

## LC–MS characterization of metoclopramide photolysis products

Aubert Maquille<sup>a,\*</sup>, Jean-Louis Habib Jiwan<sup>b</sup>

<sup>a</sup> Laboratory of Chemical and Physico-chemical Analysis of Drugs (CHAM), Louvain Drug Research Institute, Université catholique de Louvain (UCL), Avenue E. Mounier, 72 bte 7230, B-1200 Brussels, Belgium

<sup>b</sup> Laboratory of Mass Spectrometry, Université Catholique de Louvain (UCL), 1348 Louvain-La-Neuve, Belgium

### ARTICLE INFO

#### Article history:

Received 18 December 2008

Received in revised form 29 April 2009

Accepted 4 May 2009

Available online 12 May 2009

#### Keywords:

Metoclopramide  
Photolysis products  
HPLC–DAD  
LC–MS

### ABSTRACT

The aim of this study was to investigate the photodegradation of the antiemetic drug metoclopramide. Metoclopramide aqueous solutions were photoirradiated at 254 nm under argon atmosphere. Irradiated metoclopramide solutions were analyzed by high performance liquid chromatography–ion trap mass spectrometry in order to characterize photolysis products. Rapid decrease in metoclopramide purity, following first-order kinetic, was observed following irradiation. The structures of 18 photolysis products were tentatively identified based on their mass spectra and fragmentation. The main degradation mechanism was scission of the chlorine which could be followed by polymerization of the resulting products since dimeric and trimeric products were observed.

© 2009 Elsevier B.V. All rights reserved.

### 1. Introduction

A wide number of drugs are susceptible to undergo degradation when exposed to light. Light induced decomposition of pharmaceuticals could result in the loss of potency and to adverse effects due to the formation of toxic photodegradation products [1,2]. Stability tests on drugs, including light stress tests are now required by the European Agency for the Evaluation of Medicinal Products (EMA) [3] to check the stability of the active substance. However, the photostability of many existing drug has not been fully evaluated.

Furthermore, an environmental issue arises from the increasing presence of pharmaceutical compounds in surface water. Photodegradation of these drugs occurs following exposure to sun light [4]. In sewage treatment plants, photochemical processes applied to the degradation of organic compounds in aqueous medium was suggested [5]. Therefore, it is of fundamental importance to study the drugs photodegradation products to gain insight on the fate of pharmaceuticals on the environment.

Metoclopramide (4-amino-5-chloro-N-(2-(diethylamino)ethyl)-2-methoxybenzamide), is a benzamide drug that requires protection from the light during its storage [6]. It is still widely used to treat nausea and vomiting and for the relief of migraine [7,8]. Despite its known photosensitivity, metoclopramide photodegradation has not been fully studied [9] as neither the structures of

the photodegradation products and the degradation mechanisms following light exposure have been elucidated. The investigation of the photodegradation products of this drug according to the current status of art is of crucial interest for a better understanding of the photodegradation mechanisms. Liquid chromatography tandem mass spectrometry (LC–MS–MS) provides a useful tool for structure elucidation of photolysis products [10–12].

This study aims toward the photochemistry of metoclopramide, focusing on the characterization of photodegradation products from metoclopramide pharmaceutical solutions which could be useful to gain insight on the photodegradative pathway. Moreover, some photolysis products observed in this study could also be formed in photoirradiated wastewater. Photolytic degradation of metoclopramide aqueous solutions was carried out using a photoreactor equipped with 254 nm UV lamps, which is close to the drug maximal absorption wavelength. The evolution of the drug concentration and the degradation products as a function of the irradiation time were studied by HPLC–UV whilst degradation products were characterized by LC–ion trap MS. The structures of the photodegradation products were compared to those produced after gamma and electron-beam radiolysis of metoclopramide aqueous solutions [13] in order to compare radiolysis and photolysis mechanisms of this drug.

### 2. Materials and methods

#### 2.1. Materials

Metoclopramide hydrochloride (4-amino-5-chloro-N-(2-(diethylamino)ethyl)-2-methoxybenzamide) monohydrate (99%) was purchased from Sigma–Aldrich (St Louis, MO, USA) and was

\* Corresponding author. Tel.: +32 2 764 72 94; fax: +32 2 764 72 96.

