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Original Research

Yeasts in the Soils of the Alytus Region after the Fire of the Tyre Processing Plant

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Abstract

The study aimed to investigate the prevalence and biodiversity of yeasts in the soil and water samples of the city and district of Alytus after a fire at a tire processing plant. 11 soil and 3 water samples were taken each year for the periods of 2019-2021. The yeast count in the soil samples ranged from 10^2 to 10^5 CFU/g. In water samples, the count of yeasts varied between 0.00-10^3 CFU/g. A total of 114 yeast isolates were isolated from the soil samples and 46 yeast species were identified. No statistically significant changes in the phyla distribution of yeast species isolated from the soil were observed during the different study periods. A total of 25 yeast isolates were isolated from the water samples and 14 yeast species were identified. Statistically significant differences in the phyla distribution of yeast species isolated from water samples were observed. The obtained research data showed that the results of all chemical soil tests do not exceed the limit values specified in Lithuanian legal acts. The obtained results of the study allow us to state that the fire at the tyre recycling plant did not affect the prevalence and biodiversity of soil yeasts, but the possibility that the effect will be noticed after a longer period of time cannot be ruled out. We believe that this study significantly contributes to the dissemination of information about the fire at the tire recycling plant and its impact on the environment, brings more clarity and reduces doubts about the possible impact of pollution on the environment.

Keywords: yeast, prevalence, biodiversity, tyre fire, pollution

Introduction

Alytus is the largest city in southern Lithuania, the sixth largest city in the country. Alytus is located by the Nemunas river, 65 km south of Kaunas and 108 km southwest of Vilnius. Alytus is the largest city of Dzūkija, considered the informal capital of this ethnographic region. The Nemunas flows through Alytus from the southeast to the northwest. The city is divided into two parts: Alytus I is located on the right bank of the Nemunas, and Alytus II is located on the left (most of the current city is on the river's left bank) [1].

On October 16, 2019, a fire broke out at a tire recycling plant in the industrial district of the city of Alytus, Lithuania. Tires of light and heavy vehicles were collected, stored, and processed in the company's warehouses. About 5,000 tons of used and recycled tires

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were stored in the premises of this company, and the area of the premises was 2,000 square meters. The fire was extinguished only on October 18, but large areas of the fire site continued to emit harmful fumes into the environment for 7 days until October 25. About 2,000 tons of tires burned during the fire. The smoke released during the fire was carried by the wind and moved toward the village of Miklusėnai. This fire is considered one of the biggest ecological disasters in Lithuania [2].

Lithuania faces the problem of accumulation of used tire waste. In 2018 28,100 t of tires were supplied to the Lithuanian market, 14,556 t (52%) of all sorted tire waste was exported, 6220 t (22%) – was processed, 3670 t (13%) – was used for energy extraction, and 971 t (3%) – processed (i.e. used, ready for use or disposal, burned in a fire) and the rest of the tires remained unused [3].

The composition of tires includes such materials as rubber/elastomers (47% – in car tires, 45% – in truck tires), carbon black (21.5% and 22% respectively), metal (16.5% and 25% respectively), textile (5.5% in car tires), zinc oxide (1% and 2% respectively), sulfur (1% each), additives (clays, etc., 7.5% and 5% respectively). Most of the composition (74% of car tires and 67% of truck tires) consists of carbon-based materials [4].

A tire fire releases black toxic smoke and large amounts of toxic solids and gases into the environment. These substances pose a great danger to society and the environment. Also, these substances can enter the soil, groundwater and surface water. By-products of burning tires can include smoke, pyrolitic oil, ash, black carbon and many other harmful chemicals such as polycyclic aromatic hydrocarbons, benzenes, naphthalenes, toluene, ethyl benzene, anthracene, thiazoles, dioxins, furans, amines, cadmium, chromium, nickel, zinc, etc. [5].

Heavy metals released into the environment during a fire can have toxic effects and the ability to bioaccumulate. Polycyclic aromatic hydrocarbons have carcinogenic properties. These pollutant properties affect human health, soil, and water ecosystems [2, 6].

Pollution with polycyclic aromatic hydrocarbons and heavy metals significantly affects microorganisms of various systematic groups and their abundance. It has been observed that microorganisms of groups such as actinomycetes, molds, and yeasts are more resistant to the negative effects of the urbanized environment. Combined high-concentration pollution with PAHs and heavy metals significantly reduces the abundance of soil microbial communities compared to pollution with PAHs alone. This proves that high concentrations of heavy metals in soil reduce the adaptation potential of microbial communities. Also, the communities of microorganisms change depending on the sensitivity of microorganisms to these compounds. Changes in microbial communities lead to replacing more sensitive groups of microorganisms with more resistant ones [7].

