ISSN: 3062-3235

I-CRAFT AGRICULTURAL and FOOD TECHNOLOGIES

Degradation of Major Classes Of Recalcitrant Chemicals Hindering The Reuse of Wastewater (A Review)

Mustapha Yakubu DAUDA[1(0009-0006-9619-9094)], Eser CELIKTOPUZ[2(0000-0002-5355-1717)]

[1] Department of Agricultural Structures and Irrigation Engineering, University of Çukurova, Adana 01330, Turkey.

[2] Department of Agricultural Structures and Irrigation Engineering, University of Çukurova, Adana 01330, Turkey.

eceliktopuz@cu.edu.tr

Abstract. In this recent age of advanced technology and biotechnology, the existing water bodies that serve as a reservoir for lives are experiencing a surge in the occurrence of chemical contaminant. These classes of contaminants were found to include both organic and inorganic, synthetic and natural pollutants of serious environmental concern. Studies revealed that these contaminants were predominantly recalcitrant chemicals that have been grouped as either toxic, hazardous and carcinogenic. There occurrence is not limited to their numbers, types or variety but also a concentration that is alarming, leading to what is known as emerging chemical contaminants (ECC). Sudden increase in the occurrence of chemical contaminants in our water, wetlands, ponds, wastewaters and sludge were due to increasing population, high demand and consumption, while their persistent in the environment is strongly linked to their recalcitrant nature and physiochemical properties. Partial treatment of wastewater results in the accumulation of recalcitrant chemicals while incomplete degradation of recalcitrant chemicals give birth to emerging contaminant (EC). Incorporation of tertiary treatment systems to our conventional wastewater treatment system in combination with advance treatment processes becomes paramount. Advance oxidation treatment (AOP) processes, membrane systems coupled with bioreactors, biodegradations using bacteria and fungi could be used in combination to remove these recalcitrant chemicals from wastewater. Test to proof the safety and safe reuse of these treated wastewaters should also be conducted using model organism to achieve an ecofriendly treated wastewater.

Keywords: degradation, recalcitrant chemicals, wastewater, ecotoxicity, environmentally friendly.

1 Introduction

Recalcitrant chemicals belonging to both major or minor class leave negative impact in our environment. These class of recalcitrant are predominantly synthetic chemical compounds which have emerged due to high production, aimed at meeting the desired demand of the growing population. Sectors of human endeavors that inevitably release recalcitrant chemical into our water and wastewaters include feeding, health and fashion. Feeding is where food dyes are introduced into our food, chemical preservatives, flavors and seasoning chemicals, heavy metals and other carcinogens. Health sector can be divided into two basic unit beauty (aesthetics) personal care products (PCP) and medications where pharmaceutical drugs fit in. Fashion is where dyes are used on fabrics for fashion and for makeups [1]. Synthetic compounds used in the manufacture of drugs by the pharmaceutical industry to keep fit and to treat undesired health situations in humans and animals are the predominant sources of recalcitrant chemical in the wastewater [2, 3] and persists in the environment. Recalcitrant chemicals used in the food and feed sector and synthesized to take care of plant pest, herbicides and other plant diseases of significance.

Ternes and Joss [4] reported that utilization of pharmaceuticals by humans is estimated at 15g.cap^{-1}. and 50-150g.cap^{-1} in the developing countries and industrialized countries respectively. According to Ajay et al. [5] advancement in healthcare have created a swift change and awareness for the pharmaceutical industry which make the sector one of the most GDP buster of the global economy, generating about $50 billion annually. The unfortunate aspect of the industry is the generation of waste that are mostly chemical byproducts of serious environmental concern weighing up to about 200 tons per year. Animal health treatments in the veterinary sector are reported to have almost equal consumption of pharmaceuticals as in humans [4].

