Heterogeneous Photocatalysis Degradation of Wasted Drugs in Water Matrix by Combined UV/Vis/Microwave

G. Visco, F. Bellanti, M. Castrucci, C. Costanza and L. Campanella

Chemistry Department, Rome University "La Sapienza"

P.le Aldo Moro 5, 00185 Roma, Italy, Europe

Introduction

The alarms about the presence of drugs in surface waters are, by now, always present and diffuse, not only in wastewater but also in lakes and rivers [1]. This problem arose because of the pharmaceutical principles production increased during the latest years.

Following the World Health Organization definition of health, "a state of complete physical, mental and social well-being and not merely the absence of disease or infirmity" the use of drugs is widely spread.

Drug abuse has deleterious consequences both on human health and on the delicate ecosystems; infact drugs can reach streams and rivers spreading easily in to the environment and due bioaccumulation, it is possible their transfer into the food chain.

Even though the presence of drugs in waters has been already investigated for several years do not exist sufficient data regarding their recalcitrance to the natural degradation. In this research a new method for degradation of the drugs are performed; we always associate to a pool of drugs a dye and/or a pesticide to compare the different resistance to the photodegradation i.e. the recalcitrance.

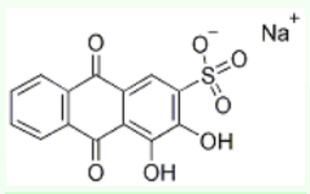

Fig. 1, Our representative dye, Alizarin Red S, CAS: 130-22-3, Colour Index Number 58005. A dye used with a variety of paints and surfaces, including oil, watercolour, tempera, dry interior plaster, and chalk.

For this research we select a stable dye, the Alizarin Red (Fig. 1). Alizarin was used for dying clothes in Asia in ancient times; also it was found at Pompeii, and in Egyptian textiles from as early of XIV seculum b.C.. In the Middle Ages, the cultivation of madder is promoted for painting and paper inks.

For a lot of drugs there are also studies about accelerated ageing [2], we hope this and similar works are useful to obtain a recalcitrance scale comparing these molecule classes.

Several European projects were dedicated to the study of persistence of organic molecules in the environment and one in particular to the drugs in waters. During one of them (Rempharmawater) were investigated the flowing in and out waters of a Sewage Treatment Plants (STP) of Latina a small town of south Lazio discovering little but significant drugs concentrations. In a previous congress [3] we show the results and a comparison of recalcitrance of drugs, pesticides and dyes.

Ecopharmacology

There are at least two definitions that can be given to ecopharmacology so we can obtain confusion on this new term.

The first definition is more used and used also in this text; the presence of pharmaceutical residuals in the environment, which always interact with the ecosystem in a negative way.

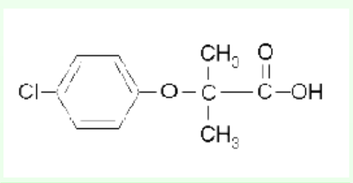

Fig. 2, Clofibric Acid, CAS: 882-09-7, a old drugs with long environmental persistance, it is an antihyperlipidemic that causes the decrease of plasmatic levels of cholesterol and triglycerides and increasing the lipase activity

Pharmaceuticals cause modifications to the ecosystem by interaction-absorption of drugs, metabolites, excipients, stabilising, thickenings, etc.. Environment-Ecopharmacology [4] defines therefore the study of the interaction with the environment of the drugs and, from the obtained results, proposes to the researchers of pharmacovigilance the remedies to reduce the environmental impact. In Ecopharmacology it is necessary to make studies on all those products: personal care, hospital cleaning, disinfectant, antibacterial, plant protection, and veterinary drugs products which are now heavily used in every field. Perhaps, some of them may be assimilated to drugs (having molecules with similar structures) and are produced in huge amounts and used almost without control.

Unfortunately for Ecopharmacology a second definition is given, tied up to the discovery of new medicines studying both native populations not civilised that they use traditional medicines drawn by plants, algae, mushrooms, etc. (rain forest for example) both extracting active principles from the biodiversity.

