Biodegradation of Diclofenac and Paracetamol Present in Waste Water through an Integrated System of Anaerobic Reactor and Constructed Wetland

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Abstract The objective of this work, that belongs to the field of alternative options for drug pollutants degradation, is to design an integrated system of anaerobic reactor and constructed wetland to eliminate paracetamol (N-acetyl-p-aminophenol) and diclofenac (2-[2-(2,6-dichloroanilino)phenyl]aceticacid) residues contained in contaminated waters. The integrated system of anaerobic reactor and constructed wetland consisted of a water tank supplying wastewater to a biodigester through a feed tank, a constructed wetland home to two species of plants *Cyperus alternifolius* and *Typha angustifolia*, and a treated water collection tank. The parameters were analyzed pursuant to NOM-001-SEMARNAT-1996, while diclofenac and paracetamol were analyzed through capillary electrophoresis. The results showed a 90% biodegradation of the organic material in the effluent. In the biodigester, the average biodegradation reaction speed was $Kt = 4.9$ days$^{-1}\pm0.870$; with $p < 0.05$. The drugs were not detected in the effluent from the wetland, and thus the integrated system of anaerobic reactor and constructed wetland represents an alternative for removing pollutants from municipal wastewaters.

Keywords Wastewater, drugs pollutants, wetlands, water treatment

Introduction

Health care requires the consumption of various drug products, and these products, once administered to the people, reach the environment through excretion and metabolization [1-2]. The occurrence of active pharmaceutical substances in the environment is of growing concern. Many drugs are released into the environment, which makes them pollutants, through industrial waste and sub-products, animal and human excretions, household garbage, etc [1. 3-4]. The administered drug product can be excreted without change, as glucuronides or sulphate conjugates, as main metabolite, or as a mixture of various metabolites. In the organism, drug products are metabolized through various mechanisms such as: oxidation, reduction, hydrolysis, conjugations, among others, and then excreted as more polar and water soluble derivatives having reduced pharmacological activity compared to the original compound [4-5].

It has been shown that drug residues are disseminated in the environment and have been detected in water supply sources, underground waters and even in drinking water, and thus it is a cause of concern because the consumption of drug products without medical prescription may lead to alterations in human beings. Conventional wastewater and drinking water treatment plants are not designed to remove these pollutants. Many of pharmaceuticals are not effectively removed by conventional treatments.

The non-planned reutilization of wastewater has had negative effects on the health of the local population and on the ecosystem in general. Both farmers and their families as well as consumers are exposed to gastrointestinal diseases caused by microorganisms present in irrigation water, besides other pollutants such as drugs, or emerging organic pollutants including pharmaceuticals [6].

Drug product concentrations in waste waters (in the order of 5g/L to 50g/L), mostly from hospitals, health centers, and population treated at home, are complex and in some cases persistent compounds, and their constant disposal in bodies of water leads to their presence in the influents to the Wastewater Treatment Plants (WWTPs) that are not designed to eliminate them, causing their accumulation in the ecosystems [4,7].

The drug products most frequently used by the Mexican population are analgesics and anti-inflammatory that are taken with or without medical prescription. Among them, diclofenac and paracetamol stand out because of their potential hepatotoxicity [4].

In some countries, the growing concern for the environmental impact of pharmaceutical products has led the regulators to recommend ways for healthcare waste handling and disposal. However, domestic wastewaters are not generally appropriately treated to eliminate drug products and thus it is necessary to look for efficient and economically viable procedures to remove this type of pollutants. It is also necessary to design standards as regards pharmaceutical waste disposal in bodies of water [6, 8-10], and thus the objective of this research was to evidence the removal of drug products residues contained in wastewaters through an integrated system of anaerobic reactor and constructed wetland (ISARCW).

Methodology

The design of the ISARCW is as follows: a feed tank supplies wastewater to a biodigester sealed with a concrete lid and equipped with a polymeric washer; said biodigester has one synthetic wastewater (SWW) inlet located at the bottom and a corresponding outlet located in the opposite upper part and is in turn fed to the constructed wetland.

The materials used as substrate for the constructed wetland were: ¾" gravel, sand, 10% fertile soil and river sand. Since gravel was the most abundant material in the constructed wetland, its porosity was determined.

The porosity of the substrate consisting of ¾" gravel was determined by the law of fluid displacement (Archimedes Law) according to Hewitt [11]. The Archimedes principle indicates that "the upward buoyant force that is exerted on a body immersed in a fluid is equal to the weight of the fluid that the body displaces".

A 20 L container was gauged with tap water at environment temperature = 33.5 °C and atmospheric pressure of 785mmHg. Gravel was then gradually added. The displaced water was collected and quantified in liters.

Then, porosity percentage was determined as follows: porosity% =(20L water- L of displaced water/20 L) X 100.

Calculations for the design and building of the ISARCW and route followed by the SWW in the biodigester and the constructed wetland

The site is located on loam soil. ARotoplas® water tank was installed at the beginning of the processing path, then two already existing concrete tanks were outfitted to form the feed tank supplying wastewater to the biodigester, and the biodigester. The floor supporting the feed tank and the biodigester was built, and inside finishing was applied to seal the biodigester. A wire mesh fence was placed around the constructed wetland for protection purposes. A treated water collection tank was build at the end of the processing path. The system of affluent and effluent conduction pipes of the integrated system of anaerobic reactor and constructed wetland was set up according to the Ramalho criteria [12].

The 450 L Rotoplas® water tank was supplied with SWW, which was fed to the 350 L feedtank, that in turn was fed to the biodigester tank of a capacity of 275 L; equipped with a polymeric washer, and having a contact surface of 2.673 m^2 equivalent to 40 L, in the central part of the biodigester, with the following dimensions: Length= 2.43 m, Width= 0.55 m and Thickness=0.000108 m, obtaining thus a working surface of 2.673 m^2.

The reactor (biodigester) dimensions were: real working volume = 0.040095 m^3, depth= 0.60 m, width= 0.50 m and length= 1.05 m, V $_{Total}$= 0.315 m^3. The average working temperature was (28.23 °C $\pm$ 2.86).

After passing through the biodigester, wastewater is directed to the sub-surface wetland (SSWL) having a hydraulic residence time (HRT) of 10.33 days, with a rate of flow of 131.96 L/d (see Figure 1).

