by A.C.R.A.F. S.p.A. (Rome, Italy).

Each molecule was dissolved in 0,6 mL D_2O containing TSP 2mM as chemical shift and concentration reference. Standard solutions were prepared dissolving an amount of analyte in D_2O up to a final concentration of 2,0 mM.

For degradation study 67,0 µl of Amuchina® were added to 0,6 ml of solution with an analyte concentration of $2{,}0 \times 10^{-4}$M.

For degradation study in ethanol, the molecules were dissolved in 0,6 mL of CD_3CD_2OD containing HMDSO 2,0 mM as chemical shift and concentration reference. The concentrations of the analytes were the same of the water stock solutions.

NMR experiments

All spectra were recorded at 298 K on a Bruker AVANCE III spectrometer operating at the proton frequency of 400,13 MHz and equipped with a Bruker multinuclear z-gradient inverse probehead. ^{1}H spectra were acquired employing the *presat* pulse sequence for solvent suppression with 96 transients, a spectral width of 6000 Hz and 64K data points for an acquisition time of 5,5 s. The recycle delay was set to 6,55 s in order to achieve complete resonance relaxation between successive scansions.

The spectra were processed with Topspin 2.0 software. The acquired FID were zero-filled, multiplied for an exponential function with a line broadening of 1 Hz, transformed, phased and then the baseline was manually corrected.

The analytical signal used for quantitative analysis (Response) was obtained from the ratio between the normalized area of the analytical signal of each drug and the normalized area of the internal standard. TSP reference area (singlet at 0,00 ppm originated from 9 hydrogen) was set to the value of 9 and the resonance areas selected for quantification are normalized by the number of hydrogen generating each resonance. The ratios of the various signals were then calculated according to the formula a):

$$\text{a) Response} = \frac{\text{Area signal } \frac{1}{n}^{\circ} \text{ of hydrogens signal 1}}{\text{Area signal } \frac{2}{n}^{\circ} \text{ of hydrogens signal 2}}$$

For degradation studies in ethanol, the same equation was employed, but the reference is HMDSO, resonating at 0,87 ppm with a reference area set to the value of 18 due to its resonance being originated from 18 hydrogens.

The least square algorithm from Excel software was employed for the calculation of the calibration lines.

Results

Resonance identification

A preliminary ^{1}H NMR study was carried out on each examined AD in order to identify the most suitable resonances for quantification. Resonance assignment was carried out on the basis of literature data (18-20). The ^{1}H spectra and the corresponding assignments are reported Figures 1-4 and Tables I-IV), while the resonances chosen for quantification are reported in Table VI.

These resonances were chosen for the degradation studies of both hypochlorite and ethanol since the ethanol spectra are analogues to the other former with minor chemical shift variations due to the solvent effect. The only difference is the presence of resonances due to the non-deuterated ethanol residue.

Validation study

The validation of the method was carried out in accordance with the Eurolab guidelines for the development and validation of NMR methods (21). The performance of the method was evaluated in terms of selectivity, linearity, precision, detection and quantification limits.

Selectivity

The method had a high degree of selectivity of since the resonances chosen for quantification do not overlap with the other resonances of the molecules, as well with as the solvent one. Moreover, no interference with the resonances of the degradation products were detected.

Table I. ***5-Fluorouracil resonance assignment***

Chemical shift (ppm)	Multiplicity	J(Hz)	Number of hydrogens	Assignment
7.55	**Doublet**	**4,86**	**1**	**6**

Table II. ***Azacitidine resonance assignment***

Chemical shift (ppm)	Multiplicity	J(Hz)	Number of hydrogens	Assignment
8,58	**Singlet**	**-**	**1**	**6**
5,79	Doublet	2,84	1	1′
4,40	Multiplet	-	1	2′
4,26	Multiplet	-	1	3′
4,15	Multiplet	-	1	4′
3,96 e 3,82	Multiplet	-	1	5′ and 5′′

Table III. ***Cytarabine resonance assignment***

Chemical shift (ppm)	Multiplicity	J(Hz)	Number of hydrogens	Assignment
7,81	**Doublet**	**7.70**	**1**	**6**
6,05	Doublet	7.70	1	5
6,21	Doublet	4.91	1	1′
4,41	Multiplet	-	1	2′
4,12	Multiplet	-	1	3′
4,01	Multiplet	-	1	4′
3,92 e 3,84	Multiplet	-	1 and 1	5 e 5′′

Table IV. ***Irinotecan resonance assignment***

Chemical shift (ppm)	Multiplicity	J(Hz)	Number of hydrogens	Assignment
7,21	Singlet		1	21
7,78	**Doublet**	**9,26**	**1**	**5**
7,38	Double doublet	9,26; 1,91	1	6
7,63	Doublet	1,91	1	8
1,70 (eq); 1,40 (ax)	Multiplet	-	2	4′′
1,83	Multiplet	-	2 and 2	3′′, 5′′
3,35 (eq); 2,97 (ax)	Multiplet	-	2 and 2	2′′,6′′
3,50 ppm	Multiplet	-	1	4′
2,26 (eq); 1,72 (ax)	Multiplet	-	2	3′
4,59 (eq); 3,15 (ax)	Multiplet	-	2	2′
4,32 (eq); 3,05 (ax)	Multiplet	-	2	6′
2,18 (eq); 1,82 (ax)	Multiplet	-	2	5′
3,13	Quadruplet	7,11	2	10-Ethyl–(CH_2)
1,31	Triplet	7,11	3	10- Ethyl–(CH_3)
5,05; 5,14	Doublets	17,79	2	12 (CH_2)
5,44; 5,62	Doublets	17,79	2	16 (CH_2)
2,01	Quadruplet	6,79	2	19- Ethyl–(CH_2)
1,01	Triplet	6,79	3	19- Ethyl–(CH_3)