E-mail addresses: [aubert.maquille@uclouvain.be](mailto:aubert.maquille@uclouvain.be) (A. Maquille), [j-l.habib@uclouvain.be](mailto:j-l.habib@uclouvain.be) (J.-L. Habib Jiwan).

stored in the dark. Ammonium acetate was from UCB (Brussels, Belgium). HPLC supra-gradient acetonitrile was supplied from Biosolve (Valkenswaard, Netherlands). Deionized water from a Millipore Milli-Q water purification system was used. Argon was supplied by Air-Liquide (Liège, Belgium).

## 2.2. Samples

Three milliliters of metoclopramide  $1 \text{ mg mL}^{-1}$  solutions prepared with deionized water were put in quartz cells (1 cm path length) closed with Teflon caps. Oxygen was removed by bubbling argon through the solution. Solutions were irradiated in a Rayonet photochemical reactor equipped with four UV fluorescent lamps RPR 2537 Å. Solutions were exposed to UV irradiation for time intervals of 15, 30, 60 and 180 min. Unirradiated control samples consisted in metoclopramide solutions filled in quartz cells wrapped in aluminum foil and therefore, protected from light exposure. The experiments were performed at room temperature ( $293 \pm 4 \text{ K}$ ). After irradiation, all samples were protected from further light exposure.

## 2.3. HPLC-DAD system

A Merck-Hitachi HPLC system composed of a L-6200 Intelligent Pump, an AS-2000 autosampler with a  $20 \mu\text{L}$  sample loop, and a L-4500 diode array detector was used. Data acquisition was performed using the Merck-Hitachi HSM 2000 software.

Chromatographic separation was achieved on a  $250 \times 4 \text{ mm}$  Merck LiChrospher® 60 RP Select B column  $5 \mu\text{m}$  particle size. The mobile phase consisted of a mixture of 14% acetonitrile and 86% ammonium acetate  $10 \text{ mM}$  aqueous solution adjusted to pH 5 with glacial acetic acid at a flow rate of  $1 \text{ mL min}^{-1}$ . The analyses were performed in triplicate at room temperature. The absorbance was measured between 200 and 700 nm. The quantification wavelength was 273 nm.

For the assessment of the drug purity following UV exposure, the percentage of drug recovery was calculated from the ratio of the areas under the curve of the drug peak between irradiated and control samples [6,14,15].

The photolysis products were quantified as a percentage of the initial drug concentration. As no standards were available, they were quantified as metoclopramide, assuming they have similar response factors as the parent drug [15]. Metoclopramide solutions in concentrations ranging from 2 to  $50 \mu\text{M}$  were injected in order to establish the calibration curve (six calibration points). The calibration curve was validated by a statistical analysis (JMP, SAS software). For peak area (measured in nominal units), the regression equation was  $y = 971.15x - 322.15$ , the value for  $R^2$  was 0.998. Standard deviation of the slope was 5.5 whilst standard deviation of the intercept was 13. Solutions with decreasing concentrations in metoclopramide were injected in order to determine the limits of detection (LOD) and quantification (LOQ) which were considered as 3 and 10 times the signal-to-noise ratio, respectively [6]. LOQ was  $2 \mu\text{M}$ . The repeatability of the method was determined by measuring intraday variation for determination of metoclopramide at  $5 \mu\text{M}$  ( $n = 3$ ). The observed RSD for intraday variation was about 2%. Intermediate precision was determined by measuring inter-day variation for the same sample. The inter-day RSD was about 2.5%.

