NEW FRONT. CHEM. (2021)
Volume 30, Number 1, pp. 25-34
ISSN 2668-9189; ISSN-L 2393-2171

Former: Ann. West Univ. Timisoara – Series Chem.
ISSN: 1224-9513

Article

PREDICT OF WATER SOLUBILITY OF THE LOW MOLECULAR WEIGH OLIGOMERS OF POLYHYDROXYALKANOATES

Alecu Ciorsac [(1)], Mădălina Filip[(2)], Adriana Isvoran[(2)] (♣)

[1] Department of Physical Education and Sport, University Politehnica Timișoara, Timișoara, Romania.
[2]Department of Biology-Chemistry and Advanced Environmental Research Laboratories, West University of Timișoara, Timișoara, Romania.

ABSTRACT

When polyhydroxyalkanoates (PHAs) are used in biomedical applications, the release of their oligomers (OHAs) occurs in the human body due to the degradation of polymers. Within this study, predictions regarding the aqueous solubility of low molecular weight oligomers of PHAs (up to 32 monomeric units) are obtained using three models: ESOL, Ali and SILICOS-IT. The investigated oligomers consist of 3-hydroxybutyrate, 4-hydroxybutyrate, 3-hydroxyvalerate, 4-hydroxyvalerate, and 3-hydroxybutyrate-co-3-hydroxyvalerate. The results illustrate the linear decrease of the aqueous solubility of OHAs with both molecular weight and partition coefficient and that the Ali model proved to be the best when predicting the aqueous solubility of these oligomers. Co-oligomers containing hydroxybuyrate and hydroxyvalerate units reflect higher molar solubility than the corresponding oligomers. It underlines the importance of the composition of PHAs designed for medical use, co-polymers seem to be more advantageous.

Keywords: oligomers; hydroxyalkanoates; molar solubility.

1. INTRODUCTION

Biocompatible and biodegradable polymers have proven to be used more and more for a wide range of products, including pharmaceutical, medical and food packaging products. Polylactic acid (PLA), polyhydroxyalkanoates (PHAs) and chitosan (CHO) are among the biopolymers that are considered promising for numerous applications. Understanding and

♣ Correspondent author: Tel:+4025592624, Fax:+40256592620, E-mail: adriana.isvoran@e-uvt.ro

controlling the properties of these polymers and those of their degradation products are important for their successful use.

Polyhydroxyalkanoates are polymers that are produced by various microorganisms, serving as intracellular carbon compounds with energy storage properties [1]. More than 150 different monomers are known that can be successfully combined within this family to provide materials with distinct properties and possessing certain characteristics such as biocompatibility, biodegradability and non-toxicity [2, 3]. These properties and the different forms of PHAs make them suitable polymers for numerous biomedical applications: drug transport systems in the body [4], implants for bone and cartilage regeneration and anticancer agents [5], therapeutic carrier, vascular grafting, cardiac, skin, liver, nerve and other tissues engineering [6]. US Food and Drug Administration (FDA) approved in 2007 the poly-4-hydroxybutyrate (P4HB) to be used in surgical sutures [7]. Other derivatives having a major potential in biomedical uses include: poly-3-hydroxybutyrate (P3HB), poly-3-hydroxyvalerate (P3HV), poly-4-hydroxybutirate (P4HB), poly-4-hydroxyvalerate (P4HV) and their copolymers represented by 3-hydroxybutyrate (3HB) and 3-hydroxyvalerate (3HV) [8].

When resorbable polymers are used in biomedical applications, the release of oligomers occurs in the human body due to the combined degradation (hydrolytic and enzymatic) of polymers. In the case of PHAs implants or PHAs drug delivery systems, hydroxyalkanoate oligomers (OHAs) and / or monomers are released [9]. The rate of degradation may be variable being strongly influenced by the size and composition of the PHAs materials [9]. There is little information on the interactions of OHAs with the human body. In a previous computational study, these oligomers were predicted as not revealing carcinogenicity, mutagenicity, cardiotoxicity and hepatotoxicity, were not considered as substrates and inhibitors of the cytochromes involved in the metabolism of drugs [10]. They may be inhibitors of the organic anion transporters OATP1B1 and OATP1B3, may produce eye and skin irritation and may have a low antagonistic effect on the androgen receptor [10]. Favourable pharmacokinetics profiles have been also revealed for derivatives and degradation products of other biopolymers: low molecular weight water soluble chitosan derivatives [11], chito-oligomers [12], and oligomers of lactic acid [13].

