Conformational modeling of the system pollutant/three-dimensional poly (2-hydroxyethyl methacrylate) (PHEMA) in aqueous medium: a new approach

Yasmina Houda Bendahma, Salah Hamri, Meriem Merad, Tewfik Bouchaour, Ulrich Maschke

Conformational modeling of the system pollutant/ three‑dimensional poly (2‑hydroxyethyl methacrylate) (PHEMA) in aqueous medium: a new approach

Yasmina Houda Bendahma[1] Salah Hamri[1,2] Meriem Merad[3] ·Tewfik Bouchaour[1] Ulrich Maschke[4]

Abstract

The polluted water, mixture of chemicals and dyes, discharged by various textile and paper industries, is a serious problem for the environment, peculiarly the water. Dyes are stable to light, heat and oxidizing agents and are usually biologically non-degradable, which makes them complicated environmental pollutants. To examine the removal of dye from water, a three-dimensional poly (2-hydroxyethyl methacrylate) (PHEMA) and eosin Y were used, respectively, as a dye retention support and pollutant. The study of the interaction between dye and a hydrophilic polymer networks by the conformational modeling using computer software represents the goal

of the present work. Both the swelling and ultraviolet–visible spectrophotometry studies show that the pollutant was well retained by the porous polymer network PHEMA. The conformational study of the system composed of polymer network/dye shows that the interaction of the oxygen (O) and bromine (Br) of eosin Y with the hydrogen (H) of the PHEMA depends on the polymer porosity that was varied by the cross-linking rate of the three-dimensional polymer networks. The results obtained by this work can be applied to improve the retention efficiency of such material for use in wastewater application.

✉ Salah Hamri

salah_hamri@yahoo.fr

Yasmina Houda Bendahma

phyhouda@yahoo.fr

Meriem Merad

merad_meriem@yahoo.fr

Tewfik Bouchaour

bouchaour@yahoo.fr

Ulrich Maschke

ulrich.maschke@univ-lille1.fr

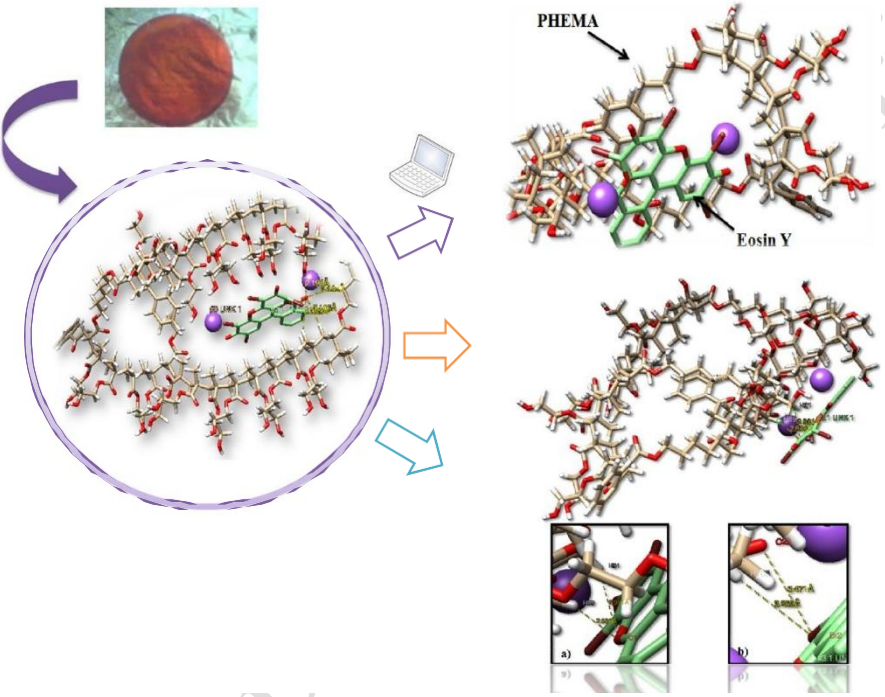
1 Laboratoire de Recherche sur les Macromolécules (LRM), Faculté des Sciences, Université Abou Bekr Belkaid, BP119, 1300 Tlemcen, Algeria

2 Centre de Recherche Scientifique et Technique en Analyses Physico-chimiques, BP 384, Zone Industrielle Bou-Ismail, 42004 Tipaza, Algeria

3 Laboratoire des Substances Naturelles et Bioactives (LASNABIO), Faculté des Sciences, Université Abou Bekr Belkaid, BP119, 1300 Tlemcen, Algeria

4 Unité Matériaux et Transformation (UMET), UMR8207(CNRS), Université de Lille 1-Sciences et Technologies, Bâtiment C6, 59655 Villeneuve d'Ascq Cedex, France

Graphical abstract



Keywords Molecular conformational · Eosin Y · Wastewater · Adsorption · Polymer networks

Introduction

Discharge disposed by various textile and paper industries is a major hazard to the environment, particularly water, due to the presence of a large number of contaminants like acids, bases, dissolved solids and dyes. These contaminated waters contain chemicals that are difficult to biodegrade, such as dye molecules that are a source of environmental degradation and affect aquatic life [1–4]. Dyes are toxic organic compounds [5]; they represent an important group of pollutants; they are used in various fields such as leather, plastic, paper industry and textile industry [6–8]. These pollution problems can be reduced by biological, chemical and physical treatment methods [9–11].