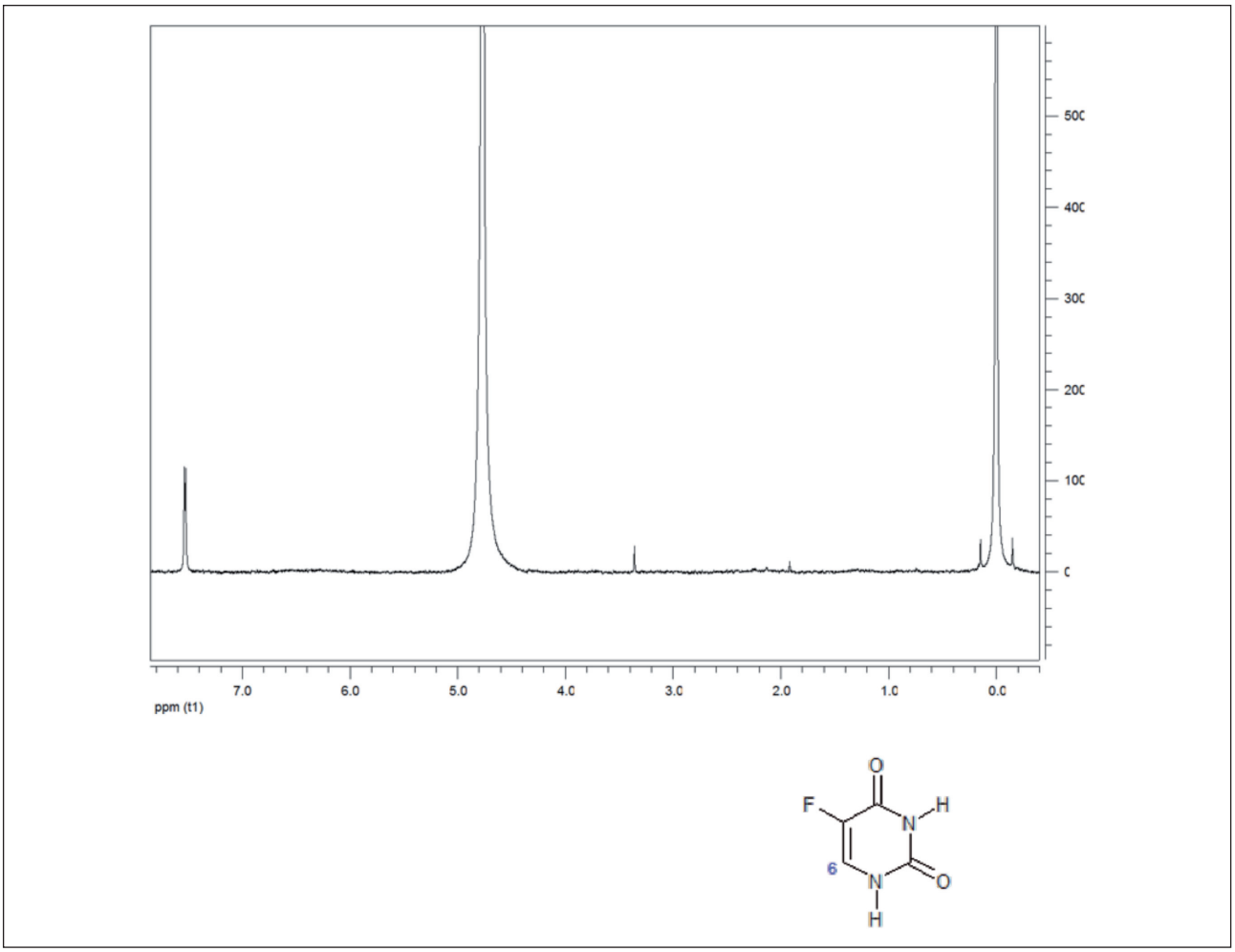

Figure 1. *^{1}H-NMR of 5-Fluorouracil*

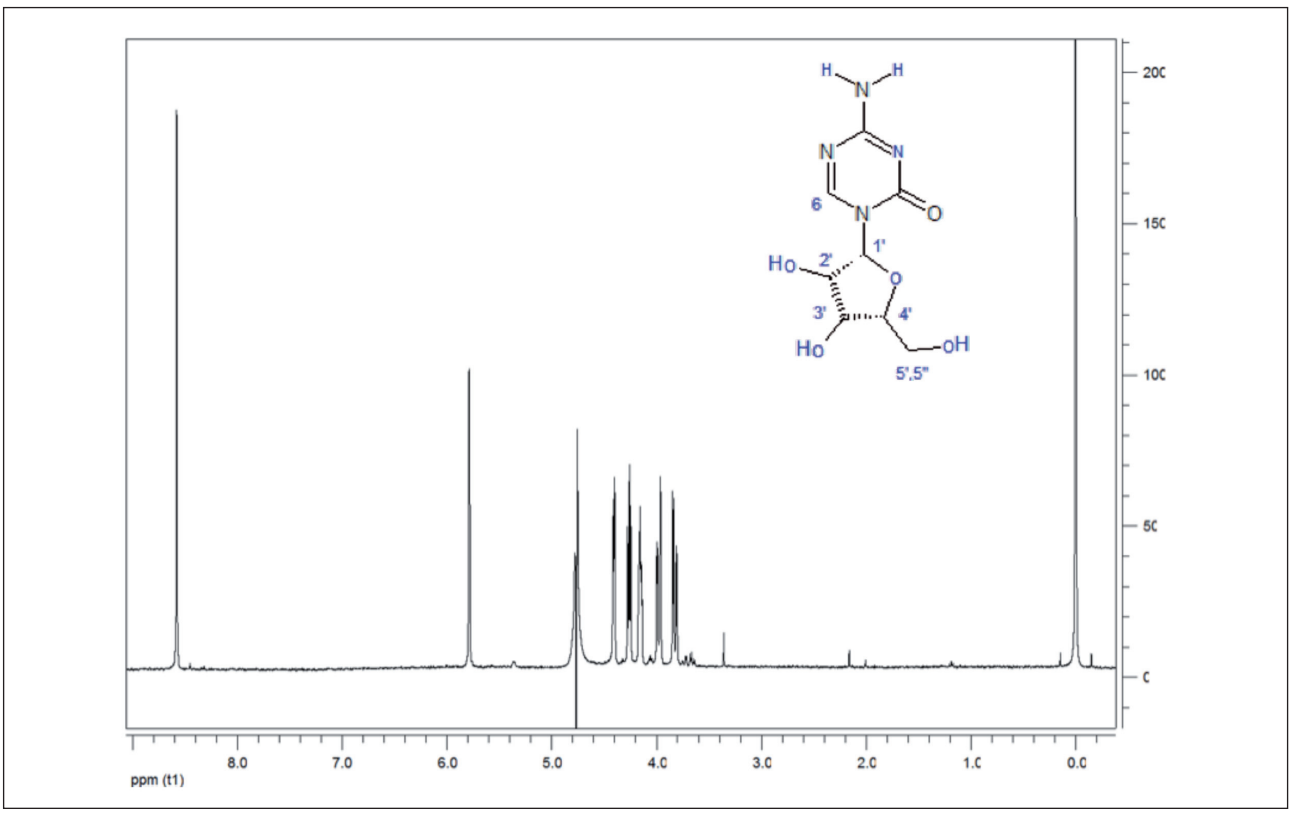

Figure 2. *^{1}H-NMR of Azacitidine*

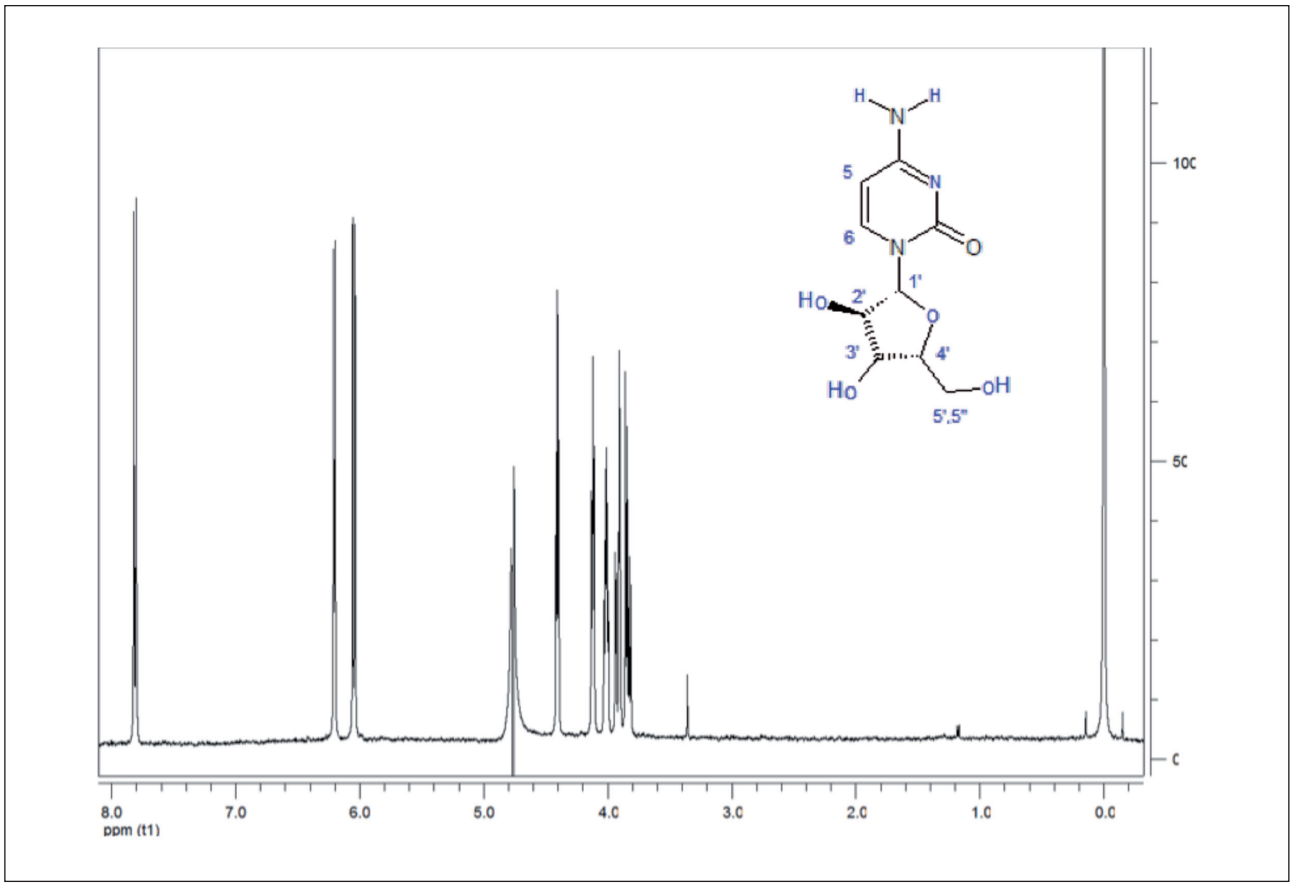

Figure 3. *^{1}H-NMR of Cytarabine*

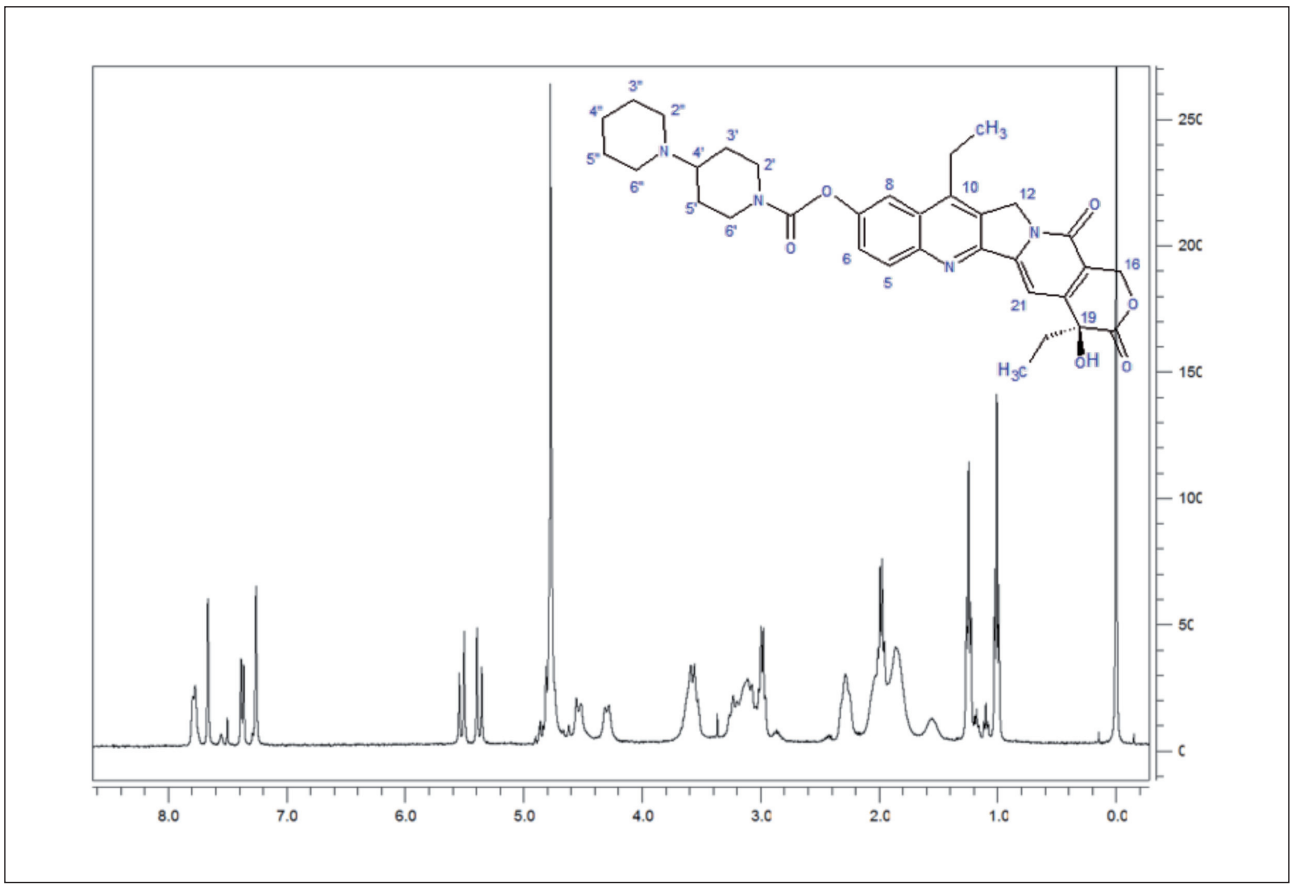

Figure 4. *^{1}H-NMR of Irinotecan*

Linearity

The range of linearity was evaluated by measuring the response of each molecule in seven dilutions of a 2,0 x 10^{-3} M stock solution (s.s.).

For each solution a NMR spectrum was acquired and the responses were reported as a function of concentration. The data were analyzed by a least square algorithm and the R^2 of all the molecules resulted to be > 0,99 (Fig. 5-8).

