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SCIENTIFIC FOUNDATIONS OF ISOLATION AND SELECTION OF INDIGE-NOUS YEAST STRAINS FOR THE PRODUCTI-ON OF WHITE AND RED WI-NES IN THE CONDITIONS OF THE REPUBLIC OF MOLDOVA

253.03 TECHNOLOGY OF ALCOHOLIC AND NON-ALCOHOLIC BEVERAGES

Summary of the doctor habilitat thesis in engineering sciences

CHIȘINĂU, 2025

The thesis was developed within the "Biotechnologies and Microbiology of Wine" laboratory, PI Scientific-Practical Institute of Horticulture and Food Technologies, and within the scientific project for Young Researchers 02.01.2019-31.12.2019: "Characteristics of indigenous yeast strains isolated from the Trifești wine center for the production of dry white and red wines".

Composition of the Doctor Habilitat Thesis Examination Committee:
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The public defense will take place **on April 9, 2025, at 10^{00} am** in the meeting of the Committee for the public defense of the DH thesis within the PI of the National Institute of Applied Research in Agriculture and Veterinary Medicine, 59 Vieru Street, room 206, MD 2070, Codru city, Chisinau municipality, Republic of Moldova.

The habilitated doctorate thesis and the summary can be consulted at the PI library of the National Institute for Applied Research in Agriculture and Veterinary Medicine (https://incaamv.md/ro/biblioteca/) and on the ANACEC website (www.anacec.md).

The summary was sent on the **7 March 2025**

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CUPRINS

REPERE CONCEPTUALE ALE CERCETĂRII

Actuality and importance of the topic addressed. To obtain quality dry white and red wines with full fermentation of carbohydrates at low temperatures, slow fermentation conditions are created, because wines fermented quickly and tumultuously at high temperatures do not possess the desired organoleptic qualities. For this, at the must fermentation stage, yeast from selected yeasts or from active dry yeasts is used [2, 4].

Currently, of particular importance in the field of biotechnology and wine microbiology is the isolation and selection for the wine industry of indigenous yeast strains with fermentative activity necessary for obtaining quality wines, the evaluation and systematization of morpho-cultural and physiological-biochemical indices of selected indigenous strains, for the efficient preservation, expansion and strengthening of the indigenous microbial gene pool specific to the wine industry [9, 11, 14].

Indigenous yeasts play a crucial role in shaping the typicality and authenticity of wines in various wine-growing centers around the world. Each wine-growing region has a specific microbiome, made up of a variety of microorganisms, including yeasts, that are present in the soil, on the grapes and in the winemaking facilities. This microbiome significantly influences the characteristics of the wine produced in that region. For example, in France, in the Bordeaux region, indigenous yeasts contribute to the formation of the complex aromatic profile of Cabernet Sauvignon, Merlot and other varieties characteristic of this region. In the Champagne region, indigenous yeasts are essential for the secondary fermentation in bottles, which gives sparkling wines a distinct aroma and fine bubbles. In Italy, in the Tuscany region, indigenous yeasts contribute to the formation of the distinct characteristics of Chianti Classico and Brunello di Montalcino wines, reflecting the terroir of these areas. In Spain, in the Rioja region, indigenous yeasts are responsible for the specific aromatic and gustatory contribution of Tempranillo, Garnacha and other varieties grown in this area [16, 18, 27, 33, 39].

These examples illustrate the importance of indigenous yeasts in creating wines with typicality and authenticity specific to each wine-growing region, reflecting the local terroir and contributing to the diversity and richness of the global wine universe.

In the Republic of Moldova, winemakers have long widely used active dry yeast (ADY) produced in France, Switzerland, Italy, Germany, etc. for wine production. The high cost of imported active dry yeasts (€50-120/kg), which leads to an increase in the cost of finished production, the poor adaptation of foreign yeasts to (local) factory conditions, the appearance of foreign nuances after must fermentation are the arguments in favor of using local yeasts for the production of local wines.

Yeasts usually enter the must from the surface of the grapes, the equipment used or by direct administration [29]. The fermentation process can be directed by administering to the must the yeast of selected yeasts or active dry yeasts (ADY) and naturally, without the administration of any yeast [19, 34]. ADY are widespread in many wine-growing countries, by using which good results are obtained [29], but it is mentioned that a quality wine is obtained when it is fermented using autochthonous yeasts [29].

Although many commercial dry yeasts are used for must fermentation, it is considered that the use of indigenous yeasts is more efficient, since they possess a dominant potential in the fermentation process in winemaking. In addition, the use of indigenous yeasts ensures the acquisition of typical and specific sensory properties for wines produced in certain winemaking centers.

The diversity of wine yeasts is a reliable source for selecting new strains, which dominate during the fermentation process and influence the organoleptic characteristics of wines [27, 33].

Oenological yeast strains have been selected for hundreds of years, permanently and systematically, due to their valuable properties and the major influence on the quality of the wine they produce. This has led to the creation of specialized oenological collections of wine yeasts, which can be accessed by winemakers to have reliable performance and diversity of wines. In winemaking practice there are many processes that can be influenced by the yeast strains used in must fermentation, but the problem of slow and incomplete fermentation of must sugars is the most common. [3, 6].

While some yeast strains are quite reliable in the fermentation process, others have difficulty adapting to environmental conditions, have a lower fermentative activity, or are inactivated before the sugars are completely fermented, contributing to the reduction of wine quality and the emergence of risks of microbiological alteration of finished wines.

Worldwide, collections of microorganism cultures have been organized in various countries under the auspices of the World Federation for Culture Collections (WFCC). In Europe, the collections of microorganisms from 17 wine-producing countries are united in the European Collection of Cell Cultures (ECCC) [28].

The largest and most recognized center for preserving collections of microbial cultures is the American Type Culture Collection (ATCC) [13].

According to the Budapest Treaty on the International Recognition of Deposits of Microorganisms and in order to ensure the protection of industrial property, the National Collection of Non-Pathogenic Microorganisms was organized in the Republic of Moldova based on the Decision of the Government of the Republic of Moldova No. 807 of July 2, 2003. Subsequently, by Government Decision No. 56 of January 26, 2004, the Regulation of the National Collection of Non-Pathogenic Microorganisms within the Institute of Microbiology and Biotechnology was approved.

The problem of studying oenological yeast strains in order to improve the quality of wines has been permanently at the center of attention of many scientists: J. Ribereau-Gayon, P. Ribereau-Gayon, P. Ungurean, V. Cotea, N. Burian, N. Saenco, A. Popa, V. Kudreavţev, S. Kişcovscaia, N. Taran, B. Gaina, Gh. Arpentin, R. Sturza, Z. Palic, E. Ivanova, G. Condo, A. Oprea, N. Moghileanschi, A. Corotchevici and al.

The National Institute of Applied Research in Agriculture and Veterinary Medicine (previous name: Scientific-Practical Institute of Horticulture and Food Technologies (SPIHFT)) is the only institution in the Republic of Moldova that holds two branch collections of microorganisms specific and characteristic for the wine industry and the milk processing industry.

The collection of microorganisms for the oenological industry (CMOI) has been kept in the Wine Biotechnology and Microbiology laboratory for over 30 years.

The main goal of the CMOI collection is to preserve the morphological and physiological properties of yeasts, ensuring the production of increased amounts of glycerol under the conditions of the formation of optimal concentrations of volatile acids and esters. At the same time, the yeasts must generate high levels of amino acids, have a high vitaminogenic potential and contribute to providing wineries in the Republic of Moldova with selected strains for must fermentation, secondary fermentation, biological deacidification and other biotechnological processes [8].

In recent years, the use of non-*Saccharomyces* yeasts in must fermentation and the formation of dry white wines has increasingly concerned the interests of winemakers both in countries with a tradition in winemaking (France, Spain, Italy, Portugal, Greece) and more novice ones (Australia, Chile, Argentina, South Africa) [23, 35].

Some researchers consider that the impact of non-*Saccharomyces* yeasts on the quality of dry white wines is negative, others discover some favorable technological capacities of this group of yeasts. Their capacity to render complexity to the final product, to produce fruity varietal aromas, to exercise an enzymatic activity of potential interest it is reported in numerous scientific publications [23, 35].

The ability of non-*Saccharomyces* yeasts to restore complexity to the final product, to produce more complex aromas, to produce higher concentrations of organic acids, glycerol, as well as 2-phenylethanol, to increase enzymatic activity by producing extracellular enzymes with technological significance, which restore sensory properties specific to the grape variety to the wine, denotes a technological interest of these yeasts, which is reported in the scientific literature [23, 35].

Currently, the first ADY co-strains proposed by 2 major ADY producers in the world, Chr. Hansen (Denmark) and Lallemand (France), have appeared on the wine market, offering a mixed culture of yeasts *Kluyveromyces thermotolerans* (10%), *Torulaspora delbrueckii* (10%) and *Saccharomyces cerevisiae* (80%) (Viniflora® HARMONY.nsac), another strain with sequential seeding of *Saccharomyces cerevisiae* and *Torulaspora delbrueckii* (LEVEL 2 BIODIVA™). Alcoholic fermentation with the sequential strain is carried out by two inoculations: first with *Torulaspora delbrueckii* yeasts, and when the sugars in the must are reduced by 15%, *Saccharomyces cerevisiae* is inoculated.

Torulaspora delbrueckii yeasts produce some specific and valuable fermentation aromatic profiles in the organoleptic appreciation of wine and are traditionally used together with *Saccharomyces* strains – yeasts, which ensure the completion of alcoholic fermentation in a reasonable time. However, it is additionally necessary to study, from an application point of view, in what quantities, as well as to establish the optimal moment of inoculation (co-inoculation, sequential inoculation). Studies on the interactions between these two yeast species are relatively few, especially from the point of view of quantifying this interaction.

In the Republic of Moldova, not enough research has been conducted in the field of valorizing the biodiversity of microorganisms found in wine, which would result in the development of new technologies offered to producers.

The purpose and objectives of the research. The purpose of the studies is to isolate, identify and select indigenous yeast strains from various wine-growing centers of the Republic of Moldova, with valuable technological properties, easily adaptable to the given environment, which completely ferment the carbohydrates in the must to obtain dry white and red wines with high organoleptic qualities, in order to guarantee the authenticity of wines of regional origin, as well as to store the highlighted yeast strains in the National Collection of Non-pathogenic Microorganisms and the Branch Collection of Microorganisms for the Oenological Industry.

To achieve the goal, the following **operational objectives** were envisaged:

1. Isolation and selection of indigenous yeast strains from different wine-growing centers (′Chișinău′, ′Purcari′, ′Trifești′).

2. Taxonomic identification of isolated yeast strains, through the study of morphological and cultural characters. Identification of isolated strains by modern PCR methods and FT-IR spectroscopy.
3. Determination of biochemical and technological indices of isolated yeast strains.
4. Study on the effect of selected indigenous strains on the fermentation process of must and pomace.
5. Study of the influence of selected autochthonous strains on the physicochemical and organoleptic indices of the obtained dry white and red wines.
6. Testing and implementation under production conditions of selected indigenous yeast strains for the production of dry white and red wines;
7. Improving the technological regimes for the production of dry white and red wines using local yeasts.
8. Study of the influence of non-*Saccharomyces* yeasts on the quality of dry white wines under laboratory, microvinification and production conditions.

Research hypothesis. The use of indigenous yeast strains in the must fermentation process has a positive impact on the quality of dry white and red wines, compared to the use of imported active dry yeasts. By isolating, identifying and selecting these indigenous strains from various wine-growing centers of the Republic of Moldova, it is anticipated that they will present valuable technological properties and adaptability to the specific local environment.

Synthesis of the research methodology and justification of the chosen research methods. To carry out the work, traditional and modern physicochemical methods were applied, such as taxonomic identification of strains by PCR method and FT-IR spectroscopy. The analysis of the physicochemical characteristics of the wines obtained was carried out by determining the alcoholic concentration; the mass concentration of: sugar, ttitrable acids, volatile acids, sulfur dioxide (total and free); determining the pH value and the oxidation-reduction potential; determining the non-reducing extract in accordance with the standard methods in force.