This other aspect of the pharmaceutical research can seem very positive but cases have been signalled of "ecorape" in which the native populations and also the States don't receive any benefit from the possible medicines that will be produced. The patentability of an already present molecule in nature complicates then the satisfaction of the legitimate expectations of the populations that suffer damages from this Ecopharmacology.

Using the first definition, before studying an effective abatement method is necessary to investigate, or at least to assume, how the pharmaceutical active principles reach the waters. Drugs have several ways to reach the surface waters:

- by incomplete metabolisation of an organism with the consequent excretion of the starting products and metabolised compounds;
- by uncontrolled draining away, also involuntary, both by the patients and by producers or distributors still unconscious of the problem;
- by incorrect use (fraudulent, doping, wrong proportions) by people and by animal breeding in farms.

European Projects

The importance of such problems is also highlighted by several initiatives of EU with the purpose to monitor the presence of drugs and other pollutant molecule in the environment and to reach methods for their correct and effective abatement:

a) ERAPharm (Environmental Risk Assessment of Pharmaceuticals): an investigation unstudied major routes leading to exposure of the terrestrial and aquatic environment, the fate of human and veterinary pharmaceuticals in surface water and sediment and the effects of antibiotics on microbial communities to the spread of genetically encoded resistance;

b) AquaStress (Mitigation of Water Stress Through new Approaches to Integrating Management): an improvement of the understanding of water stress and the development of supporting methods and tools to evaluate different mitigation options and their potential interactions with environment;

c) Rempharmawater (Ecotoxicological Assessments and Removal Technologies for Pharmaceuticals in Wastewater): a study of prevention of pollution of surface-water resources. The project also focuses on database development, *Pharmafic*, to store data (ecotoxicological, physical and chemical properties, etc) on the drugs studied;

d) Eravmis (Environmental Risk Assessment of Veterinary Medicines in Slurry): a study about exposure and distribution models for the environmental risk assessment of veterinary medicinal products at registration;

e) Poseidon: this project defines the activities of the EU in the field of research, technological development and demonstration for the period of 1998-2004;

f) WSSTP (Water Supply and Sanitation Technology Platform): an european initiative to produce a common vision document for the whole European water industry together with a strategic research agenda and an implementation plan for the short (2010), medium (2020) and long term (2030).

Following the European project for this study six molecules where used: Carbamazepine, Nifuroxazide, Clofibric Acid, Sulfamethoxazole, Ofloxacin, Alizarin Red S. In figures 1-6 are shown molecules, structures, CAS Numbers and the principal use.

Among many articles found on the subject, in which several waters are analysed measuring the drugs concentration, we signal the following one with an extensive list of active principles [5].

drugs	sample1	sample2	sample3
Gemfibrozil	0.81	0.84	4.76
Fenofibrate	0.16	0.10	0.16
Bezafibrate	n.a.	n.a.	0.91
Clofibric acid	0.68	n.a.	0.23
Ibuprofen	0.18	0.02	0.02
Flurbiprofen	n.a.	n.a.	0.34
Naproxen	0.29	0.41	5.22
Diclofenac	0.47	1.48	5.45
Phenazone	n.a.	0.37	n.a.
Acebutolol	0.04	0.02	0.11
Metoprolol	0.01	0.01	0.10
Oxprenolol	0.01	<0.01	0.03
Propranolol	0.01	0.01	0.09
Carbamazepine	0.30	0.34	0.50
Trimetoprim	0.04	0.03	0.13
Sulfamethoxazole	0.01	n.a.	0.03
Ofloxacin	0.58	0.29	0.31
Lomefloxacin	0.32	0.18	0.22
Enoxacin	0.03	0.01	0.03
Norfloxacin	0.07	0.06	0.06
Ciprofloxacin	0.07	0.06	0.04

Tab1, Drugs concentrations, in µg/L, found in STP effluents in south Lazio small city (in text method's description) by Palmer (Parco Scientifico e Tecnologico del Lazio Meridionale), Italy

Drugs in the environment

Two of the three molecule classes under study, the industrial dyes and pesticides-herbicides-parasiticides, are "selected" and/or "planned" to give persistence in environment, to resist for long periods in storage, transport and marketing so that they remain unchanged till the end of their use. They must be more resistant to the action of the solar radiations and, for pesticides, the persistence plays a fundamental role both in the utilisation and in the environmental pollution.