The sub-surface wetland with a volume of 2840 L was build on solid concrete floor, with fine finishing inside, and a slope of approximately 1%. A concrete floor was built around it, as well as a mesh booth with polycarbonate cover to allow the passage of light and offer protection against rain. A treated water collection tank was build at the end of the processing path (see Figures 1 and 2).
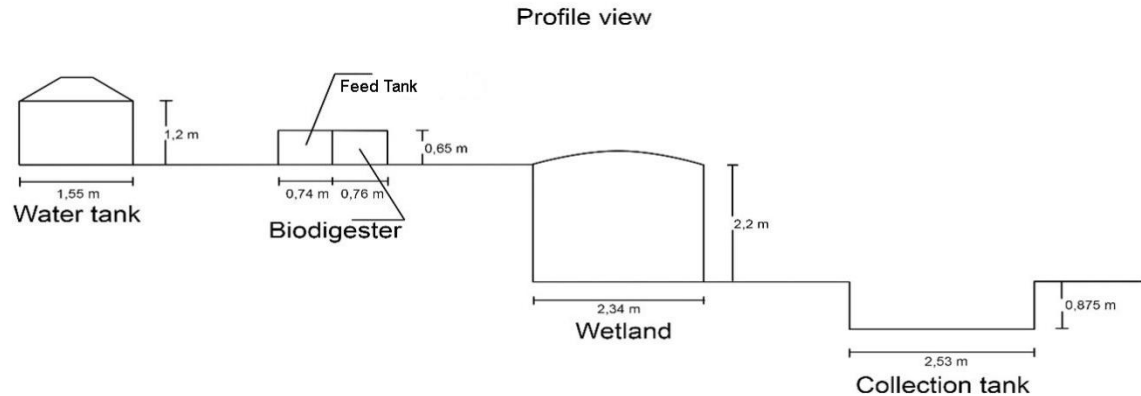
Figure 1: Integrated system of anaerobic reactor and constructed wetland with water tank, feed tank, biodigester, wetland and treated water collection tank
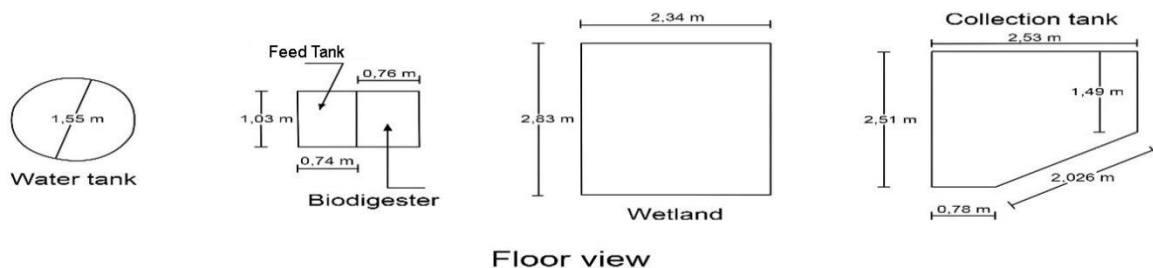

Figure 2: Plan view of the integrated system of anaerobic reactor and constructed wetland showing the dimensions of the water tank, feed tank, biodigester, wetland and treated water collection tank.

SWW preparation

Synthetic Wastewater (SWW) was prepared according to a formulation suggested by some authors such as Rodríguez and Lozano, [13], as shown in Table 1.

Table 1: Formulation of Synthetic Wastewater

Component	Quantity (mg/L)
Peptone	160
Meat extract	110
Urea	30
KH_2PO4	28
$MgSO_4.7H_2O$	2
$CaCl_2.H_2O$	4
NaCl	7

$BOD_5$was adjusted at 180mg/L according to the proposal of Metcalf and Eddy [14]. Diclofenac concentrations were kept in a range from 0.1µg/L to 28.30mg/L while paracetamol concentrations were kept in a range from 6.0mg/L to 53.0mg/L, as reported by various authors [7, 15] in WWTP effluents. In adherence to these criteria, 2.40mg/L of diclofenac and 4.8mg/L of paracetamol were added to the SWW formulation as working concentrations for the integrated system of anaerobic reactor and constructed wetland.

During the operation of the combined system at steady state during 90 days, the following parameters were analyzed weekly according to APHA, 2012: Dissolved Oxygen (DO),Chemical Oxygen Demand (COD),and

Biochemical Oxygen Demand (BOD). The parameters were measured at steady state over a period of eight weeks, samples were taken from the biodigester affluent and effluent. The results obtained were compared with the values permissible according to NOM-001-SEMARNAT-1996 [16].

The species cultivated in the ISARCW were selected according to their availability in zones close to the site to ensure their proper adaptation to this specific environment. They consist of the following aquatic plants: common cattail, *Family: Typhaceae. Scientific name: Typha angustifolia* and umbrella papyrus, *Family: Cyperaceae*, *Scientific name: Cyperus alternifolius.*

Figure 3 shows the constructed wetland, with the cultivated plants *Typhas* and *Cyperaceae*, part of the integrated system of anaerobic reactor and constructed wetland.

Family: Typhaceae
Scientific name: Typha angustifolia
Common name: Pasto salchicha, cuete

Family: Cyperaceae
Scientific name: Cyperus alternifolius
Common name: Papiro, navajuelas

Figure 3: Constructed wetland of the SWW treatment system

Registrations of parameters BOD$_5$, COD, DO, Temperature, pH of the experimental ISARCW during operation

Once the system was operating at steady state, (8 months after the start) weekly samples were taken during eight consecutive weeks, at the three following places: feed tank of synthetic wastewater plus drugs, biodigester outlet and constructed wetland outlet.

Wastewater sampling at each one of the points was performed according to conditions appropriate to ensure sample preservation pursuant to MEXICAN STANDARD NMX-AA-003-1980. The wastewater samples were transferred in rows at 4 °C to the CONAGUA laboratory in Tuxtla Gutiérrez, Chiapas where BOD$_5$, COD, DO, and pH were determined.

Operation and analysis of drug residues in the SWW treated in the ISARCW: diclofenac and paracetamol

After a three-month steady state operating period, sampling was performed to quantify drug concentrations in the system.

Samples were taken at the three above mentioned points to determine diclofenac and paracetamol contents. The purification of the drugs present in the synthetic wastewater was performed through C18 solid phase cartridges according to the EPA 1694 method [17].