Determination of the mass concentration of volatile aromatic compounds was carried out by the gas chromatography method in the laboratory "Verification of the quality of alcoholic production" of SPIHFT. Determination of the mass concentration of organic acids was carried out by the capillary electrophoresis method and using the multifunctional spectrophotometer "Bacchus 3" (France).

The microbiological study (microscopy, yeast cell count, seeding on different nutrient media, yeast biomass accumulation, etc.) was carried out according to the instruction for the microbiological control of wine production in force (IC MD 67-42582515-001:2010) and specialized bibliographic sources [5].

Theoretical importance and scientific innovation of the work: for the first time, strains of indigenous yeasts were isolated and selected from different wine-growing centers of the Republic of Moldova and the perspective of using selected indigenous yeast strains for the production of dry white and red wines was scientifically argued. The use of selected indigenous yeasts in obtaining quality dry white and red wines was argued and implemented in practice. By using selected indigenous yeasts, the specificity and authenticity of the wines were emphasized, reflecting the unique characteristics of the region of origin. Thus, terroir became an essential element in defining the organoleptic profile of the wines obtained.

The applicative value of the work:

- The indigenous yeast strains with advanced technological properties, isolated and selected from the wine-growing centers of ′Chișinău′, ′Purcari′ and ′Trifești′ were tested under production conditions at SA ”Cricova”, ÎM “Vinăria Purcari” and ”Vierul-Vin” SRL, where experimental batches of dry white wines in a total volume of 100,000 L and dry red wines in a total volume of 60,000 L were obtained.

- The technological regimes for the production of dry white and red wines using selected local yeasts have been improved. The improved technologies have been optimized based on the following criteria: low fermentation temperature under industrial conditions (minimum 13 °C for white wines); lower sulfite content of must and must (for white wines: 50-75 mg/L total SO_2 when grapes are healthy and 120-150 mg/L total SO_2 if they have been attacked by gray rot; for red wines: 40-60 mg/L total SO_2 when using healthy grapes and 120-150 mg/L total SO_2 if they have been attacked by gray rot).
- Technological recommendations were developed regarding the use of *Saccharomyces*, non-*Saccharomyces* and *Torulaspora delbrueckii* yeasts for the production of dry white wines. The use of non-*Saccharomyces Torulaspora delbrueckii* yeasts was recommended for the successive fermentation of the must with the inoculation of *Saccharomyces* yeasts upon reaching the average alcoholic concentration of 3% vol. for the production of dry white wines.
- 8 patents were obtained for yeast strains isolated from various wine-growing centers of the Republic of Moldova.

Approval of the paper at national and international scientific forums. The results obtained during the work were presented and discussed at national and international conferences: International Scientific Conference ”Microbiologic Biotechnology”, Chisinau (2014, 2016, 2018); International Conference ”Modern Technologies in the Food Industry”, Chisinau (2014, 2016, 2018, 2024); International Scientific Conference ”Problems and trends of world viticulture and winemaking: Ukrainian perspective”, Odessa, November 3, 2016; International Scientific Symposium ”Modern Horticulture- Achievements and Perspectives”, SAUM, Chisinau, October 4-6, 2018; International Conference ”Science. Education. Culture”, CSU, Comrat (2017, 2019); International Congress ”Наука, Питание и Здоровье”, R. Belarus, Minsk, June 8-9, 2017; 3rd International Agriculture Congress, Tunis, March 5-9, 2020; 2nd International Congress on Engineering and Life Sciences, Kastamonu, Turkey, April 11-14, 2019; 5th International Congress on Engineering and Life Science, Romania, Bucharest, September 10-12, 2024; International Exhibition of Inventics “Inventica”, Iași, Romania (2018, 2019, 2024); European Exhibition of Creativity and Innovation “Euroinvent”, Iași, Romania (2017 – 2024); International Specialized Exhibition “InfoInvent”, Chișinău, Moldova (2017, 2019, 2023); Innovation and Research Exhibition “UgalINVENT”, Galați, Romania (2021).

Publications on the topic of the thesis. The research results and the issues addressed in the thesis were published in 53 scientific papers, including a monograph, a chapter in a monograph, 2 articles in journals from the Web of Science and SCOPUS databases, 1 review article, 12 scientific articles in international and national journals, 10 articles in international and national scientific collections, 8 patents, 18 abstracts at international scientific events and other scientific papers.

The structure and volume of the thesis. The thesis is structured on 204 pages and includes the introduction, 6 chapters, conclusions and recommendations, bibliography with 310 sources and 7 annexes, being illustrated with 71 tables and 65 figures. The introduction presents the motivation, purpose and methodology of the research. **Chapter 1** analyzes the use of yeast strains in oenology. **Chapter 2** describes the materials, methods and experimental techniques used. **Chapter 3** focuses on the isolation and selection of autochthonous strains of the genus *Saccharomyces*. **Chapter 4** studies their influence on the quality of wines. **Chapter 5** presents the testing and implementation of the selected strains in obtaining wines under production conditions. **Chapter 6** explores the effect of non-*Saccharomyces* yeasts on white wines.

Keywords: yeasts, strains, Saccharomyces, non-Saccharomyces, wines, fermentation, physicochemical indices, quality.

I. STUDIES ON THE USE OF YEAST STRAINS IN OENOLOGY

Current scientific research shows that yeasts are an ecological component of nature and go through three development cycles: natural, close to natural, and artificial.

Native yeasts are yeasts that are found in different wine-growing regions and can be used to produce high-quality wines specific to that region.

Indigenous yeasts are adapted to specific climatic and soil conditions, making them more robust in the face of environmental changes. Their use can reduce the need for human intervention in the fermentation process and prevent the occurrence of defects in wine. They can influence the aromatic and gustatory profile of the wine, by producing compounds specific to the region. The use of these yeasts can be beneficial for the development of the local wine sector, by promoting certain characteristics specific to the region [14, 18, 27, 33, 39].

Recent studies have shown that indigenous yeasts can help reduce the environmental impact, by using these indigenous yeasts, the need to transport and purchase commercial yeasts from considerable distances is eliminated. This can help reduce the carbon footprint associated with the transport and production of commercial yeasts, thus promoting a more sustainable approach to the wine industry. Use of yeasts indigenous can be a strategy for conserving microbial biodiversity, by maintaining endemic species adapted to local conditions [14, 18].

Local yeasts can be considered a factor of cultural identity, by producing wines with specific characteristics of the respective region. Their use can be important in the process of differentiating wine products internationally, by promoting authentic wines with local specificity [14, 18].

Studies on indigenous yeasts can contribute to the development of natural treatments for the prevention of diseases in the wine sector. The use of indigenous yeasts can be a source of innovation for the wine sector, by identifying and using strains with special characteristics. Their use can contribute to the development of a circular economy, by reducing the need to purchase commercial yeasts and by valorizing waste from the production process.

Native yeasts represent a valuable natural resource for the wine sector and can be used sustainably to obtain authentic and high-quality wines [9, 10].

The analysis of the specialized literature reveals the significant influence of autochthonous yeast strains on wine quality. Thus, there is a need to evaluate and understand the biochemical and physicochemical characteristics of wines produced in wine-growing centers, based on selected autochthonous yeast strains, which could be used to guarantee the authenticity of regional wines with protected designations of origin or protected geographical indications [27, 33].

II. RESEARCH MATERIALS AND METHODS

The research was carried out within the laboratories "Biotechnologies and Microbiology of Wine" and "Quality Control of Alcoholic Production" of PI SPIHFT, in collaboration with laboratories specialized in the field of wine microbiology from the European Union, Romania (Timișoara, University of Agricultural Sciences and Veterinary Medicine of Banat "King Michael I of Romania"), Germany (Hochschule Geisenheim University), France (French Institute of Vine and Wine in Nantes), Italy (Bologna, Cesena, University of Bologna, Department of Agri-Food) and the Russian Federation (Moscow, Research Institute of Agricultural Biotechnology, SRL "Syntol"),Greece (Larissa, Department of Immunology and Histocompatibility, Faculty of Medicine, University of Thessaly, CeMIA SA Company)

Production testing and practical implementation of selected yeast strains were carried out at wine enterprises: SA "Cricova" (Chisinau municipality), ÎM "Vinăria Purcari" SRL (Purcari village, Ștefan Vodă county), SRL "Vierul-Vin" (Burlacu village, Cahul county).

Culture media used in research.

Natural media were used as culture media: of plant origin (grape must) and animal origin (broth), as well as synthetic media of various compositions, which include various nutrients: sugars, nitrogenous substances, minerals, etc.

Research objects. The following were used as research objects: grape must from the varieties Aligote, Sauvignon, Chardonnay, Feteasca Albă, Merlot, Cabernet-Sauvignon, Rară Neagră; yeast strains isolated from different wine-growing centers (′Purcari′, ′Chișinău′, ′Trifești′), yeast strains from the Branch Collection of Microorganisms for the Oenological Industry (CMOI) of SPIHFT, as well as imported industrial ADY strains, specified in table 2.1, nutrients for yeast food, raw material wines, other adjuvant materials.

Table 2.1. Yeasts used in the research process

Genus, species	**Yeast strain from SPIHFT**	**Collection number of SPIHFT (CNMN*)**
Saccharomyces cerevisiae	Rara Neagră-2	29
	Cricova-2	120 (CNMN-Y-26)
	R-N-120-P-5	127 (CNMN-Y-31)
	Ch75P-3ÎF	126 (CNMN-Y-32)
	FNFTP-1	125 (CNMN-Y-33)
	ATr.-2.3	128 (CNMN-Y-34)
	S75Tr.-4.4	129 (CNMN-Y-35)
	M100Tr-1	130 (CNMN-Y-36)
	C-S60Tr-2	131 (CNMN-Y-37)
Active dry yeasts		
Genus, species	**Tulpina**	**Producing country**
non-*Saccharomyces Torulaspora delbrueckii*	Enartis FERM	Italy
Saccharomyces cerevisiae	LittoLevure	France
	Zymaflore yeasts	France
	Anchor Alchemy I	South Africa
	Anchor NT 202	South Africa
	Oenoferm Freddo	Germany
	Oenoferm Be-Red	Germany

* deposited under the respective number in the National Collection of Non-pathogenic Microorganisms (CNMN) within the Institute of Microbiology and Biotechnology of TUM.

Research methodology and analysis methods

Isolation and selection of yeast strains. In order to select autochthonous yeast strains from the wine-growing centers of ′Chișinău′, ′Purcari′ and ′Trifești′, from the grapes and must of the white-berry varieties Feteasca Albă, Chardonnay, Aligote, Sauvignon and the black-berry varieties Rară-Neagră, Merlot and Cabernet-Sauvignon, 94 yeast strains with specific characters of the genus Saccharomyces were isolated in pure culture. To carry out the research at this stage, the isolation scheme shown in figure 2.1 was used.