Due to their high price expiring date for drugs must be long and possibly have to remain chemically active also after the package opening. Also all the molecules that must be kept at low temperature show a good environmental persistence.

Agreeing with an Rempharmawater project, PALMER [6] analysed the surface waters coming from the STP of Latina. The research was performed with two methods: liquid chromatography with electrospray detector and MS/MS, performed in SRM mode; gas chromatography with MS detector in SIM mode: at three different moments of 2003 the active principles shown in the table 1 were identified. Only the pharmaceutical principles with concentration higher than the detection limit of 0.01 µg/L among the 26 molecules under test are here shown.

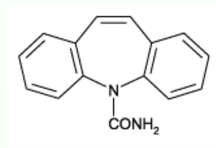

Fig. 3, Carbamazepine, CAS: 298-46-4, an anticonvulsant, antimanic agent used for epilepsy, trigeminal neuralgia, mania and bipolar disorders

Methods and Materials

There are a lot of reactions, in the environment, to remove pollutants molecules, such as:

- photolysis: by the UV rays (295-400 nm) that hit the earth surface with values between 5 and 35 W m^2 and also cross water for few centimetres. The speed of these reactions is still very low;
- oxidation: the natural and synthetic organic molecules are continuously attacked by the oxygen and degraded in presence of opportune conditions;
- ozonization: for the volatile organic molecules (VOC) the mixing up with atmosphere raises them up to 15-20 Km until they meet UV rays at low wavelength and ozone (O_3) which degrade them quickly;
- photocatalysis: catalyst increases the degradation rate and allows to obtain significant yields quickly. Catalyst as Fe_2O_3, Al_2O_3 and other metal oxides, already present in nature, accelerate very much the reactions that would spontaneously occur in very long times;
- enzymes and microorganisms, that attack the biodegradable molecules (BDOC).

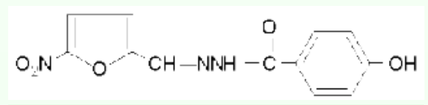

Fig. 4, Nifuroxazide, CAS: 965-52-6, it is used for the treatment of acute and chronic diarrhoea, gastro-enteritis, and colitis.
All drawings obtained from Sigma-Aldrich web site

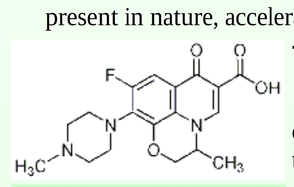

Fig. 5, Ofloxacin, CAS: 82419-36-1, it is a synthetic antibiotic, with a wide range activity, towards aerobic and anaerobic, gram-positive and gram-negative bacteria by enzyme inactivation.

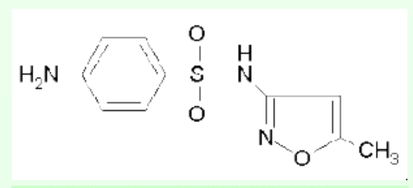

Fig. 6, Sulfamethoxazole, CAS: 723-46-6, is a sulfamide used along with trimetoprim; together they inhibit two stages in the synthesis of the acid tetra-hydrofolic in bacteria and does not affect human cell.

Many bibliographic references could be found on this subject [7-9].

The UV radiation and the solar radiations are already well known as efficient degraders; in our case it was added the radiation of microwaves that synergically improves the efficiency and allows to attack recalcitrating molecules in shorter time.

Our device (Fig. 7) is planned to use low electrical power lamps (3W Hg vapour UV-C and 20W halogen dichroic) and pulsed microwaves with duty cycle of 5% with an average value of 35W, with the continuos contribution of oxygen flow in the reaction cell containing TiO_2 Degussa P25 nanomaterials as catalyst in suspension.

The energy balance, that is the relationship between the decrease of the concentration and the electrical power used, of this tool is positive and the synergy with the microwaves places it beyond other heterogeneous photodegradation methods.

In previous presentation [3] we coined for this technique the acronym HPOP (High Performance Oxidation Process) for its efficiency, the favourable energy balance, the capacity to degrade every molecule.