The separation of pollutants by the adsorption method has a great interest in the current research [12, 13]. This method is used in a wide range of fields, ranging from petrochemical, chemical and environmental and pharmaceutical applications [14]. Many applications of adsorption can be found, such as the retention of the components at very low concentration, for example impurities or molecules and metal ions which give color products, odors or toxicity [15]. Adsorption is one of the methods of physicochemical treatment of polluted water in which a fluid mixture containing coloring

particles was attached to the surface of a solid adsorbent [16]. The adsorption process begins with a porous diffusion of coloring molecules through the solution and filling the pores of the adsorbent material, and then the dye molecules diffuse through a diffusing layer from the surface inwardly of the adsorbent material; therefore, the dye molecules attach to the material [17]. Depending on the nature of the adsorbent and the dye, several interactions can be performed [18].

Among materials that can be used as an adsorbent, hydrogel and polymer networks, these materials were considered as porous macromolecules; they can swell in solvents and do not dissolve; they have a good application of adsorption and can be used in different fields of industry, pharmacy, medicine and environment [19–21]. Eosin Y is one of the complicated pollutant dyes for environment because of its good solubility in water. It is included in the class of xanthenes dyes and it is a heterocyclic containing bromine atoms and a single carboxyl group [22, 23]. It is widely used in biological research, thanks to its intense fluorescence; it is also used in pharmacy and textile industry in the dyeing of silk, wool, nylon. The textile industry uses this colorant for these strong coloring properties and its bright color. Eosin Y has a biological degradation once it reaches the environment and can be toxic to insects, also for aquatic

life, because of its stability [24, 25].

In the present investigation, both the elaboration of porous methacrylic polymer networks by a facile technical and the photo-polymerization by UV radiation, and retention study were the first step of this work. Effectively, 2-hydroxyethylmethacrylate (HEMA), 1,6-hexanediol diacrylate (HDDA) and 2-hydroxy-2-methyl-16-phenylpropane-1 (Darocur) were used, respectively, as a hydrophilic monomer, a cross-linked agent and a photo-initiator. The final polymeric material elaborated poly (HEMA/HDDA) was applied as an adsorption support, whereas the eosin Y dye was used as a pollutant. The examination of retention dye was followed experimentally by the study of the kinetics of swelling and deswelling of the PHEMA net- work in the eosin-Y-colored solutions. The study of the absorbance variation of the solutions colored by eosin Y before and after the swelling of the polymer network was carried out by an ultraviolet–visible (UV/Vis) spectrophotometer.

The conformational study based on a molecular modeling in the aim to simulate the behavior of a selected particle system is now present in several fields of research such as biology, pharmacy, chemistry, physics and this using different computing software [26]. The Chimera calculation software (UCSF) was used in the second

step of this work to model the previous system composed of polymer/dye (PHEMA/eosin Y) in the goal to understand how macromolecular conformational modeling can affect the retention of dye molecule by the nano- pores of the methacrylic polymer network.

Experimental part

Materials

Monomer selected for this study is 2-hydroxyethylmethacrylate (HEMA) (see Fig. 1b) exhibiting a purity of 97% (from Sigma-Aldrich). The cross-linking agent is 1,6 hexanediol diacrylate (HDDA) with a purity of 98% supplied by Cray Valley (France), and the photo-initiator is 2-hydroxy-2-methyl-16-phenyl-propane-1 (Darocur 1173) from Ciba-Geigy. The dye used is a xanthene dye, eosin Y (Sigma-Aldrich); it is water-soluble, heterocyclic acid pink in color, and the chemical structure is shown in Fig. 1a. The products were used with no modification.

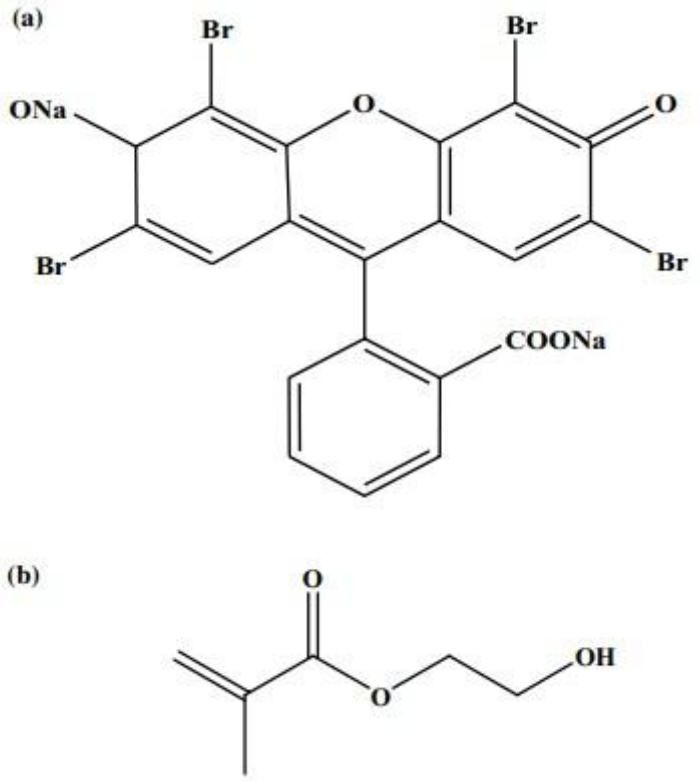


Fig. 1 Chemical structure of both **a** eosin Y and **b** 2-hydroxyethyl methacrylate (HEMA)

