USE OF MICROPAN COMPLEX AND EPARCYL PRO BIOACTIVATORS FOR PHARMACEUTICAL WASTEWATERS TREATMENT

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The paper aimed to find a practical solution for solving the problem of wastewaters with a high content of organic substances discharged from pharmaceutical industry. The influence of the main operational parameters (pH, amount of bioactivator, bioactivator type) on treatment process efficiency was investigated for comercially available Micropan complex and Eparcyl pro bioactivators. Treatment efficiencies of 88-93% were obtained for the optimal operating conditions.

Keywords: pharmaceutical wastewater, *Micropan complex*, *Eparcyl pro*, bioactivator

1. Introduction

Taking into account that in 2025 the experts are expecting that more than one third of the global population will not have access to sufficient freshwater resources we can consider that water is a limited resource, [1]. In 1960s in surface and wastewaters was identified for the first time the presence of pharmaceutical and personal care products [2]. Since then, pharmaceutical, personal care and drug industry products had experienced a tremendous growth, which led also to a growth of the generated wastewater volumes [3]. The effects of water pollution are felt directly by aquatic ecosystems and vegetation [4]. The compositional characteristics of these waters are becoming more complex (high amount of organic matter, intense color, wide pH variation domain), their treatment being a real challenge for water professionals. Low chemical oxygen demand (COD)

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removal efficiencies are caused by the presence of toxic or refractory substances in wastewaters [5].

In most of the cases, wastewaters resulting from the pharmaceutical production can not be directly discharged, because they determine aquatic environment contamination, due to their complex composition. The main organic matter removal technologies are compared and presented in Table 1.

The pharmaceutical industrial effluents are generally treated by aerobic processes, which are cost intensive in nature.

The presence of pharmaceuticals and their metabolites in surface waters and aquatic sediment was the subject of numerous studies about pharmaceuticals in the environment [6]. Results of these studies indicated that wastewater treatment plants are not enough efficient in order to remove these micropollutants from wastewaters, so they find their pass to the environment. Once entered in the environment, pharmaceutically active compounds can produce subtle effects on aquatic and terrestrial organisms, especially on the former one since they are subjected to long term exposure by wastewater effluents [7].

Bioactivators are widely used in wastewater treatment to improve settling and wastewater quality, to reduce the organic matter content, to supply influent with micro-elements, to bring specific exogenous enzymes or bacteria [8]. In the present study comercially available bioactivators *Micropan complex* and *Eparcyl pro* were used. *Micropan complex* is a mixture of bacteria (micro-organisms selected in a process of controlled fermentation with prevalence of *Bacillus spp.*,) and enzymes (cellulases, lipase, pancreatic, proteases, α-Amylolytic enzymes, β-Amylolytic enzymes, Hemicellulase, pectinase and beta-glucanase) ideal for optimization the biological degradation of organic carbon substrate present in the wastewater [9].

Eparcyl pro is a mineral additive with a large specific surface area ($68 m^2$ / g), which allows the positioning of an important number of bacteria on the carrier and thus their exponential multiplication. It contains minerals and trace elements for stimulating the activity of bacteria, and also clays embedded in micro-cavities specially designed to serve as a "nest" to the bacteria, which contributes significantly to their development [10].

The present work is a part of the current concerns in removal technologies field focused on finding viable methods for pharmaceutical industry wastewater treatment. On this line, the paper aimed to evaluate the possibility of using bioactivators to improve the removal of organic matter content and to find which is the most efficient commercially available bioactivator between *Micropan complex* and *Eparcyl pro* for the treatment of the wastewaters resulting from pharmaceutical industry. During the experimental studies, the influence of the main operational parameters (pH, amount of bioactivator, bioactivator type) on treatment process efficiency was investigated.

Table 1.