## 2.4. LC-ion trap MS system

The LC system consisted in a Waters Alliance model 2695 separation module. The chromatographic conditions were the same as for the HPLC-DAD analysis. A Thermo-Electron LCQ Advantage ion trap mass spectrometer equipped with an atmospheric pressure chemical ionization (APCI) source was used. The parameters were set as

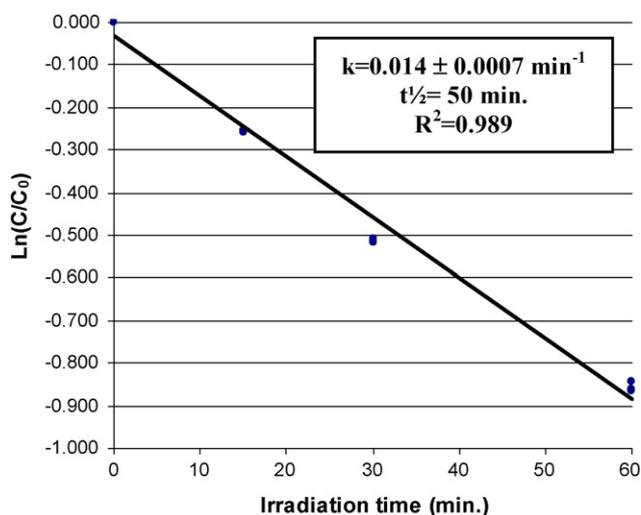


Fig. 1. Semi-logarithmic plot of relative metoclopramide concentration ( $C/C_0$ ) versus UV-irradiation time (min).

follows: APCI heater temperature,  $500^\circ\text{C}$ ; capillary voltage,  $26 \text{ V}$ ; capillary temperature,  $200^\circ\text{C}$ ; sheath gas flow,  $70 \text{ A.U.}$ ; auxiliary gas flow,  $30 \text{ A.U.}$ ; discharge current,  $5 \mu\text{A}$ . The mass determinations were performed in the positive mode in the range  $50\text{--}1000 \text{ m/z}$ . Collision-induced dissociation spectra of the main photolysis products were acquired using 35% normalized fragmentation energy. Data were acquired and processed by use of Xcalibur 1.3 software (Thermo).

## 3. Results and discussion

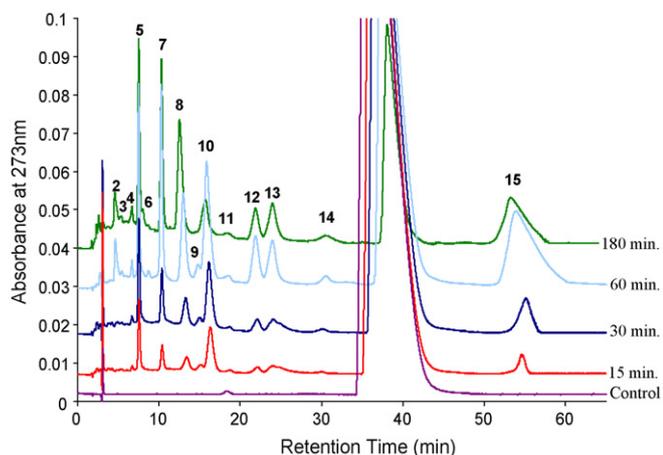
### 3.1. Changes in drug purity

Fig. 1 represents the logarithm of the relative metoclopramide concentration as a function of the irradiation duration. No significant difference was observed between control samples ( $P = 0.68$ ). For UV-irradiated samples, metoclopramide concentration decreased as a function of the irradiation time. Fast degradation of metoclopramide is observed upon  $254 \text{ nm}$  UV irradiation since after 15 min, 15% of the initial metoclopramide concentration is already consumed. Semi-log plot of the remaining metoclopramide concentration is linear at the early stage of the process (up to 60 min), indicating first-order kinetics. For the longest irradiation time, the degradation was lower than expected (only 64% of the initial concentration is degraded) since the amount of photolysis products becomes non-negligible compared to the concentration of the parent drug.

### 3.2. Analysis of degradation products

An overlay of chromatograms of metoclopramide solutions unirradiated and UV irradiated for increasing times is displayed in Fig. 2. The peaks were numbered according to their retention times in HPLC. The concentrations of the different degradation peaks expressed as percentages of the initial drug concentration are displayed in Table 1.