Anthropogenic activities and related events have a significant impact on climate change. For example, Ahmeti et al. (2022) found that coal with a calorific value of less than 6500 kJ/kg used for electricity and heat production should not be used due to its high environmental impact and promotion of climate change. Also, not only processes that use coal but also its extraction contribute to pollution [8].

Agricultural activities also have a great influence. Līcīte et al. indicate that cultivated land areas in the world occupy about 12-14% of the non-freezing land area. About 37% of the area is covered by grasslands, a significant part of which is agricultural land with organic soil. Different land management practices affect the amount of carbon stored in the soil, leading to higher carbon dioxide emissions. In some countries (Latvia, Ireland, the United Kingdom and Finland), including Lithuania, greenhouse gas emissions from managed organic soil account for more than a fifth of the country's total emissions. Also, farming strongly changes soil structure and properties (pH, fertility, amount of organic matter, etc.), which undoubtedly determines the species composition and abundance of soil microorganism communities [9].

According to Mironova et al. according to the obtained data, the restrictions introduced during the COVID-19 pandemic had a significant impact on soil quality. These restrictions led to a decrease in pollution caused by cars, industry and other anthropogenic activities. The reduction in pollution contributed to a 10% change in soil pH and the pH changed from alkaline to neutral. The amounts of nutrients (NPK) also changed, the amount of nitrogen increased by 25%, phosphorus by 30%, and potassium almost doubled. The biggest increase was observed in industrial and road areas. Changes in these parameters could also be associated with changes in soil pH, which determine the solubility and ionization of substances, and these processes affect the abundance, species composition and enzymatic activity of soil biota. Also, the amounts of lead and other heavy metals in urban soil decreased significantly (almost twice) [10].

Detecting certain yeast species in the soil can help suspect soil contamination with various pollutants. Various yeast species belonging to the genera *Rhodotorula*, *Candida*, *Rhodosporidium*, *Pichia*, *Trichosporon*, and *Exophiala* are found in soil contaminated with hydrocarbons [11]. *Aureobasidium pullulans*, *Candida albicans*, *Candida intermedia, Candida magnoliae, Candida parapsilosis, Candida sobophila, Candida tropicalis, Candida zeylanoides, Cryptococcus laurentii, Cystofilobasidium slooffiae, Diutina rugosa, Exophiala dermatitidis, Komagataella pastoris, Meyerozyma guilliermondii, Minimelanolocus obscurus, Pichia fermentans, Pichia kudriavzevii, Rhodotorula mucilaginosa* and *Saccharomyces cerevisiae* yeast species are found in soils contaminated with zinc compounds and other contaminants [12].

Lithuanian scientists, as well as the public, have doubts about the possible impact of the tire recycling plant fire on the environment and human health. To dispel these doubts, it is necessary to carry out scientific research that would confirm or deny the possible impact of pollutants released into the environment during the fire on the environment and human health.

The aim of this study was to assess the prevalence and biodiversity of yeasts in the soil of the areas of the city and district of Alytus affected by the fire of the tire processing plant. This study was necessary because there were many discussions and disagreements both in the society and among Lithuanian scientists, whether the fire at the tire processing plant had a significant impact on the environment. There are quite a few investigations of this kind in Lithuania, as events related to fires and other accidents are relatively rare. This study contributes to the general dissemination of knowledge about a tyre recycling plant fire and its possible environmental impact.

Material and Methods

Sample Collection and Isolation of Yeasts

Soil samples (100 g) from surface soil (0-15 cm depth) and water (500 ml) were collected from 14 different locations in the territories of Alytus district possibly affected by the fire at the tire processing plant (Fig. 1, Table 1).

Samples were taken from 3-4 random locations per plot, mixed, and transferred into sterile plastic bags. Samples were stored in a refrigerator at 5.0ºC before delivery to the laboratory. In the lab, stones and other unwanted soil debris were removed using 3 millimeters sieve. The cultivable yeasts were isolated and enumerated using the serial dilution plate technique. 0.1 ml of the suspension was inoculated into Rose Bengal CAF Agar (Liofilchem, Italy) plates. The plates were incubated in the incubator at 19.0±1.0ºC. Yeast colonies were counted after 3-4 days of incubation [13-15]. After incubation yeast quantity was expressed as colony forming units per gram (CFU g^{-1} and was calculated according to the formula:

$$n = \frac{a \cdot b \cdot c}{d}$$

where
n – the number of yeast colony forming units (CFU) in 1 g of dry substrate,
a – the number of grown colonies in the plate,
b – suspension dilution factor,
c – the volume of inoculated suspension (ml),
d – the weight of dry soil (g).