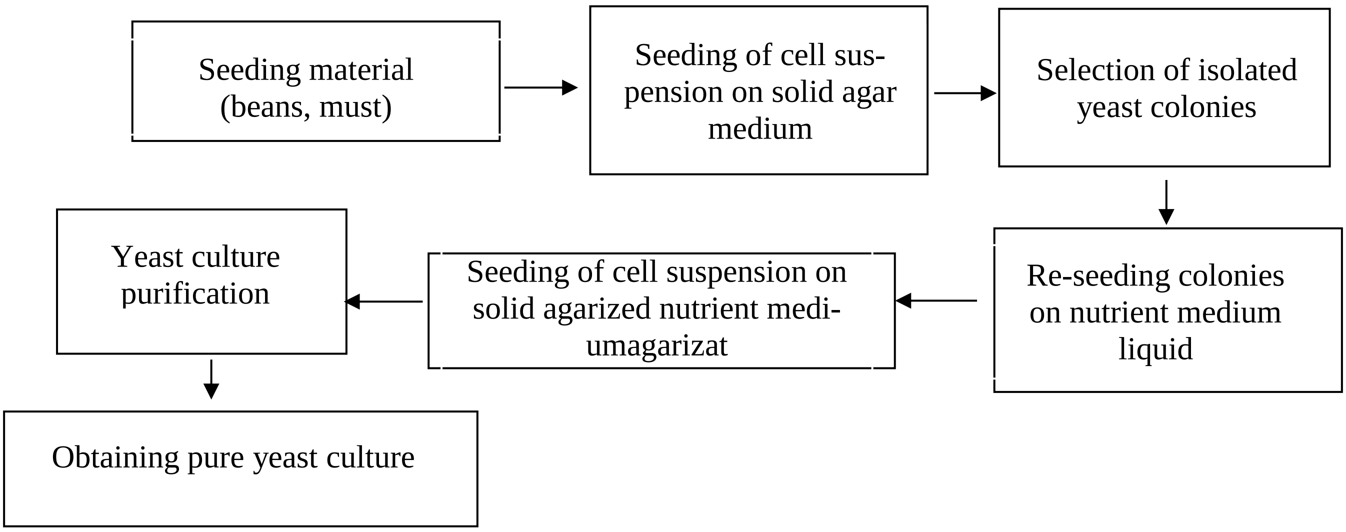

Fig. 2.1. Yeast strain isolation scheme

For the isolation of yeast cells A research methodology was developed from the berries and must, which is presented in Table 2.2.

Table 2.2. Research methodology for yeast cell isolation

Biological material from which the harvest was made	Specification	The moment of isolation
grains	Harvest time: -technical maturity	Washing with sterile water: -4 minutes -8 minutes
Grape must	**Sugar content:** 180-200 g/dm^3 (white varieties) 210-230 g/dm^3 (red varieties) **Sulfur doses applied:** White varieties: -Unsulphite must, -75 mg/dm^3, -150 mg/dm^3 Red varieties: -Unsulphite must, -60 mg/dm^3, -120 mg/dm^3	The beginning of fermentation The tumultuous fermentation End of fermentation

Determination of morphological and cultural characters of yeast strains. The determination of morphological and cultural characters of yeast strains is an important process in microbiology, providing essential information about the appearance and behavior of these microorganisms. These characteristics are useful in the classification and identification of yeast species. In the process of the study, the following were examined and determined: the shape and dimensions of the cells,cultural characters, sporulation capacities, pseudohyphae (pseudomycelia) formation [9].

Determination of technological characteristics of yeast strains. As a result of the studies carried out, the following were determined: cold resistance, alcohol resistance,resistance to SO_2, copper resistance, strain phenotype, foaming capacities, β-glucosidase activity,formation of acetic acid,volatile sulfur compounds [9, 14, 25, 26].

Identification of yeasts by PCR method. The PCR method was used to determine the genetic affiliation of yeast strains isolated from different winemaking centers.

The research was conducted in the "Biotechnologies" laboratory of the Research Institute of Agricultural Biotechnology in Moscow (Russia) and in the Department of Immunology and Histocompatibility, Faculty of Medicine, University of Thessaly, CeMIA SA Company, Larissa (Greece).

The following primers were used for PCR amplification:

ITS 1 (5'-TCCGTAGGTGAACCTGCGG –3') and ITS 4 (5'-TCCTCCGCTTATTGATATGC-3').

The amplification parameters were: denaturation at 94°C for 1.5 min., annealing at 55°C for 1.5 min., elongation at 72°C for 2 min., and final elongation for 10 min.

Taxonomic identification of yeasts by FT-IR spectroscopy. The method includes the following work steps:

- Yeasts are grown in YPD liquid culture medium or grape must at 38 °C for 72 h, until the stationary growth phase is reached,
- With the pipette, apply the yeast to the cells of the plate,
- The plate is placed in the thermostat for 1 hour at a temperature of 45°C,
- After drying the samples, the plate is inserted into the spectrometer and the analysis program is launched.

Preparing the yeast dough

Currently, oenological yeasts are used fresh on malt-agar medium, compressed, dried and lyophilized [7, 9,10].

The preparation of yeast for use in primary winemaking includes the cultivation of and the gradual accumulation of the amount of active yeast biomass necessary for the fermentation of the obtained must.

Mathematical processing of experimental results

The analysis methods used in the research were previously studied to ensure their validation by establishing the conformity of the experiment protocol. In this context, 3-5 parallel measurements were performed, and the results were subjected to statistical processing and mathematical modeling. For the statistical processing of GraphPad Prism 5.0 program was used and with the help of the online calculator on the website http://math.semestr.ru/. Mathematical processing for the purpose of optimizing experimental research was carried out based on the MS EXCEL, ANOVA and STATGRAPHICS 5.0 programs.

III. ISOLATION AND SELECTION OF YEAST STRAINS OF THE GENUS SACCHAROMYCES FROM DIFFERENT WINE GROWING CENTERS OF THE REPUBLIC OF MOLDOVA FOR THE PRODUCTION OF DRY WHITE AND RED WINES

3.1. Isolation of yeast strains from different wine-growing centers of the Republic of Moldova

To conduct studies to identify microorganisms, it is necessary to obtain a pure culture, which is descended from a single cell.

The specifics of isolating and cultivating yeast strains depend on their presence in different places (vines, must, wine, soil, etc.) and on the composition of the nutrient media.

Grape samples for the isolation of yeast strains were taken from the vineyard plantations of SA "Cricova", ÎM "Vinăria Purcari" SRL and "Vierul-Vin" SRL during the winemaking season.

Grapes were sampled from the plantations of SA "Cricova" and the must of the Chardonnay variety was obtained, which was characterized by the following initial indices: sugar concentration - 185 g/dm^3, titrable acids - 8.4 g/dm^3.

Grapes were sampled from the vineyards of the ÎM "Vinăria Purcari" SRL and obtained: must from the Feteasca albă variety with sugar concentration - 194 g/dm^3, titrable acids - 4.9 g/dm^3; must from the Chardonnay variety with sugar concentration - 202 g/dm^3, titrable acids - 7.7 g/dm^3; must from the Rară-Neagră variety with sugar concentration - 207 g/dm^3, ttitrable acids - 5.7 g/dm^3; must from the Cabernet-Sauvignon variety with sugar concentration - 220 g/dm^3, titrable acids - 6.2g/dm^3;

Grapes were taken from the vineyards of "Vierul-Vin" SRL and obtained: must from the Aligote variety with a sugar concentration of 182 g/dm^3, titrable acids of 6.2 g/dm^3; must from the Sauvignon variety with a sugar concentration of 218 g/dm^3, titrable acids of 6.5 g/dm^3; must from the Merlot variety with a sugar concentration of 230 g/dm^3, ttitrable acids of 5.4 g/dm^3; must from the Cabernet-Sauvignon variety with a sugar concentration of 220 g/dm^3, titrable acids of 6.4 g/dm^3;

Yeast cell isolation was performed starting from a single cell, using the method of successive dilutions and the method of isolation in pure culture by the loop depletion technique. From the Ascomycotina class, 34 strains (from white varieties) were isolated from the ′Chișinău′ wine center, 64 strains (34 from white varieties, 30 from red varieties) from the ′Purcari′ wine center and 61 strains (33 from white varieties, 28 from red varieties) from the ′Trifești′ wine center.

To obtain pure yeast cultures and highlight qualitative properties, the studied samples were subjected to the re-seeding process using the "Exhausted Loop" method (Fig. 3.1).

Fig. 3.1. Cultural characteristics of yeast colonies inoculated using the exhaustion loop method

In order to establish the purity of the isolated yeasts, microscopy of the cultures studied was performed. For this, the yeast strains were incubated preventively for 3 days on nutrient medium (grape must) at a temperature of 28°C. The results obtained by microscopy were confirmed and supplemented with photographs, which are presented in Fig. 3.2.

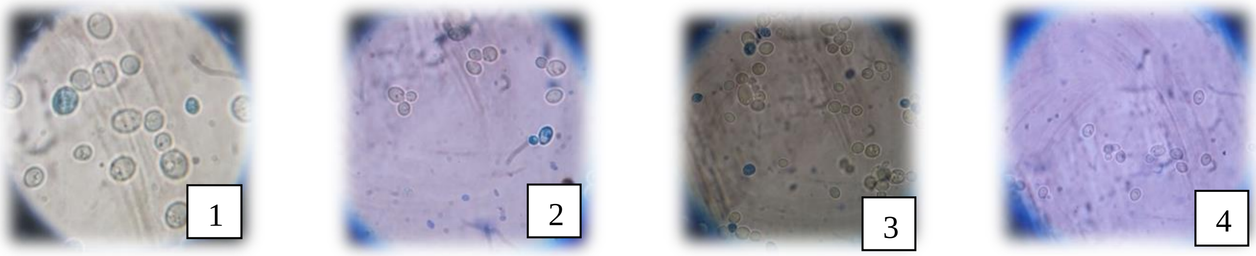

Fig. 3.2. Microscopy of yeast strains isolated from unsulfited Feteasca albă must, "Vinăria Purcari", 2016 h.y. (examples)

Microscopy of yeast strains allowed their visual evaluation and the preventive determination of morphological characters, such as: size, shape, grouping, as well as cell homogeneity.

3.2. Study of morphological and cultural characters of isolated yeast strains

In order to highlight the morphological and cultural characters of the yeasts isolated from the wine-growing centers ′Chișinău′, ′Purcari′, ′Trifești′ and to identify taxonomic indices, it is necessary to observe specific examination conditions, following which a complete evaluation of the strains can be made. This essentially contributes to the determination of the studied systematic categories (genus, species).

In order to determine the cell sizes of the isolated yeasts, microscopy of the cultures studied was performed. For this purpose, the yeast strains were pre-incubated for 3 days on nutrient medium (grape must) at a temperature of 28°C. Next, the sizes of 100 cells of each yeast strain were determined.As a result, it was established that the cell sizes of the isolated yeast strains range from 4.5 to 10 µm and differ in length and width values.

It is known that the sizes of yeast strains Saccharomycesranges from 1.5 to 25 µm, which is specific to them [9].

Microscopic analysis of the isolated yeast strains established that all yeast strains are pure, uniform, homogeneous, viable with well-defined cells of different shapes and sizes, which are in a state of budding. The process of budding of cells is unipolar or bipolar. The cells of the yeast strains isolated from the winemaking centers ′Chișinău′, ′Purcari′ and ′Trifești′ are of eukaryotic type in which the cellular components are well distinguished (fig. 3.3).

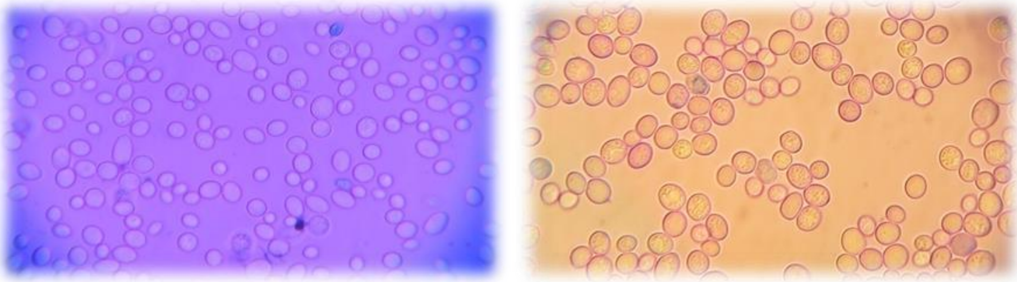

Fig. 3.3. Visual evaluation of isolated yeast strains, ("Purcari Winery", 2016 h.y.)

In order to argue the taxonomic affiliation of the studied yeast strains, identification tests according to Kudreavtsev were performed.It was established that out of 34 cultures isolated from the ′Chișinău′ wine center, 16 yeast strains belong to the genus Saccharomyces, out of 64 cultures isolated from the ′Purcari′ wine center, 31 yeast strains belong to the genus *Saccharomyces*, and out of 61 isolates from the ′Trifești wine center, 47 yeast strains belong to the genus *Saccharomyces.* The yeasts studied do not form real mycelium and all reproduce vegetatively by multilateral budding and sexually by spores, which confirms the fact that these strains belong to the genus *Saccharomyces.*

3.3. Identification of yeast strains isolated from the wine-growing centers of ′Chișinău′, ′Purcari′ and ′Trifești′ by the PCR method

The PCR method was used to determine the genetic affiliation of yeast strains isolated from different winemaking centers.