Moreso, improper use of personal care products, mishandling of pharmaceutical waste and improper disposal of expired drugs and steroids, and uncontrolled discharge of wastewater or illegal release of partially or untreated industrial wastewater into water bodies or municipal wastewater treatment plants. The raising concern of personal care products (PCPs) has been linked to its toxicological and ecological impact in the ecosystem. Recent studies have demonstrated their presence in the environment and their faith is questioned. Research conducted by [6, 7, 8] revealed the presence of recalcitrant chemicals of personal care origin at a concentration above environmental allowable limit.

The spectrum of pharmaceutical chemicals in the environment coupled with their unpredicted impact on the ecosystem are the main reason for their growing concern. The vast majority of pharmaceutical chemicals vary from each other due to their design; mode of action target tissues, hence these intentions are guided by their physical and chemical properties. Even with the existing variety of pharmaceutical chemicals more synthetic chemicals are being formulated and designed to meet higher efficiency. Due to high production of pharmaceuticals, acetaminophen (ACT) or paracetamol were reported in the environment at a concentration far above allowable limit [9].

Efforts have been made by scientist to stop accumulation of these recalcitrant chemicals and there persistent in nature by employing various methods of degradation, remediation and mineralization. Most of these recalcitrant chemicals are known to have escaped the primary and secondary treatment processes of the treatment plants. From among the treatment techniques reported include advance oxidation processes (AOP) [2, 3] bioremediation [10], biodegradation [1] and membrane removal [11].

Recalcitrant chemicals have numerous ways through which they get into wastewater and to the environment. The recalcitrant chemicals gain access to our water through direct domestic usage, while their presence in our wastewater treatment plants is mainly through discharge from the municipal waste. Persistence of recalcitrant chemicals in our environment to the ecosystem occur when the pollutants in question escape the treatment processes that are been carried out in our conventional wastewater treatment plant [12, 13].

Studies have reported reduction of the recalcitrant chemical after passing through wastewater treatment systems. The results achieved from these systems are related to some of the properties of the recalcitrant chemicals that support sorption (adsorption or absorption). Some properties of the recalcitrant chemicals that made sorption possible include physical and chemical properties, polarity, functional groups, charges and others. Even with the above-mentioned properties, recalcitrant chemicals find their ways into the environment, because some conditions need to be meet for a successful sorption to take place and subsequent removal of the recalcitrant from the wastewater.

Most conventional municipal wastewater treatment systems are not equipped or designed to handle recalcitrant chemicals of great concern hence leading to the discharge of the treated wastewater into nearby water bodies rendering the ecosystem loaded with recalcitrant chemicals.

Recalcitrant chemicals of pharmaceutical origin have been included in the group of emerging contaminants due to their dynamic and cryptic health problems impacted on humans and farm animals. These unclear nature of health problems caused by recalcitrant chemicals of both pharmaceutical origin and personal care products have raised issues of great concern in the past two decades. Guedes-Alonso et al. [14] reported the presence of recalcitrant chemical of great concern "priority pollutants" (fluoroquinolones) having a hazardous effect on the ecosystem after escaping from wastewater treatment system. This was the result of a study conducted in Spain in 2013 from the analysis of two wastewater treatment systems after post treatment and more were found to include anti-inflammatory drugs, analgesic and weight regulatory pills [15].

2 Treatment Methods

Most conventional wastewater treatment plants that are designed without tertiary treatment facilities are usually not capable of handling recalcitrant chemicals hence, treated wastewaters containing such pollutants are discharged to receiving water bodies. These water reserves will contain trace amounts of some of the classes of recalcitrant chemicals if not all, hence creating fear for the reuse of wastewater.

Guedes-Alonso et al. [14] in their research, while working with a membrane bioreactor reported a result of successful removal of the recalcitrant chemical with great success relative to sorption of the pollutants on an activated sludge at the secondary treatment stage of the conventional wastewater treatment system. Llers et al. [16] reported results with similar success [15, 17].

Miège et al. [18] demonstrated in research comparing some treatment processes for the removal of recalcitrant chemicals of personal care origin and pharmaceutical sources. The results proved that membrane bioreactors and activated sludge coupled with nitrogen treatment gave most promising results with reliable efficiency for the removal of recalcitrant chemicals of both origins [15].

