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Phototransformation of metoxuron [3-(3-chloro-4-methoxyphenyl)-1,1-dimethylurea] in aqueous solution

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Abstract: UV irradiation of metoxuron in aerated aqueous solution at 254 nm or between 300 and 450 nm led initially to an almost specific photohydrolysis of the C–Cl bond, resulting in the formation of 3-(3-hydroxy-4-methoxyphenyl)-1,1-dimethylurea (MX3) and hydrogen chloride. The quantum yield was determined to be 0.020 (±0.005) in solutions irradiated at 254 nm. Five minor photoproducts were also identified, in particular the dihydroxydimethoxybiphenyl derivatives resulting from the phototransformation of MX3. Irradiation increased the toxicity of an aqueous solution of metoxuron to the marine bacterium *Vibrio fischeri*.

Keywords: phenylurea; metoxuron; photolysis; photohydrolysis; toxicity

1 INTRODUCTION

Halogenophenylureas are a main group of herbicides. The first to be used was 3-(4-chlorophenyl)-1,1-dimethylurea—common name monuron. It was first marketed in 1952. Thereafter many mono- and dihalogenophenylureas were proposed. They are chemically stable, but they absorb light up to *c* 300 nm and direct photolysis may be involved in their elimination from natural waters, especially in summer. The photodegradation of some of them has been reported in literature. In 1968 Rosen and Strusz[1] showed that the main photoproduct formed from metobromuron [3-(4-bromophenyl)-1-methoxy-1-methylurea] was [3-(4-hydroxyphenyl)-1-methoxy-1-methylurea], but demethoxylation and demethylation also occurred. Several photoproducts of monuron were identified by Crosby and Tang.[2] They result from the oxidation or elimination of methyl groups. Hydroxylation of the ring without dechlorination and formation of 4,4′-dichlorocarbanilide have also been reported, but no dechlorination was mentioned. Rosen *et al*[3] observed the photosubstitution of Cl by OH, but did not quantify the part taken by this reaction. In methanol the main pathway is the photoreduction of C–Cl bond.[4] With chlorotoluron [3-(3-chloro-4-methylphenyl)-1,1-dimethylurea], photohydrolysis of the C–Cl bond was reported to be initially almost quantitative.[5]

To our knowledge no results have been published on the direct photolysis of metoxuron, 3-(3-chloro-4-methoxyphenyl)-1,1-dimethyl urea.

The main aim of the present work was to study the photochemical behaviour of metoxuron in order to establish whether the specificity of the photoreaction observed with chlorotoluron may be related to the position of the halogen on the ring. Secondary aims were to study the influence of irradiation on the toxicity of solutions and to check whether photochemical transformations observed in laboratory conditions may occur during sunlight irradiation.

2 MATERIALS AND METHODS

2.1 Reactants

Metoxuron (99.6%) was purchased from Riedel-de Haën. Water used for solutions was purified using a Milli-Q system and the purity monitored by measuring its resistivity (the resistivity was always >18 MΩ cm).

2.2 Irradiation

Several instruments were used for continuous irradiation. Quantum yield measurements undertaken on solutions of metoxuron were carried out at 254 nm with a device consisting of a cylindrical mirror with an elliptic base, a low-pressure mercury lamp (germicidal lamp) located along one of the focal axes and a quartz reactor (2 cm ID) located on the second focal axis. The average number of photons absorbed by the solution

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was found to be 8×10^{15} photons $s^{-1}cm^{-3}$ using uranyl oxalate as actinometer. The advantage of using light of this wavelength is that it excites metoxuron more efficiently than does light of longer wavelengths.

Solutions of metoxuron were also irradiated with six fluorescent lamps (Duke GL 20W) emitting UV light with spectral wavelengths above 275 nm and with a maximum intensity at *c* 310 nm. With these lamps mercury lines at 365, 405 and 436 nm are also emitted but not involved in the phototransformation of metoxuron. Wavelengths below 290 nm are cut off by the Pyrex™ vessel used as a reactor. In these experiments light reaching the solutions is closer in wavelength to sunlight. Dilute solutions ($c\ 10^{-4}$ M) were used for the kinetic study. For the isolation and characterisation of photoproducts, more concentrated solutions (2.2×10^{-3} M) were irradiated to obtain up to 60% of metoxuron conversion.

Some solutions were deoxygenated by argon bubbling to study the influence of oxygen on phototransformation rates.

Solutions in a Pyrex™ vessel were also exposed to sunlight in Clermont-Ferrand, France (latitude 46 °N, altitude 420 m) in late February and March.