Information presented in the literature has shown that the chemical properties of monomer units and their distribution in the polymers play an important role in terms of the properties of the resulting material and in influencing the biological activities [10, 12, 14]. The molecular properties of the oligomers resulting by polymers degradation impact on their ADMET (absorption, distribution, metabolism, excretion, toxicity) profiles [10, 12, 13]. Furthermore, the interactions of chito-oligomers with plasma proteins proved to be dependent on the deacetylation degree and pattern [15]. All these data emphasize the importance of the composition of biopolymers used for medical applications.

An important property of a substance is its solubility, especially for drugs and drug adjuvants [16]. A good solubility strongly influences the oral bioavailability, the biological activity of the molecule and it is well known that water-soluble compounds are easily excreted [17]. Water solubility also determines the environmental fate of chemical substances. A higher water solubility increases the absorption by aquatic species, absorption in soils and sediments and the degradation by photolysis or hydrolysis [18] . PHAs are not water soluble, but the hydrolysis of polymer results into water-soluble oligomers and monomers [19]. In the environment, OHAs are easily degraded via enzymatic processes. In

the human body, OHAs, as soluble degradation products, may be either metabolized or excreted [20].

To the best of our knowledge, the information regarding the molar solubility of monomers and small oligomers of hydroxyalkanoates is missing. Consequently, within this study we consider low molecular weight oligomers containing various types of monomeric acids and compute their water-solubility by using three different methods in order to assess the best model to be applied for predicting the OHAs solubility

2. METHOD

This study focuses on low molecular weight OHAs containing maximum 32 monomeric units of: 3-hydroxybutyrate (O3HB), 3-hydroxyvalerate (O3HV), 4 hydroxybutyrate (O4HB), 4-hydroxyvalerate (O4HV)). Co-oligomers of 3-hydroxybutyrate and 3-hydroxyvalerate (O(3HB3HV)) with 2, 3 and 4 different monomer units and different patterns (position of 3HB and 3HV monomers in the chain) are also considered (Table 1). The monomeric units of these oligomers are presented in Figure 1 together with their IUPAC (International Union of Pure and Applied Chemistry) nomenclature.

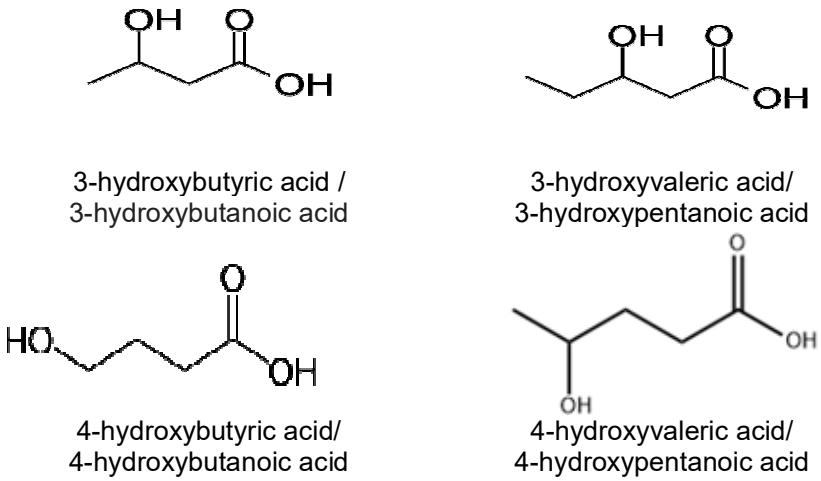
Table 1. Oligomers of hydroxyalkanoates considered in this study: O3HB – oligomers of 3-hydroxybutyrate, O3HV - oligomers of 3-hydroxyvalerate, O4HB - oligomers of 4-hydroxybutyrate, O4HV - oligomers of 4-hydroxyvalerate, O(3HB3HV) - co-oligomers of 3-hydroxybutyrate and 3-hydroxyvalerate.