Bioremediation using enzymes extracted from fungi has gained ground and the interesting part are the potentials demonstrated by the extracellular (ligninolytic) enzymes produced by white rot fungi [19, 20] which enabled them to captivate the interest of researchers. Laccase, LiP, MnP HrP are examples of the most reported ligninolytic enzymes that have demonstrated excellent biodegrading and bio-detoxifying results when use to treat industrial wastewater [1, 21,22, 23]. Studies on laccase have been on the leading front since reports on its substrate non-specificity. Laccase enzyme gave excellent results during dye decolorization [1, 19, 24, 25] biodegradation of hazardous compounds and complex micro-pollutants [26] bioremediation of xenobiotic and recalcitrant pollutants [27, 28, 29, 30] bioconversion of fuel (jet fuel) [31] and monomers crosslinking [32, 33].

Advanced oxidation processes (AOPs) are defined as technologies used in environmental management that generate hydroxyl radicals to effectively degrade complex organic pollutants in water and soil, which are often resistant to traditional treatment methods. AOPs are used similarly to other drinking water treatment processes such as membranes, granular activated carbon, air stripping, and biological degradation [3].

3 Detection and Determination of the Recalcitrant Chemical Pollutants

Studies reveal the existence of recalcitrant chemical pollutant in small concentration and in trace amounts, but their negative impact and unpredictive level of danger up to a catastrophic level is what is posing treat to both environment and human health [1, 34]. Tracing these chemical pollutants, detecting them as well as determining their concentrations in different media complex (biotic and a biotics) e.g. solids (sediments and sludge), liquids (leachates and wastewater) and others (living organisms) at different destinations becomes paramount [35].

Therefore, strategic combination of extraction techniques before instrumental analytical becomes key to achieving techniques that are more sensitive, selective, fast, and friendly to the environment. The solubility and mass transfer of any analyte is the primary driving force guiding its successful detection in any matrix [36]. Moreso, advancement in research is able to come up with more classical techniques that bring about advancement in both separation and quantification methods that have become the basics of modern standards methods of sample analysis example liquid-liquid extraction [37] for liquid samples and Soxhlet extraction methods for solid samples [38].

While the liquid-liquid extraction uses liquid, the solid extraction method (soxhlet) employs a very practical method of sample preparation that uses no solvent or a very small amount. Pawliszyn [39] classified the analyte extracting phases into; sorbent, membrane and gas also reporting that most basic steps involve in sample preparation include but not limited to careful sampling and proper homogenization, proper extraction using the right method followed by clean-up and analyte concentration before injecting or sending it to the chromatographic machine for analysis. Future research in the areas of extraction and determination should be improved to target analytes of interest using more sensitive techniques that will bring about ease of handling and less toxic and environmentally friendly.

Advancement in modern analytical instrumentation of high precision have enabled scientist to also improve on the systems and methods of analytical separation and detection techniques. To mention but a few from the advancement are such as GC–MS (Gas chromatography Mass Spectrometry), GC–MS/MS (Gas Chromatography tandem Mass Spectrometry), LC-MS (Liquid Chromatography-Mass Spectrometry) and LC-MS/MS (Liquid Chromatography-tandem Mass Spectrometry) all of which have inbuilt libraries of all the contaminants and their properties which make comparison easy. Technological evolution of instruments with high precision have helped to detect, determine and measure the concentration of recalcitrant chemicals up to nano quantities per liter level present in water and wastewater [37]. The results of a study conducted between 1999 and 2000 by the United States Geological Survey (USGS) confirmed the presence of recalcitrant chemicals, more than 50 of which are pharmaceutical chemicals in 139 streams across 30 states in the USA [12, 39]