Diclofenac and paracetamol were quantified through the capillary zone electrophoresis technique using a Beckham model MDQ equipment, with a 10-cm long fused-silica column having an internal diameter of 75µm. The supporting electrolyte was a 30 mM phosphate solution, pH 8. The sample was injected during 5 seconds at 0.5 psi, with a detection limit of 40 ng/L.

Equation 1 was used to determine the drug degradation constant.

Calculations for the design and construction of the integrated system of anaerobic reactor and constructed wetland.

$$\frac{Ln\ Ci - Ln\ Ce}{T} = K_t \qquad\qquad \textbf{Equation 1}$$

ISARCW Maintenance

The routine operation and maintenance of the constructed wetland included: control of water supply and water depth, cleaning of the inlet and outlet structures and revision of the irintegrity, handling of the constructed wetland vegetation, and routing monitoring.

Since the species *Tipha* and *Cyperus* grew at an approximate rate of 4-5 cm/month, pruning was performed every 3-4 months [18-20].

Water levels were always kept below the constructed wetland substrate. In the System, SWW was sampled at the inlet and outlet of each unit (biodigester and constructed wetland) in order to analyze the biological parameters and drug concentrations.

Results

The operation temperatures observed in the integrated system of anaerobic reactor and constructed wetland were as follows: the average environment temperature (exposed to the sun) was 35.7 °C ± 5.65, the average plant root temperature was 27.7°C ± 4.36 while the average gravel temperature was 34.5°C ± 6.20; the average temperature of the biodigester was 28.23 °C ± 2.86 and the average temperature of the constructed wetland was 27.77°C± 4.36 (Figure 4).
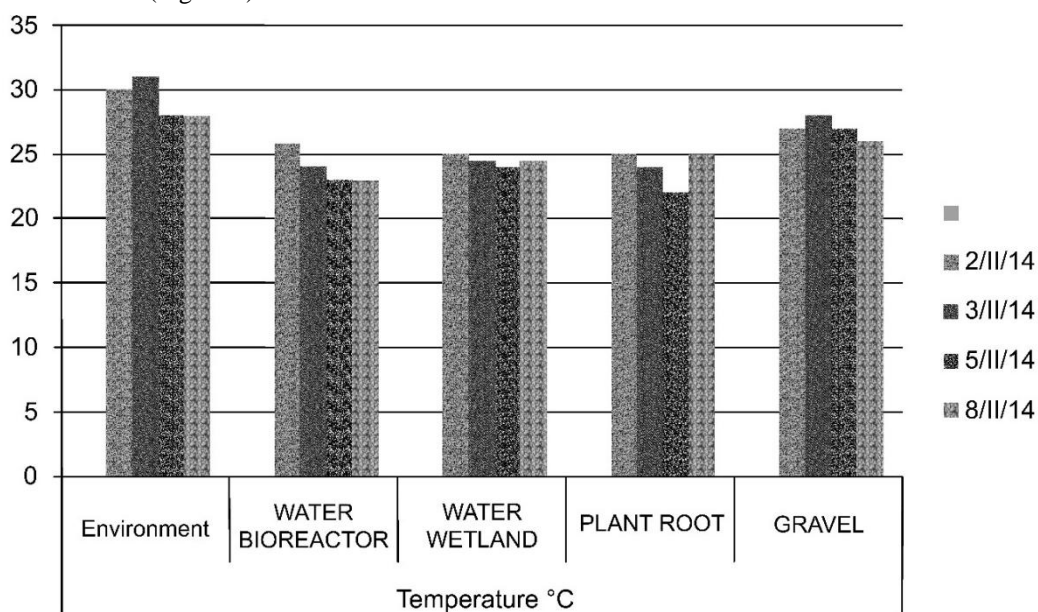

Figure 4: Comparative graph of the temperatures in the System: environment, biodigester water, wetland water, plant root, gravel

As regards porosity calculations, the results were: displaced water = 10.40 L; porosity % = (20L – 10.40 L/ 20 L) X 100 = 48%.

ISARCW operation and maintenance

The biodigester operated with an efficiency of 98 % as regards drug elimination. Paracetamol was totally degraded and was not detected anymore. According to Russo [21], 5 % of the total quantity consumed is converted inactive metabolite by the cytochrome P-450 oxidation system found in hepatic cells, leading to N-acetyl-para-benzoquinoneimine (NAPBQ). According to Russo [21], at normal paracetamol doses, the small quantity of active metabolite produced is detoxified through conjugation, preferably with reduced glutathione and excreted in urine as non-toxic conjugates of cysteine and mercapturic acid, residues that were eliminated by the biodigester. Table 1 shows the absence of drug products at the constructed wetland outlet.

Probably some diclofenac metabolites were left, according to Sisamón [22]. Table 3 shows the results obtained in the biodigester where $K_{t\ average}$ reaction speed =4.905 days^{-1}±0.870.

Table 1: Concentrations: Diclofenac Ci initial and Ce final concentrations in the ISARCW biodigester and calculation of the reaction speed constant, HRT = 24h.

Sample	Untreated water mg/L, Ci	Water from biodigester outlet mg/L, Ce	Ln Ci	Ln Ce	T(días)	Kt = LnCi – LnCe/T
1	2.2	0.021899382	0.78845736	-3.821296885	1	4.609754245
2	2.177262178	0.025542784	0.778068206	-3.667400429	1	4.445468635
3	2.185571976	0.001850027	0.781877568	-6.292555045	1	7.074432614
4	2.315141762	0.033355227	0.839470922	-3.400540787	1	4.240011709
5	2.26622879	0.036489692	0.818117124	-3.310725469	1	4.128842593
6	2.132798073	0.01687239	0.757434767	-4.082076721	1	4.839511488
7	2.246993384	0.01777578	0.809593049	-4.029918409	1	4.839511458
8	2.175117935	0.01565403	0.777082886	-4.157026887	1	4.934109773
9	2.297223501	0.0165328	0.83170122	-4.102408993	1	4.934110213
Average	2.221815289	0.020663568				4.893972525
Standard deviation=	0.062130806	0.01034226				0.870243111

