MendelNet

Conference Brno 2021

Editors:

Radim Cerkal

Natálie Březinová Belcredi

Lenka Prokešová

Proceedings of 28th International PhD Students Conference

10 November 2021, Brno, Czech Republic

Mendel University in Brno
Faculty of AgriSciences

MendelNet 2021

Proceedings of 28th International PhD Students Conference
10 November 2021, Brno, Czech Republic

Editors: Radim Cerkal, Natálie Březinová Belcredi, Lenka Prokešová

The MendelNet 2021 conference would not have been possible without the generous support of The Special Fund for a Specific University Research according to the Act on the Support of Research, Experimental Development and Innovations and the **support of**:

BIOMIN Czech s.r.o.

kontroluje.me

PELERO CZ o.s.

Profi Press s.r.o.

Research Institute of Brewing and Malting, Plc.

All contributions of the present volume were peer-reviewed by two independent reviewers. Acceptance was granted when both reviewers' recommendations were positive.

ISBN 978-80-7509-821-4

Scientific Committee:

Assoc. Prof. Ing. Radim Cerkal, Ph.D. (Chair)
Prof. Ing. Radovan Pokorný, Ph.D.
Prof. Ing. Gustav Chládek, CSc.
Assoc. Prof. Ing. Radovan Kopp, Ph.D.
Assoc. Prof. Ing. Josef Suchomel, Ph.D.
Prof. Dr. Ing. Milada Šťastná
Assoc. Prof. Ing. Šárka Nedomová, Ph.D.
Assoc. Prof. Ing. Pavel Hanáček, Ph.D.
Prof. MVDr. Zbyšek Sládek, Ph.D.
Assoc. Prof. Ing. Vojtěch Kumbár, Ph.D.
Assoc. Prof. RNDr. Ondřej Zítka, Ph.D.

Organizing Committee:

Ing. Natálie Březinová Belcredi, Ph.D. (Chair)
Ing. Lucie Melišová, Ph.D.
Ing. Lenka Prokešová
Mgr. Patrik Vacek

PREFACE

Each year, the editors of the volume you are about to read are tasked with the responsibility of putting a coherent form to the proceedings from MendelNet, the international PhD Students Conference of the Faculty of AgriSciences of Mendel University in Brno.

The event which reached, this year, on November 10, 2021, its 28th edition, is traditionally aimed at both under and postgraduate students from the Czech Republic, Europe and beyond, and proudly welcomes the participants of various professional and cultural backgrounds. And while this time the people could not gather on-site due to globally-imposed covid-19 restrictions, the conference swiftly transformed itself into a virtual and fascinating beehive of results, opinions and brand new research paths and ideas.

Here in Brno, under the spell of great genetician G. J. Mendel and the guidance of skilled senior researchers and supervisors, students can introduce, defend and discuss their scientific results while those who do not feel confident enough to present and pen their paper in English are invited to join as spectators and follow-up discussion participants.

The best submissions are, after rigorous peer-review process, collected here and range from plant and animal production to fisheries and hydrobiology to wildlife research while agroecology and rural development, food technology, plant and animal biology, techniques and technology and applied chemistry and biochemistry also belong to the core areas being investigated.

The collection as varied and huge as this can succeed only as a team effort, both on authors' and editors' side, so we would like to express our thanks and gratitude to all committees and reviewers both for their outstanding work and invaluable comments and advice.

The Editors

TABLE OF CONTENTS

PLANT PRODUCTION

ANIMAL PRODUCTION

FISHERIES AND HYDROBIOLOGY

WILDLIFE RESEARCH

AGROECOLOGY AND RURAL DEVELOPMENT

FOOD TECHNOLOGY

PLANT BIOLOGY

ANIMAL BIOLOGY

TECHNIQUES AND TECHNOLOGY

APPLIED CHEMISTRY AND BIOCHEMISTRY

Determining the phytotoxicity of rubber granulate from waste tires

Marketa Sourkova, Dana Adamcova
Department of Applied and Landscape Ecology
Mendel University in Brno
Zemedelska 1, 613 00 Brno
CZECH REPUBLIC