Review of the main organic matter removal technologies [3]

Type of treatment	Results	Remarks
Physicochemical treatment	30-50% COD reduction; 80-95% removal after combination of physicochemical technologies	Technologies include centrifugation, filtration, coagulation-flocculation, adsorption
Anaerobic digestion	60-80% COD removal for 2-5 days HRTs[*a]; up to 90% COD removal with long (25 days) HRTs or on selected supported media	Dilution, alkalinity adjustement and nutrients addition required
Anaerobic digestion after physicochemical pretreatment	50-70% increase in COD reduction with maximum removal (95%); over 90% phenol removal	Pretreatment techologies used: filtration, coagulation, GAC[b] adsorption, ozonization
Anaerobic digestion after aerobic pretreatment	40-60% COD reduction during pretreatment; 60-90% phenol reduction and toxicity reduction	Pretreatment with selected strains of aerobic microorganisms
Aerobic treatment	58-74% COD reduction depending on OLR[c] and HRT; 81-84% for longer HRTs	Technologies include activated sludge and constructed wetlands
Codigestion (anaerobic digestion with other wastewaters streams)	75-90% COD removal depending on dilution and post treatment	Biological treatment of OMW[d] with other wastes (pig manure, sewage sludge, domestic sewage, abbatoir waste)
Oxidation and advanced oxidation processes	40-60% COD removal under regular oxidation conditions; 70-99% COD removal under: oxidant excess, supercritical conditions, or after pretreatment	Processes include oxidation with ozone/H_2O_2, UV/ H_2O_2, wet air, Fenton oxidation, electrochemical oxidation
Combined processes	80-99% COD removal	Combinations of oxidation/biological processes, membrane processes
Composting	Compost with reasonable degrees of humification and Germination Index	Co-composting with sewage sludge and other agro-industrial wastes

[*a] HRT= hydraulic retention times
[*b] GAC= granular activated carbon
[*c] OLR= organic loading rates
[*d] OMW= olive mill wastewaters

2. Experimental

2.1. Materials and methods

The experimental research was done on a synthetic wastewater with characteristics similar to those of real waters discharged from the pharmaceutical industry (COD = 1000 mgO_2/L, pH = 8, turbidity = 298 UTF at 420 nm).

Micropan complex and *Eparcyl pro* used in this study were purchased from Eurovix US, respectively Eparcyl Romania and used as received without any special treatment or nutrient adding.

Micropan complex is a light brown powder with 4% moisture content, acting in the pH range 6-7 with the composition presented in Table 2.

Table 2.

***Micropan complex* bioactivator composition [9]**

Components	Characteristics
Selected microorganisms	Fucus-laminariae active principles
Enzymatic components	AGAR broth medium
Vegetal extracts	Lihothamnium and calcereum seaweed
Amino acids and oligopeptides	Mineral salts mordenite and dolomite
Carbohydrates	Mineral biocatalysts rich in trace elements
Growth natural factors	-

The main characteristics of *Eparcyl pro* bioactivator are shown in Table 3.

Table 3.

***Eparcyl pro* bioactivator composition [10]**

Parameter	Characteristics
Composition	A mixture of clay and inorganic salts, copper and zinc powder
Granulometry	70%< 100 µm, 50%<10 µm
Specific area	68 m^2 / g
Cationic exchange capacity	31meq [*a]/100g
Color	light brown
Appereance	powder
pH	7.34

[*a] meq= miliequivalent

2.2. Equipment

The pH was established using a Consort C380 pH meter. The samples were stirred using a Velp shaker.

2.3. Working procedure

In order to evaluate the process efficiency, the samples were taken after 2, 4, 6, 8, 10 and 12 hours of contact and analyzed.

The samples were filtered on white ribbon filter paper and then analyzed according to the method presented in STAS 9887-74 for COD determination. The COD value in the samples was calculated using the formula below [11] :

$$COD = \frac{[(V + V_1) - V_2] \cdot 0.316 \cdot 1000}{100} = [(V + V_1) - V_2] \times 3.16 [mg / L] \qquad (1)$$

where:

V- is the volume of 0.01N potassium permanganate solution initially added in mL;

V_1 - is the volume of 0.01N potassium permanganate solution used for titration in mL;

V_2 – is the volume of 0.01N oxalic acid solution added in the sample, in mL; 0.316 – the potassium permanganate quantity in mg, corresponding to 1mL of 0.01N potassium permanganate solution.

The process efficiency (R) was calculated using the following formula (2):

$$R(\%) = \frac{COD_i - COD}{COD_i} \cdot 100 \qquad (2)$$

where COD_i = initial COD (before treatment)

In the case of *Micropan complex,* the water samples placed in Erlenmayer vessels were maintained under continous stirring only after adding the bioactivator. Stirring is important because it supports the microbial degrading activity by means of biosurfactants and rhamnolipids production [16].