Discussion of the analyzed parameters: BOD$_5$, COD, DO, in the ISARCW

The average BOD$_5$ (mg/L) results obtained in synthetic wastewater (SWW) at the biodigester inlet are 154.16mg/L ± 17.042,while said values at the biodigester outlet are57mg/L± 31.412; moreover, an important reduction is observed at the wetland outlet, at 13.01mg/L± 5.59, as shown in Figure 5.
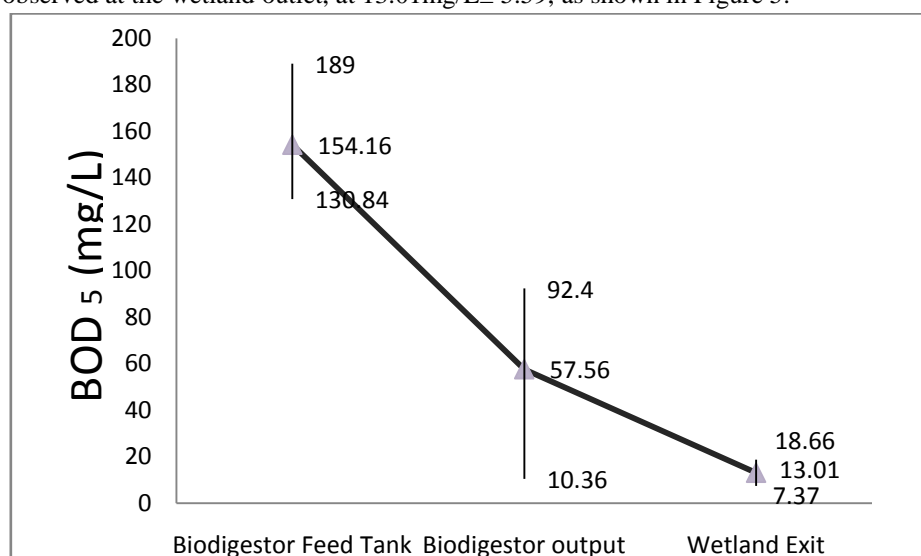

Figure 5: Results of the Biochemical Oxygen Demand of the SWW treated in the ISARCW

The results obtained as regards the chemical oxygen demand COD (mg/L) of the Synthetic Wastewater (SWW) were as follows: 131.54mg/L ± 10.859 at the biodigester inlet, 71.56mg/L± 12.42 at biodigester outlet, while an important 68% reduction was observed at the constructed wetland outlet, leaving only 41.93mg/L±4.582 (Figure 6).
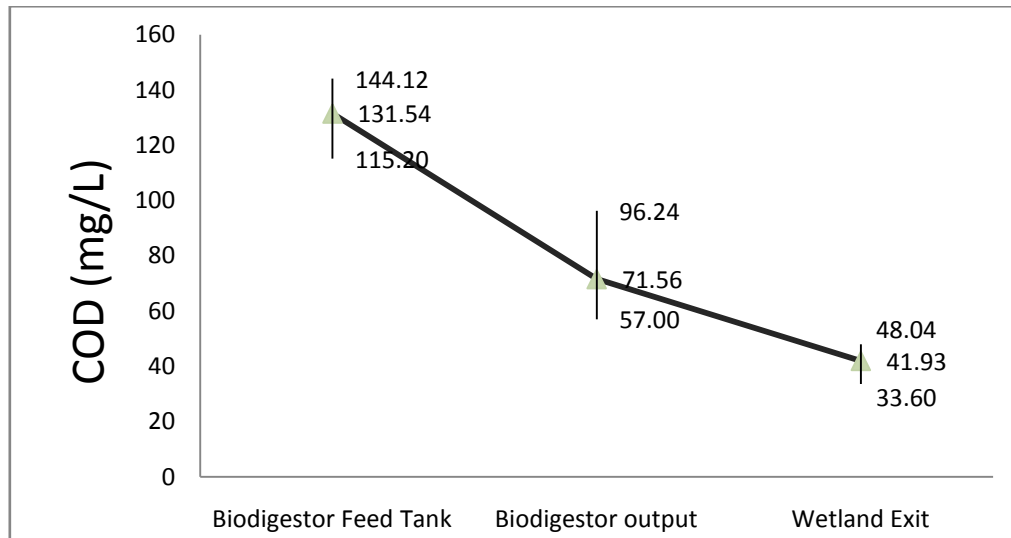

Figure 6. Graph of the Chemical Oxygen Demand of the SWW treated in the ISARCW

The average results obtained as regards dissolved oxygen (DO) (mg/L) of the synthetic wastewater (SWW) were as follows: 7.28mg/L ± 1.492 at the biodigester inlet, down to 0.38mg/L±0.142at the biodigester outlet, and up a little to 3.99mg/L± 1.613 at the wetland outlet (Figure 7).
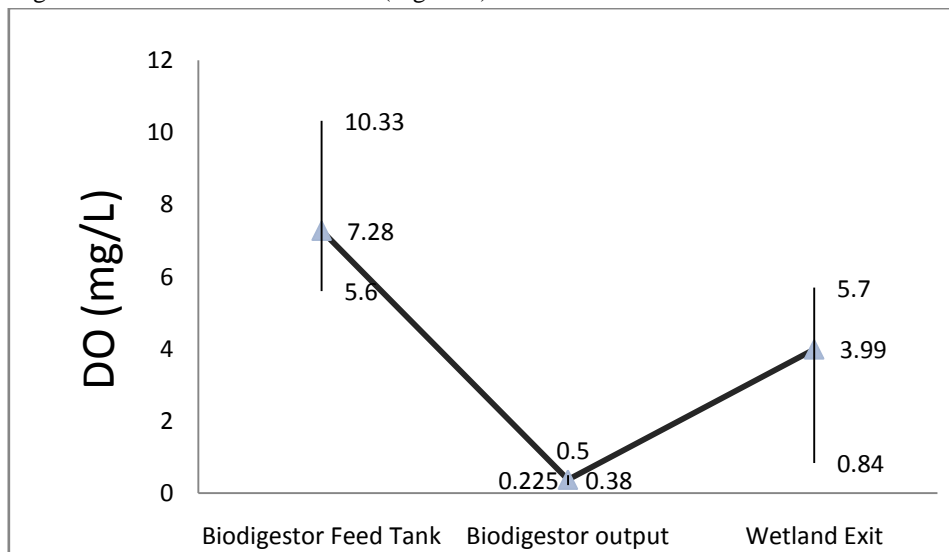
Figure 7: Results of the contents of Dissolved Oxygen of the SWW treated in the ISARCW