By using the PCR method, it was established that all the yeast strains studied belong to the genus *Saccharomyces* (Fig.3.4). Species identification was performed by comparing the nucleotide sequence obtained with the data placed in the NCBI gene bank (ncbi.nlm.nih.gov).

Fig. 3.4. Visualization of DNA fragments. 1, 12-DNA fragments of known length (marker); 2- FNFTP-1, 3- FNFTP-5, 4- FNFTP-6, 5- FNFTP-7, 6- F-75-FTP-3, 7- F-75-FTP-4, 8- F-75-FTP-5, 9- F-75-FTP-6, 10- F-150-FTP-4, 11- F-150-FTP-6:DNA fragments of some studied yeasts

Following the molecular identification analyses of the yeast species studied, 16 S.cerevisiae strains were identified from the ′Chișinău′ wine center, 22 *S.cerevisiae* strains, 6

S.pastorianus strains and 3 *S.bayanus* strains from the ′Purcari′ wine center, 32 *S.cerevisiae* strains, 12 *S.pastorianus* strains and 3 *S.bayanus* strains from the ′Trifești′ wine center.

3.4. Taxonomic identification of yeast strains isolated from the wine-growing centers of ′Chișinău′, ′Purcari′ and ′Trifești′ by FT-IR spectroscopy

Taxonomic identification by FT-IR is based on the fact that the functional groups of each molecule studied absorb infrared radiation to generate a characteristic absorption or transmission spectrum, which is rich in information and unique to the molecule. The spectra obtained are analyzed in known databases. Just as human fingerprints uniquely identify their owner, IR spectroscopy provides a spectral fingerprint that uniquely identifies a chemical compound. In the field of microbiology, an FT-IR spectrum reveals a fingerprint of the cell, reflecting its biochemical composition, which includes proteins, lipids, DNA, RNA and carbohydrates.

The high selectivity and sensitivity of the method make it possible to identify microorganisms even down to the strain level.

Taxonomic identification of the yeast strains studied by FT-IR was performed in the "Wine Microbiology" laboratory of the University of Geisenheim (Germany).

The yeast strains studied were subjected to identification analysis using FT-IR spectroscopy. As a precaution, the yeast strains were checked for purity (fig. 3.5).

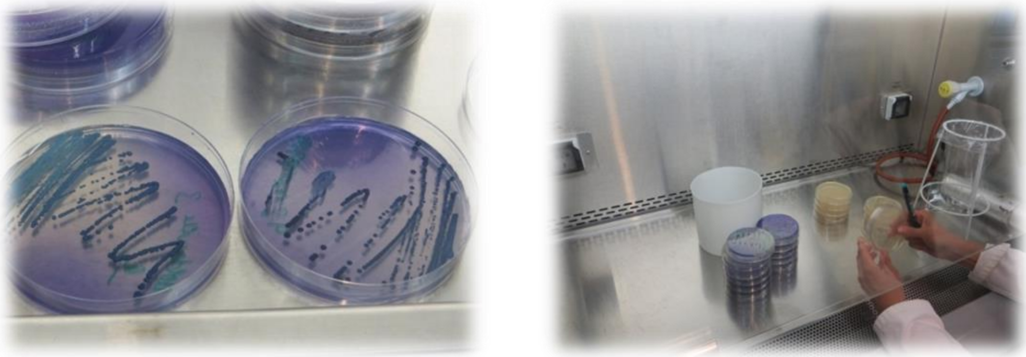

Fig. 3.5. Checking the purity of yeast strains by seeding them on the solid agarized medium YGCB-agar and YGC-agar

After checking the yeast strains for purity, each strain was taken from the Petri dish, dissolved in 0.1 ml of distilled water and transferred to the cells of the plate (ZnSe).
The plate was placed in the thermostat for 1h, at a temperature of 45°C. After drying the samples, the plate was placed in the spectrometer and the analysis program (OpusLab) was launched.
As a result of the analysis performed, based on the spectra obtained, compared with those existing in the database, it was found that all the yeast strains investigated are of three species: *Saccharomyces cerevisiae* (~75%), *Saccharomyces pastorianus* (~20%) and *Saccharomyces bayanus* (~5%).

3.5. Determination of biochemical and technological indices of isolated yeast strains

Yeast strains isolated from the winemaking centers of ′Chișinău′, ′Purcari′ and ′Trifești′ were tested in order to identify those strains that allow the production of wines with typical characteristics.

During the period 2012-2017, 16 yeast strains were isolated from the ′Chișinău′ wine center, 31 strains from the ′Purcari′ wine center, and 47 strains from the ′Trifești′ wine center.

At the first stage, for the selection of yeast strains, the following technological parameters were analyzed: alcohol resistance, SO_2 resistance, Cu resistance, cold resistance, foaming ability, β-glucosidase activity, Killer factor. As a result of the research, 5 yeast strains with advanced technological properties were selected from the ′Chișinău′ wine center, 8 yeast strains from the ′Purcari′ wine center and 10 yeast strains from the ′Trifești′ wine center.

In the qualitative characteristics evaluation test, carried out under laboratory conditions, selective culture media were used and the ability of selected yeast strains to produce hydrogen sulfide, acetic acid and β-glucosidase activity was established.

It has been established, that most of the studied yeast strains are resistant to alcohol (from 10% vol. to 14% vol.), the exception being ~30% of yeasts, which do not develop at an alcohol concentration of 14% vol. Resistance to high SO_2 concentrations was detected in 95% of the studied yeasts, the exception being strains No. 3,4,7,11,15 (′Chișinău′), No. 17,20 (′Purcari′) and No. 2,21,39 (′Trifești′) (fig.3.6).

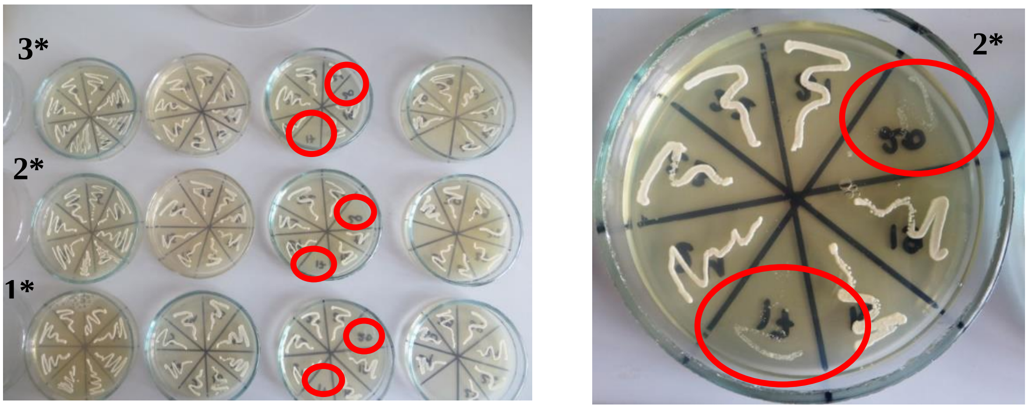

Fig. 3.6. Resistance of some yeast strains to high concentrations of SO_2 (example)

Legend: *1*- SO_2-100 mg/L; 2*- SO_2-150 mg/L; 3*- SO_2-200 mg/L.*

The resistance of yeast strains to high concentrations of copper demonstrated that its presence does not negatively influence the fermentative activity of the studied strains.

It was determined that 75% of the strains studied formed very little foam in the first 24-48 hours, with 38 strains being included in the "foam-free" yeast category (fig.3.7).

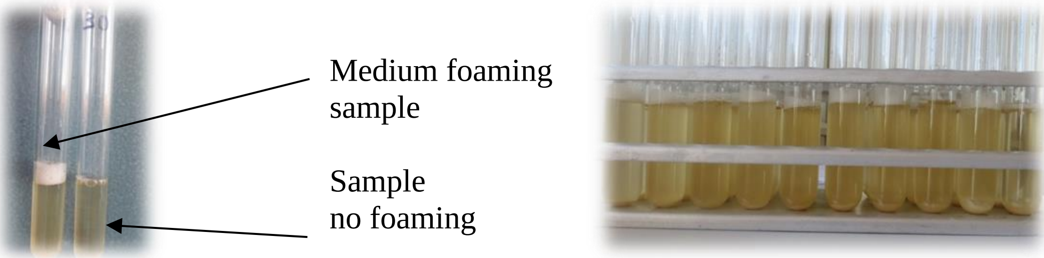

Fig. 3.7. The ability of some yeast strains studied to form foam (example)

The research conducted and the results obtained based on this study demonstrated that 85% of the yeast strains isolated from the winemaking centers of ‘Chișinău’, ‘Purcari’ and ‘Trifești’ are of the Neutral phenotype, meaning they do not die in the presence of strains of the Killer phenotype and do not inhibit the activity of strains of the Sensitive phenotype.

Evaluation of technological characteristics demonstrated that 100% of yeasts possess β-glucosidase activity, but this activity decreases by ~33% at pH between 2.8-3.5 [14].

As a result of the research carried out, it was established that 56% of the yeasts isolated from the ′Chișinău′ winemaking center, 58% of those isolated from the ′Purcari′ winemaking center and 68% from the ′Trifești′ winemaking center produce minimal concentrations of acetic acid, 38%, 32% and respectively 21% form increased concentrations, and approximately 10% of yeasts produce exaggerated concentrations of this acid (fig. 3.8). The concentration of acetic acid varies depending on the strain, the concentration of sugars in the must and the fermentation temperature.

The formation or non-formation of H_2S is related to the presence or absence of sulfur amino acids in the must. The results obtained demonstrated that about 30% of the yeasts isolated from the ′Chișinău′ wine center, 23% of the yeasts isolated from the ′Purcari′ wine center and about 25% of the yeasts isolated from the ′Trifești′ wine center produce H_2S. (Fig.3.9)

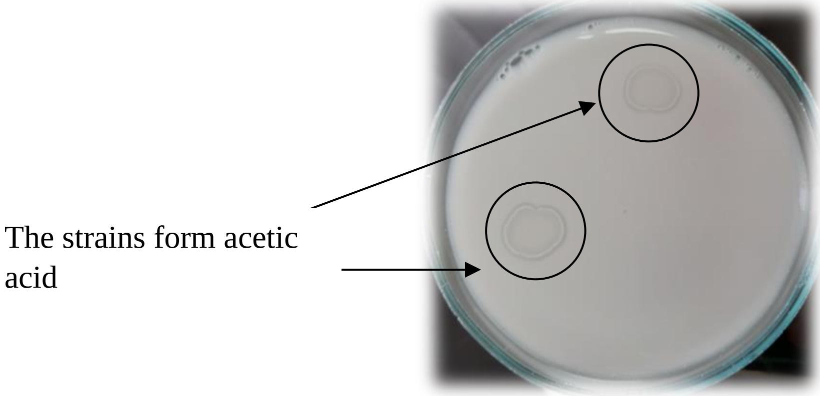

Fig. 3.8. Acetic acid formation by the yeast strains studied (example)

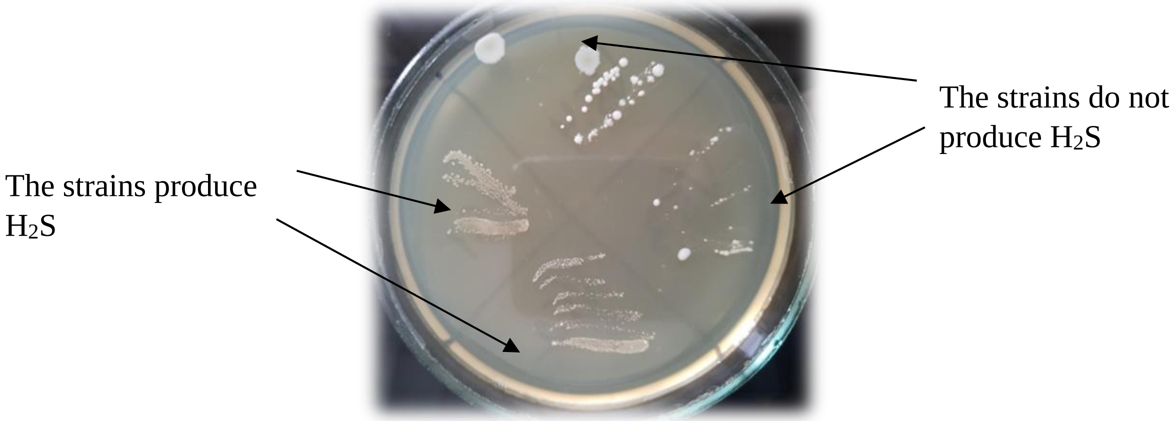

Fig. 3.9. H_2S formation by the yeast strains studied (example)

Thus, for the production of dry white and red wines, yeast strains capable of fermenting under specific conditions, with fermentative vigor [41] in optimal time, without the formation of hydrogen sulfide, with the ability to flocculate the precipitate, which eliminates the need for additional clarification or filtration of the wine and low or medium foam formation capacity, which indicates the presence of surface-active substances, were selected [15].