Sample preparation

Polymer network poly (HEMA/HDDA)

The polymer networks were prepared from a photo-reactive solution consisting of 99% HEMA monomer, 0.5% HDDA cross-linking agent and 0.5% photo-initiator. The solution was prepared and placed in a stirrer for 30 min at room temperature, and then it was placed in circular molds in the polymerization chamber exposed to a UV radiation (TL80 UV lamp) for 30 min. The reaction was in a nitrogen atmosphere. The polymer network obtained after photo-polymerization had the circular shape of the mold (pellet).

Pollutant solutions

The colored solutions were prepared with a mixture of distilled water and eosin Y. Two solutions of different concentrations of eosin Y were prepared from the initial solution. There were therefore three different concentrating solutions C_1 = 0.1 mg/mL, C_2 = 0.05 mg/mL and C_3 = 0.025 mg/mL.

Swelling and deswelling measurements

The polymer network obtained was weighted in the initial state (dry state) and then immersed in colored solutions with different concentrations of eosin Y (C_1, C_2 and C_3). The mass of the polymer network PHEMA was taken in a time interval until the maximum of its weight was obtained. The swelling kinetics of the polymer network PHEMA was therefore followed, and the swelling ratio is calculated by Eq. (1).

$$\tau = \frac{w_s - w_d}{w_d} \times 100 \quad (1)$$

where τ represents the swelling ratio (%), w_d is the weight of the dried polymer network, and w_s is the weight of the swollen polymer network at different time.

In the case of the deswelling measurements, the swollen sample was taken out of the solution and left under air atmosphere, and it was weighted in a time interval until the minimum of its weight was obtained.

Software of the conformational study

ChemDraw is the most complete tool of choice for chemists and biologists who want to create intelligent scientific drawings ready for use in electronic laboratory notebooks, publications and databases. ChemDraw Ultra version (8.0) was used in this study to draw the chemical structure of the model polymer network PHEMA/eosin Y dye.

The UCSF Chimera calculation software, version (2.3), was used with the AutoDock Vina interface to model the polymer/dye system (PHEMA/eosin Y). This software allows us to determine the most stable conformation of the chosen system by minimizing the energy of the system and then determining the existing and most important interactions between the atoms of the polymer and that of the dye.

AutoDock Vina uses a sophisticated optimization gradient method in its local optimization procedure. Its calculation parameters were [27]:

- The names of the files contain the model of polymer network and dye.
- The initial state of the model composed of polymer network/dye (random con- formation).
- Introduction of the dimension of the virtual (3-D) grid which depends

on the dimension of the model studied.

- Finally, the docking process starts and at the end of each scan cycle, AutoDock Vina records the most important interactions.

To allow the execution of the polymer network/dye calculations using the UCSF Chimera software [28], we must represent the model PHEMA/eosin Y with its polar hydrogen, as well as the partial charges of all its atoms.

Results and discussion

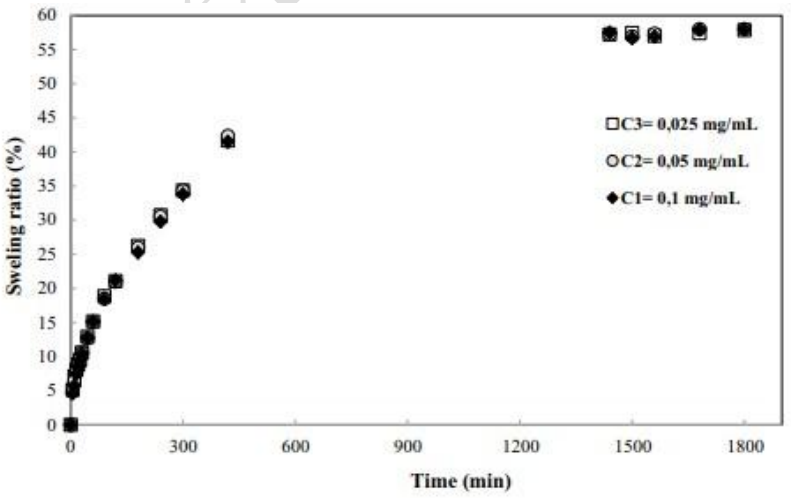
Swelling and deswelling studies

The swelling kinetics of polymer network PHEMA in colored distilled water with three different concentration (C_1 = 0.1 mg/ml, C_2 = 0.05 mg/ml and C_3 = 0.025 mg/ ml) is presented in Fig. 2. The polymer network PHEMA swelled and followed two steps: In the first step, the diffusion of colored distilled water in the polymer network followed an exponential behavior in the time interval [0–24] h. The second step is what called a tray; in this step, during 6 h, from 24 to 30 h, a constant swelling ratio was noted, because the polymer networks was saturated and could not uptake more water. At this time, we note the equilibrium swelling ratio considered as a maximum of

swelling ratio. For the three colored solutions as you can see in Fig. 2, the maximum swelling ratio was almost stable about 58%.