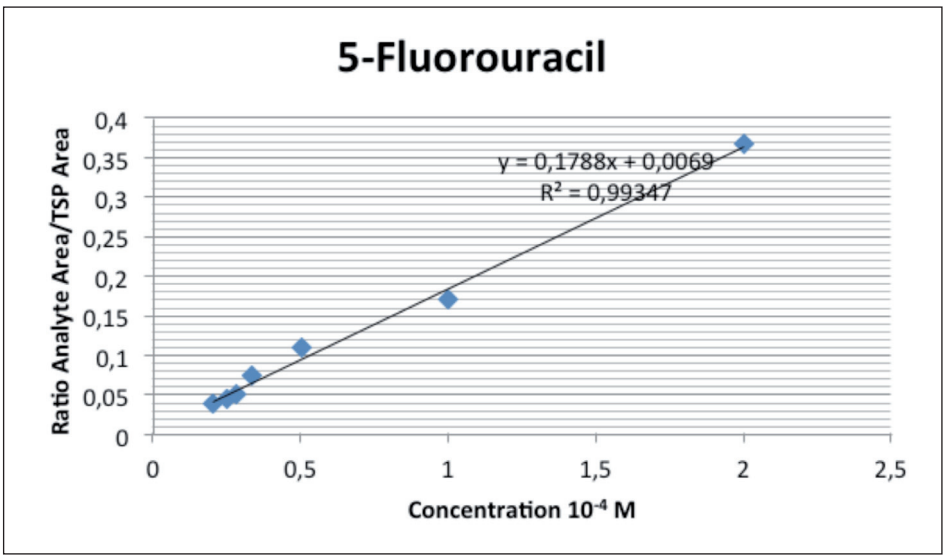

Figure 5. *5-Fluorouracil linearity study*

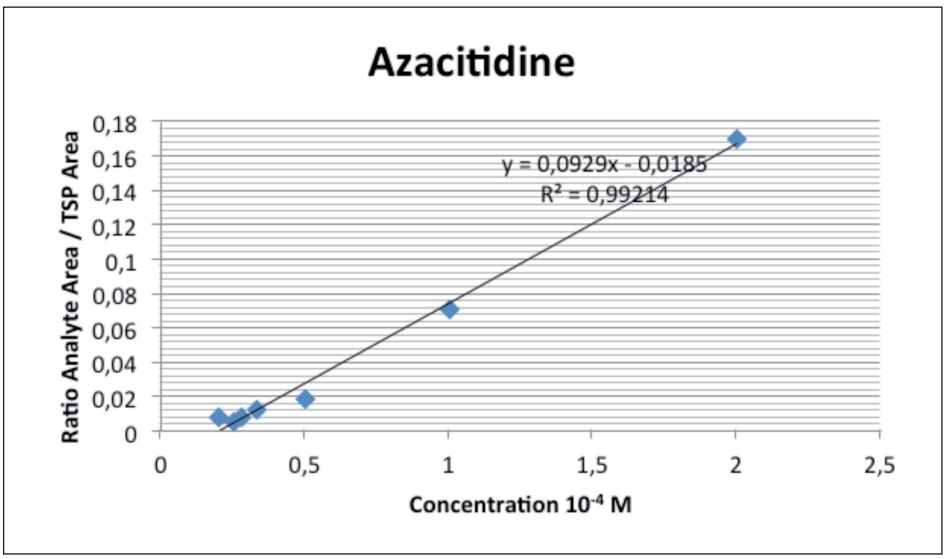

Figure 6. *Azacitidine linearity study*

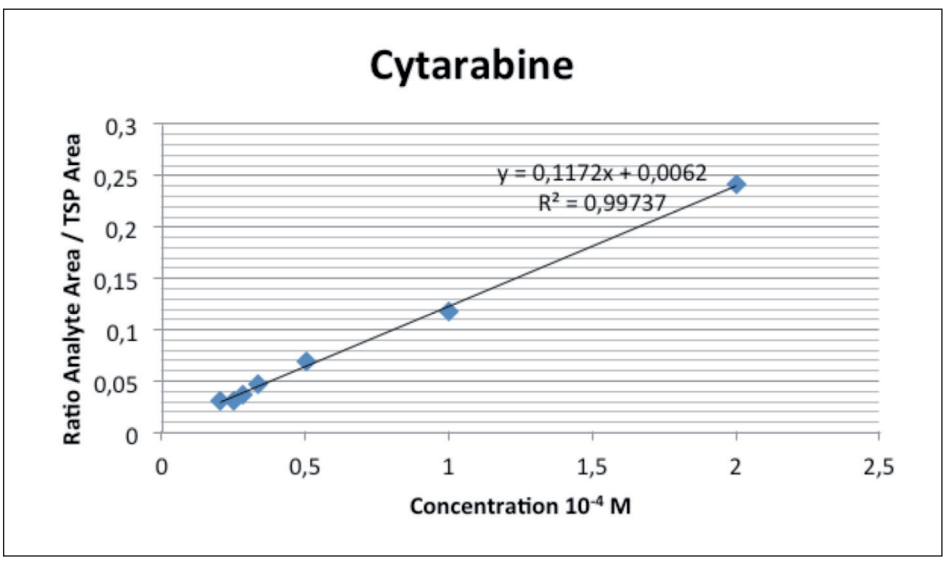

Figure 7. *Cytarabine linearity study*

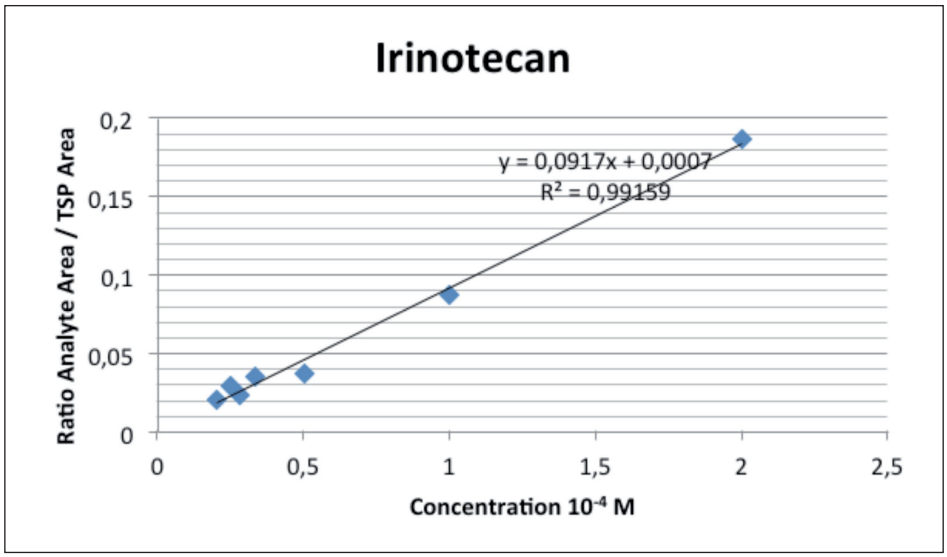

Figure 8. *Irinotecan linearity study*

Precision

Six consecutive spectra at the lowest concentration (2,0 x10^{-5}M) were acquired for all analytes and the method precision was evaluated by the mean and relative standard deviation of the responses.

Detection and quantification limits

The limit of detection (LOD) and the limit of quantification (LOQ) were estimated as the average of six replicas at the lowest concentration (2,0 x10^{-5}M).

LOD was calculated as the amount of analyte providing a peak three time stronger than the baseline noise (S/N =3) while LOQ was calculated as the amount of analyte providing a peak ten time stronger than the baseline noise (S/N =10). Results are reported in Table V.

Table V. *Limit of detection and quantification*

Analyte	LOD (mM)	LOQ (mM)
5-Fluorouracil	0,00251	0,00838
Azacitidine	0,00410	0,01367
Cytarabine	0,00669	0,02229
Irinotecan	0,00418	0,01395

Degradation studies

In order to evaluate the effect of hypochlorite (0,115% of NaClO), 67 µl of concentrated Amuchina® (1,15% of NaClO) were added to the NMR tube (volume 0,6 ml) with an analyte concentration of 2x10^{-4} M. ^{1}H NMR experiments were performed before (t=0 min) and at 15 minutes after Amuchina® addiction (t=15 min). In each spectra (Fig. 9-12) the response was calculated and the ratio of the responses between the two times was employed to evaluate the drug degradation (Table VI). It is possible to observe a % degradation of more than 90% for all molecules after 15 minutes.