The results are presented after calculating the average from the sample repetitions.

Yeast Identification

Yeast isolates were identified by standard phenotypic methods and using the Matrix-assisted laser desorption/ionization-time of flight (MALDI-TOF) mass spectrometry method (Bruker, USA). Samples were prepared by formic acid extraction protocol [16, 17].

Chemical Parameters of the Soil

Chemical assays of the soil were performed at the Agrochemical Research Laboratory of the Lithuanian Research Centre for Agriculture and Forestry, according

Fig. 1. Map of sampling locations.

Table 1. Soil sampling locations, their descriptions and coordinates.

Sample No.	Sampling location	Sample type	Location of the sampling points
1	Alytus city, Kindergarten "Obelėlė", Šaltinių str. 3	Urban soil	54.408696, 24.021765
2	Alytus city, Kindergarten "Putinėlis", Kalnėnų str. 2	Urban soil	54.415644, 24.021698
3	Alytus Medical Rehabilitation and Sports Center, Pramonės str. 9	Urban soil	54.421715, 24.009385
4	Alytus, air intake shaft, at the scene of the fire, Pramonės str. 1	Water used to extinguish the fire	54.418812, 24.011488
5	Alytus city, Textile chemical cleaning facility, Naujoji str. 114	Water from decommissioned textile chemical treatment facilities used to extinguish the fire	54.419887, 24.007460
6	Alytus city, Gulbynė lake, Putinų str. 1	Lake water used to extinguish the fire	54.415157, 24.013166
7	Alytus city, Private yard, Putinų str. 1	Agricultural soil	54.415843, 24.014347
8	Raudonikiai village, private nursery, Sodžiaus str.	Agricultural soil	54.454886, 23.976735
9	Taukotiškės village, private homestead, Taukotiškių str. 6	Agricultural soil	54.484511, 23.996358
10	Mardosai village, private farm, Šaltinių str.	Agricultural soil	54.497112, 23.922139
11	Užubaliai village, private homestead, Pavasario str.	Agricultural soil	54.425951, 23.979384
12	Alytus city, Kindergarten "Nykštukas", Studentų str. 4	Urban soil	54.402114, 24.063791
13	Venciūnai village, Prie-school education department, Alyvų str. 4	Urban soil	54.385507, 24.134704
14	Klausiškės village, Varsos str.	Forest soil	54.361864, 24.215388

to the requirements of ISO 10390:2021, ISO 10694:1995, ISO 11261-1995, LVP D-13:2021, LVP D-09:2021 standards [18-20].

Statistical Analysis

Data analysis was performed with IBM SPSS Statistics 27 software (IBM Corporation, USA). The obtained results were considered statistically significant when $P<0.05$. The frequency of detection (i.e. the percentage expression between the samples in which the yeast species was detected and the total number of tested samples, AD) was calculated according to the formula [21]:

$$A = \frac{B}{C} \cdot 100\%$$

where
A - the frequency of detection (FD, %),
B - the number of samples in which the species was detected,
C - total number of samples.

The proportion of yeast species among all yeast species isolated was calculated according to formula:

$$A = \frac{B}{C} \cdot 100\%$$

where
A - the proportion of yeast species among all yeast species isolated (%),
B - the number of isolates of a specific yeast species,
C - total number of yeast isolates.

Results and Discussion

Changes in the Number of Yeasts during Different Study Periods

The yeast count in the soil samples ranged from 10^2 to 10^5 CFU/g. The highest number of yeasts in 2019 was determined in samples No. 2 ($4.80\pm0.11 \times 10^5$ CFU/g) and No. 10 ($9.54\pm0,36 \times 10^5$ CFU/g), in 2020 – in samples No. 10 ($2.97\pm0.12 \times 10^5$ CFU/g) and No. 11 ($2.63\pm0.11 \times 10^5$ CFU/g), and in 2021 – in sample No. 1 ($1.60\pm0.00 \times 10^5$ CFU/g). The lowest number of yeasts in 2019 was determined in sample No. 3 ($(2.51\pm0.11) \times 10^3$ CFU/g), in 2020 – in sample No. 1 ($1.00\pm0.00 \times 10^3$ CFU/g), in 2021 – in sample No. 10 ($9.00\pm0.12 \times 10^2$ CFU/g)