2.3 Analyses

UV spectra were recorded on a Varian Cary 3 spectrophotometer.

HPLC analyses were carried out using a Waters 990 (Millipore) chromatograph equipped with a photodiode array detector. Separation was achieved on a Spherisorb ODS2 5 µ column (250 mm × 4.6 mm ID) with a methanol + water (40 + 60 by volume) eluent. The same HPLC system was used for both analysis and isolation of photoproducts. The following procedure was used. The pH of the irradiated solutions (2.2×10^{-3} M; 200 ml) was increased to 11 with sodium hydroxide (0.1 M) in order to selectively extract unreacted metoxuron with chloroform. The solution was then acidified to pH = 3 with a few drops of hydrochloric acid (1 M) and evaporated to about 2 ml. Sub-samples (50 µl per injection) were injected in HPLC and isolated by fraction collection.

Structural elucidation of isolated photoproducts was achieved by a combination of MS and ^{1}H NMR. Direct introduction of samples into a Hewlett-Packard 5989-B mass spectrometer running in chemical ionisation mode (methane) was used for the determination of molecular masses. Ions corresponding to M + 1 and M + 29 were generally obtained using this technique.

Samples of isolated photoproducts were prepared for NMR analysis by dissolving in hexadeuteroacetone. ^{1}H NMR spectra were obtained from the prepared samples using a Bruker AC 400 spectrometer.

2.4 Test of toxicity

The toxicities of solutions were determined by applying a Microtox® test.[6] This test consists of determining the concentration (EC_{50}) of a toxicant that inhibits 50% of the natural luminescence of a marine bacterium *Vibrio fischeri* (Bejerinck) Lehman & Neumann (*Photobacterium phosphoreum* Ford). The luminescence emission is measured at two or three exposure times, usually 5, 15 and 30 min. Values of EC_{50} are often not significantly different for these exposure times. A decrease of EC_{50} corresponds to an increase of toxicity. This test is rapid and gives standardized values that can be compared with those of a large database.[6] However, the scale of toxicity obtained with this bacterium is not necessarily the same as scales obtained with other micro-organisms.

3 RESULTS

3.1 Spectrophotochemical study

The UV spectrum of metoxuron between 210 and 320 nm is given in Fig 1 (annotated the 'initial' time-point). Absorption maxima are located at 241 and 285 nm, with measured molar absorption coefficients corresponding to 13 900 (±200) and 4600 (±100) $M^{-1}cm^{-1}$ respectively.

The trend in spectral change produced over the successive irradiation time-points, shown in Fig 1, was observed to be the same for both air-saturated and deoxygenated solutions. This trend was also observed whether the incident UV was 254 nm (as in Fig 1) or within the range 290–350 nm (spectra not shown). In all instances irradiation resulted in a reduction in the spectral absorption between 232 and 257 nm accompanied by an increase in the spectral absorption between 260 and 300 nm. The presence of two isosbestic points for all the experiments indicated a specific chemical transformation, the chemical outcome of which was not significantly influenced by the presence of dissolved oxygen. (Isosbestic points correspond to wavelengths where the substrate and its products have the same molar absorption. The presence of isosbestic points in a photochemical transformation implies that the stoichiometry is not modified during the transformation). Furthermore,

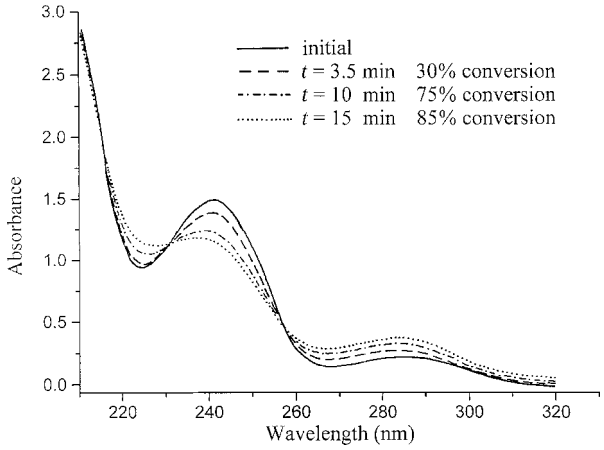


Figure 1. UV spectrum of a 1.1×10^{-4} M solution of metoxuron irradiated at 254 nm.

the relatively minor modifications observed in the spectra over the time of irradiation would imply that metoxuron and its principal photoproducts have broadly similar UV spectral characteristics.