Based on the results obtained and the comparative assessment of different indices, the following yeast strains isolated from the ′Chișinău′ wine center were selected: No. 2, No. 8, No. 9, No. 10, No. 14 and No. 16; from the ′Purcari′ wine center: No. 1, No. 3, No.

7, No. 12 – for the production of white wines, No. 21, No. 24, No. 29, No. 30 – for the production of red wines; from the ′Trifeşti′ wine center: No. 1, No. 10, No. 15, No. 19, No. 22 – for the production of white wines and No. 27, No. 32, No. 35, No. 41, No. 43 – for the production of red wines.

IV. THE INFLUENCE OF DIFFERENT YEAST STRAINS ON THE QUALITY OF DRY WHITE AND RED WINES

For the technological assessment of yeast strains, especially those used on an industrial scale, one of the basic indicators is the ability of yeasts to form various secondary compounds during fermentation, which play a very important role in the wine production process, because they contribute significantly to the formation of the physicochemical and organoleptic indices of dry white and red wines

4.1. Study of the influence of different yeast strains selected from the ′Chişinău′ wine center on the quality of dry white wines

4.1.1. Influence of selected yeast strains on the physicochemical indices of dry white Chardonnay wines

Dry white wines obtained by fermentation with different yeast strains from the Chisinau wine center were subjected to physicochemical analyses, and the results obtained are presented in table 4.1.

Thus, we can mention that dry white wines fermented with the use of yeast strains No. 2 - Cricova-2 and No. 14 - 1VT are characterized by a higher concentration of ethyl alcohol of 11.5 and 11.4% vol. respectively. The use of yeast strains No. 8 - Cricova Chardonnay(3), No. 9 - Cricova Chardonnay(4), and ADY (LittoLevure, France) contributes to obtaining dry white wines with a lower alcoholic strength: 11.0-11.1% vol. (table 4.1).

Table 4.1. Physico-chemical indices of dry white wines obtained with different strains of yeast (y.h. 2011)

№	Strain	Alcohol concentration, % vol.	The mass concentration of		pH	Potential OR, mV	Residual sugars, g/dm^3	Note organoleptic, points
			titrable acids, g/dm^3	volatile acids, g/dm^3				
1	Nr.29 - Rara – Neagră 2 (control)	11,2±0,1	8,4±0,1	0,39±0,03	3,15±0,02	214,2±0,02	1,9 ±0,1	7,9
2	ADY (LittoLevure, (control)	11,1±0,1	8,2±0,2	0,52±0,03	3,15±0,03	214,5±0,03	3,0 ±0,1	7,9
3	Nr.2 - Cricova-2	11,5±0,1	8,2±0,1	0,48±0,03	3,15±0,02	214,2±0,02	1,1 ±0,1	8,0
4	Nr.8 - Cricova Chardonnay(3)	11,1±0,1	8,4±0,1	0,33±0,04	3,15±0,01	214,5±0,04	1,7 ±0,1	7,9
5	Nr.9 - Cricova Chardonnay(4)	11,0±0,1	8,6±0,1	0,62±0,03	3,13±0,02	215,2±0,05	3,0 ±0,1	7,8
6	Nr.10 - 1S	11,2±0,1	8,4±0,2	0,59±0,04	3,14±0,02	214,8±0,04	1,9±0,1	7,9
7	Nr.14 - 1VT	11,4±0,1	8,5±0,1	0,39±0,04	3,15±0,05	214,0±0,01	2,3 ±0,1	8,0
8	Nr.16-3VT	11,3±0,1	8,6±0,1	0,73±0,02	3,12±0,02	215,7±0,05	1,9 ±0,1	7,8

The concentration of titrable acids in dry white wines obtained under microvinification conditions varies insignificantly and, depending on the strain used, varies within the limits of 8.2-8.6 g/dm^3 (table 4.1).

Variation of pH index values in dry white wine samples obtained using different yeast strains, is in a narrow range and is 3.12-3.17 depending on the strain used.

The lowest redox potential was recorded in dry white wines fermented with yeast strains No. 2 - Cricova-2, No. 14 - 1VT and No. 29 - Rara-Neagră-2. At the same time, these 3 yeast strains are even more resistant to cold, compared to others. Strict adherence to technological processes in grape processing allowed obtaining dry white wines with a low redox potential, which ranges from 214.0 to 215.7 mV(table 4.1).

The mass concentration of volatile acids varies in the wines obtained in the range of 0.33-0.73 g/dm^3, which can be explained by the course of different enzymatic reactions.

The residual sugar values in dry white wines do not exceed the permissible limits of 4 g/dm^3, which is characteristic for this category of wines (table 4.1).

An important component of dry white wines is glycerol, which participates in the formation of the wine taste and gives it a sweet and soft sensation [17]. The results regarding the amount of glycerol in white wines, obtained using yeast strains selected from the ′Chisinau′ winemaking center, are presented in figure 4.1. Thus, all the investigated wines contain about 6 g/dm^3 of glycerol, but maximum amounts of 7.2-7.3 mg/dm^3 were established in those fermented with strains No.2 - Cricova-2 and No.16 - 3VT. The lowest amounts of glycerol were recorded in dry white wines fermented with strains No.8 - Cricova Chardonnay(3), No.9 - Cricova Chardonnay(4), No.10 - 1S, No.14 - 1VT and No.29 - Rara-Neagră-2 (figure 4.1.).

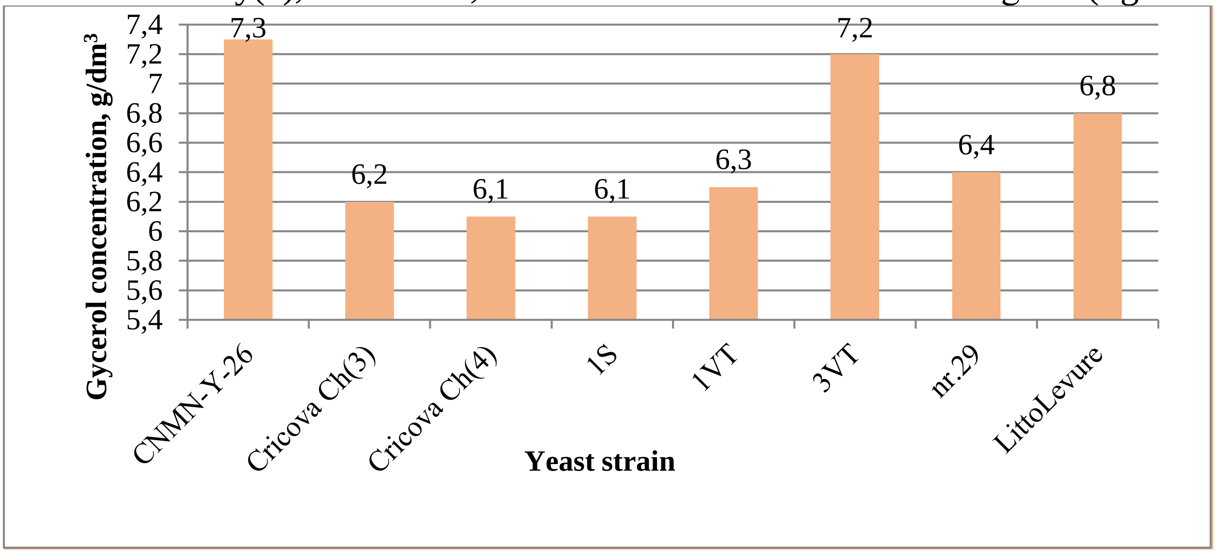

Fig. 4.1. Glycerol concentration in dry white Chardonnay wines obtained with using different selected yeast strains

Thus, we can mention the significant influence of yeast strains on the glycerol content in the dry white wines obtained. These findings highlight the importance of choosing the right yeast strains in the wine production process, having a direct impact on the organoleptic characteristics of the final product.

In order to establish specific aromas in dry white wines, characteristic of each selected yeast strain,the organoleptic assessment of dry white wines was carried out, obtained by fermentation with different strains of yeast from the 'Chisinau' wine center.

The analysis of the obtained results indicates that when using the yeast strain No. 2 - Cricova-2 (CNMN-Y-26) the dry white wine acquires a floral and fruity aroma, white fruit aromas such as apple predominate, offering a sensation of freshness and vitality, and when using the yeast strain No. 14 - 1VT, citrus aromas predominate. Faint herbaceous nuances were highlighted in the wine fermented with the yeast strain No. 16 - 3VT, and yeasty nuances were detected in the wine sample with the use of ADY. The other dry white wines were appreciated with a clean varietal aroma.

In this way, research conducted under micro-vinification conditionsÞie in the 2011 wine campaign they demonstrated that the use of the selected yeast strain No. 2 - Cricova-2 (CNMN-Y-26) in the fermentation of must from the Chardonnay grape variety allows the production of high-quality dry white wines, both in terms of physico-chemical indices and organoleptic notes, and does not yield to the quality of wine obtained using imported ADY.

The results obtained indicate that the physicochemical composition and quality of dry white wines depend largely on the yeast strain used in the must fermentation process. To obtain high-quality dry white wines in the must fermentation process, it was recommended to use the yeast strain isolated from the Chișinău wine center No. 2 - Cricova-2 (CNMN-Y-26) in production.

4.2. Study of the influence of different yeast strains selected from the ′Purcari′ wine center on the quality of dry white and red wines

4.2.1. Influence of selected yeast strains on the physicochemical indices of dry white Aligote and dry red Cabernet-Sauvignon wines

Dry white and red wines obtained by fermentation with different yeast strains from the ′Purcari′ wine center were subjected to physico-chemical analyses, the results of which are presented in tables 4.2 and 4.3.

Table 4.2. Physico-chemical indices of dry white Aligote wines obtained with different strains of selected yeasts (y.h. 2017)

№	Strain	Alcohol concentration, % vol.	The mass concentration of		pH	Potential OR, mV	Residual sugars, g/dm^3	Note organoleptic, points
			titrable acids, g/dm^3	volatile acids, g/dm^3				
1	Oenoferm Freddo (martor)	12,4±0,1	6,3±0,1	0,39±0,04	3,15±0,03	204,0±0,9	2,1±0,1	7,95
2	Nr.1 - FNFTP-1	12,4±0,1	6,2±0,1	0,39±0,03	3,15±0,02	204,2±1,1	2,3±0,1	7,90
3	Nr.3 - FNFTP-6	12,3±0,1	6,3±0,1	0,36±0,04	3,15±0,02	204,2±1,7	1,5±0,1	8,00
4	Nr.7 - F-75-FTP-5	12,4±0,1	6,2±0,1	0,39±0,03	3,13±0,01	205,2±0,8	2,5±0,1	7,90
5	Nr.12 - Ch75P-3ÎF	12,4±0,1	6,3±0,2	0,39±0,04	3,14±0,03	204,8±0,9	3,2±0,1	8,00

Thus, we can mention that dry white and red wines fermented using selected yeast strains are characterized by a high ethyl alcohol concentration of 12.4% vol. for dry white wines and 13.5% vol. for dry red wines.