xsourkov@mendelu.cz

Abstract: The annually increasing global production of tires ranges around 1 billion and the number of waste tires is growing too. A question comes to the fore how to handle the waste to prevent environmental risks. The most advanced recycling facilities can process waste tires by sophisticated methods leading to their further use. One of products is a so-called rubber granulate which enjoys great interest of gardeners who use it instead organic mulch. In relation with this research, phytotoxicity of rubber granulate made from waste tires was studied in laboratory conditions using a test kit (Phytotoxkit^{TM}) for the determination of inhibitory/stimulating effect. The testing was made on the reference soil and tested seeds were the seeds of white mustard (*Sinapis alba* L). The granulate was applied onto the soil at rates of 5%, 10%, 25%, 50% and 75%. Research results demonstrated the inhibition of *Sinapis alba* L. seeds already from the rate of 5%, i.e. 6.50%. In the other rates, the inhibition ranged between 26.37% and 62.36%. The granulate is therefore considered phytotoxic and should not be used on the soil.

Key Words: white mustard, reference mixture soil, Phytotoxkit^{TM}, mulch, environmental risks

INTRODUCTION

At the moment when a tire is at the end of its service life, it becomes waste which has to be handled appropriately (Llompart et al. 2013, Birkholz et al. 2012). In the past, waste tires were disposed in landfills without any use. However, they captured methane due to their large volume, which caused upward pressure and subsequent damage to the sealing of landfills that was to prevent substances from leaking into the environment (Price and Smith 2006). Waste tires are valorized for energy, namely in cement works as a replacement fuel for brown coal which has a calorific value three times lower than tires (Kizlink 2014, Kuraš 2014). Apart from that, tires can be converted into valuable products (active coal, fuel) and energy source by means of pyrolysis (Sathiskumar and Karthikeyan 2019, Ramos et al. 2011). Waste tires can be also used as gravel or as admixture to asphalt roadbeds where they improve adhesion and considerably reduce traffic noisiness (Kuraš 2014, Kumaran et al. 2008, Cao 2007).

Waste tires are currently increasingly used for recycling. The produced granulate is most frequently utilized as rubber mulch in gardens, non-slip material of filling for artificial turfs. Its use is still popular for the floor surface of fitness centers, sports grounds and children’s playgrounds (Llompart et al. 2013, Birkholz et al. 2012, Li et al. 2010). Although some studies claim the tires to be inert and non-toxic (Kizlink 2014, Nelson et al. 1994, Schnick et al. 1982), it is already known today that they contain heavy metals (arsenic, cadmium, chromium, lead and zinc), polycyclic aromatic hydrocarbons and other toxic additives (Šourková et al. 2021, Wink and Dave 2005). Tire consists of three basic components: rubber polymers (ca 50%), soot (ca 28%) and softeners (ca 18%). Other additives are anti-degrading agents, activators, accelerators and vulcanizing agents (Šourková et al. 2021, Wink and Dave 2005). In this sense, it is important to monitor impacts of rubber granulate on the environment and human health.

Tests of phytotoxicity were conducted to determine the effect of rubber granulate made from waste tires. Contaminants in the rubber granulate represent environmental risks and the assessment of their impact on the environment is therefore very important.

The goal of this study was to determine the phytotoxicity rate of rubber granulate from waste tires in laboratory conditions using the Phytotoxkit^{TM} testing kit.

MATERIAL AND METHODS

Phytotoxicity of rubber granulate from waste tires with the use of micro-biotests

The fraction of rubber granulate tested in our study was 1–3 mm and the granulate was produced of rubber for general use (SBR) by the technology of crushing and sowing. Rubber granulate has been recently ever more used as a replacement of organic mulch as it does not attract insects and is almost indecomposable. Nevertheless, harmful substances can gradually be released from the granulate into the soil and plants despite all these advantages. The goal of this study was to determine the rate of its phytotoxicity.