3.2 Analytical study

It is clear from the HPLC chromatogram of a 1.1×10^{-4} M aqueous solution of metoxuron irradiated at 254 nm (Fig 2) that 3-(3-hydroxy-4-methoxyphenyl)-1,1-dimethylurea (MX3) is the major product formed. Some minor photoproducts, MX1–MX6, appeared only after relatively long irradiation periods.

In order to identify the photoproducts, a more concentrated solution (2.2×10^{-3} M) was irradiated and treated as described in Section 2.3. Several products were obtained in sufficient amounts to allow ^{1}H NMR and MS analyses. Results are summarised in Table 1.

3.2.1 Influence of oxygen

At 254 nm metoxuron phototransformation was initially about 20% faster in deoxygenated solutions than in air-saturated solutions. Thus oxygen had only a slight inhibiting effect on the overall rate of conversion. Whilst the qualitative profile of photoproducts formed in deoxygenated solutions was almost the same as in aerated solutions, the quantitative profile was different. The reaction was more specific towards MX3 formation in the presence of oxygen, where *c* 90% of metoxuron conversion was accounted for as MX3. This contrasted with the *c* 60% of metoxuron conversion as MX3 in the absence of oxygen. Besides this, the absence of oxygen favoured the production of MX4 and MX6, but reduced that of MX5. An unidentified photoproduct, MX7, was only observed in deoxygenated solutions (Fig 2).

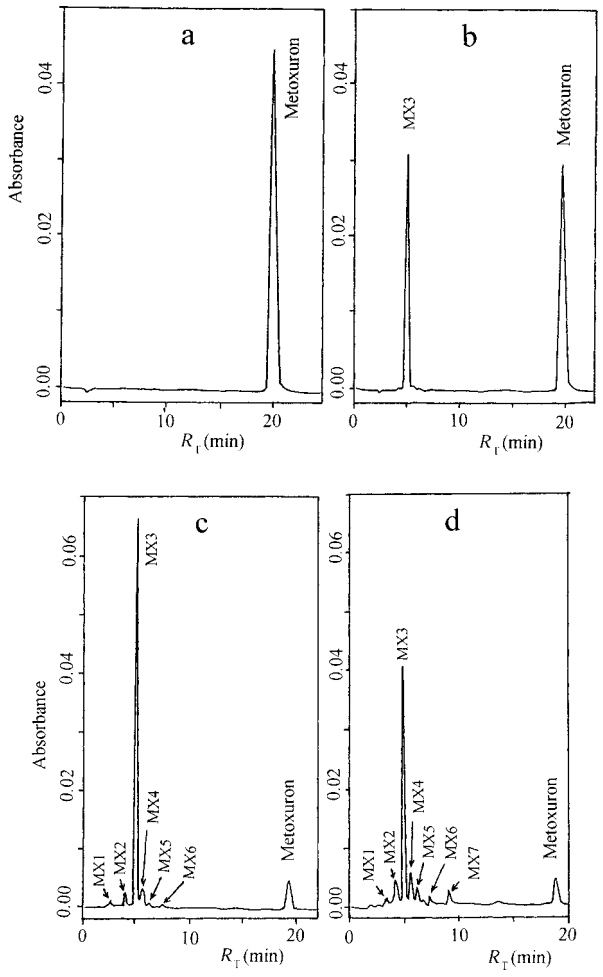


Figure 2. HPLC chromatograms of a solution of metoxuron 1.1×10^{-4} M irradiated at 254 nm: a: Initial; b: air-saturated solution irradiated for 4 min (35% conversion); c: air-saturated solution irradiated for 17 min (89% conversion); d: deoxygenated solution irradiated for 15 min (92% conversion).

3.2.2 Influence of wavelength

The transformation of metoxuron in aqueous solutions irradiated in the range 290–350 nm was significantly slower than at 254 nm. This is mainly due to the weaker absorption at the longer wavelength range, as metoxuron does not appreciably absorb beyond 313 nm and absorptivity is much lower between 290–313 than at 254 nm (Fig 1). The same photoproducts were formed in both high- and low-wavelength experiments, but MX3 accounted for a slightly higher proportion of the photoproducts in the longer wavelength experiment.

3.3 Kinetic study

The quantum yield of phototransformation was $\phi = 0.020$ (± 0.005) for an air-saturated solution (10^{-4} M) irradiated at 254 nm. In our experimental conditions it was assumed that all photons reaching the solution were absorbed. The determination of quantum yield in polychromatic light ($\lambda > 290$ nm) was thought to be less accurate.