Discussion of the results of the analysis of drugs treated in the ISARCW

Figure 8 shows the results of untreated water, obtaining values that are very close to the ones initially fed to the system. At the biodigester outlet, the presence of a residue that does not correspond to diclofenac was observed, possibly some of the metabolites generated by the diclofenac molecule in its biological degradation. At the constructed wetland outlet, no presence of diclofenac was detected, but traces of residues (probably metabolites) were observed, that must be identified through other analytical methods. Figure 8 shows the results of the paracetamol and diclofenac removal from the ISARCW.
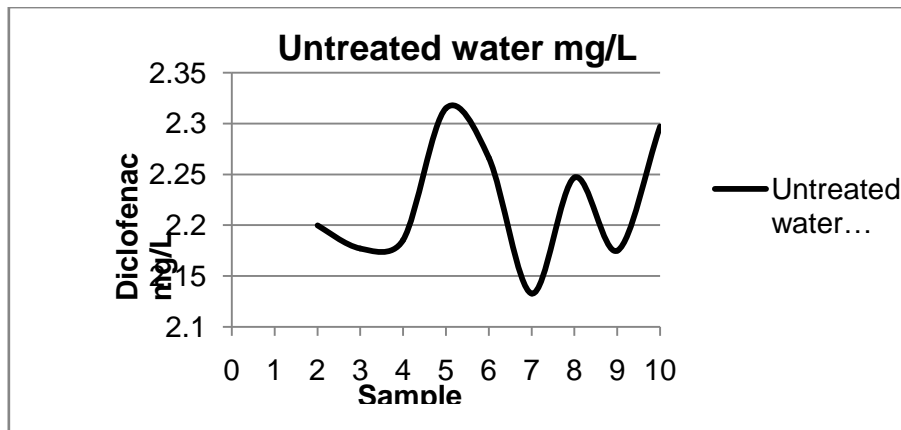
Figure 8: Graph of diclofenac concentration in the SWW fed to the biodigester of the ISARCW

Figure 9 shows the concentrations of diclofenac residues in the SWW treated by the biodigester, with a 98% removal rate.
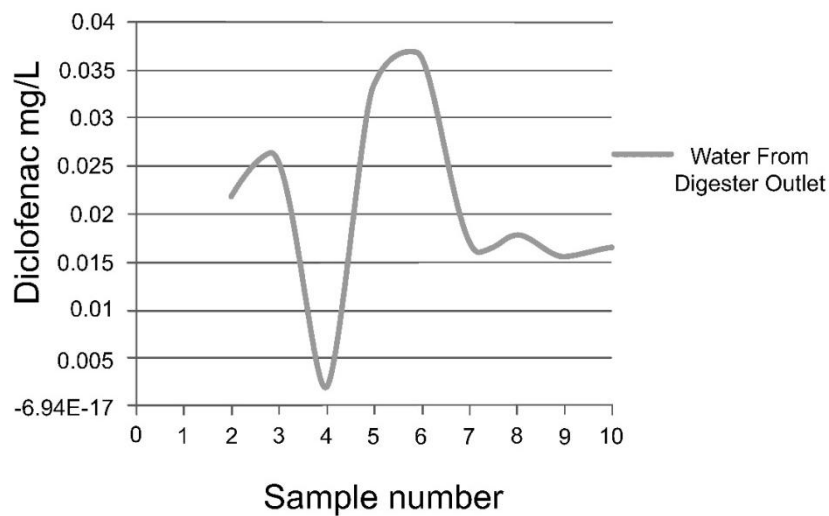

Figure 9: Diclofenac concentrations at biodigester outlet

Figure 10 shows the compared concentrations at the ISARCW; it can be seen that the concentration at the constructed wetland outlet is nil.
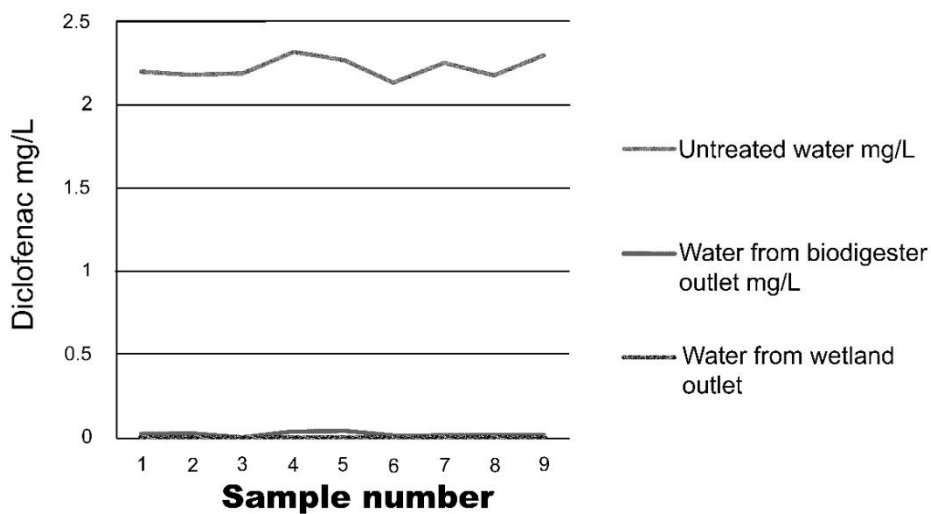

Figure 10: Diclofenac concentrations in untreated SWW, at the biodigester outlet, and at the constructed wetland outlet of the ISARCW.

With the information shown in Table 1 and Equation 1, diclofenac degradation constant K_t was determined, averaging 4.905436668 days^{-1} $\pm$ 0.8702 at an average temperature of 28.23 °C$\pm$ 2.86.

On the other hand, as reference to the typical values of the kinetic parameters reported in works performed by other authors such as Zhang et al., [23] and Nowrotek et al., [20], no Kt values have been especially referred to for diclofenac or paracetamol or for any specific drug.

A repetitive pattern was observed as regards diclofenac concentrations. In the electropherogram, peaks of the diclofenac sample in untreated water were observed, migrating at 9 minutes according to diclofenac standard sample. In the electropherogram of effluent water from the biodigester, the peak was observed migrating at 12 minutes, the 3-minutedifference possibly corresponding to another compound.