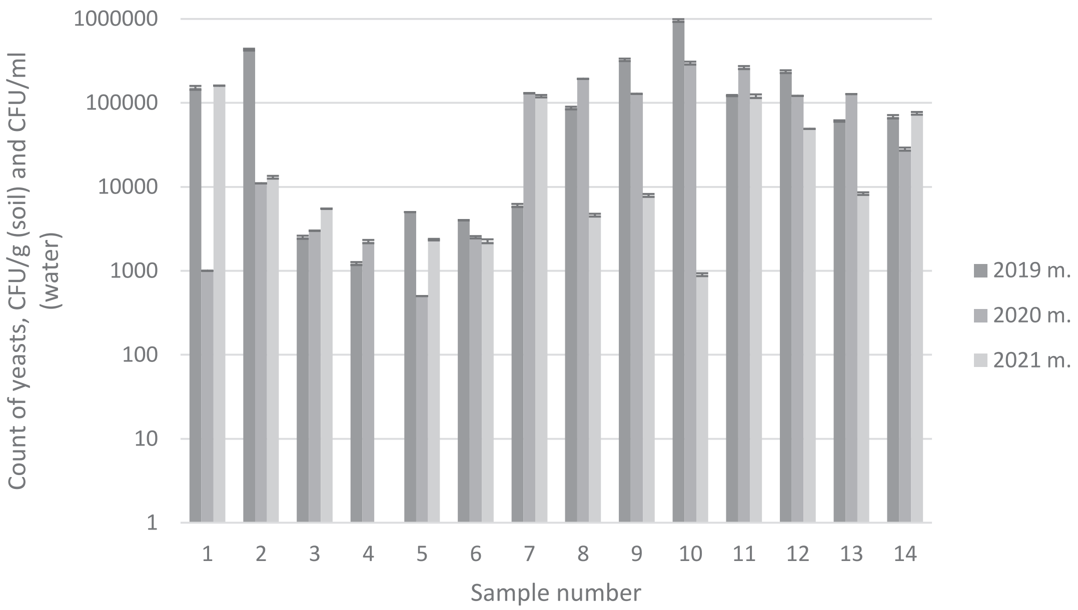

Fig. 2. Changes in the number of yeasts in the soil samples analyzed in 2019-2021 (the description of the soil samples and explanations of the numbering are listed in Table 1).

(Fig. 2). There were no statistically significant differences between the distance of the soil sampling site to the epicenter of the fire and the count of yeasts. Therefore, it can be stated that the distance to the epicenter of the fire and the fire itself did not affect the changes in the count of yeasts.

According to literature sources, the direct effect of fire on soil microorganisms is significant, it causes a decrease in their biomass. This usually happens at the expense of yeast and microscopic fungi. Some authors describe a greater effect on bacteria in the top 15 cm of soil. However, these observations apply only to soil at the site of the fire and not to the soil exposed to airborne pollutants [22].

According to the literature, the count of yeast in soil usually does not exceed a few thousand cells per gram of soil, but higher cell numbers reaching millions of cells per gram of soil are sometimes found. More yeasts are known to be found in the soil near plants that have fruits, as decaying fruit enters the soil and can become a nutrient-rich medium for yeast growth. Also, various organic compounds secreted through plant roots are assimilated by yeast. For these reasons, higher yeast populations are found in the rhizosphere [23].

In water samples (No. 4-6), the number of yeasts varied between 0.00-10^3 Yeast counts were similar in all samples except sample No. 4 (0.00) in 2021 and sample No. 5 ($5.00 \pm 0.00 \times 10^2$) in 2020. According to the literature, the number of yeasts in various water bodies reaches up to 10^1-10^2 CFU/ml and more, depending on the type of water body and its level of contamination [24]. A slightly higher yeast count was obtained in this study. This could have been caused by a large amount of soil entering the water during firefighting.

Yeast Biodiversity during the Study Period

In this study, a total of 114 yeast isolates were isolated from the soil samples and 46 yeast species were identified.

The results of the study showed that in 2019 the majority of yeast species isolated from the soil belonged to the *Ascomycota* phylum (58.3%), in 2020 to the *Basidiomycota* phylum (51.2%), and in 2021 the species were equally distributed in both phyla. Overall data for the entire study period showed that the majority of isolated yeast species belonged to the *Ascomycota* phylum (53,7%). No statistically significant changes in the distribution of yeast species by phyla were observed during the different study periods (Fig. 3).