O3HB	O3HV	O4HB	O4HV	O(3HB3HV)
O3HB 1u-O3HB 32 u	O3HV 1u-O3HV 32 u	O4HB 1u - O4HB 32 u	O4HV 1u - O4HV 32 u	O3HVB, O3HBV, O3HVBV, O3HBVB, O3HVBVB, O3HBVBV

The SMILES (Simplifies Molecular Input Line Entry System) formulas of the OHAs were obtained using ACD / ChemSketch (https://chemicalize.com, accessed in April 2021) [21]. SMILES formulas were used to calculate the molecular weight (MW) and the partition coefficient (logP) and to predict the aqueous solubility for every of these oligomers. Both computations of the molecular properties and of the aqueous solubility were done using the SwissADME computational tool [22]. In the case of logP, the consensus values provided by SwissADME tool are considered. SwissADME uses three calculation methods to estimate the solubility: the ESOL method [23], the Ali method [24], and SILICOS-IT method (a method based on a system of 16 fragmentary contributions modulated by the square root of the molecular weight) [25]. These methods are shortly presented in the following.

Estimating SOLubility (ESOL) method allows to estimate the solubility in water starting directly from the structure of the compound, being used on a large scale in the field of computer science [23].

Figure 1. Structural formulas of the monomers of hydroxyalkanoates: common and IUPAC names



The equation based on which the water solubility is calculated in the case of the ESOL method is

$$\log S = 0.16 - 0.63C \log P - 0.0062MW + 0.066RB - 0.74TPSA \quad (1)$$

where MW is the molecular weight, RB represents the number of rotatable bonds and TPSA is the topological polar surface area, these being the molecular properties of the investigated compound [23].

Ali method for calculating the solubility of chemical compounds uses the equation

$$\log S = -1.0377 \log P - 0.021TPSA + 0.4488 \quad (2)$$

that takes into account the topological polar surface area (TPSA) and the partition coefficient (logP) as molecular descriptors [24].

SILICOS-IT method performs the filtering of molecules with undesired properties through a filtering program (Filter-IT) holding a series of pre-programmed molecular properties that can be used for filtering. The equation used in this case for the calculation of logS is a polynomial equation of order 3,

$$\log S = 0.898 + 0.104\sqrt{MW} + wi.ci \quad (3)$$

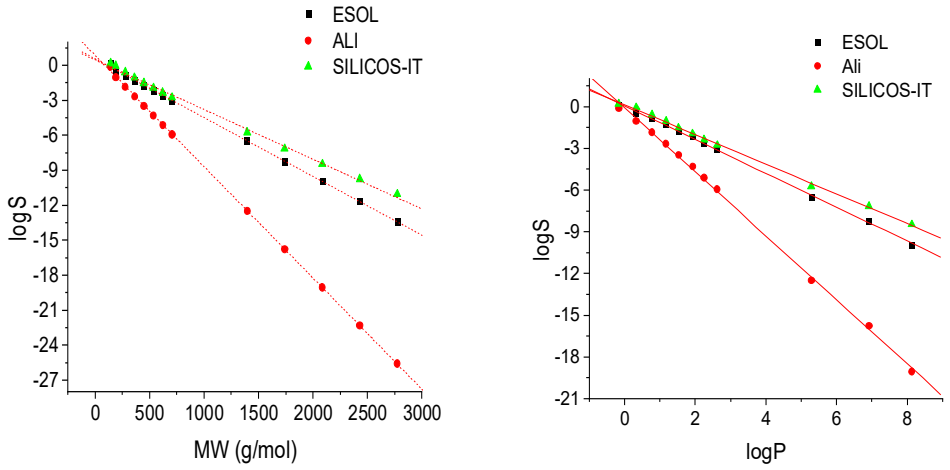
which gives the variation logS with the molecular weight (MW) and where wi and ci are the respective weights and counts for the fragment i (http://silicos-it.be.s3-website-eu-west-1.amazonaws.com/software/filter-it/1.0.2/filter-it.html, accessed in September 2021) [25].
Data obtained using these equations are compared in order to assess which is the best model to be used for predicting the solubility of OHAs.