The experiments done using *Eparcy pro* were performed in Erlenmayer vessels provided with glass stopper and covered with aluminum foil in order to provide anaerobic conditions proper for bioactivator action. All experiments were made in duplicate at constant temperature, 25± 2°C, and the mean result was given.

3. Results and discussion

The main objective of the paper was to reduce COD below the limit imposed by the legislation in force for the treated water which will be discharged into the environment, using two bioactivators.

During the experimental procedure, several operational parameters (Table 4) with potential influence on the treatment process efficiency were investigated.

Table 4

Main operational parameters

Parameter	Value
pH of the intial solution	4, 6, 8
contact time between phases (h)	1-12
bioactivator concentration (g/L)	2, 4, 6
bioactivator type	*Micropan complex* *Eparcyl pro*

The pH variation in a wide range represents a characteristic of the wastewaters generated from pharmaceutical, diary and textile industry.

The initial pH of the synthetic wastewater was adjusted to 4, respectively 6, using 1N H_2SO_4 solution, and to 8, using 1N NaOH solution.

3.1. Influence of the bioactivator concentration on process efficiency

Preliminary investigations were made in order to establish the optimal variation domain for the bioactivator concentration, which is an important operational parameter determining the efficiency of the bioactivator for an initial pollutant.

The data obtained using *Micropan complex* and *Eparcyl pro* are shown in figure 1 and 2, respectivelly.

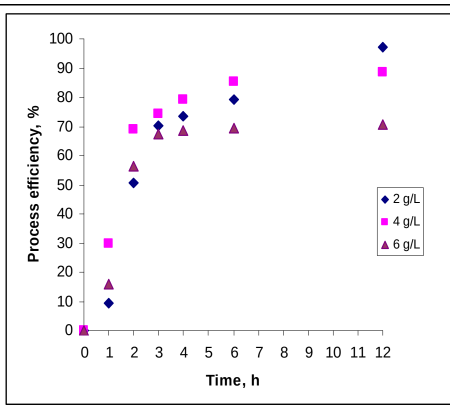

Fig. 1. *Influence of* Micropan complex *concentration on process efficiency,* COD_i *= 1000* mgO_2/L*,* $pH_i = 6$

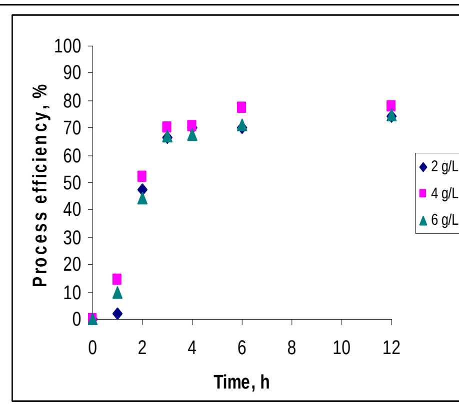

Fig. 2. *Influence of* Eparcyl pro *concentration on process efficiency,* COD_i *= 1000* mgO_2/L*,* $pH_i = 6$

From fig.1 it can be seen that after an hour of contacting the wastewater sample with *Micropan complex*, the best results were obtained for 4g/L, the efficiency being 30%. For 2g/L, respectively 6g/L the process efficiencies were

10% and 16%. Between 2-6 hours, the system behaviour was approximately the same for all concentrations. The best results were obtained for 4g/L. However, for longer times, after12 hours, the highest process efficiency (95%) was obtained for 2g/L. From fig. 2, it can be said that within 0-4 h, the results were rather different for all *Eparcyl pro* concentrations, the process efficiencies atteining 68-70% after 4 hours. Between 4-12 hours, the results were similar for all bioactivator concentrations, the process efficiency being almost 78%.

The results can be rationalized taking into account the number of adsorption sites. Normally, it is expected an increase of the process efficiency when increasing the bioactivator concentration, due to the fact that more biosorption sites are provided and, consequently, the biosorption capacity is increased. The results obtained showed that increasing the bioactivator concentration above 4g/L lead to a decrease or a stagnation of the process efficiency. This behaviour could be related to the fact that, in the case of high bioactivator concentrations, a *screening effect* to the cell wall appears, protecting the binding sites, and thus causing lower treatment efficiencies as previously reported for other systems [12,15].