It was found that the most abundant isolated yeast species consisted of *Solicoccozyma terrea* (45.45%), *Komagataella pastoris* (30.30%), *Candida tropicalis* (18.18%), *Cutaneotrichosporon mucoides* (18.18%), *Issatchenkia orientalis* (15.15%) *Saccharomycopsis schoenii* (15.15%), *Candida glabrata* (12.12%) and *Rhodotorula mucilaginosa* (12.12%) yeast species (Table 2). There were no statistically significant differences between the distance of the soil sampling site to the epicenter of the fire and the species composition of yeasts. Therefore, it can be stated that the distance to the epicenter of the fire and the fire itself did not affect the yeast biodiversity.

Yeasts of the genera *Cystobasidium*, *Rhodotorula*, *Rodosporidiobolus*, *Sporobolomyces* and *Vishniacozyma* are found in the soil and are associated with plant surfaces. Indigenous soil yeasts can include *Barnettozyma*, *Cyberlindnera*, *Kazachstania*, *Schwanniomyces*, *Cryptococcus*, *Trichosporon*, *Goffeauzyma*, *Naganishia*,

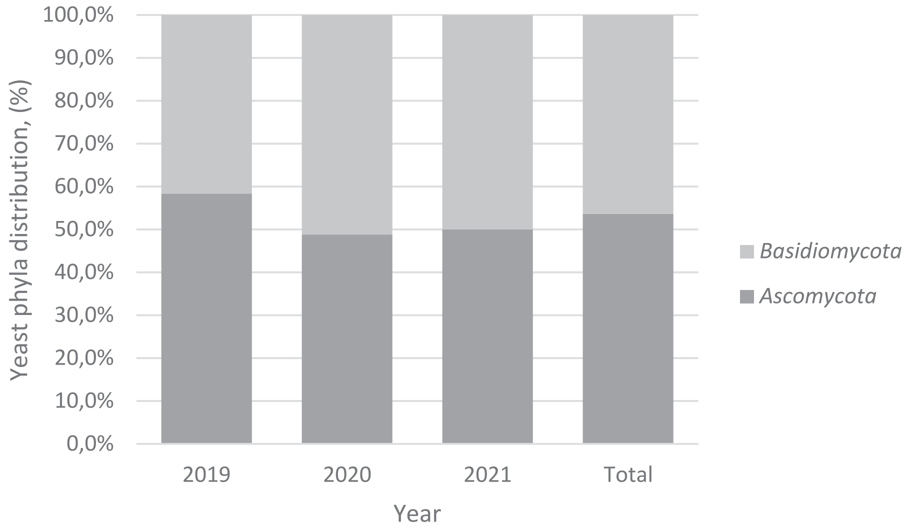

Fig. 3. Affiliation of yeast phyla distribution from the research period.

Solicoccozyma, *Vanrija* and *Tausonia*. Yeasts of the *Basidiomycetes* class are more commonly found in forest soils. The yeast species of this class most often found in soil belong to the *Cryptococcus* genus. Yeasts of the genera *Barnettozyma*, *Cyberlindnera*, *Kazakhstania* are mostly found in grassland and agricultural soils. Various species of *Candida* yeasts are also found in grassland and agricultural soils. *Papiliotrema laurentii, Apiotrichum dulcitum, Apiotrichum laibachii, Vanrija humicola, Naganishia albida, Goffeauzyma gastrica, Solicoccozyma terrea, Tausonia pullulans* are the most common yeast species isolated from different types of soil [23].

Most of the yeast species we isolated from the soil coincided with the yeast genera and species indicated by other scientists. Based on the data provided by this author, it can be said that we managed to isolate both indigenous yeast species and yeast species associated with anthropogenic activities. The most common yeast species isolated in this study was *Solicoccozyma terrea* (45.45%). This species of yeast is also classified by other scientists as one of the most commonly found species in the soil [23].

A total of 25 yeast isolates were isolated from the water samples and 14 yeast species were identified. Statistically significant differences ($P<0.05$) were observed in the phyla distribution of yeast species isolated from water at different study periods. In 2019 and 2020, the largest part of isolated yeast species were *Basidiomycota* phylum yeasts (92.3% and 75.0% respectively), and in 2021 - *Ascomycota* phylum yeasts (80.0%). Examining the overall distribution of yeast species by phyla in all study periods, it was observed that the majority of yeast species belonged to the *Basidiomycota* phylum (73.1%).