The disappearance of metoxuron in an irradiated solution (1.1×10^{-4} M) at 254 nm or in the range 290–350 nm obeyed apparent first-order kinetics. In our experimental conditions the rate constants were 0.148 and 0.05 min^{-1} respectively.

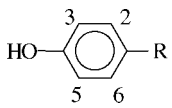
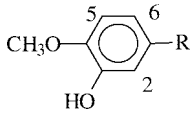
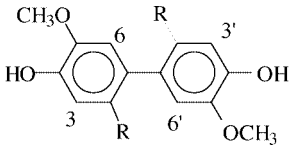
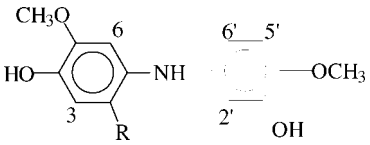
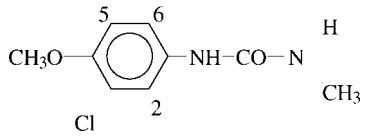
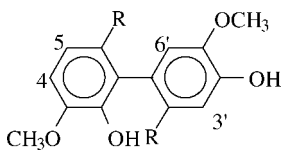
The main photoproduct (MX3) was obtained in sufficient quantities to enable preparation of solutions of defined concentrations and to plot an HPLC peak *versus* MX3 concentration calibration curve. It was then possible to evaluate the concentration of MX3 formed for each irradiation time and to compare it with that of transformed metoxuron. Results obtained in air-saturated solutions irradiated in the range 290–350 nm are plotted in Fig 3. It appeared that, in the early stages of the reaction (up to 35% of metoxuron transformation), MX3 accounted for *c* 95% of metoxuron converted. The specificity was a little lower at 254 nm.

The formation of photoproducts in air-saturated solutions irradiated at 254 nm is summarised in Fig 4 in relative units of absorbance (detection at 260 nm). Only MX3 showed the kinetics of a primary photoproduct. The other products did not appear in the early stages of the reaction. It could then be deduced

that they did not result directly from the phototransformation of metoxuron, ie they are secondary photoproducts.

Solutions were also exposed to sunlight from mid-February to the end of March. After 40 days of exposure, metoxuron was approximately 90% transformed. Metoxuron converted almost exclusively into MX3 during the first few days, but thereafter MX3 did not accumulate as much as in shorter wavelength laboratory experiments. This may be due merely to slow thermal oxidation of MX3 during long experiments in sunlight or to degradation of MX3

Table 1. Identification of metoxuron photoproducts

Product	*MS (m/z) chemical ionisation (methane)*	*^{1}H NMR (δ ppm) in acetone-d_6*
MX2 	181 ($M^+ + 1$); 209 ($M^+ + 29$) 136 protonated isocyanate* 72 $CON(CH_3)_2$	7.96 (s) OH; 7.45 (s) NH; 7.29 (m) H_2 and H_6; 6.69 (m) H_3 and H_5; 2.95 (s) $(CH_3)_2$
MX3 	211 ($M^+ + 1$); 239 ($M^+ + 29$); 195 (loss of CH_3) 166 protonated isocyanate* 72 $CON(CH_3)_2$	7.44 (s) OH or NH; 7.40 (s) NH or OH; 7.14 (d, J = 2.50 Hz) H_2; 6.90 (dd, J = 2.50 and 8.76 Hz) H_6; 6.77 (d, J = 8.76 Hz) H_5; 3.75 (s) OCH_3; 2.95 (s) $N(CH_3)_2$
MX4 	419 ($M^+ + 1$); 447 ($M^+ + 29$) 374 protonated isocyanate* 402 isocyanate, $M^+ + 29$ 329 protonated double isocyanate	7.68 (s) 2 × OH; 7.44 (s) H_6, $H_{6'}$ or H_3, $H_{3'}$; 6.71 (s) H_3, $H_{3'}$ or H_6 $H_{6'}$; 6.64 (s) 2 × NH; 3.80 (s) 2 × OCH_3; 2.74 (s) 2 × $N(CH_3)_2$
MX4′ 	303 protonated isocyanate* 331 isocyanate ($M^+ + 29$)	7.49 (d, J = 8.70 Hz) $H_{5'}$; 7.35 (s) H_3 or H_6; 7.25 (s) H_6 or H_3; 6.81 (s) NH; 6.79 (dd, J = 2.90 and 8.70 Hz) $H_{6'}$; 6.71 (d, J = 2.90 Hz) $H_{2'}$; 3.25 (s) 2 × OCH_3 Product not resolved from MX4 Some signals are merged
MX5 (unidentified)	372, 344, 327, 299	8.40 (s broad); 7.58 (s broad); 7.54 (s broad); 7.43 (s sharp); 7.42 (dd, J = 2.70 and 8.60 Hz); 7.28 (s broad); 7.25 (d, J = 2.70 Hz); 6.90 (d, J = 8.60 Hz); 6.74 (s sharp); 3.56 (s sharp); 3.00 (s sharp)
MX5′ 	215, 217 ($M^+ + 1$) chlorinated product 243 ($M^+ + 29$)	7.64 (d, J = 2.50 Hz) H_2; 7.24 (dd, J = 2.50 and 8.80 Hz) H_6; 6.86 (d, J = 8.80 Hz)H_5 Product not resolved from MX5 Some signals are merged
MX6 	419 ($M^+ + 1$); 447 ($M^+ + 29$) 374 protonated isocyanate* 402 isocyanate ($M^+ + 29$) 329 double isocyanate (protonated) 357 double isocyanate ($M^+ + 29$)	7.71 (s) OH; 7.45 (s) OH; 7.39 (s) $H_{6'}$ or $H_{3'}$; 7.33 (d, J = 8.80 Hz) H_5; 6.92 (d, J = 8.80 Hz) H_4; 6.80 (s) NH; 6.69 (s) NH; 6.68 (s) $H_{3'}$ or $H_{6'}$; 3.85 (s) OCH_3; 3.78 (s) OCH_3; 2.72 (s) $N(CH_3)_2$; 2.70 (s) $N(CH_3)_2$