Only one impurity peak was present in the unirradiated control samples. The amount of this product did not increase following UV irradiation. Chromatograms of UV-irradiated solutions exhibited numerous degradation peaks. The concentrations of the degradation peaks increased as a function of the UV exposure duration, although after prolonged UV exposure (for 180 min), a decrease in their concentration was observed, with the exception of some peaks that continued to increase, such as 5 and 8. The decrease in the con-



**Fig. 2.** HPLC–UV chromatograms of metoclopramide solutions unirradiator and UV irradiated for 15, 30, 60 and 180 min.

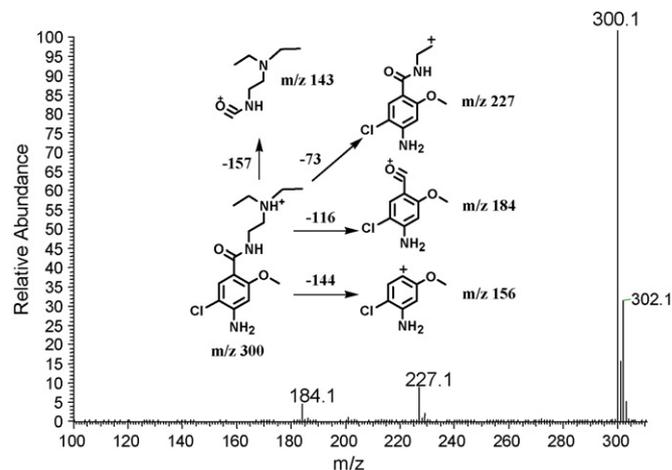
centration of these photolysis products over time might be due to their photolysis, which generates secondary photolysis products.

### 3.3. Characterization of degradation products

In the positive mode with an atmospheric pressure chemical ionization source (APCI), protonated molecules ( $[M+H]^+$ ) are obtained for each product. The mass to charge ratios of the products along with the corresponding collision-induced dissociation (CID) fragments are listed in Table 2. Insights on the structures of photolysis products are provided by the mass spectrometer. The presence of the chlorine is indicated in the MS spectrum as well as the number (odd or even) of nitrogen atom. The comparison between the MS–MS spectrum of the degradation product with that of the parent drug allows to locate the modification. The MS spectrum of metoclopramide and the structure of its major fragments are presented in Fig. 3. Metoclopramide, with a mass to charge ratio ( $m/z$ ) of 300 fragments into a  $m/z$  227 product, obtained after the loss of diethylamine from the lateral chain ( $-73$ ), a  $m/z$  184 fragment results from the cleavage of the amide bond. The structure of metoclopramide and those proposed for the photolysis products are in Fig. 4.

The mass spectrometer revealed that several chromatographic peaks resulted from a coelution of different degradation products. For example, two products are detected within peak 9 with mass to charge ratios of 529 and 563, respectively.

Although products with lower  $m/z$  than metoclopramide such as 266 and 282 predominate after short irradiation periods, prolonged



**Fig. 3.** Mass spectrum and structure of major MS–MS fragments of metoclopramide.

irradiation results in increased amounts of higher molecular weight products such as  $m/z$  563, 529, and even the appearance of products with  $m/z$  of 792, 790 and 808 after a 3 h UV exposure, demonstrating the formation of polymerization products.

A  $m/z$  217 product was only detected by the mass spectrometer as it shows no absorbance in the UV range. CID of this product leads to fragments corresponding to the loss of methanol ( $m/z$  185) whilst the presence of the other fragments such as 144 ( $-73 = \text{diethylamine}$ ) and 117 ( $=(\text{CH}_3-\text{CH}_2)_2-\text{NH}^+-\text{CH}_2-\text{CH}_2-\text{NH}_2$ ) shows that this molecule possessed a similar lateral chain as metoclopramide. Therefore the modification has occurred on the ring. The odd mass suggests the loss of the nitrogen from the ring. This molecule might correspond to product (1), resulting from the cleavage of the ring.

The  $m/z$  266 product has an unmodified lateral chain and results from the loss of the chlorine. This product has been identified previously in gamma and electron-beam irradiated metoclopramide [13]. Product (7) might be obtained after homolytic scission of the C–Cl bond following by dismutation of the resulting radical.

Product  $m/z$  282 has no chlorine and its CID spectrum shows no modification on the lateral chain. Therefore, it might be due to the loss of the chlorine by homolytic scission followed by the attack of the resulting radical on adjacent water molecule to yield an hydroxylated product (5a). As irradiations were carried out in the absence of oxygen, this product was most likely generated by the attack of the carbocation obtained after heterolytic scission of the C–Cl bond on adjacent water molecules [17].