This is explained by the architecture of the polymer network. Forever, the chains of the polymer network are entangled in the initial state (dry state) and then the polymer network is immersed in the solutions stained by eosin Y, so there are a rapid diffusion of the colored solution and relaxation of the chains of the polymeric network; therefore, there is progressive adsorption on the surface and then progressively on the inside until the chains reach their maximum.

The deswelling kinetics of PHEMA was monitored once the polymer network reached its maximum swelling. Then, it was removed from eosin solutions and left at free air and room temperature (see Fig. 3).



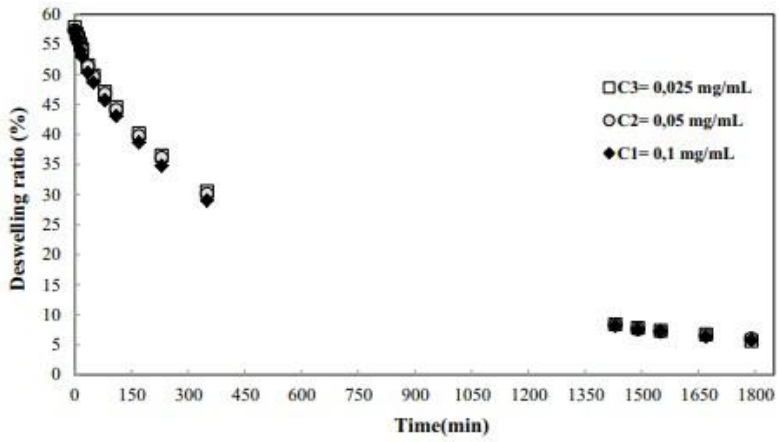


Fig. 3 Deswelling kinetics of PHEMA at free air and room temperature

The results show that the polymer network PHEMA returns almost to its initial weight after 24 h, so the PHEMA remains colored. It is concluded that the dye was adsorbed and trapped by the polymer network; which can be explained by the fact that there has been the retention of the dye molecule in the nano-pore of polymeric network.

Spectral analysis of colored solutions

The spectral analysis of the solutions colored by eosin Y was done by ultraviolet–visible spectrophotometer as shown in Fig. 4. The colored solutions were placed in the apparatus to determine the absorbance

spectra in the initial and final states and to see the variation in the absorbance with the variation of dye concentration.

The results show that the concentration of eosin Y dye for each solution (C_1, C_2 and C_3) decreases after swelling of the polymer network PHEMA. Thus, the dye was well experimentally retained by the polymeric support.

Conformational study of polymer network/dye system (PHEMA/eosin Y)

The objective of this part is to consider a molecular model of the polymer network/dye system and then to determine the most stable conformation of the chosen system, using the molecular modeling program UCSF Chimera, to simulate dye retention eosin Y, by the polymer network PHEMA.

Chimera is a highly usable program for interactive visualization and analysis of molecular structures, including supramolecular assemblages; it is widely used by biologists and biochemists to study protein/enzyme systems where proteins and enzymes are directly taken from database [29].

The principle is therefore to insert a coloring molecule eosin Y in a

simplified model of the polymer network PHEMA (Fig. 5). The program permits to calculate the minimum energy of the model and then to determine the possible interactions between the atoms of the dye and the polymer network.

The introduction of data consists of drawing a model of the polymer network and the dye molecule using the ChemDraw software which gives conforming but random structures. The Chimera program permits to choose a parallelepiped-shaped virtual cavity that may contain the network/dye model.

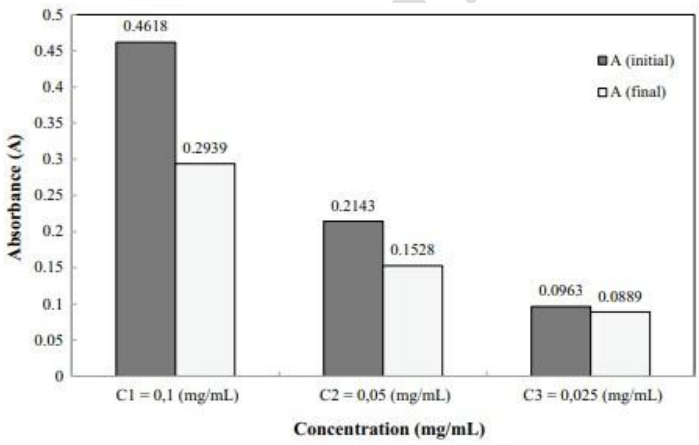


Fig. 4 Variation of absorbance of the eosin Y dye in the medium for both initial states without polymer (filled square) and after the swelling of the polymer networks PHEMA (square)

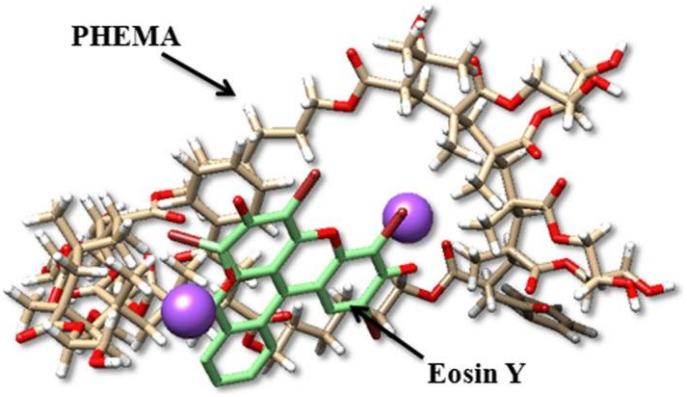


Fig. 5 Simplified model of a system composed of polymer network PHEMA and dye eosin Y

The calculation of the energy minimization gives the most stable structure of the model and therefore a deduction of the most important interactions between the atoms of the dye and those of the polymer network [30].