In order to evaluate the degradation effect of ethanol on the analytes, ^{1}H NMR experiments were performed after the initial solubilization (t=0 min) and after 15 minutes (t=15 min).

In each spectra (Fig. 13-16) the response was calculated and the ratio of the responses between the two times was employed to evaluate the drug degradation (Table VII). It is possible to observe the absence of any change between the two observation times.

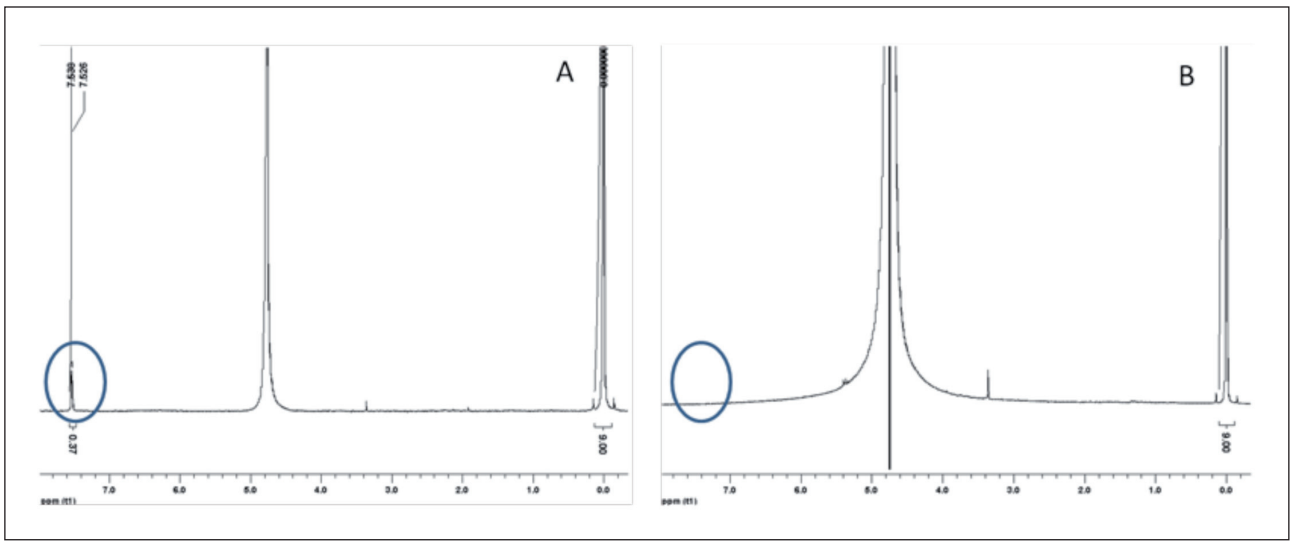

Figure 9. *^{1}H spectrum of 5-Fluorouracil in D_2O A) t=0 min, B) after hypochlorite addiction*

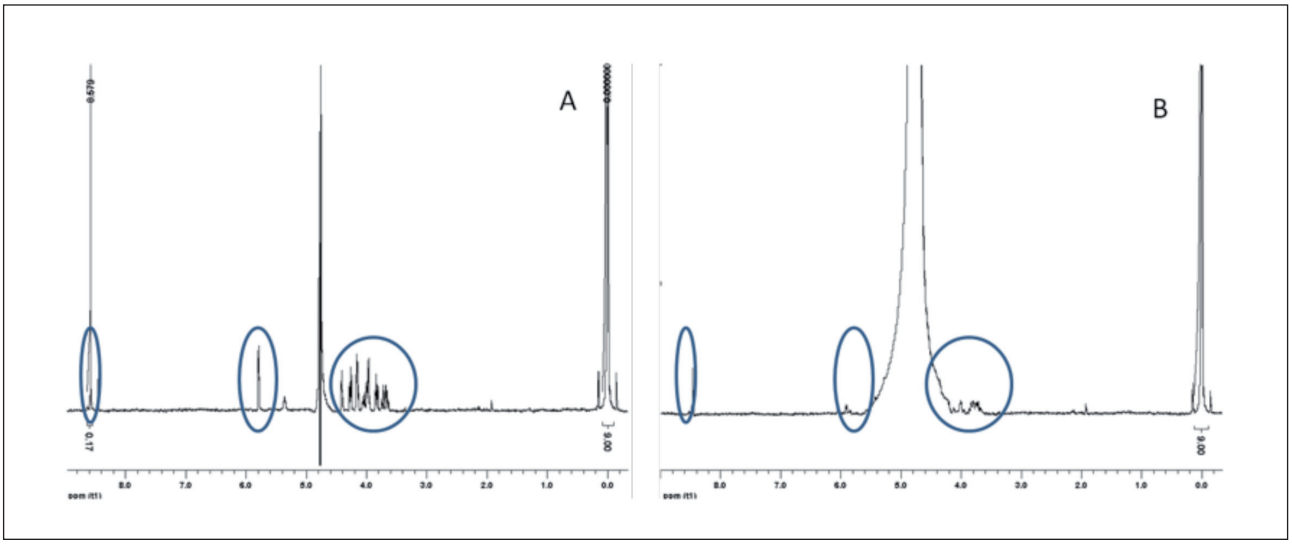

Figure 10. *^{1}H spectrum of Azacitidine in D_2O A) t=0 min, B) after hypochlorite addiction*

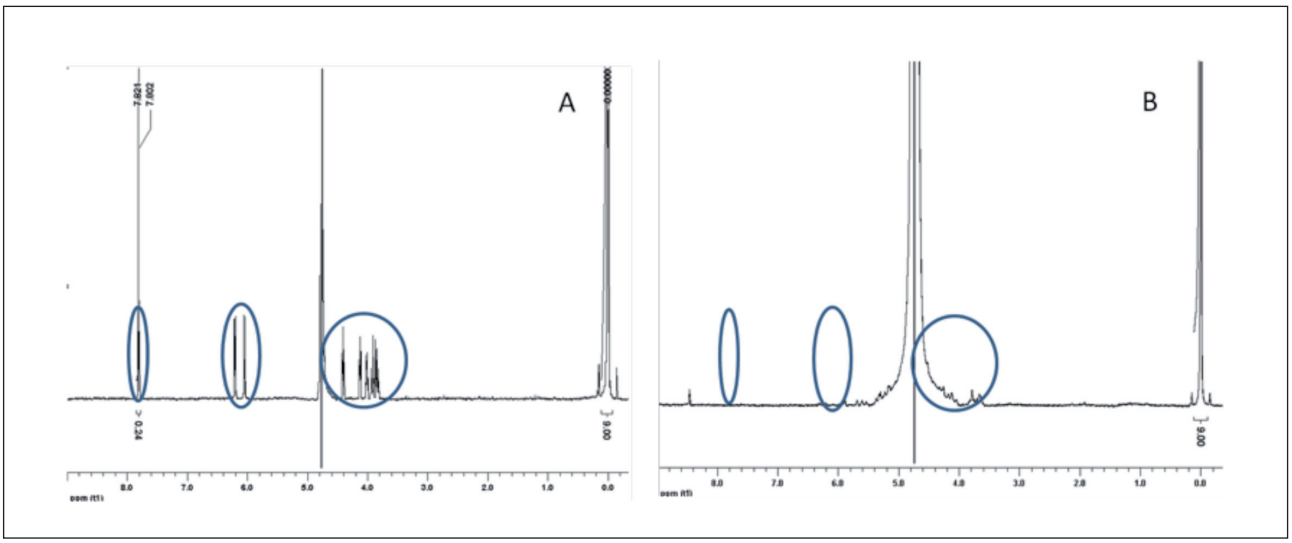

Figure 11. *^{1}H spectrum of Cytarabine in D_2O A) t=0 min, B) after hypochlorite addiction*