It was found that the most abundant isolated yeast species consisted of *Cutaneotrichosporon mucoides* (12.12%), *Cystofilobasidium macerans* (9.09%) and *Rhodotorula mucilaginosa* (9.09%) (Table 3).

Mostly *Candida*, *Clavispora*, *Cyberlindnera*, *Cryptococcus*, *Debaryomyces*, *Hanseniaspora*, *Kluyveromyces*, *Metschnikowia*, *Meyerozyma*, *Pichia*, *Rhodotorula*, *Saccharomyces*, *Torulaspora* and *Yarrowia* are found in surface water bodies of the temperate climate zone. Yeasts found in water bodies can be associated with anthropogenic water pollution [24].

Assessment of Soil Chemical Parameters and Their Influence on Yeast Abundance and Biodiversity in the Context of Fire-Induced Pollution

According to soil agrochemical tests, the pH of the tested soil samples ranged from 4.4 (very acidic soil) to 7.6 (close to neutral soil) [25]. The amounts of mobile phosphorus ranged from 50 mg/kg (very low phosphorus soil) to 1525 mg/kg (very high phosphorus soil), and mobile potassium from 68 mg/kg (low phosphorus soil) to 1050 mg/kg (very high phosphorus soil) (Table 4).

The carbon to nitrogen ratio (C:N) in this study ranged from 9:1 to 18:1. The highest C:N ratio was determined in samples no. 3, 8, 13 and 14. Since these soil samples were taken in the territories of a flower garden or fields of cultivated soil the higher C:N ratio could be due to the higher amount of organic matter in the soil due to the due to organic materials, compost and peat used for fertilization and soil improvement purposes. The count of yeasts in soil sample no. 3 was around 10^3 CFU/g in all periods of the study and was slightly lower than in the other studied soil samples (from 10^3 to 10^5 CFU/g in whole period of the study). Yeast count in samples no. 8, 13 and 14 was similar to other soil samples studied and varied from 10^3 to 10^5 CFU/g in different study periods. No statistically significant differences in the count of yeasts and their biodiversity were observed in comparison with other studied soil samples. A typical soil C:N ratio is around

Table 2. Proportion of yeasts species among all yeast species isolated from soil samples and their frequency of detection (%).

Yeast species	The proportion of yeast species among all yeast species isolated (%)	Frequency of detection (%)
Apiotrichum laibachii	1.75	6.06
Aureobasidium pullulans	0.88	3.03
Candida albicans	0.88	3.03
Candida blankii	0.88	3.03
Candida boidinii	0.88	3.03
Candida glabrata	**3.51**	**12.12**
Candida intermedia	1.75	6.06
Candida nemodendra	1.75	6.06
Candida parapsilosis	2.63	9.09
Candida tropicalis	**5.26**	**18.18**
Cyberlindnera jadinii	0.88	3.03
Clavispora lusitaniae	1.75	6.06
Goffeauzyma gastrica	1.75	6.06
Cryptococcus gattii	0.88	3.03
Cryptococcus saitoi	2.63	9.09
Cutaneotrichosporon jirovecii	0.88	3.03
Cutaneotrichosporon mucoides	**5.26**	**18.18**
Diutina catenulata	0.88	3.03
Cystofilobasidium macerans	0.88	3.03
Filobasidium magnum	0.88	3.03
Intersonilia perplexans	2.63	9.09
Issatchenkia orientalis	**4.39**	**15.15**
Kazachstania unispora	0.88	3.03
Komagataella pastoris	**8.77**	**30.3**
Lachancea thermotolerans	0.88	3.03
Lecytophora hoffmanii	0.88	3.03
Meyerozyma guilliermondii	2.63	9.09
Metschnikowia kunwiensis	1.75	6.06
Metschnikowia pulcherrima	0.88	3.03
Naganishia albida	1.75	6.06
Naganishia diffluens	2.63	9.09
Naganishia uzbekistanensis	1.75	6.06
Papiliotrema flavescens	0.88	3.03
Pichia fermentans	1.75	6.06
Pichia occidentalis	1.75	6.06
Rhodotorula diobovata	0.88	3.03
Rhodotorula mucilaginosa	**3.51**	**12.12**
Saccharomyces cerevisiae	0.88	3.03
Saccharomycopsis schoenii	**4.39**	**15.15**
Solicoccozyma terrea	**13.16**	**45.45**
Trichosporon asahii	0.88	3.03
Trichosporon inkin	0.88	3.03
Udeniomyces puniceus	0.88	3.03
Vanrija humicola	0.88	3.03
Zygoascus hellenicus	0.88	3.03
Zygosaccharomyces rouxii	1.75	6.06

10:1 - 12:1. It depends on the type of soil, the fertilizers used, and the amount of organic matter entering the soil. The C:N ratio is an important indicator for determining soil quality and microbiological activity and affects the decomposition of organic matter. The C:N ratio directly controls the transformation of organic matter in the soil. Organic matter with a higher C:N ratio remains in the soil for a longer time. As this ratio decreases, the processes shift towards mineralization [26, 27].