Figure 6 shows a model of interaction between oxygen (O_1) of eosin Y and hydrogen (H_{139}) of the polymer network PHEMA which has a strong interaction (2.839 Å), and interaction between the brome (Br_2) of the dye and the hydrogen (H_{41} and H_{43}) of the polymer network which has an average interaction (3.236 and 3.518 Å).

Another PHEMA/eosin Y system is made by adding a second

cavity to the polymer network PHEMA, so the model has with two equal cavities. The same calculation steps of the first system are followed; the possible interactions between the molecules of the dye and the polymer network are observed as shown in Fig. 7.

The results show that the molecule of dye eosin Y is directed toward the part of the polymer network PHEMA where there are more atoms and therefore more interactions.

Strong interactions were observed between the oxygen of the dye (O_1) and hydrogen of the polymer network (H_{89} and H_{91}) with values (2.636 and 2.981 Å) shown in Fig. 7a and also less strong interactions between the brome (Br_2) of the dye and hydrogen of the polymer network PHEMA (H_{106}) shown in Fig. 7b with values (3.253 Å).

In another model, a PHEMA/eosin Y system was formed from two cavities with different sizes, the same calculation steps in the first and second systems were followed, and the interactions existing between the polymer network PHEMA and the dye were observed as shown in Fig. 8.

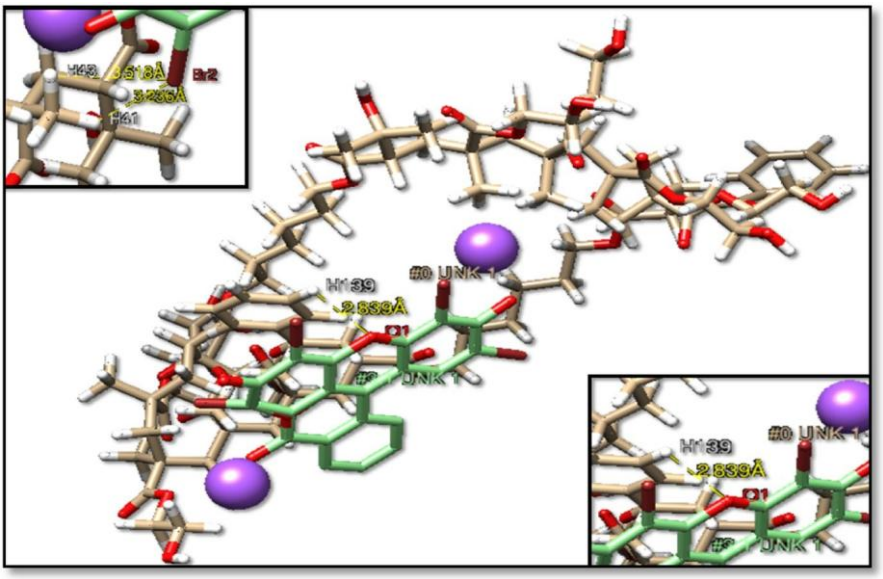


Fig. 6 Model of interactions between atoms of eosin Y dye and polymer network PHEMA; aggrandizements shows an example of interaction of oxygen (O_1) and brome (Br_2) atoms of eosin Y dye with hydrogen atoms of polymer network PHEMA