**Table 1**

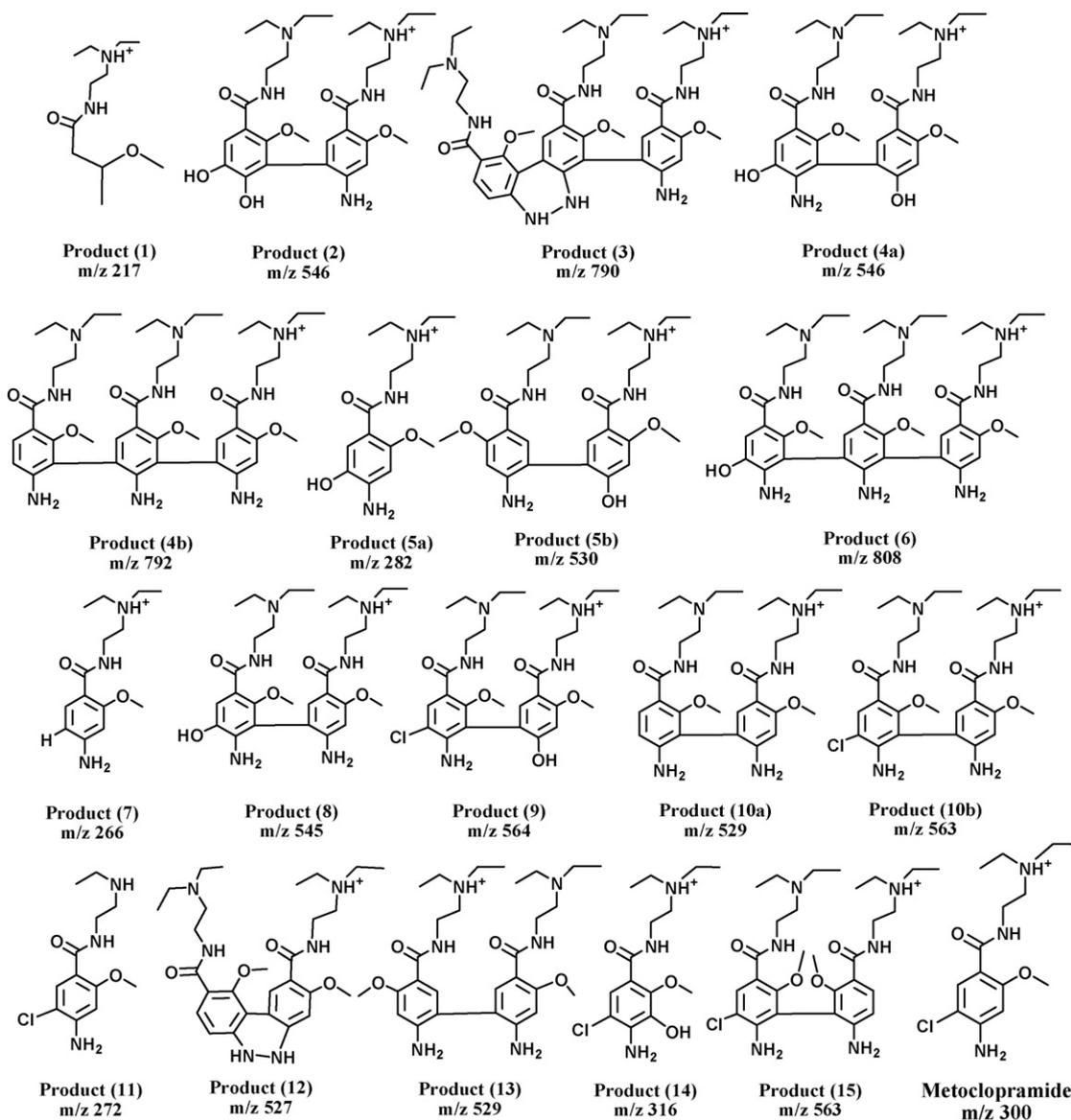
HPLC–DAD quantification of degradation peaks from UV-irradiated metoclopramide solutions. The concentrations of degradation peaks are expressed as percentages of the initial drug concentration.