There is no quantitative recording of wetland effluent because no characteristic peak was observed (Figure 11).
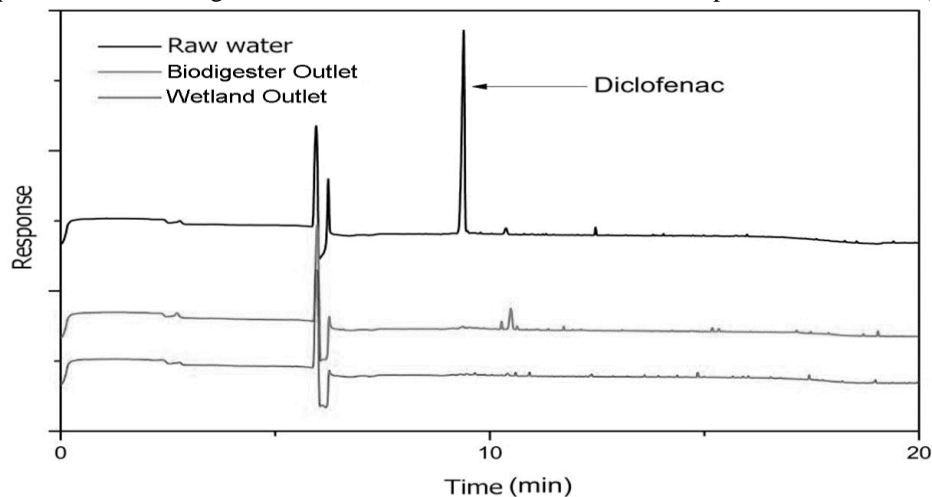
Figure 11: Electropherogram of diclofenac degradation in the ISARCW

Paracetamol may have been eliminated by the solvents during the extraction process because of its solubility in them, [24] and could not be detected. As control, an analysis was conducted without extracting the untreated water, SWW, in order to check whether it was present, and, once verified, a Spiking (standard paracetamol aliquot) was added to confirm its presence in the sample, which was corroborated (Figure 12). It is not possible to run the samples in these conditions, because the equipment would be damaged if the analyses were performed directly without the extraction requested by the technique. Moreover, paracetamol presents some difficulty because of its water solubility due to the possible in stability of its polarity [25] (Figure 12).
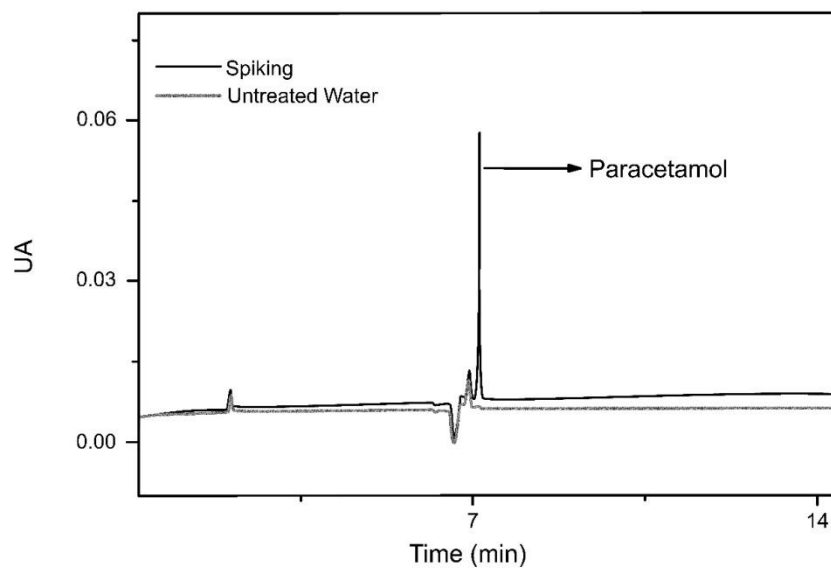
Figure 12: Electropherogram of paracetamol in untreated SWWof the ISARCW

For the above reasons regarding paracetamol, a statistical analysis was performed through the ***One-way anova*** method on the data obtained with diclofenac.

As regards the results of the diclofenac concentrations in untreated water (prior to treatment in the biodigester), an X means of 2.2 and a standard deviation, $\sigma = 6.21308E^{-02}$ were obtained. The statistical report showed calculated T (Tc)= 107.28 withp=0.0000; in this case, a critical T was calculated (Tcr)= 2.31,according to the decision Law that if Tc >Tcr, p< 0.05,95% removal was recorded.

The diclofenac concentration at the biodigester outlet was obtained for diclofenac having an average X = $2.066357E^{02}$ and a standard deviation, $\sigma = 1.034226E^{02}$.

In statistical analysis, a calculated T (Tc)=5.9939withp = 0.000326 was reported; in this case, critical T (Tcr)=1.86.

SWW plus drugs without treatment through the ISARCW. Graphs of the statistical analysis through the ***One-way anova*** method:
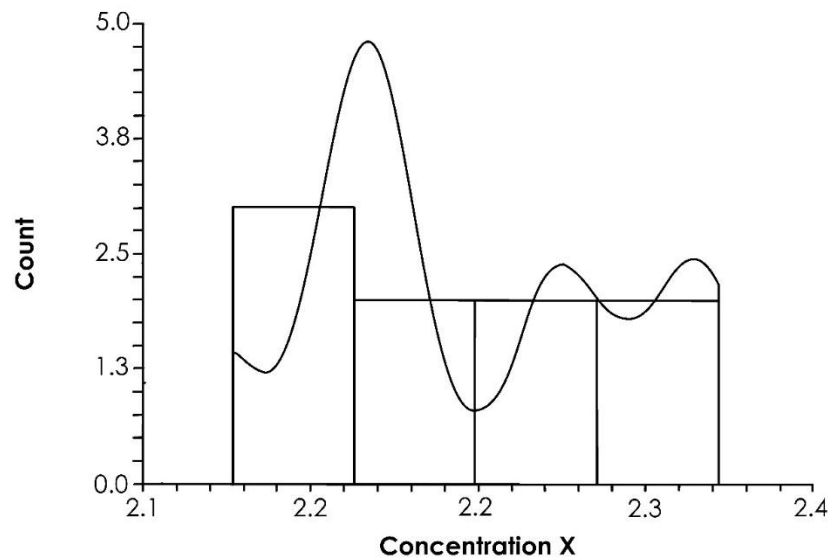