3.2. Influence of initial pH value on process efficiency

The initial wastewater pH value was the most important operational parameter which influenced the bioremediation process efficiency. In general, enzymes are strongly affected by the pH value change, the impact varying from a change of enzyme function to its total inhibition. [13] Taking into account these aspects and knowing that the best results are obtained at pH value for which the enzyme is most active, we choose for the study three values, namely: 4, 6 and 8 at 4g/L bioactivator concentration (considered to be the optimum concentration).

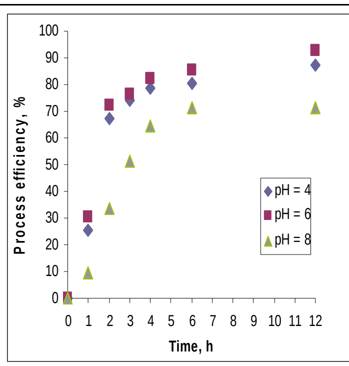

Fig. 3. Influence of pH value on process efficiency in the presence of Micropan complex (4g/L), COD_i = 1000 mgO_2/L

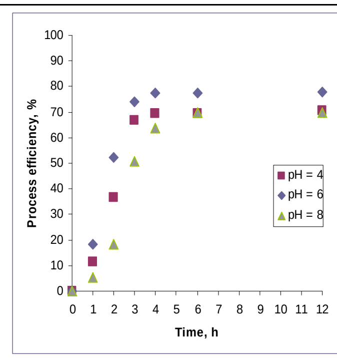

Fig. 4. Influence of pH value on process efficiency, in the presence of Eparcyl pro (4g/L) COD_i = 1000 mgO_2/L

The process efficiency evolution in time for *Micropan complex* (fig. 3) reveals a relatively similar behaviour of the system in the first 4 hours of contact at pH = 4 and pH = 6, slow variations appearing after 6 hours, the final process efficiency being achived at pH = 6 (89%). Regarding the results obtained at pH = 8, they were the lowest; after 12 hours of contact, the maximum efficiency was 72%.

From fig. 4 it can be noted that for *Eparcyl pro* after 1 hour, at pH = 6, the process efficiency was about 12%, and 6% at pH = 8 . Between 3 and 6 hours, at pH = 6, the process efficiency recorded an increase up to almost 80%,.

At pH = 4 after 6, respectively 12 hours the process efficiency, reached 70%, the same results being obtained for pH = 8.

The results can be rationalized by taking into account that the pH is a stability factor. At pH values situated in the range 2.0-6.0, the surface of the bioactivator is highly protonated, and, as a result, a strong attraction exists between organic compounds and bioactivator positively charged surface according to the previously publised studies [14].

The decrease in organic matter removal efficiency with pH increase beyond 6 may be due to the fact that, at higher pH the substrate may be negatively charged by adsorbing hydroxyl ions on the surface or by ionization of very weak acidic functional groups of the bioactivator. At lower pH (<6) the bioactivator regeneration process predominates over the removal process [15,16]. Also, it can be considered that organic matter is decomposed at low pH either by oxidation, reduction, or with the help of hydrolytic enzymes. According to the literature studies, there is no significant change of the pH value durring the treatment process, reason for which, this evolution was not followed in this research [17].

3.3. Influence of bioactivator type on process efficiency

The results obtained revealed that, the use of *Micropan complex* lead to process efficiencies superior to those obtained using *Eparcyl pro* (fig. 5) for all contact times. After 2 hours the process efficiencies were 72% and 52% with *Micropan complex* and *Eparcyl pro*, respectively. The reason for this behaviour can be related to the composition of the two bioactivators, namely: *Eparcyl pro* (mixture of clay and inorganic salts), and *Micropan complex* (natural concentrate of enzymes and bacteria). It can be concluded that the better results obtained in the case of *Micropan complex* are due to the fact that it acts as a catalyst and degradation agent for the organic matter [12,18].