The concentration of titrable acids in wines obtained under microvinification conditions varies insignificantly within the limits of 6.2-6.3 g/dm^3 for dry white wines and, respectively, 7.4-7.5 g/dm^3 for dry red wines (tables 4.2 and 4.3).

The mass concentration of volatile acids varies in dry white wines obtained in the range of 0.36-0.39 g/dm3 and in dry red wines obtained in the range of 0.46-0.52 g/dm3. The lowest mass concentration of volatile acids was determined in dry white wine fermented using strain No. 3 -FNFTP-6, and the highest in dry red wine fermented using strain No. 21 - CS-120-P-2.

Table 4.3. Physico-chemical indices of dry red Cabernet-Sauvignon wines obtained with different selected yeast strains (y.h.2017)

№	Strain	Alcohol concentration, % vol.	The mass concentration of		pH	Potential OR, mV	Residual sugars, g/dm^3	Note organoleptic, points
			titrable acids, g/dm^3	volatile acids, g/dm^3				
1	Oenoferm Be-Red (martor)	13,5±0,1	7,4±0,2	0,46±0,03	3,28± 0,09	211,1±1,1	3,2±0,1	7,95
2	Nr.21 - C-S-120-P-2	13,5±0,1	7,4±0,1	0,52±0,02	3,28± 0,11	211,1±0,9	3,1±0,1	7,80
3	Nr.24 - R-NNP-2	13,4±0,1	7,5±0,1	0,46±0,03	3,25± 0,08	212,0±0,8	4,2±0,1	7, 95
4	Nr.29 - R-N-120-P-4	13,5±0,1	7,4±0,2	0,52±0,03	3,25± 0,09	212,0±1,1	3,1±0,1	7,85
5	Nr.30 - R-N-120-P-5	13,5±0,1	7,4±0,2	0,46±0,03	3,28± 0,05	211,1±1,2	1,2±0,1	8,00

The variation in the pH value in dry white wines using different yeast strains is within a narrow range and is 3.13-3.15, and for dry red wines 3.25-3.28 depending on the strain used.

Strict adherence to technological procedures in grape processing allowed the production of experimental wines with a low oxidation-reduction potential, which ranges from 204.8 to 205.2 mV in white wines and from 211.1 to 212.0 mV in red wines.

The residual sugar values in dry wines do not exceed the permissible limits of 4 g/dm^3, which is characteristic for this category of wines, the exception being red wine fermented using yeast strain No. 24 - R-NNP-2, where the mass concentration of residual sugars is 4.2 g/dm^3.

The results regarding the amount of glycerol in dry white and red wines, obtained using yeast strains selected from the ′Purcari′ winemaking center, are presented in figure 4.2. Thus, all the wines investigated contain about 7 g/dm^3 of glycerol, and the highest values were determined in the samples fermented by strains No. 1 - FNFTP-1, No. 7 - F-75-FTP-5, No. 12 - Ch75P-3ÎF and No. 30 RN-120-P-5, which vary within the limits of 8.0 - 8.3 g/dm^3. The lowest glycerol values were determined in dry red wines, obtained using yeast strains No. 21 - CS-120-P-2 and No. 29 - RN-120-P-4 (figure 4.2). Thus, we can see a great influence of yeast strains on the glycerol content in the dry white and red wines obtained.

These results clearly highlight the significant influence of yeast strains on the glycerol content in dry white and red wines, thus highlighting the importance of careful selection of strains in the winemaking process to obtain the desired organoleptic characteristics.

In order to establish specific aromas in dry white and red wines, characteristic of each yeast strain selected from the ′Purcari′ wine center, the organoleptic assessment of the wines obtained was carried out.

The analysis of the obtained results indicates that when using yeast strains No. 1 - FNFTP-1 and No. 12 - Ch75P-3ÎF, dry white wines acquire a floral and fruity aroma, a fresh taste, and when using Oenoferm Freddo yeast, citrus aromas predominate. Yeast nuances were detected in wines fermented using yeast strains No. 3 - FNFTP-6 and No. 7 - F-75-FTP-5. The other dry white wines were appreciated for their clean and typical varietal aroma.

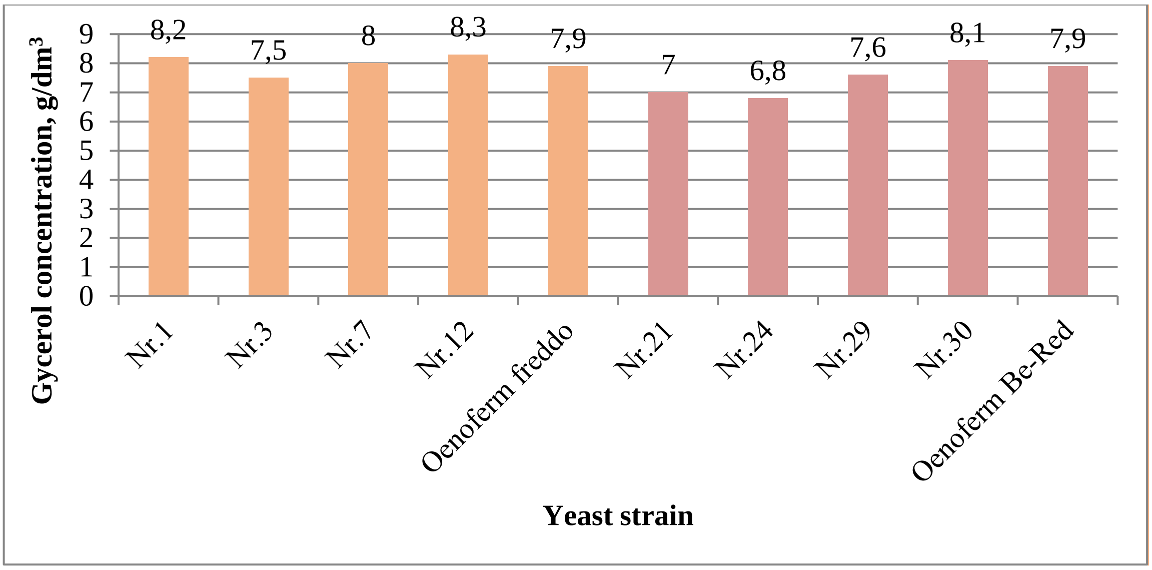

Fig. 4.2. Mass concentration of glycerol in dry white and red wines obtained with different selected yeast strains

The experimental dry red wines were characterized by rich red fruit aromas, AS blackberries and black cherries. Dry red wines fermented using yeast strains No. 24 - R-NNP-2, No. 30 - RN-120-P-5 and Oenoferm Be-Red were highlighted.The organoleptic analysis of dry red wines obtained under microvinification conditions demonstrated that the quality of the wine samples is high, and the wine fermented using the yeast strain No. 30 - RN-120-P-5 was rated with the highest score of 8.0 points (out of 10 max.).

Thus, research conducted under microvinification conditions in the 2017 wine campaign demonstrated that the use of yeast strains selected from the Purcari wine center No. 1 - FNFTP-1, No. 24 - R-NNP-2 and No. 30 - RN-120-P-5 allows obtaining high-quality dry white and red wines, both in terms of physicochemical indices and organoleptic notes, and does not yield to the quality of wine obtained using imported ADY.

4.2.2. Influence of yeast strains selected for the production of dry red wines on the concentration of phenolic substances and color indices of wines

An important factor in the production of dry red wines is played by yeast strains, which can directly influence the physicochemical indices of the wine and can contribute to the improvement of quality parameters, such as the content of phenolic substances, anthocyanins and organoleptic note. Yeasts, through their enzymatic system, particularly active during fermentation, catalyze the condensation of these compounds, which provides stability to the color of the wine.

For these reasons, the influence of different selected yeast strains on phenolic substances and color indices was studied. The results obtained are shown in table 4.4.

The analysis of the results presented in table 4.4 indicates that the yeast strains used in must fermentation have a major impact on the color indices of red wines.

The maximum color intensity was established in the dry red wine fermented with the control strain Oenoferm Be-Red (15.0), followed by the wines fermented with strains No. 21 - CS-120-P-2 and No. 30 - RN-120-P-5.

It was also established that the yeast strain No. 30 - RN-120-P-5 essentially favored the extraction of phenolic substances, and the maximum content of phenols is 1542 mg/dm³ and anthocyanins 298 mg/dm³. Red wine, obtained using the yeast strainOenoferm Be-Redalso obtained an advanced content of phenolic substances (1513 mg/dm³), but also having the lowest content of anthocyanins (266 mg/dm³), since the yeasts adsorb in significant quantities during the fermentation-maceration process. Dry red wines obtained with strains No. 21 - CS-120-P-2, No. 24 - R-NNP-2 and No. 29 - RN-120-P-4 were characterized by lower amounts of phenolic substances (1385-1420 mg/dm³) and anthocyanins (table 4.4).

To determine the influence of the yeast strain on the content of phenolic substances and anthocyanins in red wines, the extraction yield of these compounds was calculated compared to the technological reserve from grapes, which constitutes 2916 mg/dm^3 of phenolic compounds and 516 mg/dm^3 of anthocyanins. The results of determining the yield of phenolic substances and anthocyanins from grapes depending on the yeast strain used in the production of dry red wines are presented in figure 4.3.

Table 4.4. Phenolic substance content and color indices of dry red wines Cabernet-Sauvignon obtained using different yeast strains (2017 h.y.)

№	Yeast strain, No.	The sum of phenolic compounds, mg/dm³	Anthocyanin concentration, mg/dm³	Color intensity, (Ic=A420+A520+A620)	The hue of the color, (Nc=A420nm/A520nm)
1	Oenoferm Be-Red (martor)	1513±7	266±2	15,0±0,8	0,61±0,02
2	Nr.21 - C-S-120-P-2	1406±5	278±4	14,2±0,5	0,50±0,02
3	Nr.24 - R-NNP-2	1385±8	284±2	13,9±0,9	0,48±0,03
4	Nr.29 - R-N-120-P-4	1420±4	276±1	13,7±0,5	0,54±0,02
5	Nr.30 - R-N-120-P-5	1542±3	298±3	14,6±1,1	0,57±0,05

Based on the results obtained, we can conclude that yeast strains have a major influence on the content of phenolic substances in wines. The extraction yield of phenolic substances during the fermentation-maceration process of red wines varied between 47.5% (strain No.21 - CS-120-P-2) and 52.9% (strain No.30 - RN-120-P-5). The maximum extraction yield of anthocyanins (η=57.6 %) was established in dry red wine fermented with strain No.30 - RN-120-P-5.Anthocyanins are adsorbed in significant quantities on the cell walls of yeasts during fermentation-maceration depending on their nature and the structure of the cell wall.