3. Results and Discussions

Using the SwissADME tool we computed the molecular weight (MW), partition coefficients (logP) and aqueous solubility logS (with S expressed in mg/mL) values for the different types of OHAs. The molecular properties for the investigated OHAs are presented

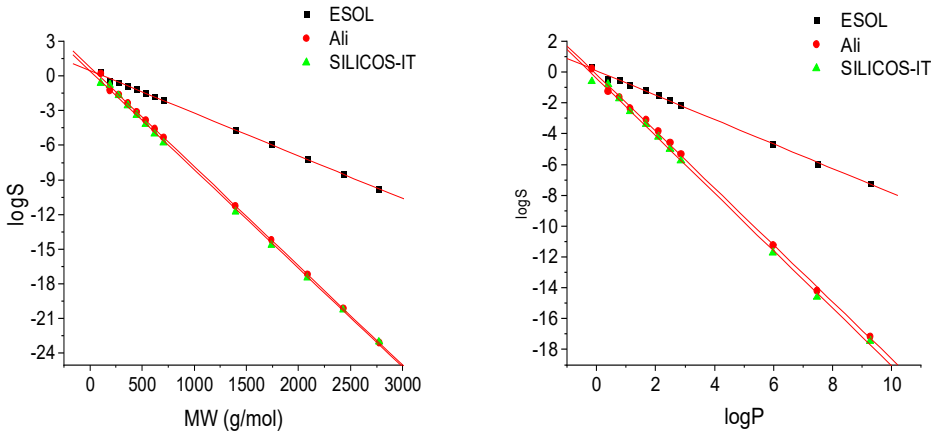
elsewhere [10]. The dependence of the logS values on the molecular weight (MW) and respectively on the partition coefficient (logP) for various types of OHAs are shown in Figures 2-6.

Figure 2. Dependence of logS values on MW (a) and logP (b) values for oligomers of 3-hydroxybutyrate (O3HB)



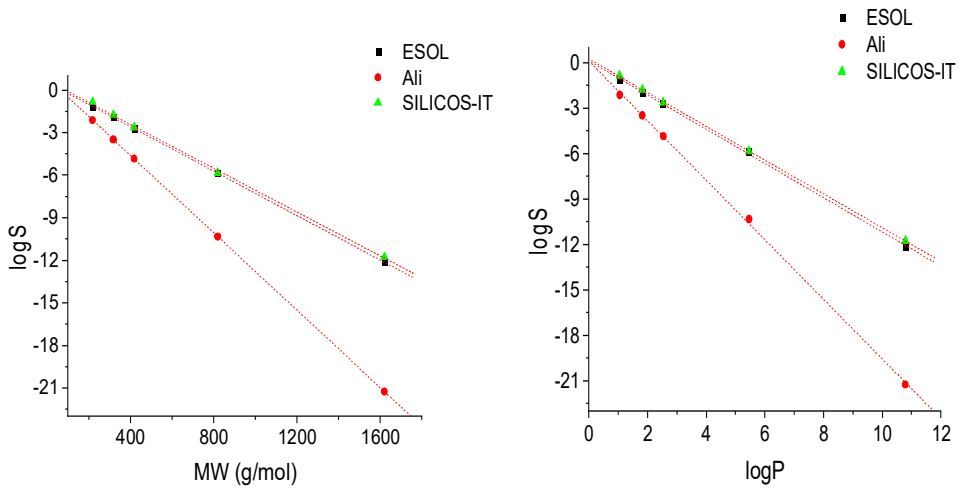
a b

Figure 3. Dependence of logS values on MW (a) and logP (b) values for oligomers of 4-hydroxybutyrate (O4HB)