R = —NH—CO—$N(CH_3)_2$.

s = singlet, d = doublet, dd = double doublet, m = multiplet.

* Isocyanate resulting from the pyrolysis of urea derivative in the mass spectrometer.

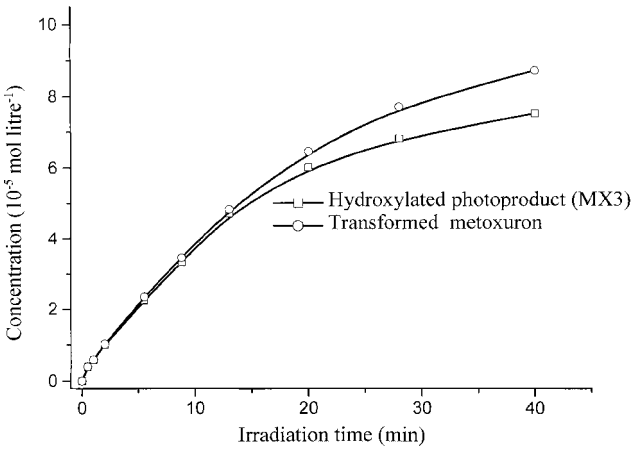


Figure 3. Specificity of the phototransformation of metoxuron into the hydroxylated product MX3 in air-saturated solution 1.1×10^{-4} M irradiated in the range 290–350 nm.

photoinduced by some minor photoproducts. Phototransformation is expected to be faster in summer, since the sunlight UV spectrum starts at shorter wavelengths in that season (*c* 290 nm) than in winter (*c* 300 nm) and the total photon flow is higher. Metoxuron and its main photoproduct should not accumulate much, at least in summer.

3.4 Test of toxicity

Solutions (2.2×10^{-3} M) irradiated at 254 nm were used to study the influence of irradiation on the toxicity to *V fischeri* (the Microtox test). It is more important to study, as a first step, the influence of irradiation on the overall toxicity of the solution by subjecting the bacterium directly to irradiated solutions containing the crude mixture of products, rather than to test the individual toxicities of the main photoproducts, since minor photoproducts, not detected or not identified, may have a significant influence on toxicity. In these conditions calculations are based on the total concentration of all products, assuming that this concentration is not significantly modified by irradiation. The concentration of toxicant that inhibited 50% of the luminescence of the bacterium (EC_{50}) was determined after a 5-min exposure. The relationship between EC_{50} and percentage of metoxuron transformation is represented graphically in Fig 5. As metoxuron is replaced by its photoproducts, the EC_{50} of the solution decreases, which implies that at least one of the photoproducts is more toxic to *V fischeri* than is metoxuron. The rate of increase in toxicity during the early irradiation period correlates well with the rate of disappearance of metoxuron and the corresponding rate of formation of the principal photoproduct, MX3. This would imply that MX3 is the main cause of toxicity. However the possibility of a contribution from other photoproducts to the overall toxicity cannot be excluded.