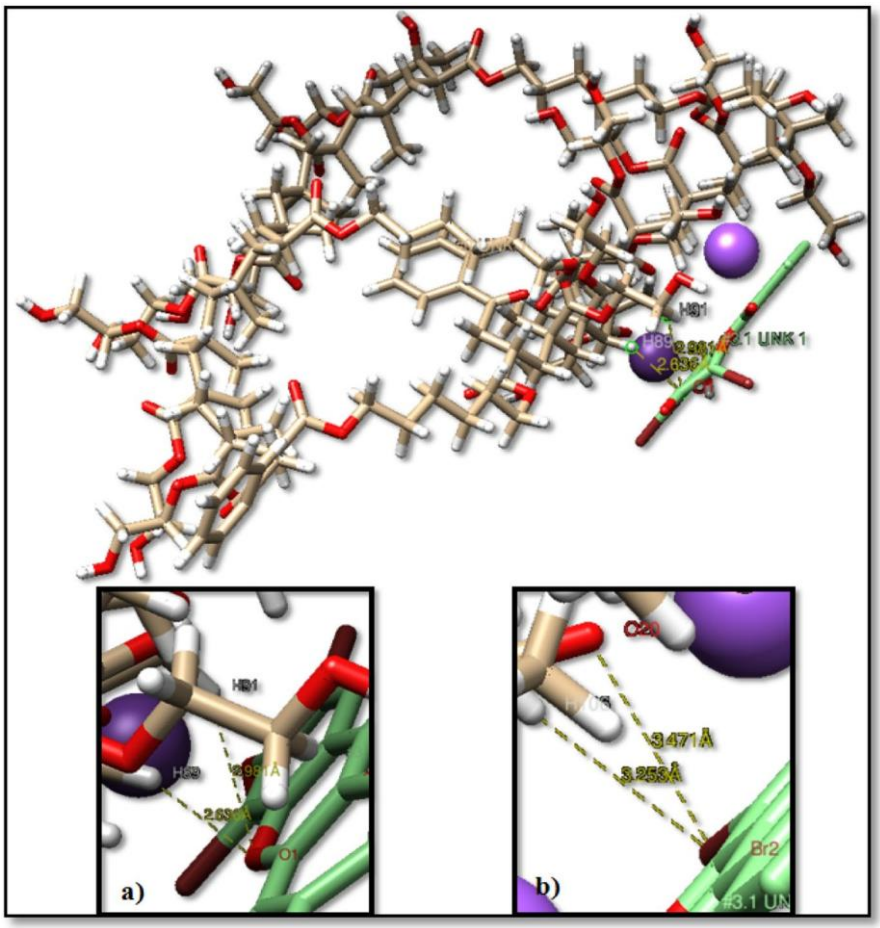


Fig. 7 Model of interactions between polymer network PHEMA with two equal cavities and eosin Y dye. Aggrandizements: **a** interaction between oxygen (O_1) atoms of eosin Y and hydrogen atoms of the PHEMA. **b** Interaction between brome (Br_2) atoms of eosin Y and hydrogen atoms of the PHEMA

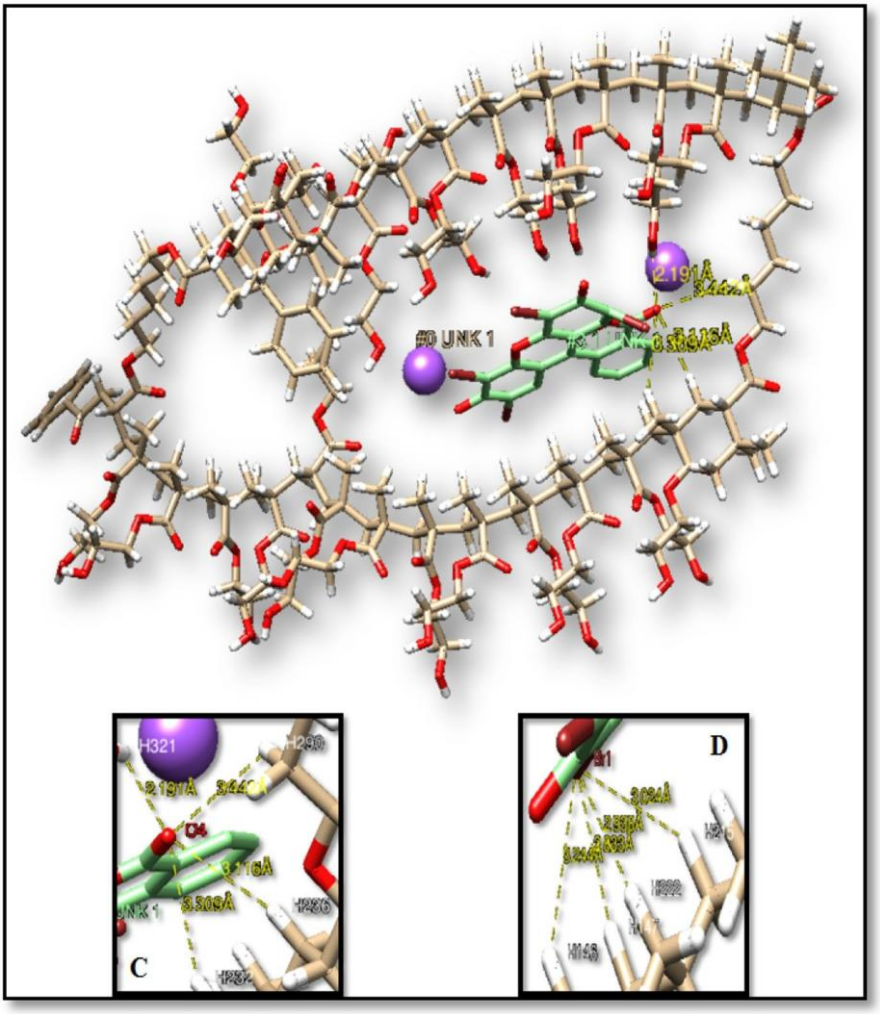


Fig. 8 Model of interactions between polymer network PHEMA with two different cavities and eosin Y dye. Aggrandizements: **c** interaction between oxygen (O_4) atoms of eosin Y and hydrogen atoms of the PHEMA. **d** Interaction between brome (Br_1) atoms of eosin Y and hydrogen atoms of the PHEMA

The results show that the polymer networks were directed toward the large cavity where there were more interactions.