In all studied soils, the values of chemical research results can be associated with the influence of farming and other anthropogenic activities. This was also confirmed by chemical tests of the soil carried out by the Lithuanian Geological Service [28]. Therefore, it can be stated that the fire at the tyre processing plant did not have a significant impact on the soil microorganism communities and chemical composition.

A higher C:N ratio in some soil samples did not significantly affect the abundance of yeasts in the studied soil. The count of yeasts remained similar in all soils, only slight variations were observed.

According to the Lithuanian Geological Service data, no pollution with harmful products from tyre fires and other possible sources was found in the soil of the city and district of Alytus. The amounts of all tested chemicals in the soil do not exceed the maximum permissible limit values valid in the residential areas of Lithuania [28].

After a landfill fire in Poland, significant concentrations of harmful polycyclic aromatic hydrocarbons were found in soil samples. This has been observed both in the soil and in the burnt waste left after the fire, which poses a risk to soil micro-organisms, plants and animals. No increase in the concentration of heavy metals was detected in the soil. This may be due to the fact that during a tire and black rubber fire, due to the high temperature and oxidation processes, organic pollutants are mostly released [29]. In the case

Table 3. The proportion of yeasts species among all yeast species isolated from water samples and their frequency of detection (%).

Yeast species	The proportion of yeast species among all yeast species isolated (%)	Frequency of detection (%)
Aureobasidium pullulans	8.00	6.06
Candida haemulonii	4.00	3.03
Clavispora lusitaniae	4.00	3.03
Cystofilobasidium macerans	**12.00**	**9.09**
Cryptococcus gattii	4.00	3.03
Cutaneotrichosporon mucoides	**16.00**	**12.12**
Issatchenkia orientalis	4.00	3.03
Meyerozyma guilliermondii	4.00	3.03
Moesziomyces aphidis	8.00	6.06
Rhodotorula diobovata	8.00	6.06
Rhodotorula mucilaginosa	**12.00**	**9.09**
Saccharomycopsis schoenii	4.00	3.03
Solicoccozyma terrea	8.00	6.06
Vanrija humicola	4.00	3.03

of the fire in Alytus, all the effects of the fire and waste were removed as soon as it was extinguished, only smoke and water used to extinguish the fire entered the environment. The conducted soil tests did not show any contamination with compounds formed during the fire.

According to the data of our research and the research conducted by the Lithuanian Geological Service, pollution by harmful substances that entered the environment during the fire and its negative effect on the soil microbiota were not detected in the soil. On the contrary some studies state that according to the obtained data, soil pollution with large amounts of polycyclic aromatic hydrocarbons, especially of low molecular weight, was observed at the fire site. The same situation was observed in the village of Miklusėnai, which was probably the most affected by the pollutants released during the fire. Also, zinc was found at the fire site and in the village of Miklusėnai, which confirms the hypothesis that this village was the most affected. All the data obtained by the authors suggests that the fire contributed to the environmental pollution of the surrounding areas [2]. During the period of this study, no tangible impact of the pollution caused by the fire of the tire processing plant on the soil microbiocenosis was detected, but the possibility that such an impact will be observed and recorded in the future cannot be ruled out.

Since there is still the potential for adverse environmental effects from fire releases, it is crucial that more studies like ours are conducted to monitor the long-term effects of fire on the environment. We believe that the results of our research could encourage other Lithuanian scientists to more actively study the effects of the fire that occurred in the city of Alytus on the microbiological, chemical and other properties of the soil.

The strength of this study is that not many such studies have been conducted in Lithuania, but this causes problems when comparing the obtained results at the national level. We would recommend conducting more research related to the fire in Alytus and thereby increasing the availability of information. In the future, it is planned to continue the research of the soil and water bodies of the Alytus region, to carry out a wider microbiological and chemical analysis of them.