4 DISCUSSION AND MECHANISMS

4.1 Formation of the main photoproduct MX3

The photoexcitation of metoxuron leads almost exclusively to the formation of MX3 (substitution of Cl by OH) in the presence of dissolved oxygen (about 90% yield) and somewhat less exclusively in its absence. The oxygen in OH is necessarily provided by water and the reaction cannot be explained by a homolytic scission of the C—Cl bond, since this requires the homolytic scission of water, which is highly unlikely under these conditions. Similar reactions have previously been observed with 3-halogenophenols. Comparable values for transformation efficiency were obtained in the case of 3-fluorophenol and 3-chlorophenol.[7] The latter reaction occurs even in very acidic solutions.[8] It is explained by a mechanism of photohydrolysis involving the concerted heterolytic scission of the C—Cl bond and a H—OH bond in a molecule of water. It can be deduced from the partial inhibition by oxygen that the reaction may occur from the excited triplet state of metoxuron. This assumption is consistent with the possible sensitization of the photohydrolysis of halogenophenols[7] (Fig 6(a)).

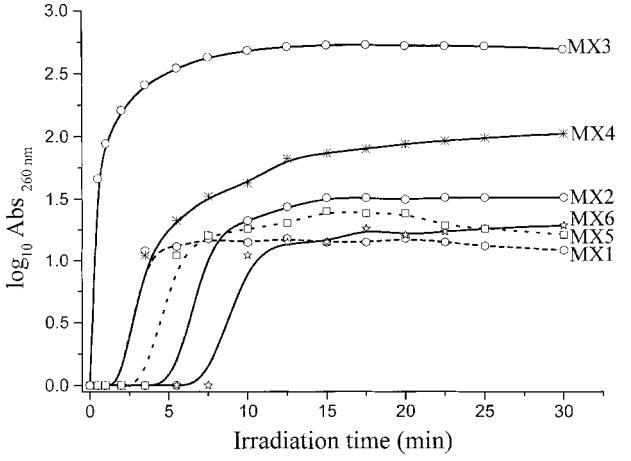


Figure 4. Kinetics of formation of the main photoproducts in a deoxygenated solution 1.1×10^{-4} M of metoxuron irradiated at 254 nm. Values of absorbance at 260 nm are given in arbitrary units.

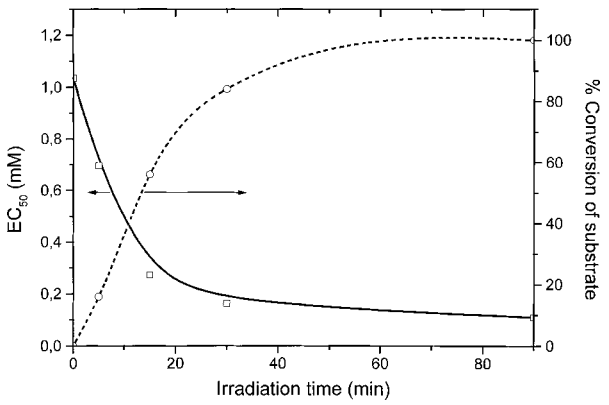


Figure 5. Toxicity of the photoproducts of metoxuron: evolution of EC_{50} with the course of transformation.