4 Toxicity and Ecofriendly Test

Toxicity analysis is a major analysis that can restore or give room for wastewater reuse by end users. Toxicity test are experiments design to test the impact of chemicals on a set of microorganisms at the list allowable concentration that will impact minimal effect on the environment or the minimum concentration of toxic waste that can allow for a period of time that the environment can take care of it with minimal or no threat. Different reference organisms were used for different test pending on the available test method. Toxicity analysis is practically achieved by exposing some series of organism in a medium which could either be soil or water in other to evaluate the effect of the contaminant at varying concentration. The effect of the chemical contaminant could be monitored in terms of survival of the organism in the contaminated medium relative to a control (free and uncontaminated media). Various other survival activities could be monitored which ae not limited to growth, reproduction and other behavioral patterns. In the case of recalcitrant, toxic and carcinogenic chemicals sometimes hormonal changes and behavioral changes are also put into consideration during toxicity test. Some of the most reported toxicity test include the use of fluorescence bacteria (Vibrio fischeri), model organism such as crustaceans and algae and sometimes E. coli or cell cultures are also employed. Toxicity analysis conducted by [1,34] reported a successful toxicity test result with Microtox toxicity test kits which were based on the monitoring of luminous intensity of Vibrio Fischeri luminescent microorganisms before, after and at intervals during the degradation experiments.

Dauda and Erkurt [1] demonstrated the use of model organisms for testing the toxicity of intermediate compounds and final products generated during degradation of anthraquinone. Toxicity results from research conducted by [1, 40, 41, 42, 43] revealed that the microorganism responded differently to different toxicity levels during the degradation experiments. Toxicity test carried out at different stages of degradation showed an increasing toxicity level prior to reduction then mineralization. Reasons for the increased toxicity were due to the formation of toxic intermediates formed during degradation processes as proven by [34] and during degradation of anthraquinone dye [1]. Hence drawing the attention of researchers towards avoiding partial degradation or incomplete mineralization, as this might result in the production of even more toxic chemical compounds than the original mother compound.

Results from toxicity tests after biodegradation with enzymes and mineralization with advanced oxidation systems revealed that the treated medium containing the chemical pollutants contain a smaller number of intermediate products which implies presence of chemicals of less toxicity to the environment making them environmentally friendly.

5 Conclusions and Future Prospectives

Conventional system for the treatment of polluted water and wastewater from municipalities are not capable of removing recalcitrant chemical of high environmental concern. The presence of recalcitrant chemicals of compounded origins with different characteristic properties ranging from organic to inorganic, natural and synthetic, simple to complex make it very hard for the conventional system to treat efficiently. Therefore, these conventional systems will inevitably discharge the treated wastewater containing these recalcitrant chemicals into the receiving water bodies hence making the water bodies a reservoir for contaminant and recalcitrant chemicals. These array of properties are a reason for including the recalcitrant chemicals into hazardous, carcinogenic, mutagenic, biogenic hormone, endocrine disrupting compounds and many other classes of pollutants including the recently discovered pollutant as emerging contaminants. Moreso, both sludge and activated sludge that are generated from both primary and secondary treatment systems respectively are often loaded with high concentration of recalcitrant chemicals serving as reservoir or sink for the pollutant. Unfortunately, when these sludges are used as fertilizers, they serve as intermediary sink for the recalcitrant chemicals.

The discharge of inadequately treated wastewater in to the nearby wet lands, pond streams and other water bodies in the name of proper disposal of wastewater is the primary source of carcinogen and environmental contamination. Alternatively, the direct use of the same ill-treated wastewater leaded with recalcitrant chemicals will directly or indirectly affect the bioactivities and performance of our agricultural crops. The need for tertiary treatment systems coupled with advanced wastewater treatment technologies becomes a priority if we have to save ourselves and the environment.

No single independent treatment system is perfect for the removal of recalcitrant chemicals, but researches have proven that a combination of the advance systems will give the perfect result [6, 7, 8]. Researches have demonstrated the combination of membrane bioreactors and systems that uses both oxidative and reductive processes were found effective for the removal of a number recalcitrant chemical compound [17, 18].

Declaration of Competing Interest: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported.