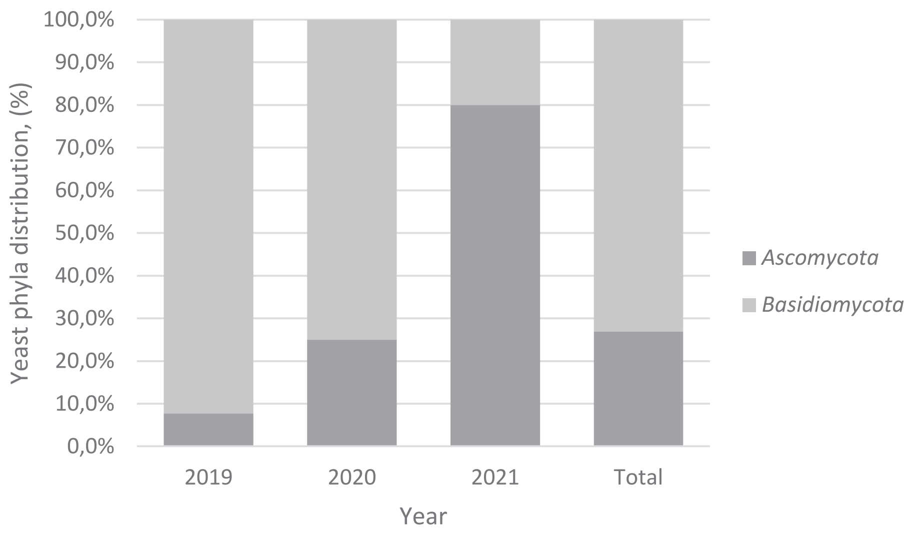

Fig. 4. Affiliation of yeast phyla distribution from the research period.

Table 4. Results of soil chemical tests.

Sample No.	pH	Mobile phosphorus (P_2O_5) mg/kg	Mobile potassium (K_2O) mg/kg	Organic carbon (C) mg/kg	Total nitrogen (N) %	Total sulfur (S) mg/kg	C:N ratio
1	6.4	184	121	4.60	0.476	767	10:1
2	6.8	254	56	0.86	0.080	136	11:1
3	6.8	445	112	6.55	0.360	580	18:1
7	6.8	855	126	1.98	0.181	229	11:1
8	7.6	353	630	2.78	0.208	279	13:1
9	6.8	1525	1050	3.37	0.343	441	10:1
10	6.6	905	141	2.00	0.193	224	10:1
11	7.1	270	310	1.64	0.178	197	9:1
12	7.2	491	229	2.25	0.214	322	10:1
13	6.8	608	145	4.32	0.300	512	14:1
14	4.4	50	68	1.84	0.140	141	13:1

Conclusions

The results of the research showed that the yeast count in the soil samples ranged from 10^2 to 10^5 CFU/g. In water samples the count of yeasts varied between 0.00-10^3 CFU/g. All of the yeast counts obtained are consistent with normal soil yeast counts, so it can be said that the tire recycling plant fire did not affect the yeast counts.

A total of 114 yeast isolates were isolated from the soil samples and 46 yeast species were identified. No statistically significant changes in the phyla distribution of yeast species isolated from the soil were observed during the different study periods. It was found that the most abundant isolated yeast species were *Solicoccozyma terrea*, *Komagataella pastoris*, *Candida tropicalis*, *Cutaneotrichosporon mucoides*, *Issatchenkia orientalis, Saccharomycopsis schoenii*, *Candida glabrata* and *Rhodotorula mucilaginosa*.

A total of 25 yeast isolates were isolated from the water samples and 14 yeast species were identified. Statistically significant differences in the phyla distribution of yeast species isolated from water samples were observed. Gradually, the *Basidiomycota* phylum's yeasts species replaced the *Ascomycota* phylum's yeast species. However, it is not possible to directly associate these changes with possible water pollution by hazardous substances released during the fire, nor to rule out the possible influence of this pollution. It was found that the most abundant isolated yeasts were *Cutaneotrichosporon mucoides*, *Cystofilobasidium macerans* and *Rhodotorula mucilaginosa.* These yeast species are commonly found in normal soils and water. Therefore, it can be said that the chemicals and other particles that entered the environment during the fire did not affect the yeast biodiversity.

The obtained research data showed that the results of all chemical soil tests do not exceed the limit values specified in Lithuanian legal acts. The results of agrochemical tests show that the soil has normal structure and properties, and deviations from the limit values can be attributed to local human agro-cultural activities.

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Conflicts of Interest

The authors declare no conflict of interest exists.

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