In accordance with the Phytotoxkit^{TM} methodology, the phytotoxicity of rubber granulate was tested in combination with the seeds of white mustard (*Sinapis alba* L.) at rates 5%, 10%, 25%, 50% and 75%. A mixture of reference (OECD) soil certified for the tests of phytotoxicity was applied on the testing Phytotoxkit in combination with distilled water and the determined rate of rubber granulate from waste tires. A control sample was OECD soil (85% of silica sand, 10% of kaolin clay, 5% of peat and $CaCO_3$) prepared with distilled water. Ten seeds of *Sinapis alba* L. were placed in a row on the filter paper. Each rate of rubber granulate was given three repetitions. A summary of phytotoxicity tests is presented in Table 1. The prepared sample container was sealed, installed in a holder in vertical position and incubated for 72 hours in the Ecocell dryer without light access at a controlled temperature of 23±1 °C. After the lapse of the set-up time, partial lengths of individual roots of *Sinapis alba* L. were measured by means of Image Tool 3.0 for Windows, and the percentage of plant growth inhibition/stimulation (IR) was calculated from the following equation (Šourková et al. 2021, Vaverková et al. 2019) (1):

$$IR\ (\%) = \frac{(L_C - L_S)}{L_C} \times 100 \qquad (1)$$

where L_C is an average value of plant root length in the sample without the rubber granulate (mm) and L_S is an average value of plant root length in the sample with the rubber granulate (mm).

The resulting value is considered growth inhibiting when IR is >0 or growth stimulating when IR is <0. A scheme of the experiment is presented in Figure 1.

Figure 1 Phytotoxicity of rubber granulate, Brno, Czech Republic, 2021

Legend: 1 – Waste tires; 2 – Transport to the recycling line; 3 – Tire recycling line; 4 – Granulate from waste tires;
5 – Phytotoxicity test

Table 1 Phytotoxicity tests and their labelling

Designation	Addition of granulate from tires	Number of repetition
Control	-	3×
1	5%	3×
2	10%	3×
3	25%	3×
4	50%	3×
5	75%	3×

Legend: Control – control sample, tests without granulate, 3 replicates; 1 – tests with 5% granulate, 3 replicates; 2 – tests with 10% granulate, 3 replicates; 3 – tests with 25% granulate, 3 replicates; 3 – tests with 50% granulate, 3 replicates; 4 – tests with 75% granulate, 3 replicates

RESULTS AND DISCUSSION

Results of the growth inhibition of *Sinapis alba* L. for the tested rates of rubber granulate were determined using the equation of percentage inhibition/stimulation of plant growth (IR) and are presented in Table 2.

The average value of plant root length in the sample without the rubber granulate (Control) was 92.40 mm (L_C). Average values of root length (mm) in the sample with the rubber granulate (L_S) were calculated for each percentage rate separately.

Table 2 Results of Sinapis alba L. growth inhibition in the phytotoxicity tests of rubber granulate

Designation	Addition of granulate from tires	L_S (mm)	IR (%)
1	5%	86.39	6.50
2	10%	68.03	26.37
3	25%	57.53	37.73
4	50%	47.37	48.73
5	75%	34.78	62.36

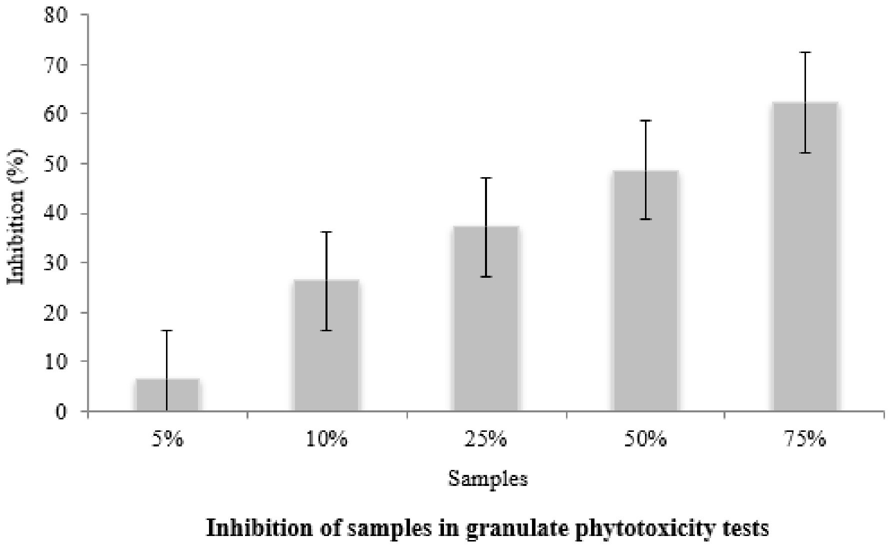
Legend: 1 – tests with 5% granulate; 2 – tests with 10% granulate; 3 – tests with 25% granulate; 3 – tests with 50% granulate; 4 – tests with 75% granulate; L_S – average values of root length in the sample with rubber granulate (mm); IR – results of percentage inhibition/stimulation of plant growth (%)