Acknowledgments: This article was not funded by any funding agencies nor was it for profit purposes.

6 References

1. Ajay S., Rahul G., Anjali C. (2025). A review on sustainable management of hazardous, nonhazardous, and chemo-waste in the pharmaceutical sector and its correlation with UNSDGs 3, 6, 9, and 11–15. Environ Monit Assess (2025) 197:1002 https://doi.org/10.1007/s10661-025-14428-1

2. Arica, M.Y., Salih, B., Celikbicak, O., Bayramoglu, G., (2017). Immobilization of laccase on the fibrous polymer-grafted film and study of textile dye degradation by MALDI–ToFMS. Chem. Eng. Res. Des. 128, 107–119. https://doi.org/10.1016/j.cherd.2017.09. 023.

3. Asadgol, Z., Forootanfar, H., Rezaei, S., Mahvi, A.H., Faramarzi, M.A., 2014. Removal of phenol and bisphenol-A catalyzed bylaccase in aqueous solution. J. Environ. Health Sci. Eng. 12, 93. https://doi.org/10.1186/2052-336X-12-93.

4. Asmita Gupta and Indu Shekhar Thakur; (2016). Treatment of Organic Recalcitrant Contaminants in Wastewater Submitted: 31 March 2016 Reviewed: 14 October 2016 Published: 29 March 2017 DOI: 10.5772/66346)

5. Barceló D., (1993). J Chromatogr 643:117–143

6. Bayramoglu, G., Salih, B., Akbulut, A., Arica, M.Y., (2019). Biodegradation of Cibacron Blue 3GA by insolubilized laccase and identification of enzymatic byproduct using MALDI-ToF-MS: toxicity assessment studies by Daphnia magna and Chlorella vulgaris. Ecotoxicol. Environ. Saf. 170, 453–460. https://doi.org/10.1016/j.ecoenv. 2018.12.014.

7. Bilal, M., Rasheed, T., Nabeel, F., Iqbal-Hafiz, M.N., Zhao, Y., (2019). Hazardous contaminants in the environment and their laccase-assisted degradation – a review. J.Environ. Manage. 234, 253–264. https://doi.org/10.1016/j.jenvman.2019.01.001.

8. Dauda, M. Y., & Erkurt, E. (2019). Investigation of reactive Blue 19 biodegradation and byproducts toxicity assessment using crude laccase extract from Trametes versicolor. Journal of Hazardous Materials 393 (2020) 121555 https://doi.org/10.1016/j.jhazmat.2019.121555

9. Dogan, S., & Kıdak, R. (2013). Degradation of Isoproturon by Advanced Oxidation Processes and Analysis of Toxicity of Byproducts. Istanbul International Solid Waste, Water And Wastewater Congress (2013) Pg 237 https://www.academia.edu/download/71672353/Istanbul3WCongAbstracts2013EngInteraktif.pdf#page=237

10. Dogan, S., & Kıdak, R. (2016). A plug flow reactor model for UV-based oxidation of amoxicillin, Desalin. Water Treat. 57 (2016) 13586–13599.

11. Dogan, S., & Kıdak, R. (2018). Medium-high frequency ultrasound and ozone based advanced oxidation for amoxicillin removal in water, Ultrasonics Sonochemistry 40 (2018) 131–139.

12. Environmental Protection Agency, (2012). Estimation Programs Interface SuiteTM for Microsoft Windows, <http://www.epa.gov/oppt/exposure/pubs/episuite. htm>, 2012 (accessed 12.12.2012).

13. Erkurt, E.A., Unyayar, A., Kumbur, H., (2007). Decolorization of synthetic dyes by white rot Fungi. Involving Laccase Enzyme in the Process, Process Biochem. 42, 1429–1435. https://doi.org/10.1016/j.procbio.2007.07.011.