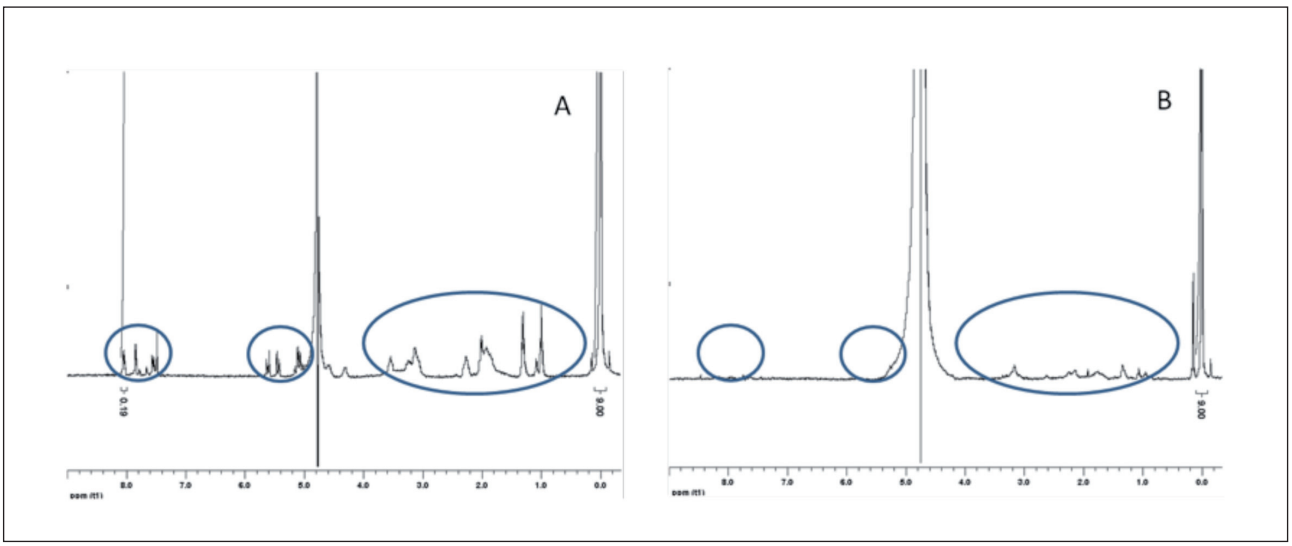

Figure 12. *^{1}H spectrum of Irinotecan in D_2O A) t=0 min, B) after hypochlorite addiction*

Table VI. ***Degradation study of the analytes in hypochlorite***

Analyte	Analytical signal (ppm)	Analyte Area/ Reference Area t=0	Analyte Area/ Reference Area t=15	% of degradation (after 15 min)
5-Fluorouracil	7,55 ppm	0,3681	0,0040	>90
Azacitidine	8,58 ppm	0,1704	0,0036	>90
Cytarabine	7,81 ppm	0,2420	0,0238	>90
Irinotecan	8,05 ppm	0,1871	0,0112	>90

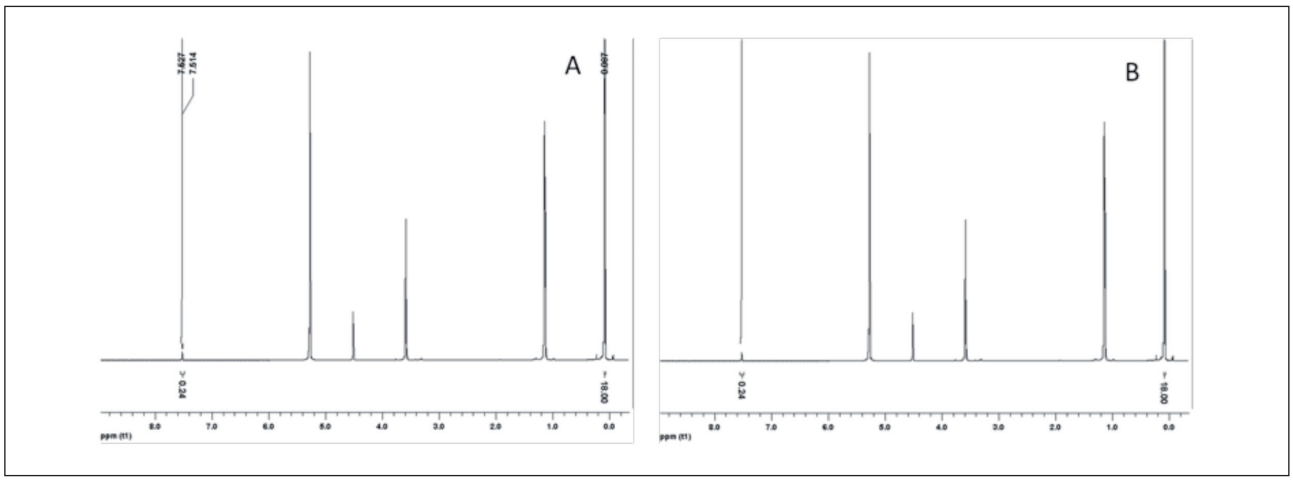

Figure 13. ***^{1}H spectrum of 5-Fluorouracil in ethanol A) t=0 min, B) t=15 min***

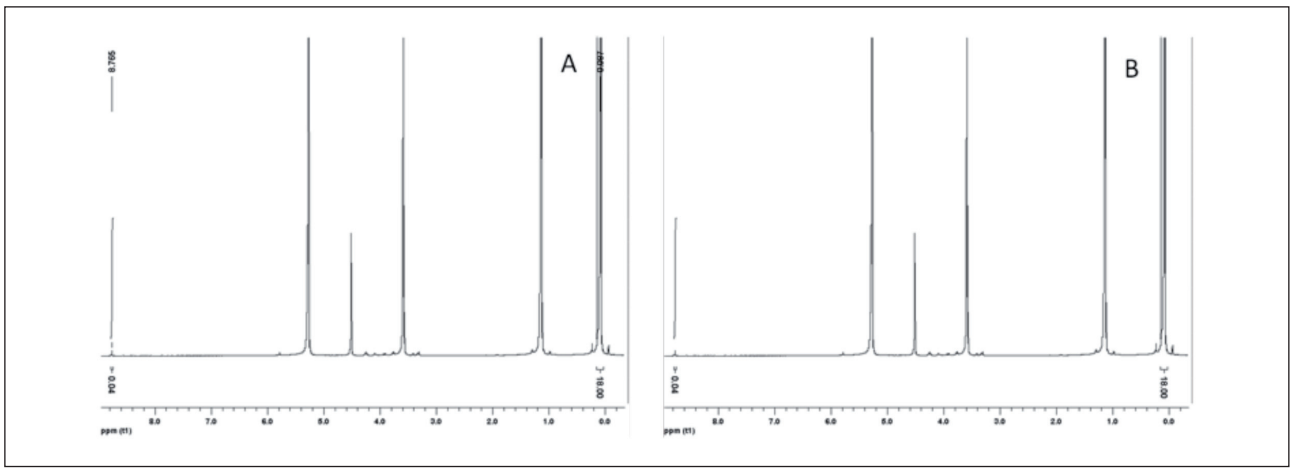

Figure 14. ***^{1}H spectrum of Azacitidine in ethanol A) t=0 min, B) t=15 min***