The lowest inhibition values were recorded at testing the 5% rubber granulate (6.50%). In contrast, the highest inhibition values were recorded at testing the 75% rubber granulate (62.36%). In the other rates of rubber granulate, the values of inhibition ranged from 26.37% to 48.73%. The values were related to the control sample and were also used to plot the diagram in Figure 2. The use of higher rates of rubber granulate on the OECD soil significantly inhibited the growth of *Sinapis alba* L. An error analysis of the experimental procedure in the following graph was plotted by the application of error lines in the Microsoft Excel application. The analysis helps evaluate the measurement accuracy and reliability despite the random nature of the measurement.

The results demonstrate that in terms of phytotoxicity, rubber granulate from waste tires with the size of fractions 1–3 mm has an environmental impact. Residues of chemical substances (most frequently excess zinc) from the original tires remain also in the rubber granulate which may impair the quality of soil and plants if applied on the soil in the form of mulch. In addition, rubber granulate has a very long decomposition time and does not provide nutrients to plants unlike organic mulch.

Chalker-Scott (2018) and Jared (2001) say that rubber mulch is offered by the manufacturer as a durable, safe and non-toxic plant material that retains irrigation (Chalker-Scott 2018, Jared 2001). But the opposite is true. People do not realize the risks associated with their use. Toxic substances (mentioned in the Introduction) contained in rubber leach out during decomposition and contaminate soil and plants. However, some substances bioaccumulate and act gradually over time (Chalker-Scott 2018). This is confirmed in their study by William and Shenker (2016), who performed a hydroponic experiment with cucumbers, to which rubber granulate was added. They tested at 0%, 5% and 10% granulate concentration. The soil to which the granulate was applied showed a significant reduction in plant growth (toxicity) compared to the control sample. This condition was mainly caused by the leachability of heavy metals in granulate (especially zinc), which accumulated both in plants and in the soil itself (William and Shenker 2016).

Figure 2 Graphical illustration of the results of Sinapis alba L. root growth inhibition in the tests of rubber granulate phytotoxicity, Brno, Czech Republic, 2021



Legend: 5% – 75% samples with the given rates of granulate

In spite of its advantages mentioned in the chapter of Material and Methods, rubber granulate should not be therefore used as a substitute for organic mulch. As to the disposal of waste tires, the most appropriate option of its valorization is its energy use as fuel in cement works instead of brown coal.

CONCLUSION

Rubber granulate which arises from recycling waste tires represents a potential environmental risk in terms of phytotoxicity. In the production of tires, no technologies exist so far that would be able to reduce the toxicity of rubber and maintain at the same time all singular functions of tires (resistance, direction guidance, permanence irrespective of climatic conditions, shock absorption etc.). Tire manufacturers currently try to make tires that would be more sustainable for the environment and to reduce the consumption of natural resources for their production.

The phytotoxicity of rubber granulate from waste tires was determined in laboratory conditions according to the methodology of Phytotoxkit^{TM}. All tested rates of the rubber granulate (5%, 10%, 25%, 50% and 75%) exhibited the growth inhibition of *Sinapis alba* L. Being phytotoxic already at low rates, rubber granulate should not be used as a substitute for organic mulch.

REFERENCES

Birkholz, D.A. et al. 2003. Toxicological Evaluation for the Hazard Assessment of Tire Crumb for Use in Public Playgrounds. Journal of the Air & Waste Management Association [Online], 53(7): 903–907. Available at: https://pubmed.ncbi.nlm.nih.gov/12880077. [2021-08-08].

Cao, W. 2007. Study on properties of recycled tire rubber modified asphalt mixtures using dry process. Construction and Building Materials [Online], 21(5): 110–116. Available at: https://www.sciencedirect.com/science/article/pii/S095006180600016X?via%3Dihub. [2021-08-08].

Chalker-Scott, L. 2018. Is Rubber Mulch Safe for My Garden? [Online], Available at: https://www.motherearthnews.com/nature-and-environment/is-rubber-mulch-safe-zm0z14onzsor. [2021-10-23].