14. Erkurt, H.A., (2015). Biodegradation and detoxification of BPA: involving laccase and a mediator clean soil. Air, Water. 43, 932–939. https://doi.org/10.1002/clen. 201400628

15. Fatone F, Di Fabio S, Bolzonella D, Cecchi F. (2011). Fate of aromatic hydrocarbons in Italian municipal wastewater systems: an overview of wastewater treatment using conventional activated-sludge processes (CASP) and membrane bioreactors (MBRs). Water Res. 2011; 45(1):93–104.

16. Fent. K, Weston AA, Caminada D. (2006). Ecotoxicology of human pharmaceuticals. Aquat. Toxicol. 2006; 7:122–159.

17. Gao, D., Du, L., Yang, J., Wu, W.M., Liang, H., (2010). A critical review of the application of white rot fungus to environmental pollution control. Crit. Rev. Biotechnol. 30, 70–77. https://doi.org/10.3109/07388550903427272.

18. Garric J, Ferrari B. (2005). Pharmaceuticals in aquatic ecosystems. Levels of exposure and biological effects: A review. Revue des Sciences de l'Eau/Journal of Water Science. 2005; 18(3): 307–330.

19. Gassara, F., Brar, S.K., Verma, M., Tyagi, R.D., 2013. Bisphenol a degradation in water by ligninolytic enzymes. Chemosphere. 92, 1356–1360. https://doi.org/10.1016/j.chemosphere.2013.02.071

20. Guedes-Alonso R, Afonso-Olivares C, Montesdeoca-Esponda S, Sosa-Ferrera Z and Santana-Rodríguez JJ. (2013). An Assessment of the Concentrations of Pharmaceutical Compounds in Wastewater Treatment Plants on the Island of Gran Canaria (Spain). Springer Plus. 2013; 2:24. http://www.springerplus.com/content/2/1/24

21. Hatakka, (1994). Lignin-modifying enzymes from selected white-rot fungi: production and role in lignin degradation. FEMS Microbiol. Rev. 13, 125–135. https://doi.org/10. 1111/j.1574-6976.1994.tb00039.x.

22. José Juan Santana Rodríguez & Zoraida Sosa Ferrera & Daura Vega Moreno & M. Esther Torres Padrón & Cristina Mahugo Santana (2008). Recent trends in the use of organized molecular systems combined with chromatographic techniques in environmental analysis. Anal Bioanal Chem (2008) 391:725–733 DOI 10.1007/s00216-008-1838-x

23. Kummerer K. (2001). Drugs in the environment: emission of drugs, diagnostic aids and disinfectants into wastewater by hospitals in relation to other sources – a review. Chemosphere. 2001; 45:957–969.

24. Lamia Ayed, Kamel Chaieb, Abdelkarim Cheref, Amina Bakhrouf. (2010). Biodegradation and decolorization of triphenylmethane dyes by Staphylococcus epidermidis. Desalination 260 (2010) 137–146. doi:10.1016/j.desal.2010.04.052.

25. Lao, R. C., Thomas, R. S., Monkman, J. L., (1975). J Chromatogr 112:681– 700

26. Li, X.Z., Cheng, Q., Wu, Y.C., Feng, Y.Z., Liu, W.W., Liu, X.G., (2014). Influencing factors and product toxicity of Anthracene Oxidation by fungal laccase. Pedosphere 24, 359–366. https://doi.org/10.1080/23311843.2017.1339841

27. Llers, Ö. S, Singer HP, Fässler P, Müller SR. (2001). Simultaneous quantification of neutral and acidic pharmaceuticals and pesticides at the low-ng/l level in surface and waste water. J. Chromatogr. A. 2001; 911:225–234.

28. Miège C, Choubert JM, Ribeiro L, Eusèbe M, Coquery M. (2009). Fate of pharmaceuticals and personal care products in wastewater treatment plants – Conception of a database and first results. Environ. Pollut. 2009; 157:1721–1726.