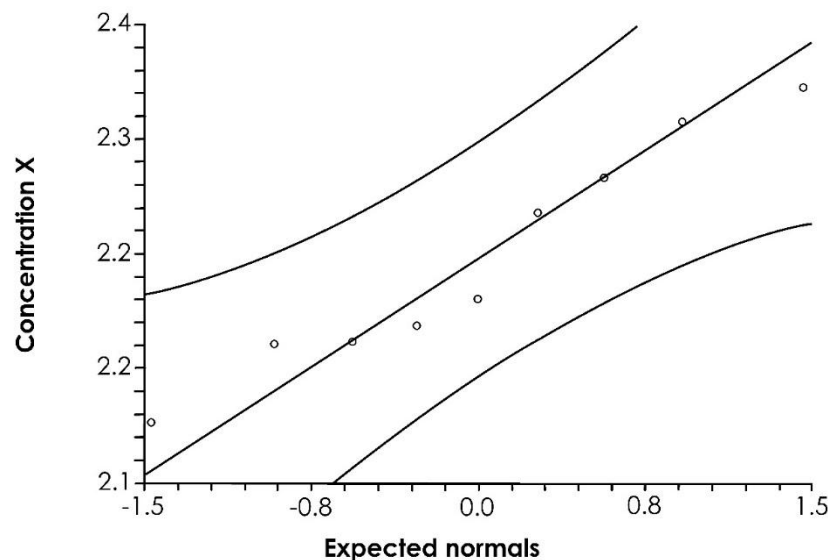*Figure 13: A) Concentration histogram*

Figure 13: B) Normal distribution of diclofenac concentration in SWW plus drugs without treatment through the integrated system of anaerobic reactor and constructed wetland

SWW plus drugs at the biodigester outlet: Graphs of the statistical analysis through the *One-way anova* method*:*
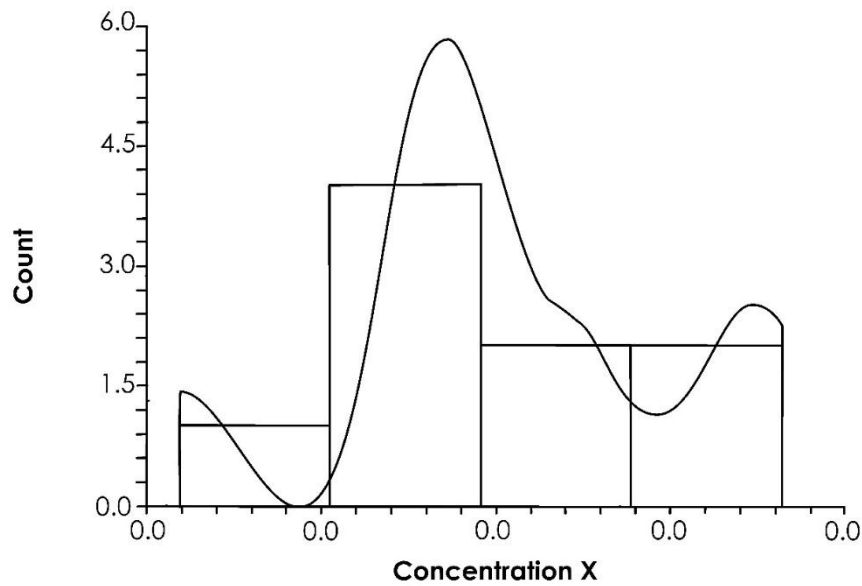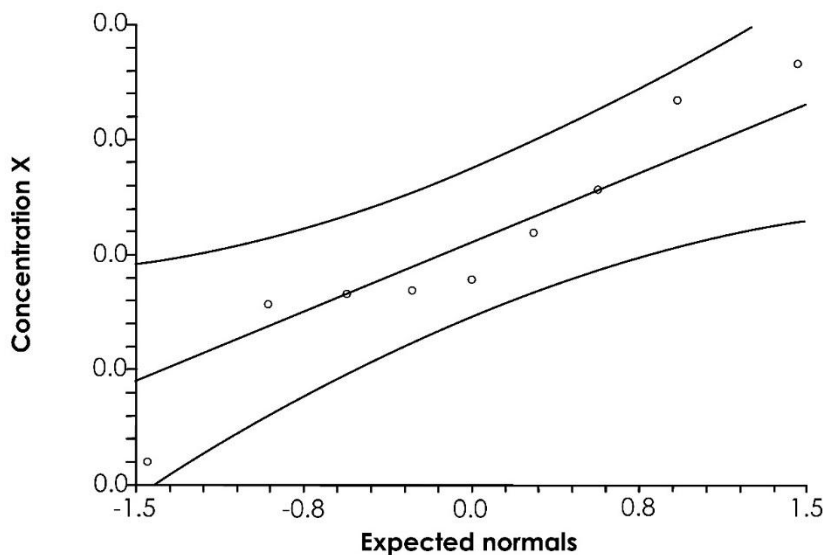

Figure 14: A) Concentration histogram

Figure 14: B) Normal distribution of diclofenac concentration in SWW plus drugs treated in the ISARCW

Conclusions

The integrated system of anaerobic reactor and constructed wetland adequately built and operated eliminates drugs. No presence of diclofenac or paracetamol was analytically detected from the constructed wetland effluent. A biodegradation constant in the biodigester of $K_{t\ average}$ **reactions peed=4.905 days^{-1}±0.870** was obtained**;** with $p < 0.05$.

The typical values of the anaerobic process constant k_{tdel}, in wastewater were: for acetates, from 3.6 to 4.8mg/mg d; for propionates, 9.8mg/mg d; for palmitates, 3.85mg/mg d, resulting from an anaerobic biodegradation.

With the wetland expressly built for this study, a 98% efficiency was obtained as regards anti-inflammatory and analgesic drugs from wastewaters having passed through the biodigester.

Plants of the species *Typha spp.*and *Cyperus spp.*used in the wetland of this study were extremely well adapted to the environment, and generated excellent results.

Thus, this depuration technique for all types of contaminants is valid, economical, requires low maintenance and no energy, is effective and should be taken into account as an alternative or complement to conventional WWTPs.

References

[1]. SJ Khan; DJ Rose; CM Davies; GM Peters; RM Stuetz; R Tucker; NJ Ashbolt. Chemical contaminants in feedlot wastes: concentrations, effects and attenuation. Environ Int. 2008; 34(6):839-859.

[2]. C García-Gómez; P. Gortáres-Moroyoqui; P Drogui. Contaminants emergentes: efectos y tratamientos de remoción Química Viva [en línea] Agosto 2011, año10: [Fecha de consulta: 19 de enero de 2017] Disponible en: <http://www.redalyc.org/articulo.oa?id=86319141004> ISSN

[3]. AB Boxall; LA Fogg; PA Blackwell, P Kay; EJ Pemberton, A. Croxford. Veterinary medicines in the environment. *Rev Environ Contam Toxicol.*2004; 180:1-91.