Jared, D.M. 2001. Evaluation of Recycled Rubber Mulch Products. Georgia DOT, Office of materials and research [Online], 1–38. Available at:

http://g92018.eos-intl.net/ELIBSQL14_G92018_Documents/9808.PDF. [2021-10-23].

Kizlink, K. 2014. Waste: collection, processing, recovery, disposal, legislation. 3rd ed., Brno: Academic Publishing house CERM, pp. 183–204.

Kumaran, G.S. et al. 2008. A Review on Construction Technologies that Enables Environmental Protection: Rubberized Concrete. American Journal of Engineering and Applied Sciences [Online], 1(1): 40–44. Available at: https://thescipub.com/abstract/ajeassp.2008.40.44. [2021-08-08].

Kuraš, M. 2014. Odpady a jejich zpracování. 1st ed., Chrudim: Vodní zdroje Ekomonitor.

Li, X. et al. 2010. Characterization of substances released from crumb rubber material used on artificial turf fields. Chemosphere. [Online], 80(3): 279–285. Available at: https://www.sciencedirect.com/science/article/pii/S0045653510004285?via%3Dihub. [2021-08-07].

Llompart, M. et al. 2013. Hazardous organic chemicals in rubber recycled tire playgrounds and pavers. Chemosphere [Online], 90(2): 423–431. Available at: https://www.sciencedirect.com/science/article/pii/S0045653512009848. [2021-08-07].

Nelson, S.M. 1994. Identification of tire leachate toxicants and a risk assessment of water quality effects using tire reefs in canals. Bulletin of Environmental Contamination and Toxicology [Online], 52: 574–581. Available at: https://link.springer.com/article/10.1007%2FBF00194146#citeas. [2021-08-10].

Ramos, G. et al. 2011. The recycling of end-of-life tyres. Technological review. Revista De Metalurgia [Online], 47(3): 273–284. Available at: https://revistademetalurgia.revistas.csic.es/index.php/revistademetalurgia/article/view/1197/1208. [2021-08-07].

Price, W. et al. 2006. Waste tire recycling: environmental benefits and commercial challenges. International Journal of Environmental Technology and Management [Online], 6: 362–374. Available at: http://www.inderscience.com/offer.php?id=9001. [2021-08-05].

Sathiskumar, C. et al. 2019. Recycling of waste tires and its energy storage application of by-products–a review. Sustainable Materials and Technologies [Online], 22. Available at: https://www.sciencedirect.com/science/article/pii/S2214993719300636. [2021-08-06].

Schnick, R.A. et al. 1982. Mitigation and enhancement techniques for the Upper Mississippi River system and other large river systems. Resource publication. Washington, DC: U.S. Department of the Interior, Fish and Wildlife Service, pp. 102.

Šourková, M. et al. 2021. Phytotoxicity of Tires Evaluated in Simulated Conditions. Environments [Online], 8(6): 49. Available at: https://www.mdpi.com/2076-3298/8/6/49#cite. [2021-08-03].

Vaverková, M.D. et al. 2019. Landfill leachate effects on germination and seedling growth of hemp cultivars (*Cannabis Sativa* L.). Waste and Biomass Valorization [Online], 10: 369–376. Available at: https://link.springer.com/article/10.1007/s12649-017-0058-z. [2021-08-11].

Wik, A., Dave, G. 2005. Environmental labeling of car tires—toxicity to Daphnia magna can be used as a screening method. Chemosphere [Online], 58(5): 645–651. Available at: https://www.sciencedirect.com/science/article/pii/S004565350400760X. [2021-08-10].

William, M., Shenker, M. 2016. Pulverized Tires as Soil Amendment for Plant Growth. International Journal of Scientific & Engineering Research [Online], 7(6): 161–167. Available at: https://www.ijser.org/researchpaper/Pulverized-Tires-as-Soil-Amendment-for-Plant-Growth.pdf. [2021-10-23].

Name of publication:	MendelNet 2021
	Proceedings of 28th International PhD Students Conference
Editors:	Assoc. Prof. Ing. Radim Cerkal, Ph.D. Ing. Natálie Březinová Belcredi, Ph.D. Ing. Lenka Prokešová
Publisher:	Mendel University in Brno Zemědělská 1665/1 613 00 Brno Czech Republic
Year of publication:	2021
Number of pages:	492
ISBN:	978-80-7509-821-4

Contributions are published in original version, without any language correction.