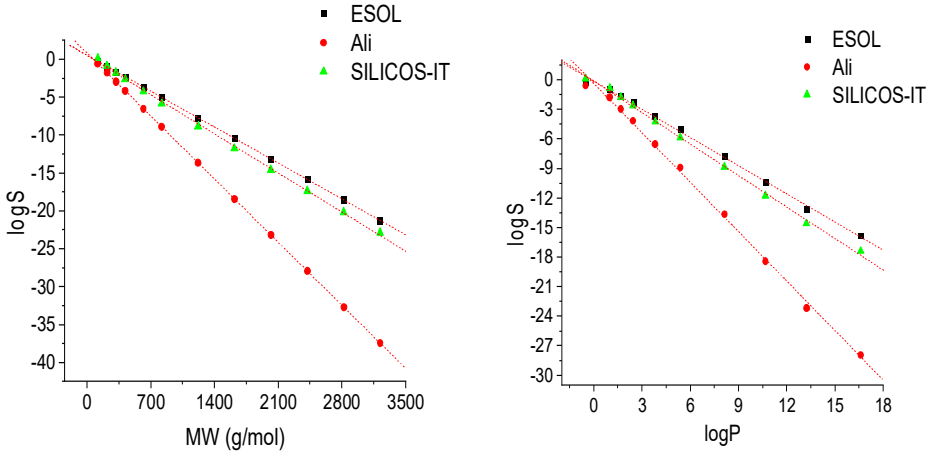
a b

Figure 4. Dependence of logS values on MW (a) and logP (b) values for oligomers of 3-hydroxyvalerate (O3HV)



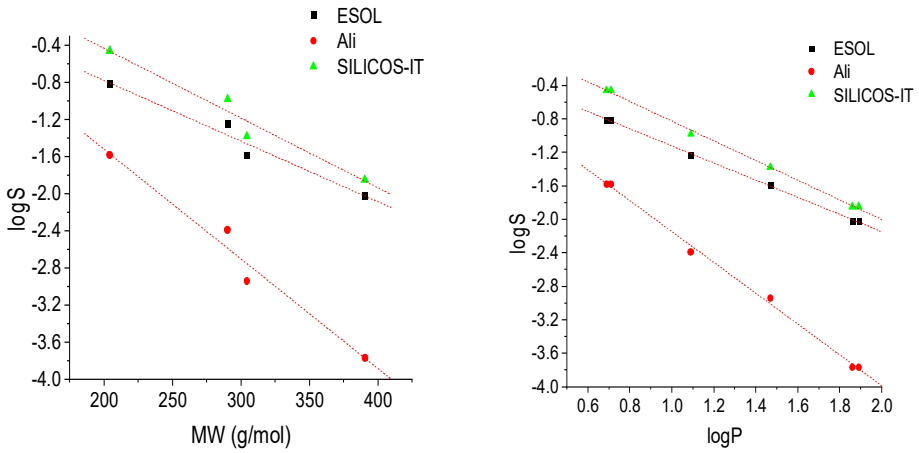
a b

Figure 5. Dependence of logS values on MW (a) and logP (b) values for oligomers of 4-hydroxyvalerate (O4HV)



a b

Figure 6. Dependence of logS values on MW (a) and logP (b) values for co-oligomers of 3-hydroxybutyrate and 3-hydroxyvalerate O(3HB3HV).



a b

Table 2. Equations corresponding to the fitting of the dependences of values of molar solubility in water (log S) with molecular weight (MW) and partition coefficient (logP) for oligomers of hydroxyalkanoates (OHAs) using the ESOL, Ali and SILICOS-IT methods. The R-squared (R2) values are also indicated for the fitted regression lines.

OHA	Methods/ Parameters	MW		logP	
		Equation	R^2	Equation	R^2
O3HA	ESOL	logS = 0.55 - 0.005 MW	0.9998	logS= 0.06 – 1.22 logP	0.9995
	Ali	logS = 0.85- 0.009 MW	0.9999	logS =-0.08 -2.31 logP	0.9995
	SILICO-IT	logS = 0.47- 0.004 MW	0.9985	logS= 0.12 – 1.06 logP	0.9991
O4HA	ESOL	logS = 0.48 - 0.004 MW	0.9996	logS= 0.09 – 0.79 logP	0.9992
	Ali	logS = 0.77- 0.008 MW	0.9998	logS =-0.14 -1.84 logP	0.9995
	SILICO-IT	logS = 0.45- 0.009 MW	0.9997	logS= 0.04 – 1.86 logP	0.9991
O3HV	ESOL	logS = 0.55 - 0.008 MW	1.0000	logS= 0.12 – 1.13 logP	0.9997
	Ali	logS = 0.85- 0.013 MW	1.0000	logS = 0.11 -1.96 logP	0.9997
	SILICO-IT	logS = 0.67- 0.008 MW	0.9996	logS= 0.25 – 1.11 logP	0.9995
O4HV	ESOL	logS = 0.53 - 0.007 MW	1.0000	logS= 0.14 – 0.95 logP	0.9993
	Ali	logS = 0.82- 0.010 MW	1.0000	logS = 0.36 -1.67 logP	0.9996
	SILICO-IT	logS = 0.48- 0.007 MW	0.9994	logS= -0.13 – 1.06 logP	0.9991
O(3HB 3HV)	ESOL	logS = 0.53 - 0.006 MW	0.9889	logS= - 0.09 – 1.13 logP	0.9984
	Ali	logS = 0.85- 0.011 MW	0.9992	logS = 0.30 -1.84 logP	0.9986
	SILICO-IT	logS = 1.07- 0.007 MW	0.9889	logS= 0.34 – 1.75 logP	0.9991