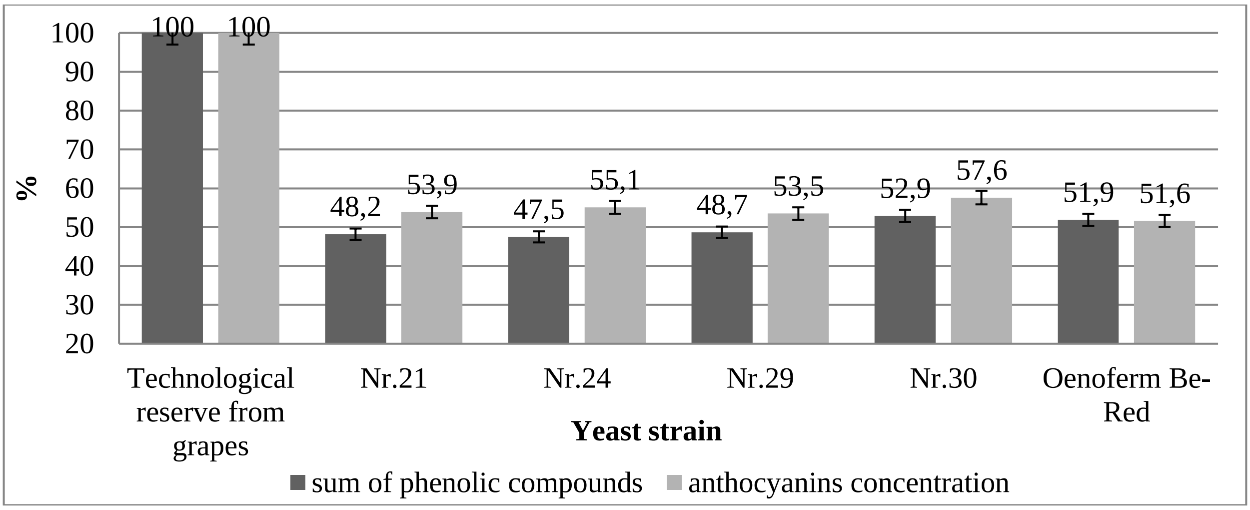

Fig. 4.3. The yield of phenolic substances and anthocyanins from grapes depending on the yeast strain used in the production of dry red wines (Cabernet-Sauvignon variety, 2017)

Thus, for the production of red wines with high anthocyanin content, it is necessary to select yeasts with low anthocyanin adsorption capacity, to minimize the loss of anthocyanins from wines and avoid compromising the color of the wine.

4.3. Study of the influence of different yeast strains selected from the ′Trifești′ wine center on the quality of dry white and red wines

4.3.1. Influence of yeast strains selected on the physicochemical indices of dry white Aligote and dry red Cabernet-Sauvignon wines

After completion of alcoholic fermentation, the dry white and red wines, obtained using different yeast strains selected from the ′Trifești′ wine center, were subjected to physicochemical analyses, and the results obtained are presented in tables 4.5 and 4.6.

Thus, we can mention that dry white and red wines fermented using selected yeast strains are characterized by a high ethyl alcohol concentration of 13.0% vol. for dry white wines and 15.0% vol. for red wines.

The concentration of titrable acids in wines obtained under microvinification conditions varies insignificantly within the limits of 6.6-6.7 g/dm^3 for dry white wines and 5.4-5.5 g/dm^3 for dry red wines, respectively.

The mass concentration of volatile acids varies in all wines obtained in the range of 0.36-0.52 g/dm^3, with maximum values being characteristic of red wines 0.46-0.52 g/dm^3.

Table 4.5. Physico-chemical indices of the dry white Aligote wines obtained with the use of different yeast strains (y. h. 2018)

№	Strain	Alcohol concentration, % vol.	The mass concentration of		pH	Potential OR, mV	Residual sugars, g/dm^3	Note organoleptic, points
			titrable acids, g/dm^3	volatile acids, g/dm^3				
1	Oenoferm Freddo (martor)	12,9±0,1	6,7±0,1	0,39±0,04	3,15±0,05	211,9±0,8	2,1±0,1	8,0
2	Nr.1 - STr-1	12,8±0,1	6,7±0,1	0,39±0,03	3,15±0,04	211,1±1,1	2,3±0,1	7,9
3	Nr.10 - S75Tr-2	12,9±0,1	6,7±0,1	0,36±0,04	3,15±0,05	212,0±1,3	1,5±0,1	7,9
4	Nr.15 - S75Tr-4.4	13,0±0,1	6,6±0,1	0,39±0,03	3,13±0,03	212,0±0,8	1,5±0,1	8,0
5	Nr.19 - ATr-2	12,9±0,1	6,7±0,2	0,39±0,04	3,14±0,03	211,7±0,9	3,2±0,1	7,9
6	Nr.22 - ATr-2.3	12,9±0,1	6,7±0,2	0,39±0,04	3,14±0,04	211,1±1,2	2,2±0,1	8,0

The pH values in the control wine samples and those obtained using the selected strains are practically identical and vary between 3.13-3.15 for the white ones and 3.25-3.28 for the red ones. Rigorous adherence to the technological procedures in the grape processing process led to the production of dry white and red wines with a low red-oxidation potential, located in the range from 211.1 to 215.2 mV.

The residual sugar values in dry white and red wines do not exceed the permissible limits of 4 g/dm^3, which is characteristic for this category of wines.

Table 4.5. Physico-chemical indices of dry red Cabernet-Sauvignon wines obtained using different yeast strains (2018 h.y.)

№	Strain	Alcohol concentration, % vol.	The mass concentration of		pH	Potential OR, mV	Residual sugars, g/dm^3	Note organoleptic, points
			titrable acids, g/dm^3	volatile acids, g/dm^3				
1	Oenoferm Be-Red (martor)	14,8±0,1	5,4±0,2	0,46±0,03	3,28±0,03	214,8±0,6	3,5±0,1	8,0
2	Nr.27 MTr-4	14,8±0,1	5,4±0,1	0,52±0,02	3,28±0,02	214,0±0,2	3,0±0,1	8,0
3	Nr.32 M100Tr-1	15,0±0,1	5,5±0,1	0,46±0,03	3,25±0,03	214,0±0,4	2,2±0,1	8,0
4	Nr.35 M100Tr-4	14,9±0,1	5,4±0,2	0,52±0,03	3,25±0,04	215,2±0,8	2,5±0,1	7,9
5	Nr.41 C-S60Tr-2	15,0±0,1	5,4±0,2	0,46±0,03	3,28±0,04	214,8±0,9	2,1±0,1	8,1
6	Nr.43 C-S60Tr-4	14,9±0,1	5,4±0,1	0,52±0,02	3,28±0,01	214,8±0,8	3,1±0,1	8,0

An important component of dry wines is glycerol, and according to the data presented in Figure 4.4, all the studied wines have a glycerol concentration higher than 7 g/dm^3. The highest values were determined in the samples using yeast strains No. 15 - S75Tr-4.4, No. 22 - ATr-2.3, No. 32 - M100Tr-1 and No. 41 - C-S60Tr-2 and vary within the range of 8.1- 8.3 g/dm^3.

The lowest glycerol values were determined in dry white wines obtained using yeast strains No. 1 - STr-1, No. 19 - ATr-2 and in dry red wines obtained using yeast strains No. 27 - MTr-4 and Oenoferm Be-Red. Thus, we can conclude that the influence of selected yeast strains on glycerol formation in dry white and red wines is significant.

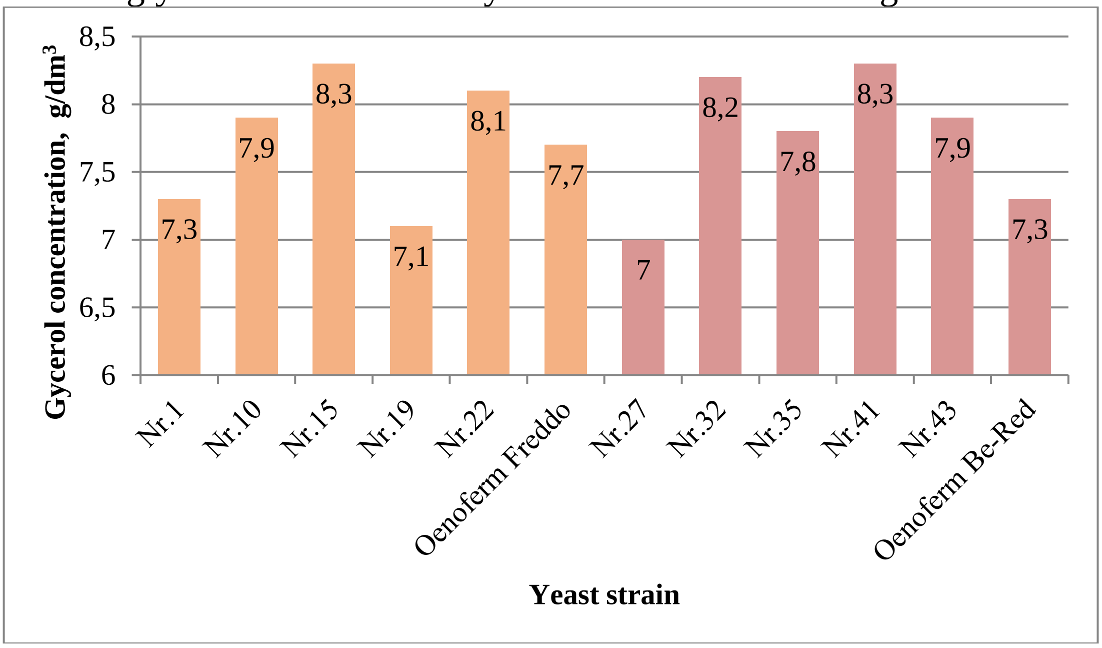

Fig. 4.4. Mass concentration of glycerol in dry white and red wines obtained using different selected yeast strains

In order to establish specific aromas in dry white and red wines, characteristic of each yeast strain selected from the ′Trifești′ wine center, an organoleptic assessment of the wines obtained was carried out.
The organoleptic analysis showed that when using yeast strains No.15 - S75Tr-4.4 and No.22 - ATr-2.3, the dry white wines acquire a floral and fruity aroma, and when using Oenoferm Freddo yeast, some citrus aromas are present. Yeast nuances were detected in the dry white wines fermented using yeast strains No.10 - S75Tr-2 and No.19 - ATr-2. The other dry white wines were appreciated for their clean and typical varietal aroma.

Dry red wines were characterized by rich aromas of red fruits, such as currants, blackberries and black cherries. Dry red wines fermented using yeast strains No.32 - M100Tr-1, No.41 - C-S60Tr-2 and Oenoferm Be-Red were highlighted. The taste of the wines was characterized by black fruits, including plums and black strawberries, and the aromas of cocoa, spices and sometimes dark chocolate contributed to the complexity of the wines obtained.The organoleptic analysis of dry red wines obtained under micro-vinification conditions demonstrated that the quality of the wine samples is high, and the wine fermented using the yeast strain No. 41 - C-S60Tr-2 was rated with the highest score of 8.1 points.

Thus, research conducted under micro-vinification conditionsÞie in the 2018 wine campaign they demonstrated that the use of yeast strains selected from the ′Trifești′ wine center No.15 - S75Tr-4.4, No.22 - ATr-2.3 for white wines and No.32 - M100Tr-1, No.41 - C-S60Tr-2 for red wines, it allows the production of high-quality dry white and red wines, both in terms of physico-chemical indices and organoleptic quality, and does not yield to the quality of wines obtained using imported ADY.

4.3.3. Influence of yeast strains selected for the production of dry red wines on the concentration of phenolic substances and color indices of wines

An important factor in the production of dry red wines is played by yeast strains, which directly affect the physicochemical indices of the wine and can contribute to improving quality parameters, such as the content of phenolic substances, anthocyanins and organoleptic note.

For these reasons, the influence of different selected yeast strains on phenolic substances and color indices was studied. The results obtained are shown in table 4.7.

The analysis of the results presented in table 4.7 demonstrates that the yeast strains used in must fermentation have a major impact on the chromatic indices of red wines.

The maximum color intensity was determined in dry red wine fermented using the yeast strainNo.41 - C-S60Tr-2(15.4), followed by the wine fermented with the control strain Oenoferm Be-Red. The lowest values were recorded in the wines fermented with the use of strains No.27 - MTr-4 and No.35 - M100Tr-4.

It was also established that the yeast strain No. 41 - C-S60Tr-2 essentially favored the extraction of phenolic substances, and the maximum content of phenols is 1520 mg/dm³ and anthocyanins 302 mg/dm³. Red wine, obtained using the yeast strainOenoferm Be-Redalso obtained an advanced content of phenolic substances (1496 mg/dm³), but also having the lowest content of anthocyanins (275 mg/dm³), because the yeasts adsorb significant amounts during fermentation-maceration. Dry red wines obtained using yeast strains No.27 - MTr-4, No.32 M100Tr-1, No.35 - M100Tr-4 and No.43 - C-S60Tr-4 have a lower content of phenolic substances (1436-1466 mg/dm³) and a moderate content of anthocyanins.