[4]. V Moreno-Ortiz; J Martínez-Núñez; J Kravzov-Jinich; L Pérez-Hernández; C Moreno-Bonett; M Altagracia-Martínez. Los medicamentos de receta de origen sintético y su impacto en el medio ambiente Revista Mexicana de Ciencias Farmacéuticas, 2013; vol. 44, núm. 4, 2:17-29 Asociación Farmacéutica Mexicana, A.C. Distrito Federal, México. Disponible en: http://www.redalyc.org/articulo.oa?id=57930578003

[5]. S Raju; ZM Xiaochun; H Benjamin; J Bethlyn; B Sloey; F Sarah; O Wilson; J. Greg; J Xiao, L Gary. Skiles Pharmacokinetics, Biotransformation, and Excretion of [14C] Etelcalcetide (AMG 416) Following a Single Microtracer Intravenous Dose in Patients with Chronic Kidney Disease on Hemodialysis. February 2017; Volumen 56, Issue 2, pp 179–192.

[6]. Wells, M. J. M.; Bell, K. Y.; Traexler, K. A.; Pellegrin, M. L.; Morse, A. Emerging Pollutants. Water Environ. Res., 2010, 82 (10), 2095 – 2170.

[7]. V Henriquez. "Presencia de Contaminantes Emergentes en aguas y su impacto en el ecosistema. Estudio de caso: productos farmacéuticos en la cuenca del Río Biobío", región del Biobío, Tesis: Universidad de Chile, Chile. 2012, 387

[8]. M Petrovic; D Hernando;S Díaz-Cruz; D Barceló. Liquid chromatography–tandem mass spectrometry for the analysis of pharmaceutical residues in environmental samples. A review of *Journal of Chromatography A, 1067*, 2005, 1-14.

[9]. MJ Benotti; RA Trenholm; BJ Vanderford; JC Holady; BD Stanford. Snyder Pharmaceuticals and endocrine disrupting compounds in U.S. drinking water. Environ Sci Technol. 2009; 43(3):597-603.

[10]. Taggart; K Senacha; RG Green; YV Jhala; B Raghavan; AR Rahmani; R Cuthbert; DJ DJ Pain, AA Meharg. Diclofenac residues in carcasses of domestic ungulates available to vultures in India. Environ Int. 2007; 33(6):759-756.

[11]. PG Hewitt. Conceptos de Física, Editorial Limusa S.A. de C.V. Editorial. 2009, 682.

[12]. RS Ramalho. Tratamiento de aguas residuales. Edit. Reverté, S.A. Barcelona España. 2003; 503-533.

[13]. SIM Rodríguez; RWA Lozano. Preparación, composición y uso de agua residual sintética para alimentación de reactores prototipo y de laboratorio. Año 8, Número 11, Revista Didáctica de Ambiental. Bogotá D.C., Colombia. 2012,10 -16.

[14]. Metcalf and Eddy, 1998. Ingeniería de aguas residuales: tratamiento, vertido y reutilización. Tercera edición. Edit. Mc. Graw-Hill, México. Pp.271-345.

[15]. TA Ternes. La aparición de las drogas en las plantas de tratamiento de aguas residuales y ríos alemanes. Investigación del agua., Volumen 32, Número 11,1 de Noviembre 1998, 3245- 3260 (16). Editorial Elsevier.

[16]. NOM-001-SEMARNAT-1996,Que establece los límites máximos permisibles de contaminantes en las descargas de aguas residuales en aguas y bienes nacionales.

[17]. EPA. Method 1694: Pharmaceuticals and Personal Care Products in Water, Soil, Sediment, and Biosolids by HPLC/MS/MS. 2007, 27-43, 42-45, 65-68.

[18]. A Dordio; AJP Carvalho; D.M. Teixeira; CB Dias; AP Pinto.Removal of pharmaceuticals in microcosm constructed wetlands using *Typha spp.* and LECA. *Bioresource Technology,*2010; 101 (3), pp. 886-892. Cited 78 times. DOI: 10.1016/j.biortech.2009.09.001.

[19]. A Dordio; R Ferro; D. Teixeira; AJ Palace; AP Pinto; CMB Dias. Study on the use of *Typha spp.* for the phytotreatment of water contaminated with ibuprofen.*International Journal of Environmental Analytical Chemistry,* 2011; 91 (7-8), pp. 654-667. Cited 24 times. DOI: 10.1080/03067311003782708

[20]. M Nowrotek; A Sochacki; E Felis; K Miksch. Removal of diclofenac and sulfamethoxazole from synthetic municipal waste water in microcosm downflow constructed wetlands: Start-up results. International Journal of Phytoremediation, 2016; 18 (2), pp. 157-163. DOI: 10.1080/15226514.2015.1073669

[21]. DK Russo. 2012. Paracetamol, [en línea]. [Consultado en marzo 2013]. Facultad de Ciencias Exactas y Naturales.Universidad de Belgrano. Disponible en: <www.ub.edu.ar/revistas_digitales/Ciencias/Vol8 Numero6/Articulo.pdf>

[22]. IA Sisamón. Acerca de la hepatotoxicidad del paracetamol. Revista del Hospital Privado de Comunidad 2003; vol 6, nº 2 Hospital Privado de Comunidad. Córdoba 4545. (B7602CBM) Mar del Plata. Argentina. Pags.: 2-6 MA

[23]. DQ Zhang; T Hua; RM Gersberg; J Zhu; WJ Ng; SK Tan. Fate of diclofenac in wetland mesocosms planted with *Scirpus validus. Ecological Engineering*, (2012) 49: 59-64.

[24]. C Tejada; E Quiñones;M Peña. Contaminantes Emergentes en Aguas: metabolitos de fármacos. Una revisión. Revista Facultad de ciencias básicas. Universidad Politécnica de Granada. 2014; 10 (1):80-101

[25]. CI Bolaños; EE Alonso; P Luengas;H Barbosa; F Martínez. Solubilidad de Acetominofén en los sistemas agua-alcohol USP- jarabe USP y Agua-Alcohol USP- Sorbitol USP. Revista Colombiana de Ciencias Químico farmacéuticas. 1999, 71-75.