Figures 2-6 and Table 2 illustrate the linear decrease of logS with the molecular weight and partition coefficient respectively, regardless of the computational model approached. Also, there is a stronger dependence of logS values with logP rather than with molecular weight. The linear fit with the highest values of the R-

squared (R^2) coefficient corresponds to the Ali model for almost all the studied cases.

The results illustrating the linear decrease of the aqueous solubility of OHAs with both molecular weight and partition coefficient are in good agreement with those obtained for short oligomers of lactic acid, Ali method being also considered the best for modelling the solubility of lactic acid oligomers [13].

Another observation resulting from data presented in Figures 2-6 is that OHVs have lower solubility when compared to OHBs with O4HVs being the oligomers emphasizing the lowest molar solubility, and O4HBs have a higher solubility than O3HBs. The co-oligomers O(3HB3HV) have higher molar solubility than the corresponding oligomers. Several dissimilarities were also noticed in the predicted pharmacokinetics properties of the oligomers by comparison to the co-oligomers and of the co-oligomers containing the same number of monomeric units but having distinct sequence patterns [10]. It underlines that the composition of the PHAs is important when the polymer is used in medical purposes and that the composition and properties of these biopolymers can be ameliorated to enhance their medical applications.

4. CONCLUSION

Following the computational study performed in order to determine the aqueous solubility of the oligomers of hydroxyalkanoates, a linear decrease in their molar solubility was observed with increasing molecular weight and with partition coefficient values. The Ali model proved to be the best when predicting the water solubility of the oligomers of hydroxyalkanoates. This model can be used as a predictive tool to evaluate the behavior of these oligomers in aqueous media. Co-oligomers O(3HB3HV) reflect higher molar solubility than the corresponding oligomers. It underlines the importance of the composition of PHAs designed for medical use, co-polymers seem to be more advantageous than pure polymers.

REFERENCES

1. Zinn, M.; Witholt, B.; Egli, T. Occurrence, synthesis and medical application of bacterial polyhydroxyalkanoate. *Advanced Drug Delivery Reviews* **2001**, *53*, 5-21.

2. Koller, M. Biodegradable and Biocompatible Polyhydroxy-alkanoates (PHA): Auspicious Microbial Macromolecules for Pharmaceutical and Therapeutic Applications. *Molecules* **2018**, 8-23.

3. Samrot, A.V.; Samanvitha, S.K.; Shobana, N.; Renitta, E.R.; Senthilkumar, P.; Kumar, S.S.; Abirami, S.; Dhiva, S.; Bavanilatha, M.; Prakash, P.; Saigeetha S.; Shree, K.S.; Thirumurugan, R. The Synthesis, Characterization and Applications of Polyhydroxyalkanoates (PHAs) and PHA-Based Nanoparticles. *Polymers* **2021**, *13*, 19-38.

4. Radu, B.M.; Epureanu, F.B.; Radu, M.; Fabene P.F.; Bertini, G. Nonsteroidal anti-inflamatory drugs in clinical and experimental epilepsy. *Epilepsy research* **2017**, *131*, 15-27.

5. Ray, S. and Kalia, V.C. Biomedical Application of Polyhydroxyalkanoates. *Indian Journal of Microbiology* **2017**, *57*, 261-269.

6. Shahid, S.; Razzaq, S.; Farooq, R.; Zill I.; Nazil, H. Polyhydroxyalkanoates: Next generation natural biomolecules and a solution for the worlds future economy. *Int J Biol Macromol* **2021**, *166*, 297-321.

7. Williams, S.F.; Rizk, S.; Martin, D.P. Poly-4-hydroxybutyrate (P4HB): a new generation of resorbable medical devices for tissue repair and regeneration. *Biomedizinische Technik/Biomedical Engineering* **2013**, *58*, 5.