For all analysis were used Analytical Grade reagents, ultrapure water, and if necessary, High Purity Grade materials principally from Merck and Sigma. For the six molecules were used pure reagents and pharmaceutical principles.

Results

For each of the 6 molecules a 10^{-5} mol/L solution was prepared in order to both simulate a concentration comparable to the one actually present in the environment and easy to be detected by our method even after a reduction to 10%.

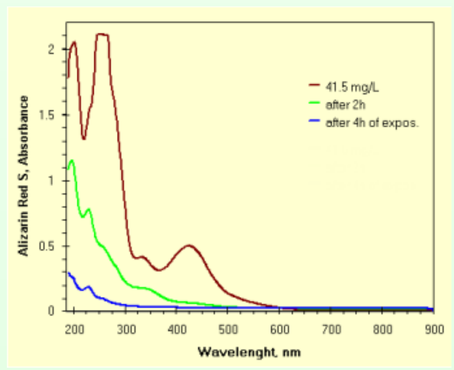

Fig. 8, Alizarin Red`S, 41.5 mg/L, UV-Vis absorbance after 2, 4 hours of exposure time in HPOP photodegrad. system

The concentration decrease, due to the photodegradation process, was tested by UV-Vis spectrophotometry, in particular the area subtended by the absorption curve was measured, in arbitrary units (a.u.), before (A_0) and after (A_t) the photodegradation and their percentage rate ($100*A_t/A_0$) calculated. For each molecule the median value of three tests was used to evaluate the effettiveness of the process.

Figs. 8 and 9 show, respectively, the absorption spectra of Alizarin Red S and Nifuroxazide at different exposure time; in both the cases the concentration decrease is well evidenced. Table 2 and fig 10 show the results obtained for all the analysed molecules.

molecule \ hours	one	two	four
Alizarin Red S	47.4	70.7	89.3
Carbamazepine	n.a.	26.7	33.9
Clofibric Acid	n.a.	62.9	76.5
Nifuroxazide	68.8	n.a.	73.7
Ofloxacin	n.a.	71.6	84.2
Sulfamethoxazole	n.a.	63.4	69.3

Tab2, computing area under UV-Vis spectrum in a.u. before and after 1, 2, 4 hours of exposure time for the six molecules and obtaining % ratio

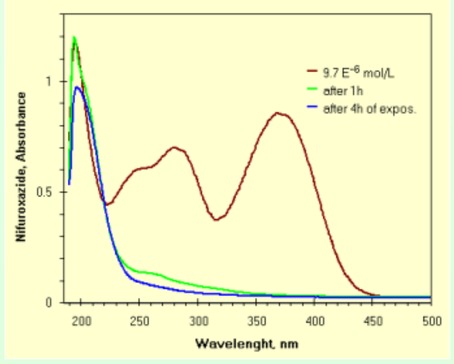

Fig. 9, Nifuroxazide, 9.7 µmol/L, UV-Vis absorbance after 1, 4 hours of exposure time in HPOP photodegrad.system

Fig. 7, Photodegradation system: 1= laboratory microwave digesting system modified, by CEM Italy, both in the firmware and hardware to obtain a pulsated operation and also a window needed by the internal lamp; 2= quartz dichroic internal lamp overvoltaged in C.C. to obtaining a continuos emission (but with loss of life time) and also "1 sun" (with spectrum, intensity and focusing near the summer sun in the Mediterranean places); 3= fused silica cell with UV-Vis transmittance, volume of 50 mL and holes for the circulation tubing and air flows; 4= counter of magnetron impulses, useful to obtain the reproducibility of the MW supply; 5= dual circuit peristaltic circulation pump, by means of four Teflon tubes it allows circulation of solution (105 mL) in about 6 minutes and maintains the catalyst suspension; 6= the cell in PMMA, above the magnetic stirrer, containing Hg vapors lamp mainly emitting at 254 nm, the thermometer, the stirring bar, a syringe needle for the continuos air insufflation and a hole for sampling during the degradation; 7= the digital thermometer allows to avoid the thermal microwave effect, monitoring the temperature (always under 40 °C) at different duty cycles; 8= membrane air pump, with splitter, to insufflate the filtered (0.47 µm) room air, by Teflon tubes, in the outside and inside cells, with the triple purpose to remove produced CO_2, to guarantee an oxygen excess in solution, to keep the catalyst in suspension; 9= two power supplier to feed UV lamp in A.C. and quartz lamp in C.C..