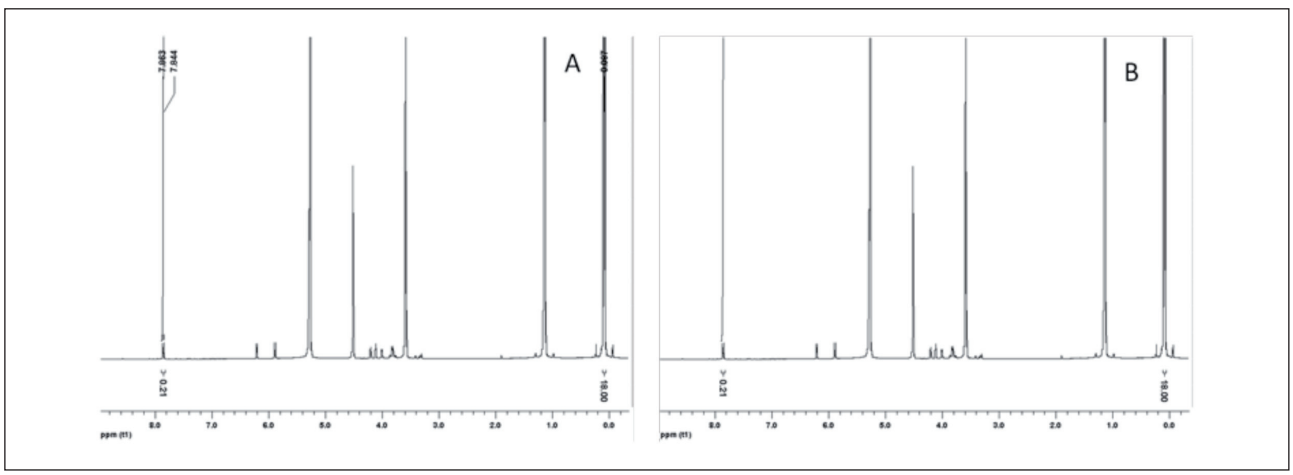

Figure 15. ***^{1}H spectrum of Cytarabine in ethanol A) t=0 min, B) t=15 min***

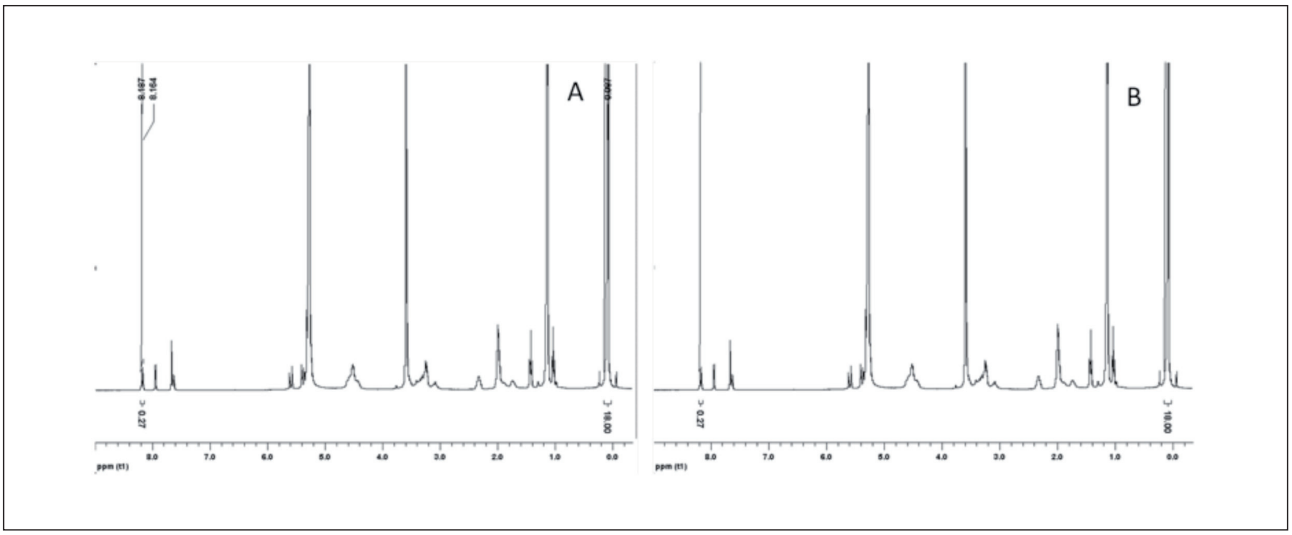

Figure 16. ***^{1}H spectrum of Irinotecan in ethanol A) t=0 min, B) t=15 min***

Table VII. ***Degradation study of the analytes in ethanol***

Analyte	Analytical signal (ppm)	Analyte Area/ Reference Area t=0	Analyte Area/ Reference Area t=15	% of degradation (after 15 min)
5-Fluorouracil	7,52	0,24	0,24	<0
Azacitidine	8,77	0,04	0,04	<0
Cytarabine	7,85	0,21	0,21	<0
Irinotecan	8,17	0,27	0,27	<0

Discussion

National and international scientific studies evaluate different interesting decontamination solutions for some antineoplastic agents of work surfaces in order to validate the efficiency of cleaning procedures in hospital oncology pharmacy. In particular, seven different cleaning procedures after controlled contamination of the work surface of a biological safety cabinet workbench in an Italian hospital oncology pharmacy (22); moreover eight hospitals were investigated by means of wipe sampling for surface residue determination and tested for five ADs considered suitable exposure markers (23). A large-scale study was carried out in order to determine the contamination level of some antineoplastic drugs in pharmacies and to investigate the suitability and effects of a large number of wipe samples monitoring at regular intervals (24). Finally a work has evaluated health care worker exposure to antineoplastic drugs by cross-sectional study examined environmental samples from pharmacy and nursing areas (25). These studies were performed analyzing wipe samples of work surfaces, by using liquid chromatography tandem mass spectrometry.

The present work has been focused on the study of the efficacy of decontamination media, by using Amuchina® and Ethanol, following the chemical degradation of the active principle of ADs by NMR spectroscopy. The tested solutions for decomposition were Amuchina® and Ethanol, at concentration ranged from 2 X 10 $^{-4}$M to 2 X10 $^{-5}$ M.

The final aim of the study was the comparison of efficacy of both decomposition solutions in the degradation of some ADs, as 5-fluorouracil, azacitidine, cytarabine and irinotecan. At present, the decomposition solutions examined are the most useful in the cleaning procedures of contaminated surfaces in the areas dedicated to the manipulation and administration of chemioterapic drugs.