8. Bonartsev, P.; Bonartseva, G.A.; Reshefov, I.V.; Kirpichnkov, M.P.; Shaitan, K.V. Application of Polyhydroxyalkanoates in Medicine and the Biological Activiti of Natural Poly(3-Hydroxybutyrate). *Acta Naturae* **2019**, *11*, 4-16.

9. Zhang, J.; Shishatskaya, E.I.; Volova, T.G.; Silva, L.F.; Chen, G.Q. Aqueous Solubility Methods of Estimation for Organic Compounds. *Mater* **2018**, *86,* 144-150.

10. Roman, D.L.; Isvoran, A.; Filip, M.; Ostafe, V.; Zinn, M. In silico Assessment of Pharmacological Profile of Low Molecular Weight Oligo-Hydroxyalkanoates. *Front. Bioeng. Biotechnol* **2020**, *8*.

11. Isvoran, I.; Ciorsac, A.; Ostafe, V. ADME-Tox profiling of some low molecular weight water soluble chitosan derivatives. *ADMET & DMPK* **2017**, *5*, 192-200.

12. Roman, D.L.; Roman, M.; Som, C.; Schmutz, M.; Hernandez, E.; Wick, P.; Casalini, T.; Perale, G.; Ostafe, V.; Isvoran, A. Computational Assessment of the Pharmacological Profiles of Degradation Products of Chitosan. *Frontiers in Bioengineering and Biotchnology* **2019**, *214*, 7-47.

13. Dascalu, D.; Roman, D.; Filip, M.; Ciorsac, A.; Ostafe, V.; Isvoran A. Solubility and ADMET profiles of short oligomers of lactic acid. *ADMET & DMPK* **2020**, *8*, 425-436.

14. Meereboer, K.W.; Misra, M.; Mohantym, A.K. Review of recent advances in the biodegradability of polyhydroxyalkanoate (PHA) bioplastics and their composites. *Green Chemistry* **2020**, *22*, 5.519-5.558.

15. Roman, D.L.; Ostafe, V.; Isvoran, A. Computational Assessment of Chito-Oligosaccharides Interactions with Plasma Proteins. *Mar Drugs* **2021**, *120*, 3-19.

16. Ritchie, T.J.; Macdonald, S.J.F.; Peace, S.; Pickett, S.D.; Luscombe, C.N. Increasing small molecule drug developability in sub-optimal chemical space. *Med. Chem. Commun* **2013**, *4,* 673-680.

17. Vemula, V.R.; Lagishetty, V.; Lingala, S. Solubility enhancement techniques. *International Journal of Pharmaceutical Sciences* **2010**, *5*, 41-51.

18. Mamy, L.; Patureau, D.; Barriuso, E.; Bedos, C.; Bessac, F.; Louchart, X.; Martin-laurent, F; Miege, C.; Benoit, P. Prediction of the Fate of Organic Compounds in the Environment from Their Molecular Properties: A Review. *Environmental Science and Technology* **2015** *45*, 1.277-1.377.

19. Utsunomia, C.; Ren, Q.; Zinn, M. Poly(4-Hydroxybutyrate): Current State and Perspectives. *Frontiers in Bioengineering and Biotechnology* **2020**, *8*, 8-26.

20. Barreto, E.F.; Larson, T.R.; Koubek, E.J. Drug Excretion. *Biomedical Sciences* **2021**, 10-12.
21. *Chemicalize homepage*. https://chemicalize.com (accessed: April 2021).
22. Daina, A.; Michielin, O.; Zoete, V. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules, *ScientificReports* **2017**, *7*, 1-13.23. Delaney, J.S. ESOL: estimating aqueous solubility directly from molecular structure. *Journal of Chemical Information and Computer Sciences* **2004**, 4*4,* 1000-1005.
24. Ali, J.; Camilleri, P.; Brown, M.B.; Hutt, A.J.; Kirton, S.B. Revisiting the general solubility equation: in silico prediction of aqueous solubility incorporating the effect of topographical polar surface area. *Journal of Chemical Information and Modeling* **2012**, *52*, 420-428.
25.*SILICOS-IT* homepage. http://silicos-it.be.s3-website-eu-west-1.amazonaws.com/software/filter-it/1.0.2/filter-it.html (accessed: April 2021).