Conclusion

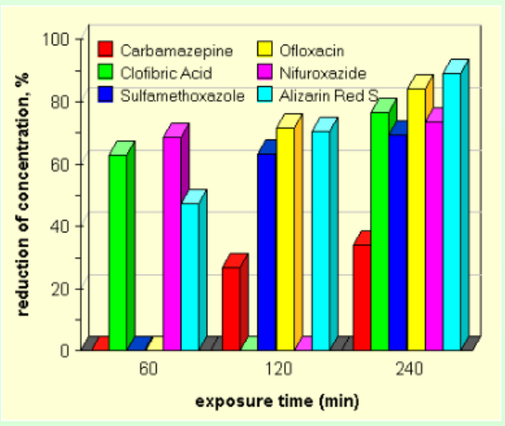

Fig. 10, Photodegradation of drugs compared with a dye. Concentration reduced by means of Three Waves Photodegradation System measure with UV-Vis area under spectra at different exposure times. TWPS = UV-C + 1 sun + pulsed microwave

This preliminary study shows the efficiency of the Three Waves Photodegradation System to destroy recalcitrant molecules. A longer irradiation time probably can let to a complete mineralisation while, at a fixed time, a recalcitrance scale can be done.

By a comparison of the results here presented with those by previous work [3], pharmacological principles shows a recalcitrance more similar to that of dyes than those of herbicide-pesticides, using our HPOP etherogenous photocatalysis.

Hystogram in Fig. 10 shows that Carbamazepine is the most recalcitrant molecule while for the other an analogous photodegration efficiency is evidenced.

Running researches concerning the use of new lamps and a modified flow system would improve the method. A new catalyst photoactivity scale as well as a new recalcitrancy scale could be obtained and shorter exposure time could be needed for the mineralisation. The lab prototype could be transformed in an industrial model able to destroy not only the toxicant but also its intermediate often more dangerous.

We wish that EU will continues to support these kind of projects and launches a campaign for the "first 100 drugs (F100D)", the 100 pharmaceutical molecules (or assimilable) more discovered in the European surface waters.

References

1) Scott Streater, Drug found in stirs concern fish area, Star-Telegram Journal, at http://www.dfw.com/mld/startelegram/news/state/7036372.htm?1c (© Reuters Limited, Dallas, Oct. 23 2003)

2) K. C. Waterman, R. C. Adami, Accelerated aging: Prediction of chemical stability of pharmaceuticals, International Journal of Pharmaceutics, 293 (2005)

3) G. Visco, M. Ricci, F. Bellanti, M, Castrucci & L. Campanella, Photodegradation of Drugs in Wastewater, Comparative Correlated Recalcitrance Analysis and Related Problems, 32° Congresso Nazionale della Societa' Italiana di Farmacologia, Centro Congressi Royal/Continental, Napoli, Italy, 1-4 Giugno 2005, http://w3.uniroma1.it/chemo/congress/napoli05/nastart.html

4) K. Kümmerer, The Effects of Drugs on the Environment-Ecopharmacology, 6^ Congress of the European Association for Clinical Pharmacology and Therapeutics (EACPT), Istanbul, 24-28. June 2003, Towards Drug Development and Rational Drug Therapy 35

5) R. Andreozzi, R. Marotta, N. Paxeus, Pharmaceuticals in STP effluents and their solar photodegradation in aquatic environment, Chemosphere, 50 (2003). **6**) Palmer web site. http://www.pst-palmer.it/

7) R. G. W. Norrish, Some fast reactions in gases studied by flash photolysis and kinetic spectroscopy, Nobel Price Lecture, December 11, 1967

8) I. Salem, Recent Studies on the Catalytic Activity of Titanium, Zirconium, and Hafnium Oxides, Catalysis Reviews, 45-2 (2003)

9) A. Fujishima, K. Honda, Electrochemical Photolysis of Water at a Semiconductor Electrode, Nature, 238 (5358), 37.(1972)

by_Dr_G.Visco
visco@uniroma1.it