Peaks	Mean percentages of impurity peaks relative to the initial drug concentration (%) ( $n = 3$ )				
	0 min	15 min	30 min	60 min	180 min
2	<LOD	<LOQ	<LOQ	0.73	0.56
3	<LOD	<LOQ	<LOQ	<LOQ	<LOQ
4	<LOD	0.21	0.23	0.28	0.29
5	<LOD	0.80	1.60	1.96	2.20
6	<LOD	<LOQ	LOQ	0.26	0.33
7	<LOD	0.45	0.77	2.07	2.29
8	<LOD	0.45	0.78	1.92	3.24
9	<LOD	0.27	0.38	0.45	<LOD
10	<LOD	1.31	2.02	3.11	1.63
11	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ
12	<LOD	0.31	0.56	1.67	1.22
13	<LOD	0.51	0.74	1.81	1.80
14	<LOD	<LOQ	0.27	1.18	0.50
15	<LOD	1.97	4.16	7.27	3.51

**Table 2**  
m/z values and fragments of major photolysis products of metoclopramide.

Peak number according to elution order	Retention time (min)	Products	m/z	Major fragment ions						
1	3.5	(1)	217	185	144	117	100			
2	4.6	(2)	546	528	473	430	402	359	329	288
3	5.4	(3)	790	717	674	648	601	560	532	
4	6.8	(4a)	546	528	473	430	402	359	329	288
4	6.8	(4b)	792	719	676	603	562	489	446	
5	7.5	(5a)	282	209	166	143	117	100		
5	7.5	(5b)	530	457	414	343	272			
6	8.1	(6)	808	735	692	666	619	578	505	462
7	10.3	(7)	266	193	150	100				
8	12.6	(8)	545	472	429	399	358	315		
9	15.0	(9)	564 <sup>a</sup>	491	448	418	377	349	313	306
10	15.8	(10a)	529	456	413	383	340	299		
10	15.8	(10b)	563 <sup>a</sup>	490	447	417	376	348	333	
11	18.6	(11)	272 <sup>a</sup>	227	184	156				
12	22.1	(12)	527	454	411	381	338	297		
13	24.0	(13)	529	456	413	383	340	299		
14	31.0	(14)	316 <sup>a</sup>	243	200	143	117	100		
15	39.1	Metoclopramide	300 <sup>a</sup>	227	184	156	143			
15	53.8	(15)	563 <sup>a</sup>	490	447	417	376	333		

<sup>a</sup> Presence of a chlorine.



**Fig. 4.** Chemical structures (protonated) of metoclopramide and its degradation products.

$m/z$  272 product was previously identified as resulting from the loss of the ethyl on the diethylamino group from the lateral chain [16]. Product (11), already present in unirradiated metoclopramide, is not a photodegradation product as its concentration is not influenced by UV irradiation.

The  $m/z$  316 product corresponded to a hydroxylated version of metoclopramide which was also observed after gamma and electron-beam irradiation of metoclopramide solutions [16]. The addition of the hydroxyl most probably occurred on the ring since fragments 143, 117 and 100 demonstrated that the lateral chain was intact. The exact location of the hydroxyl on the ring cannot be determined by the mass spectrometer and the most probable structure according to organic chemistry rules is displayed for product (14).

Several isobaric products with  $m/z$  529 were detected. These products had no chlorine and exhibited similar CID spectra with fragments  $m/z$  456 (–73),  $m/z$  413 (–116),  $m/z$  383 (–146 = loss of 2 diethylamine), revealing the presence of two lateral chains similar to that of metoclopramide. Therefore, these products might be obtained from coupling of two metoclopramide molecules following chlorine losses, corresponding either to product (10a) or (13). Product (5b) with  $m/z$  530 had two intact lateral chain although its even mass indicated further modification on the nitrogen located on the ring. It might result from products (10a) or (13), following the substitution of one of the aniline  $\text{NH}_2$  groups by a hydroxyl.