Strong and medium interactions were observed between the oxygen (O_4) of eosin Y and the hydrogen of the PHEMA polymer network (H_{232}, H_{236}, H_{290} and H_{321}) with values (3.309, 3.116, 3.442 and 2.191 Å). There are also strong and medium interactions between eosin Y and hydrogen atoms of the polymer network PHEMA (H_{146}, H_{147}, H_{215} and H_{222}) with values (3.244, 2.833, 3.024 and 2.938 Å).

The interactions between the atoms of the system where the distances between 2.5 and 3.1 Å were considered as strong interactions, those between 3.1 and 3.55 Å were classified as a medium interactions ,and a very weak or absent interactions for distances greater than 3.55 Å [31].

Conclusion

The hydrophilic methacrylic polymer network poly (HEMA/HDDA) elaborated by UV photo-polymerization was used as a retention support of the dye, whereas the eosin Y was considered as a pollutant. Both swelling and deswelling kinetics show that the polymer networks became colored and eosin Y was well encapsulated inside

the polymeric support. The decrease in the absorbance from the initial to the final polluted solutions justified that the hydrogel polymer network PHEMA has retained an amount of the eosin Y dye molecule.

The conformational study of the polymer network/dye system (PHEMA/eosin Y) shows the presence of strong, medium and weak interactions in the selected system. In the case of one cavity in the polymer network PHEMA, the oxygen and brome of the eosin Y dye have interactions with the hydrogen atoms of PHEMA. When the polymer network has two cavities of the same size, the dye is directed to the part where there are more interactions; the oxygen and brome of the dye interact with the hydrogen of the polymer networks. For the last system where the selected polymer network has two different cavities in sizes, the dye is directed toward the part where there are more interactions (the large cavity); the oxygen atoms and the bromine of the dye interact with the hydrogen atoms of the polymer network PHEMA. So for the three chosen systems the dye takes the most stable conformation with the network and goes toward the part where there are more interactions.

The distance between atoms has an important effect on the interactions of the system, and it is found that the interactions can be

classified as strong and medium interactions for a distance interval [2.5, 3.1] and [3.1, 3.55] Å, respectively, whereas there are very weak or absent interactions for distances between selected atoms greater than 3.55 Å.

Acknowledgements I thank very much Pr. Tewfik Bouchaour for his support to accomplish this work and also all authors who participated in this investigation.

References

1. George ZK, Dimitrios NB, Athanasios CM (2017) Chitosan adsorbents for dye removal: a review. Polym Int 66:1800–1811. https://doi.org/10.1002/pi.5467
2. İnal M, Erduran N (2015) Removal of various anionic dyes using sodium alginate/poly(*N*-vinyl- 2-pyrrolidone) blend hydrogel beads. Polym Bull 72:1735–1752. https://doi.org/10.1007/s0028 9-015-1367-7
3. Aderonke AA, Idowu OA, Olugbenga SB (2017) Adsorption of dyes using different types of clay: a review. Appl Water Sci 7:543–568. https://doi.org/10.1007/s13201-015-0322-y
4. Simphiwe PB, Ademola OO, Balakrishna P (2012) Textile dye removal from wastewater effluents using bioflocculants produced by indigenous bacterial isolates. Molecules 17:14260–14274. https:// doi.org/10.3390/molecules171214260
5. Ramaraju B, Reddy PMK, Subrahmanyam C (2013) Low cost absorbent from agricultural waste for removal of dyes. Environ Prog Sustain Energy 33:38–46. https://doi.org/10.1002/ep.11742
6. Vieira SS, Magriotis ZM, Santos NAV, Cardoso MG, Saczk AA (2012) Macauba palm (Acrocomia aculeata) cake from biodiesel

processing. An efficient and low cost substrate for the adsorption of dye. Chem Eng J 183:152–161. https://doi.org/10.1016/j.cej.2011.12.047

7. Hamri S, Bouchaour T, Maschke U (2014) Erythrosine/triethanolamine system to elaborate crosslinked poly(2-hydroxyethylmethacrylate): UV-photopolymerization and swelling studies. Mac- romol Symp 336:75–81. https://doi.org/10.1002/masy.201300018
8. Fleischmann C, Lievenbrück M, Ritter H (2015) Polymers and dyes: developments and applications. Polymers 7:717–746. https://doi.org/10.3390/polym7040717
9. Tobiason JE, Bazilio A, Goodwill J, Mai X, Nguyen C (2016) Manganese removal from drinking water sources. Curr Pollution Rep 2:168–177. https://doi.org/10.1007/s40726-016-0036-2
10. Ravi VK, Narayana SKV, Khasim BS, Gopinath R (2016) Dye removal by adsorption: a review. J Bioremediat Biodegrad 7:371. https://doi.org/10.4172/2155-6199.1000371
11. Elhami S, Abrishamkar M, Esmaeilzadeh L (2013) Preparation and Characterization of diethylen- triamine-montmorillonite and its application for removel of eosinY dye: optimization, kinetic and isotherm studies. J Sci Ind Res 72:461–466