29. Mitra S., (2003). Sample preparation techniques in analytical chemistry. Wiley–Interscience, New Jersey

30. Nguyen, L.N., van de Merwe, J.P., Hai, F.I., Leusch, F.D.L., Kang, J., Price, W.E., Roddick, F., Magram, S.F., Nghiem, L.D., (2016). Laccase–syringaldehyde-mediated degradation of trace organic contaminants in an enzymatic membrane reactor: removal efficiency and effluent toxicity. Bioresour. Technol. 200, 477–484. https://doi.org/10.1016/j. biortech.2015.10.054.

31. Parra Guardadoa, A.L., Bellevillea, M.-P., Alanisb, M.J.R., Saldivarb, R.P., Sanchez-Marcano, J., (2019). Effect of redox mediators in pharmaceuticals degradation by laccase: a comparative study. Process. Biochem. 78, 123–131. https://doi.org/10.1016/j.procbio.2018.12.032.

32. Pawliszyn J (1997). “Solid phase microextraction: Theory and practice”. Wiley-VCH, 247 pp

33. Pulate, V.D., Bhagwat, S., Prabhune, A., 2013. Microbial oxidation of medium chain fatty alcohol in the synthesis of sophorolipids by Candida bombicola and its physicochemical characterization. J Surfact Deterg 16, 173–181. https://doi.org/10.1007/s11743-012-1378-4

34. Qutob, M., Doğan, S., & Rafatullah, M. (2022). Heterogeneous Activation of Persulfate by Activated Carbon for Efficient Acetaminophen Degradation: Mechanism, Kinetics, Mineralization, and Density Functional Theory. Chemistry Select 2022, 7, e202201249 (11) doi.org/10.1002/slct.202201249

35. Salazar-Lopez, M., Rostro-Alanis, Mde J., Castillo-Zacarias, C., Parra-Guardado, A.L., Hernandez-Luna, C., Iqbal, H.M.N., Parra-Saldivar, R., (2017). Induced degradation of anthraquinone-based dye by laccase produced from pycnoporus sanguineus (CS43). Water Air Soil Pollut. 228, 469. https://doi.org/10.1007/s112700173644-6.

36. Shraddha, R., Shekher, S., Sehgal, M., Kamthania, A., (2011). Kumar Laccase: microbial sources, production, and potential biotechnological applications. Enzyme Res. 2011, 11. https://doi.org/10.4061/2011/217861

37. Su, J., Noro, J., Fu, J., Wang, Q., Silva, C., Cavaco-Paulo, A., (2019). Coloured and low conductive fabrics by in situ laccase-catalysed polymerization. Process. Biochem. 77, 77–84. https://doi.org/10.1016/j.procbio.2018.11.007

38. Ternes T. A. (1998) Occurrence of drugs in German sewage treatment plants and rivers. Water Res. 1998;32(11):3245–3260.

39. Ternes, T. A., Joss, A., (2006). Human Pharmaceuticals, Hormones and Fragnences: The Challenge of Micropollutants in Urban Water Management, IWA Publishing, London, 2006.

40. Unyayar, A., Mazmanci, M.A., Atacag, H., Erkurt, E.A., Coral, G., (2005a). A Drimaren blue X3LR dye decolorizing enzyme from Funalia trogii: one step isolation and identification. Enzyme Microb. Technol. 36, 10–16. https://doi.org/10.1016/j.enzmictec. 2004.02.008.

41. Unyayar, A., Mazmanci, M.A., Erkurt, E.A., Atacag, H., Gizir, A.M., (2005b). Decolorization kinetics of the azo dye drimaren blue X3LR by laccase. React. Kinet. Catal. Lett. 86, 99–107. https://doi.org/10.1007/s11144-005-0300-8.

42. World Health Organization, (2012). Pharmaceuticals in drinking-water. http: <www. who.int/water_sanitation_health/publications/2012/pharmaceuticals/en/>,2012 (accessed 20.03.13).

43. Viswanath, B., Rajesh, B., Janardhan, A., Kumar, A.P., Narasimha, G., (2014). Fungal laccases and their applications in bioremediation, Review Article. Enzyme es.2014 https://doi.org/10.1155/2014/163242. ID 163242.