The fragmentation characteristic of the presence of two intact lateral chains is exhibited by product (12) with  $m/z$  527. The difference of two mass units compared to the  $m/z$  529 products, (10a) and (13), indicated the loss of hydrogen on the ring part.

Compared to both products (10a) and (13), product (8), with  $m/z$  545, had a mass difference of 16 units which is due to the addition of a hydroxyl. According to the fragmentation pattern, the addition was on the ring part. This product was most probably a secondary photolysis product as it continued to increase for longest UV-irradiation time. Products (2) and (4a) that differed from product (8) and by one mass unit and had similar fragmentation patterns might result from further photolysis of this products, following substitution of the aniline  $\text{NH}_2$  by a hydroxyl group.

Both products (10b) and (15) with  $m/z$  563 showed similar MS–MS fragmentation. The presence of one chlorine was shown by the  $^{37}\text{Cl}$  peak in the mass spectrum. MS–MS fragmentation pattern revealed the presence of two unmodified lateral chain. One of this product (corresponding to peak 10) was also formed after radiolysis [13].

Product (9) with  $m/z$  564 exhibited a very similar fragmentation pattern as products (10b) and (15), with a difference of one mass unit. Its even mass is due to a difference in the number of nitrogen atoms located on the ring part. This product could result from the substitution of a nitrogen by an hydroxyl radical.

The fragmentation patterns of products (3), (4b) and (6), indicated the presence of three lateral chains as fragments corresponding to the loss of either 346 or 303 mass units are observed. Therefore, these products might result from the coupling of a dechlorinated metoclopramide molecule with one of the existing photoproducts. Product (8) might be generated from product (6), whilst product (12) might be obtained from product (10a), and product (13) might result from product (4b) photolysis.

#### 3.4. Summary of photodegradation processes

Most photoproducts share similar structure as the parent drug. The major photolysis reaction occurring for metoclopramide is the loss of the chlorine. Breakage of this bond might either occur through homolysis, providing two radicals or heterolysis that leads to a carbocation and a chloride [18]. The formation of the hydroxylated products most likely occurred through nucleophilic attack

of adjacent water molecules on the carbocation. The formation of hydroxylated photoproducts after photolysis in the absence of oxygen has already been described for several halogen substituted drugs such as fluoroquinolones [18,19]. Dimerization products are obtained from the reaction of the radical obtained after homolytic scission with other metoclopramide molecules. The substitution of the  $\text{NH}_2$  from the aniline ring might be due to the homolysis of the C–N bond that results in the formation of an amino radical and a carbon centered radical which can react with adjacent water molecules to give a hydroxylated product. The ejection of an electron from the excited molecule, has been observed following photolysis of 2-chloro aniline [20]. Such mechanism might explain the formation of the photoproducts (12) and (3).

Many products seem to be secondary photoproducts resulting from further photolysis of primary photoproducts.

The formation of product (1) shows that fragmentation of the metoclopramide ring is possible following UV irradiation.

#### 3.5. Comparison between photolysis and radiolysis processes

The radiolysis pathway of metoclopramide may be found in Ref. [13]. Although photolysis and radiolysis mechanisms are different, some products generated after radiolysis were also formed following photolysis. For example, product (5a), which was formed by nucleophilic substitution of a hydroxyl radical on the chlorine during the radiolysis process, is generated following homolytic scission and attack on water in the photolysis process. However, after photoirradiation, larger amounts of higher molecular weight products were formed compared to radiolysis where only the  $m/z$  563 product was observed.

## 4. Conclusion

Metoclopramide is highly photolabile in solution. A rapid decrease in metoclopramide purity is observed after UV irradiation. Structural characterization of the photolysis products of metoclopramide indicated that degradation occurred through scission of the carbon–chlorine bond. Some degradation products are similar between photolysis and radiolysis. However, different formation routes are followed to generate them. Dissociation of the chlorine is the major photodegradation pathway of metoclopramide and is generally followed by coupling of the products to generate high molecular weight products. Alternatively, hydroxylated products could also be generated.