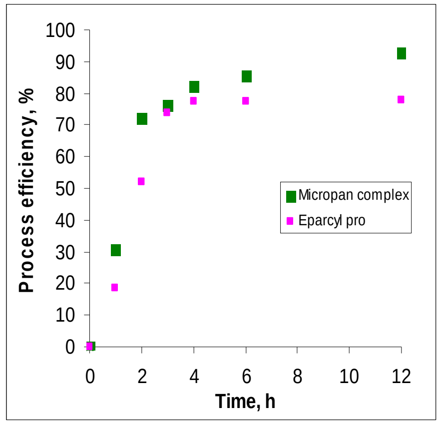

Fig. 5. Influence of bioactivator type on process efficiency,
COD_i = 1000 mgO_2/L, pH_i = 6, bioactivator concentration: 4g/L

Based on the results shown in figure 5 it can be also stated that by increasing the contacting time from 4 to 6 hours the process efficiency increased from 70% to about 85%, in the case of *Micropan complex*, respectively from 60% to 75% for *Eparcyl pro*. Therefore, in order to get good treatment efficiencies and economically viabile results, it is required to have a contact time between bioactivator and wastewater for about 4 hours.

4. Conclusions

The paper proposed a practical solution for solving the problem of wastewaters with a high content of organic substances discharged from pharmaceutical industry. It consisted in using the comercially available products *Micropan complex* and *Eparcyl pro* as bioactivators for the improvement of the pharmaceutical wastewater treatment efficiency.

The influence of the main operational parameters (pH, amount of bioactivator, bioactivator type) showed that *Micropan complex* was the best bioactivator leading to removal efficiencies of 93%, for 4g/L concentration and pH = 6. The results obtained revealed that the final effluent presents characteristics that are consistent with the drains discharge standards, NTPA 002/2002.

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REFERENCES

[1] *A.R. Miron*, *C. Modrogan, O. D. Orbuleț, C. Costache, I. Popescu*, U.P.B. Sci. Bull., Series B, **vol. 72**, 2010, p.93-100

[2] *R. Petruy, G. Lettinga*, Biores. Technol. **vol.61**, 1997, p. 141-149

[3] *P. Paraskeva, E. Diamadopoulos*, J. Chem. Technol. Biotechnol. **vol.81,** 2006, p.1475–1485

[4] *L. Loperena, M.D. Ferrari, V. Saravia, D. Murro, C. Lima, L. Ferrando, A. Fernández, C. Lareo*, Biores. Technol., **vol. 98**, 2007, p.1045-1051

[5] S. Renou, J.G. Givaudan, S. Poulain, F. Dirassouyan, P. Moulin, J. Haz. Mat., **vol. 150**, 2008, p. 468–493

[6] *P. Vazquez-Roig, R. Segarra, C. Blasco, V. Andreu, Y. Picó* , J. Chromat. A, 2010, vol. 16, p.2471-2483

[7] *R. Reif, S. Suárez, F. Omil and J. M. Lema*, Desalination, **vol. 221**, 2008, p.511-517

[8] *** http://www.tehnix.ro

[9] ***Micropan complex – technical data sheet

[10] *** http://www.eparcyl.com

[11] *C.M. Costache, C. Modorgan,. O.D. Butucea, L. Boborică*, Lucrări practice de laborator, Bucureşti, Ed. Cartea universitară, 2003, p. 106-108

[12] *S.K. Singh, A. Bansal , M.K. Jha, A. Dey*., Bioresour. Technol., **vol.104,** 2012, p.257-265

[13] *S. Şimon (Niste), T. Rusu, A. Borlea (Tiuc),* Analele Universității „Constantin Brâncuşi”, Tg. Jiu, Seria Inginerie, **vol. 2**, 2011, p.226-231

[14] *V.M. Boddu, K. Abburib, J.L. Talbottc, E.D. Smitha and R. Haaschd, Water Res.*, **vol. 42**, 2008,p. 633-642

[15] *A. D. Costa, A. Carlos, and F. Pereira*, Braz. J. Microbiol., vol. 32, 2001, p.1-5

[16] *S. Şimon (Niste), T. Rusu, A. Borlea (Tiuc),* Analele Universității „Constantin Brâncuşi”, Seria Inginerie, **vol. 3**, Tg. Jiu, 2010, p. 523 – 531

[17] *A. K. Giri*, Removal of arsenic (III) and chromium (VI) from the water using phytoremediation and bioremediation techniques, PhD Thesis, Department of Chemistry National Institute of Technology, India, 2012

[18] *S. Şimon (Niste), T. Rusu*, Sebeş, a X-a Conferință Națională multidisciplinară cu participare internațională a ”Profesorului Dorin Pavel - fondatorul hidroenergeticii româneşti”, 2010, p. 133-138