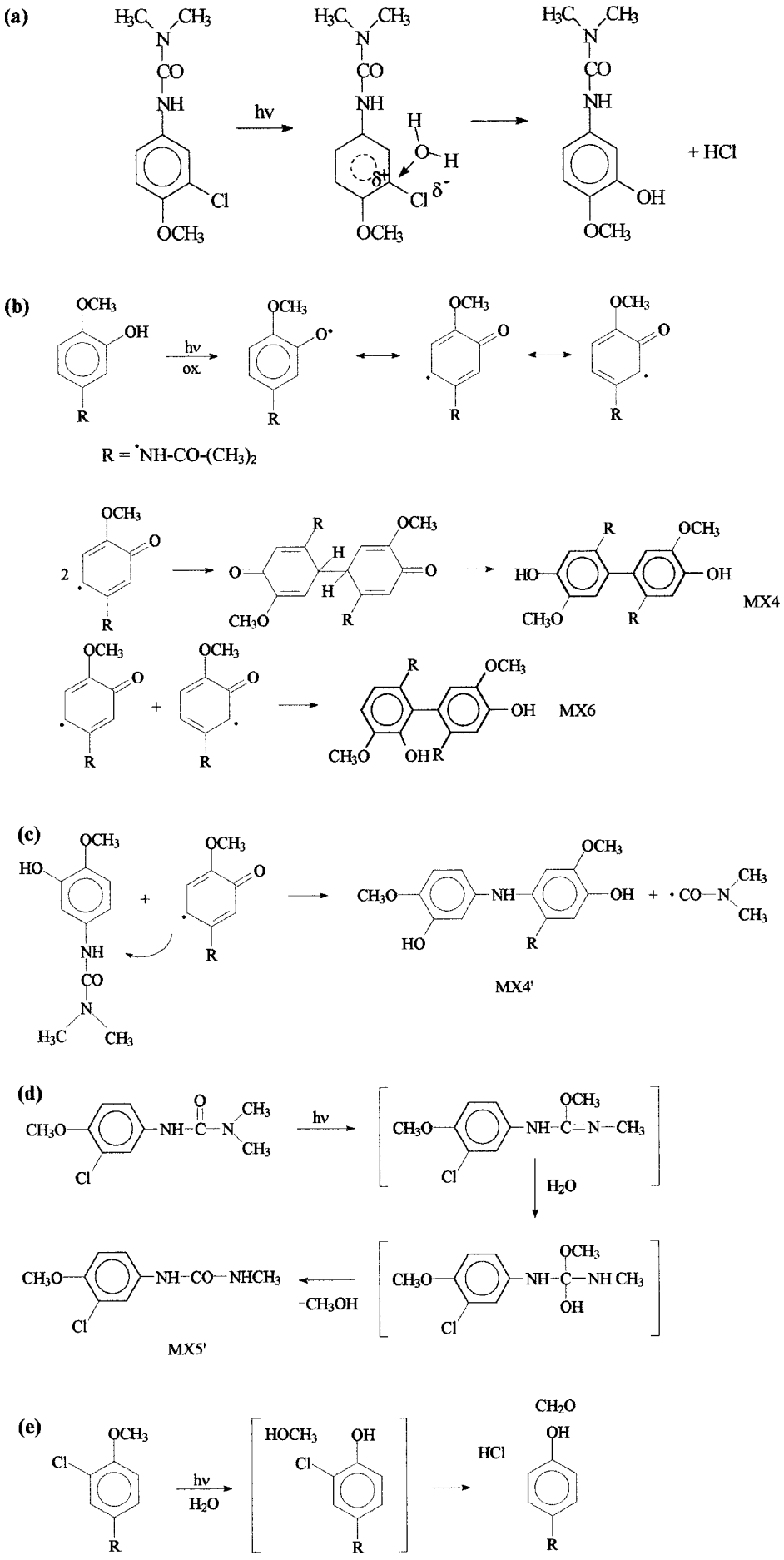


Figure 6. Mechanism of formation of photoproducts of metoxuron (a) MX3, (b) MX4 and MX6, (c) MX4′, (d) MX5′, (e) MX2.