12. Sasaki T, Lizuka A, Watanabe M, Hongo T, Yamasaki A (2014) Preparation and performance of arsenate (V) adsorbents derived from concrete wastes. Waste Manag 34:1829–1835. https://doi. org/10.1016/j.wasman.2014.01.001
13. Debora LP, Carla AD, Francielle Z, Danielle CCM, Clovis AR (2016) Adsorption of rhodamine B and methylene blue dyes using waste of seeds of *Aleurites Moluccana*, a low cost adsorbent. A E J 55:1713–1723. https://doi.org/10.1016/j.aej.2016.03.017
14. Ebrahimi A, Arami M, Bahrami H, Pajootan E (2013) Fish Bone as a low-cost adsorbent for dye removal from wastewater: response surface methodology and classical method. Environ Model Assess 18:661–670. https://doi.org/10.1007/s10666-013-9369-z
15. Kamran R, Rouhollah M, Ahmad A, Nourollah M (2018) Preparation of nanoparticle-modified polymeric adsorbent using wastage fuzzes of mechanized carpet and its application in dye removal from aqueous solution. J Clean Prod 178:373–383. https://doi.org/10.1016/j.jclepro.2017.12.213
16. Kokkarachedu V, Tippabattini J, Emmanuel RS (2017) Removal of dye by carboxymethyl cellu- lose, acrylamide and graphene oxide via a free radical polymerization process. Carbohydr Polym 164:186–194. https://doi.org/10.1016/j.carbpol.2017.01.094

17. Nidheesh PV, Minghua Z, Mehmet AO (2018) An overview on the removal of synthetic dyes from water by electrochemical advanced oxidation processes. Chemosphere 197:210–227. https://doi.org/10.1016/j.chemosphere.2017.12.195
18. Fan H, Zhanghui W, Baoku Z, Liping Z, Youyi X (2018) Poly (N-vinyl imidazole) Gel-filled membrane adsorbers for highly efficient removal of dyes from water. J Chromatogr A. https://doi.org/10.1016/j.chroma.2018.05.075
19. Hamri S, Bouchaour T (2016) pH-dependent swelling behaviour of interpenetrating polymer net- work hydrogels based on poly(hydroxybutyl methacrylate) and poly(2-hydroxyethyl methacrylate). Int J Plast Technol 20:279–293. https://doi.org/10.1007/s12588-016-9155-2
20. Boudraa K, Bouchaour T, Maschke U (2008) Swelling of acrylic interpenetrating polymer networks in liquid crystals. Macromol Symp 273:33–37. https://doi.org/10.1002/masy.200851305
21. Dali-Youcef B, Bouchaour T, Bouberka Z, Bigan M, Maschke U (2017) Swelling behavior of poly(n-butyl acrylate/1,6-hexane-diol-di-acrylate)/nematic liquid crystal E7 systems: experimental measurements and modeling by factorial design method. J Appl Polym Sci 134:45230–45236. https://doi.org/10.1002/app.45230

22. Ernesto MA, Carlos MP, Sonia GB (2016) Photochemical study of Eosin-Y with PAMAM dendrim- ers in aqueous solution. J Lumin 180:369–375. https://doi.org/10.1016/j.jlumin.2016.08.017

23. Fan Z, Feng S, Wei M, Fei G, Yang J, Hui L, Jingchuan W, Xinyan S, Xinghua L, Sheng M (2013) Controlling adsorption structure of eosin Y dye on nanocrystalline TiO2 films for improved photo- voltaic performances. J Phys Chem C 117:14659–14666. https://doi.org/10.1021/jp404439p

24. Leonardo GF, Ricardo NDS, Glaucius O, Adriano DA (2015) Molecular docking and structure- based drug design strategie. Molecules 20:13384–13421. https://doi.org/10.3390/molecules200713 384

25. Sandhya G, Diptisoni Rakshitameta, Surbhi B (2015) Use of barium chromate in photocatalytic degradation of eosin yellow. Chem Sci Trans 4:851–857. https://doi.org/10.7598/cst2015.1069

26. Emmanuel OO, Johannes AMA, Sylvester KT (2017) Equilibrium, kinetic and thermodynamic study of removal of eosin yellow from aqueous solution using teak leaf litter powder. Scientific Reports 7:12198. https://doi.org/10.1038/s41598-017-12424-1

27. Pettersen EF, Goddard TD, Huang CC, Couch GS, Greenblatt DM, Meng EC, Ferrin TE (2004) UCSF Chimera–a visualization system

for exploratory research and analysis. J Comput Chem 25:1605–1612. https://doi.org/10.1002/jcc.20084

28. Li-Hua M, Bo W, Hao-Yang R, Ping L, Dai-Hong G, Fu-Meng W, Lin B, Yan-Shen G (2012) Syn- thesis and inhibitory effect of piperine derivates on monoamine oxidase. Bioorg Med Chem Lett 22:3343–3348. https://doi.org/10.1016/j.bmcl.2012.02.090
29. Miller FP, Vandome AF, Brewster JM (2010) Gebhart Factors. VDM Publishing House Ltd, Eng- land. ISBN: 6134096326
30. Ayachi H, Merad M, Ghalem S (2013) Study of interaction between dipeptidyl peptidase-4 and prod- ucts extracted from the Stevia plant by molecular modeling. Int J Pharm Sci Rev Res 23(1):87–90
31. Soufi W, Merad M, Boukli F, Ghalem S (2011) the Complementarity Effect for Cdc25 Phosphatase Inhibitors. Adv Mol Imag 1:17–23. https://doi.org/10.4236/ami.2011.12003