The efficacy of the 0,115% sodium hypochlorite solution was compared to a 99,9% ethanol solution in the same experimental conditions and tested on ADs solutions, prepared by diluting a stock solution of 2,0 x 10^{-3} M.

From the comparison of the degradations processes, it is clearly observable the greater efficiency of the sodium hydrochloride over ethanol.

For all the examined analytes, it is possible to observe a complete degradation (> 90%) within 15 minutes of hypochlorite addictions. On the contrary, no degradation (< 0%) was evidenced by ethanol addictions. The details are reported in the Figures 9-16.

This behavior can be explained on the basis of the characteristic of ethanol. In fact it is largely employed as antiseptic and disinfectant, its antimicrobial action is linked to its ability to denaturize proteins and to disrupt lipid associations present in bacteria and fungi, but the same mechanism do not cause degradation of the chemical bonds of molecules. On the other hand the degradation efficiency of hypochlorite can be attributed to its oxidizing properties (E°=1,63 V), which can easily disrupt the aromatic moieties of the examined ADs.

Conclusions

Our study shows how it is important a routine check of antineoplastic drugs in order to ensure the efficacy of exposure risk management procedures. The choice of NMR for checking this procedures is due to the fact that it is an extremely reproducible analytical method, able to uniquely identify the presence/absence of the chemical investigated and able to reduce the costs and time of analyses for sampling preparation.

This work represents a first preliminary study with NMR technique in the field of risk exposure to chemioterapics and could be extended to other classes of antineoplastic drugs in order to be considered a tool for checking the management of exposure of hospital personnel.

References

1) Agents classified by the IARC monographs, volumes 1-109. (http://monographs.iarc.fr/ENG/Classification/ClassificationsAlphaOrder.pdf)
2) Falck K, Gröhn P, Sorsa M, et al. Mutagenicity in urine of nurses handling cytostatic drugs. Lancet 1979; 1: 1250-51.
3) Cloak MM, Connor TH, Stevens KR, et al. Occupational exposure of nursing personnel to antineoplastic agents. Oncol Nurs Forum 1985; 12: 33-9.
4) Fishbein L. Perspectives on occupational exposure to antineoplastic drugs. Arch Geschwulstforsch 1987; 57: 219-48.
5) Gruppo di lavoro dell'Associazione Italiana di Medicina Preventiva dei Lavoratori della Sanità. Summary of recommendations for a rational implementation of the ministry of health guidelines on the prevention of occupational risks in handling antineoplastic agents. Med Lav 2001; 92: 137-48.
6) Center for disease control and prevention. NIOSH alert: preventing occupational exposure to antineoplastic and other hazardous drugs in health care settings. DHHS - NIOSH Publication Number 2004-165.
7) Petrelli G, Palmi S. Esposizione professionale a chemioterapici antiblastici: rischi per la riproduzione e strategie per la prevenzione. Roma: Istituto Superiore di Sanità; 2002 (Rapporti ISTISAN 02/16).
8) Lancharro PM, De Castro-Acuña Iglesias N, González-Barcala FJ, et al. Evidence of exposure to cytostatic drugs in healthcare staff: a review of recent literature. Farm Hosp 2016; 40: 604-21.
9) World Health Organization. Laboratory biosafety manual. 3rd ed. 2004, rev. 2014. (https://www.who.int/csr/resources/publications/biosafety/Biosafety7.pdf)
10) American Society of Heath-System Pharmacists – ASHP Guidelines on Handling Hazardous Agents. 2004. (https://www.ashp.org/-/media/assets/policy-guidelines/docs/guidelines/handling-hazardous-drugs.ashx).
11) I.S.P.E.S.L. Documento di Linee Guida per la Sicurezza e la salute dei lavoratori esposti a chemioterapici in ambiente ospedaliero. GU n. 236 del 7 ottobre 1999.
12) Perico A, Lorini C, Biliotti T, et al. Valutazione dell'esposizione professionale a chemioterapici antiblastici in ambiente ospedaliero in Toscana Ann Ig 2003; 15: 363-71.
13) Royal Free Hospital NHS Trust and Royal Free and University College Medical School of University College London (royal free campus). Spillage management arrangements and guidance. 2003. (https://www.scribd.com/document/107490302/Spillage-Management).
14) Gallo V, Intini N, Mastrorilli P, et al. Performance assessment in fingerprinting and multi component quantitative NMR analyses. Analytical Chemistry 2015; 87: 6709-17.
15) Gödecke T. Napolitano JG, Rodríguez-Brasco MF, et al. Validation of a Generic Quantitative ^{1}H NMR Method for Natural Products Analysis Phytochem Anal 2013; 24: 581-97.
16) Cheenou H, Aaron P, Alonzo Justin Y. Vang et al. Real-Time Enzyme Kinetics by Quantitative NMR Spectroscopy and Determination of the Michaelis–Menten Constant Using the Lambert-W Function. J Chem Educ 2015; 92: 1943-8.
17) Maiwald M, Grützner T, Ströfer E, et al. Quantitative NMR spectroscopy of complex technical mixtures using a virtual reference: chemical equilibria and reaction kinetics of formaldehyde-water-1,3,5-trioxane. Anal Bioanal Chem 2006; 385: 910-7.
18) Wishart DS, Jewison T, Guo AC, et al. HMDB 3.0 - The Human Metabolome Database in 2013. Nucleic Acids Res 2013; 41: D 801-7.
19) Sułkowska A, Bojko B, Równicka J, et al. Competition of cytarabine and aspirin in binding to serum albumin in multidrug therapy. Biopolymer. 2006; 81: 464-72.
20) D'Amelio N, Aroulmoji V, Toraldo A, et al. Aggregation properties and structural studies of anticancer drug Irinotecan in DMSO solution based on NMR measurements. J Mol Str 2012; 1013: 26-35.
21) Schönberger T, Monakhov YB, Lachenmeier et al. Guide to NMR Method Development and Validation – Part 1: Identification and Quantification. Eurolab Technical Report number 01/2014. (www.eurolab.org).
22) Negri S, Oddone E, Morandi F, et al. Validation of cleaning procedures used in an Italian Hospital Pharmacy for antineoplastic drug decontamination: a new tool for industrial hygiene. Med Lav 2019; 110(2): 93-101.
23) Sottani C, Grignani E, Oddone E, et al. Monitoring Surface Contamination by Antineoplastic Drugs in Italian Hospitals: Performance-Based Hygienic Guidance Values (HGVs) Project. Ann Work Expo Health. 2017;61(8):994-1002.
24) Kiffmeyer TK, Tuerk J, Hahn M et al. Application and assessment of a regular environmental monitoring of the antineoplastic drug contamination level in pharmacies - the MEWIP project. Ann Occup Hyg. 2013; 57: 444-55.
25) Connor TH, DeBord D G, Pretty J R et al. Evaluation of antineoplastic drug exposure of health care workers at three university-based US cancer centers. J Occup Environ Med 2010; 52: 1019-27.

Correspondence: *Prof. Maurizio Delfini, University of Rome La Sapienza, Department of Chemistry (S. Cannizzaro Building), P.le Aldo Moro 5, 00185 Rome, Italy, Tel. and Fax -39 6 49913124, mauriziodelfini@uniroma1.it*