## References

- [1] V. Andrisano, P. Hrelia, R. Gotti, A. Leoni, V. Cavrini, Photostability and phototoxicity studies on diltiazem, *J. Pharm. Biomed. Anal.* 25 (2001) 589–597.
- [2] G. Costa, Photodegradation and photosensitization in pharmaceutical products: assessing drug phototoxicity, *Pure Appl. Chem.* 76 (2004) 263–275.
- [3] European Agency for the Evaluation of Medicinal Products (EMA), Photostability Testing of New Active Substances and Medicinal Products, ICH Topic Q1B, step 5 (CPMP/ICH/279/95), EMA, London, 1995.
- [4] R. Andreozzi, R. Marotta, P. Nicklas, Pharmaceuticals in STP effluents and their solar photodegradation in aquatic environment, *Chemosphere* 50 (2003) 1319–1330.
- [5] A.F. Martins, T.G. Vasconcelos, C. da Silveira Frank, D.M. Henriques, K. Kümmerer, in: Kümmerer (Ed.), *Pharmaceuticals in the Environment Sources, Fate, Effects and Risks*, Springer, Berlin, 2008.
- [6] European Pharmacopoeia, 5th ed., Council of Europe, Strasbourg, 2005.
- [7] B.G. Katzung, *Basic and Clinical Pharmacology*, 7th ed., Appleton & Lange/McGraw-Hill, New York, 1998.
- [8] I. Colman, M.D. Brown, G.D. Innes, E. Grafstein, T.E. Roberts, B.H. Rowe, Parenteral metoclopramide for acute migraine: meta-analysis of randomized controlled trials, *Br. Med. J.* 329 (2004) 1369–1373.
- [9] K. Weclawska, E. Siminska, The effect of visible and ultraviolet radiation on metoclopramide solutions, *Acta Pol. Pharm.* 42 (1985) 555–560.
- [10] M.A. Dearth, T.J. Korniski, J.L. Gerlock, The LC/MS/MS characterization of photolysis products of benzotriazole-based ultraviolet absorbers, *Polym. Degrad. Stabil.* 48 (1995) 111–120.

- [11] S. Perez, P. Eichhorn, D. Barcelo, Structural characterization of photodegradation products of enalapril and its metabolite enalaprilat obtained under simulated environmental conditions by hybrid quadrupole-linear ion trap-MS and quadrupole-time-of-flight-MS, *Anal. Chem.* 79 (2007) 8293–8300.
- [12] M. Petrovic, D. Barcelo, LC-MS for identifying photodegradation products of pharmaceuticals in the environment, *Trends Anal. Chem.* 26 (2007) 486–493.
- [13] A. Maquille, J.L. Habib Jiwan, B. Tilquin, Radiosterilization of drugs in aqueous solutions may be achieved by the use of radioprotective excipients, *Int. J. Pharm.* 349 (2008) 74–82.
- [14] United States Pharmacopeia National Formulary 28, United States Pharmacopeial Convention, Rockville, MD, 2005.
- [15] European Agency for the Evaluation of Medicinal Products (EMA), Note for Guidance on Impurities in New Medicinal Products, Topic Q3B, step 4 (CPMP/ICH/282/95), London, 1996.
- [16] A. Maquille, C. Slegers, J.L. Habib Jiwan, B. Tilquin, Electron beam and gamma radiolysis of solid-state metoclopramide, *Pharm. Res.* 23 (2006) 1343–1349.
- [17] B. Arnold, L. Donald, A. Jurgens, J.A. Pincock, Homolytic versus heterolytic cleavage for the photochemistry of 1-naphthylmethyl derivatives, *Can. J. Chem.* 63 (1985) 3140–3146.
- [18] M.C. Cuquerella, F. Bosca, M.A. Miranda, Photonucleophilic aromatic substitution of 6-fluoroquinolones in basic media: triplet quenching by hydroxide anion, *J. Org. Chem.* 69 (2004) 7256–7261.
- [19] Y.N. Li, D.E. Moore, B.N. Tattam, Photodegradation of amiloride in aqueous solution, *Int. J. Pharm.* 183 (1999) 109–116.
- [20] E.K. Winarno, N. Getoff, Photo-induced decomposition of 2-chloroaniline in aqueous solution, *Z. Naturforsch.* 57 (2002) 512–515.