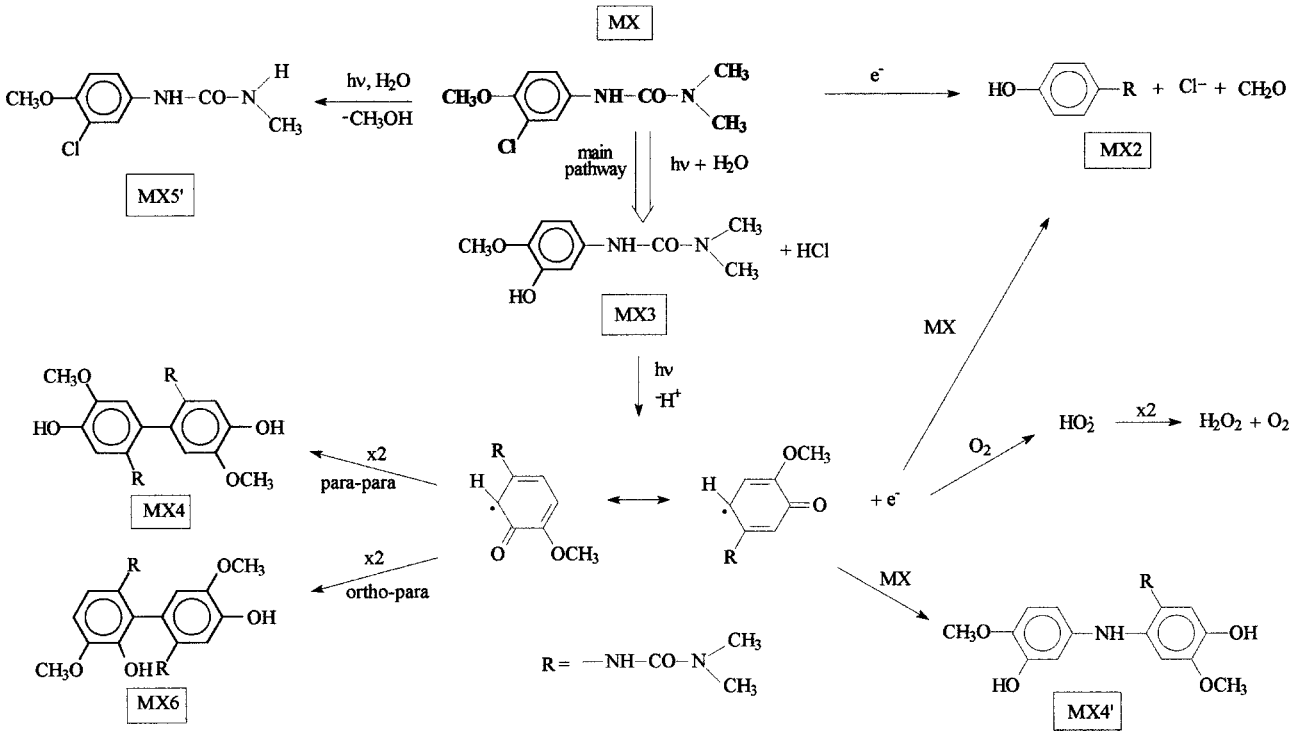


Figure 7. Main pathways for the photochemical transformation of metoxuron (MX).

4.2 Photoproducts MX4, MX6 and MX4′

MX4 and MX6 have kinetics of secondary products. Their structures imply their formation from MX3. The formation of similar biphenyl derivatives has previously been reported in the case of the photolysis of phenol.[9] The intermediate formation of a phenoxyl-like radical is favoured by the presence of an oxidant which traps the ejected electron or the hydrogen atom released (Fig 6(b)). Formation of the third expected product (2-2′ coupling) is probably unfavourable because of steric hindrance.

The formation of MX4′ is explained by the reaction of the radical with the NH—CO bond of metoxuron (MX) (Fig 6(c)).

4.3 Formation of MX5′ and MX2

MX5′ results from the elimination of a methyl group from nitrogen. A similar reaction has previously been observed with several halogenophenylureas.[1,2] This was tentatively explained in the case of diuron by the intermediate formation of a methyl enolate.[10] A similar mechanism might therefore be proposed for metoxuron, as shown in Fig 6(d).

The formation of MX2 is not affected by oxygen. It may be explained by hydrolysis of the methoxy bond concerted with C—Cl scission (Fig 6(e)).

The main routes for the photochemical transformation of metoxuron are summarised in Fig 7.

5 CONCLUSIONS

The phototransformation of metoxuron, 3-(3-chloro-4-methoxyphenyl)-1,1-dimethylurea, in aqueous aerated solution proceeds initially almost exclusively to one product, 3-(3-hydroxy-4-methoxyphenyl)-1,1-dimethylurea, (MX3) via a mechanism of photohydrolysis. By comparing the phototransformation of metoxuron with that of chlorotoluron [3-(3-chloro-4-methylphenyl)-1,1-dimethylurea] and monuron [3-(4-chlorophenyl)-1,1-dimethylurea], it appears that photohydrolysis is more likely to occur exclusively when the chlorine atom is in the *meta* position. This conclusion is consistent with previously reported observations from the phototransformation of chlorophenols and chloroanilines. Reactivity is often more complex with *para* halogenated derivatives.

In the course of the metoxuron study several other minor photoproducts were identified:

3-(4-hydroxyphenyl)-1, 1-dimethylurea (MX2),
3-(3-chloro-4-methoxy)-1-methylurea (MX5′),
two biphenyl derivatives (MX4 and MX6) derived from the main photoproduct MX3,
a substituted diphenylamine (MX4′) also formed from MX3.

The formation of the last three compounds (MX4, MX6 and MX4′) is expected to be negligible in diluted environmental conditions since their formation involves bimolecular reactions.

The irradiation of metoxuron significantly increases the toxicity of solutions to *V fischeri*. This is an important feature concerning the environmental impact of this herbicide. However the scale of toxicity may be different according to the test micro-organisms and it is probable that the photoproducts identified would photo- or biodegrade in further stages.

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