Table 4.7. Phenolic substances content and color indices of dry red Cabernet-Sauvignon wines obtained using different yeast strains (2018 h.y.)

№	Yeast strain, No.	The sum of phenolic compounds, mg/dm³	Anthocyanin concentration, mg/dm³	Color intensity, (Ic=A420+A520+A620)	The hue of the color, (Nc=A420nm/A520nm)
1	Oenoferm Be-Red (martor)	1496±5	275±2	14,8±0,5	0,60±0,05
2	Nr.27 - MTr-4	1439±4	287±3	13,6±0,4	0,47±0,03
3	Nr.32 - M100Tr-1	1442±5	292±2	13,8±0,4	0,48±0,03
4	Nr.35 - M100Tr-4	1436±2	284±4	13,3±0,3	0,45±0,04
5	Nr.41 - C-S60Tr-2	1520±7	302±7	15,4±0,6	0,62±0,05
6	Nr.43 - C-S60Tr-4	1466±5	310±8	13,9±0,4	0,55±0,02

To determine the influence of the yeast strain on the content of phenolic substances and anthocyanins in red wines, the extraction yield of these compounds was calculated compared to the technological reserve from grapes, which constitutes 2988 mg/dm^3 of phenolic compounds and 532 mg/dm^3 of anthocyanins. The results obtained are presented in figure 4.5.

Based on the results obtained, we can conclude that the selected yeast strains have a major influence on the content of phenolic substances in wines. The extraction yield of phenolic substances during the fermentation-maceration process of red wines varied between 48.1% (yeast strain No.35 - M100Tr-4) and 50.9% (yeast strain No.41 - C-S60Tr-2). The maximum extraction yield of anthocyanins (η=58.3 %) was established in dry red wine fermented with strain No.43 - C-S60Tr-4.

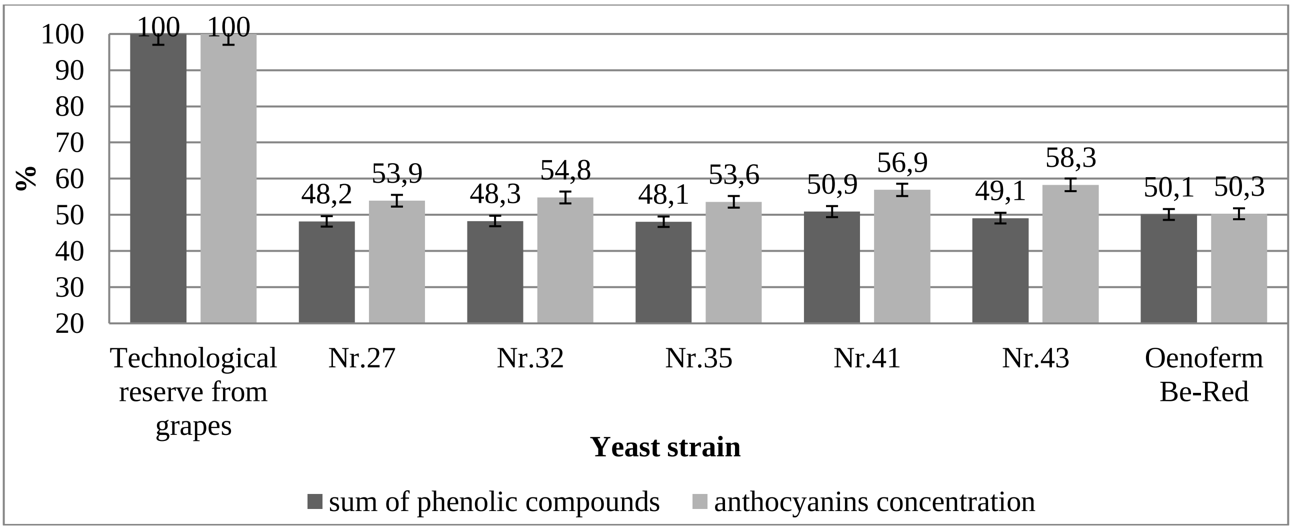

Fig. 4.5. The yield of phenolic substances and anthocyanins from grapes depending on the yeast strain used in the production of dry red wines (Cabernet-Sauvignon variety, fall 2018)

The advanced concentration of anthocyanins is the main parameter, which is responsible for the color of the wine and therefore the ratio (anthocyanins/phenolic substances) is the highest.

Based on the scientific results obtained, the yeast strain No. 2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 for the production of dry white wines was recommended for implementation in production conditions at SA "Cricova", at ÎM "Vinăria ′Purcari′" SRL the yeast strains No. 1-FNFTP-1- *S.cerevisiae* CNMN-Y-32, No. 12-Ch75P-3ÎF - *S.cerevisiae* CNMN-Y-33 for the production of dry white wines and the yeast strain No. 30-RN-120-

P-5 - *S.cerevisiae* CNMN-Y-31 for the production of dry red wines, at "Vierul-Vin" SRL the yeast strains No. 15-S75Tr-4.4 - *S.cerevisiae* CNMN-Y-34, No. 22-Atr-2.3 - *S.cerevisiae* CNMN-Y-35 for the production of dry white wines and yeast strains No.32-M100Tr-1- *S.cerevisiae* CNMN-Y-36, No.41-C-S60Tr-2 - *S.cerevisiae* CNMN-Y-37 for the production of dry red wines for the purpose of obtaining industrial batches of wines.

V. IMPLEMENTATION OF SELECTED YEAST STRAINS IN CONDITIONS PRODUCTION

5.1. Testing and implementation of yeast strains isolated and selected from the ′Chișinău′ wine center under production conditions

After completion of alcoholic fermentation, the dry white wines, obtained using the selected yeast strain and the control strain, were subjected to physicochemical analyses, and the results obtained are shown in table 5.1.

Dry white wine fermented using the yeast strain No.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 is characterized by an ethyl alcohol concentration of 11.7% vol., and the use of the ADY (Zymaflore yeast) yeast strain contributes to obtaining a wine with an alcoholic strength of 12.2% vol. (table 5.1), which can be explained by the differences in the initial sugar indices in the must.

The data in table 5.1 indicate that dry white wine fermented using the yeast strain No. 2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 and ADY (Zymaflore yeasts) is characterized by complete fermentation of sugars.

The mass concentration of residual sugars in dry white wines does not exceed the permissible limit of 4 g/dm^3 for this category of wines.

The concentration of titrable acids in the experimental dry white wines does not vary significantly and is 6.6-6.7 g/dm^3 for the wine fermented with the selected autochthonous strain and the control strain, respectively.

The mass concentration of volatile acids differs slightly, which is explained by the course of different enzymatic reactions and the high initial concentration of sulfurous anhydride, which directly influences the acetic acid content.

The use of yeast strain No.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 contributes to obtaining white wines with a higher content of non-reducing dry extract and constitutes 21.6 g/dm^3. According to the Regulation on the organization of the wine market, the mass concentration of non-reducing dry extract, expressed in grams per cubic decimeter, must be at least 15 g/dm^3 for white wines.

Table 5.1. Physico-chemical indices of dry white Chardonnay wines obtained with different yeast strains under production conditions (h.y. 2011)

№	Grape variety	Yeast strain	Physico-chemical indices					
			concentrations alcohol content, % vol.	Concentrația în masă a:				pH
				residual sugars, g/dm^3	titrable acids, g/dm^3	volatile acids, g/dm^3	non-reducing dry extract, g/dm^3	
1	Chardonnay	*Nr.2-Cricova-2 - S.cerevisiae CNMN-Y-26*	11,7±0,1	2,9±0,2	6,7±0,1	0,52±0,03	21,6±0,6	3,15±0,02
2	Chardonnay (control)	*Zymaflore yeast*	12,2±0,1	2,3±0,2	6,6±0,2	0,46±0,03	20,8±0,3	3,17±0,01

The pH value in experimental wine samples using different yeast strains varies within a narrow range and is 3.15-3.17 depending on the strain used.

In this way, the researched yeast strains contribute to the formation of dry white Chardonnay wines with a clean, fine aroma, the taste being full, full-bodied, harmonious and typical.

5.1.1. Determination of the content of volatile substances in dry white wine obtained using the yeast strain selected from the ′Chișinău′ winemaking center under production conditions

In order to study the influence of the selected yeast strain and the control strain (ADY) on the aromatic potential of dry white wines, obtained under production conditions, some volatile substances were determined: higher alcohols, fatty acids, esters, aldehydes, etc. [20, 31]. The results obtained are presented in table 5.2.

From the results presented in table 5.2, it can be concluded that the content of volatile substances in the dry white wines studied varies depending on the yeast strain used in must fermentation.

According to Rapp and Versini, a total concentration of higher alcohols lower than 300 mg/dm^3 for sure contribute to a complex aroma of the wine. Increasing the total concentration of these substances up to 400 mg/dm^3, influences negatively affects quality of wine.

In both samples of dry white Chardonnay wines studied, the sum of higher alcohols does not exceed it is the permissible limit and constitutes 236.9 mg/dm^3 in wine fermented with the use of yeast strain No. 2-Cricova-2 - *S.cerevisiae* CNMN-Y-26, and 268.4 mg/dm^3 in wine fermented using the Zymaflore yeast strain.

Isopentanol (isoamyl alcohol) is a specific higher alcohol that accounts for more than 50% of the total higher alcohols and predominates in the composition of wines [35]. In both samples of dry white wines, the concentration of isopentanol is quite low, but its content is 8.4 mg/dm^3 higher in the wine fermented using the Zymaflore yeast strain (Table 5.2).

The concentrations of isobutanol and phenyl-2-ethanol in fermented wines indicate that the Zymaflore yeast strain has a higher potential for producing these alcohols as a result of alcoholic fermentation of the must.

The concentration of hexanol in white wines can vary from 0.3 to 12 mg/dm^3 [12]. In the research conducted, dry white Chardonnay wine obtained using the selected autochthonous yeast strain contains 0.86 mg/dm^3 of hexanol, and wine fermented using the Zymaflore yeast strain contains 1.06 mg/dm^3, which is a positive factor on the quality of the wine.

1-Propanol was not detected in large quantities in the obtained wines, but its content is quite considerable, reaching 27.4 mg/dm^3 in the dry white wine fermented using the Zymaflore yeast strain, which is 9.2 mg/dm^3 more than in the wine fermented with the Nr.2-Cricova-2 yeast strain – *S. cerevisiae* CNMN-Y-26.

A significant difference is observed in the concentration of ethyl acetate, which influences the organoleptic properties, giving the wines a fresh apple aroma. In the case of using the indigenous yeast strain No.2-Cricova-2 - S.cerevisiae CNMN-Y-26, the concentration of ethyl acetate is lower than in the dry white wine that was fermented using ADY. But, it is necessary to mention, high concentrations of ethyl acetate negatively influence the organoleptic qualities of dry white wines.

Table 5.2. Content of volatile substances in dry white Chardonnay wines, (mg/dm^3)

№	Name of the sub-stance	Yeast strain	
		Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26	Zymaflore yeasts (control)
Higher alcohols			
1	Propanol-1	18,2±0,9	27,4±1,1
2	Hexanol	0,85±0,06	1,06±0,05
3	Isobutanol	21,4±1,0	31,5±1,09
4	Phenyl-2-ethanol	50,48±2,1	54,08±2,4
5	Isopentanol	146,0±3,7	154,4±5,8
6	∑ Higher alcohols	236,9±5,9	268,4±6,1
Fatty acids			
7	Butyric acid	0,67±0,03	0,71±0,02
8	Isovaleric acid	0,65±0,02	0,63±0,02
9	Caproic acid	4,27±0,09	4,80±0,08
10	Caprylic acid	2,48±0,07	2,52±0,05
11	Capric acid	0,050±0,004	0,09±0,007
12	∑ Fatty acids	8,12±0,21	8,75±0,32
Esters			
13	Ethyl acetate	25,7±1,2	47,8±2,6
14	Isobutyl acetate	0,010±0,001	0,010±0,001
15	Isoamyl acetate	1,66±0,05	1,82±0,04
16	Hexyl acetate	0,030±0,001	0,040±0,001
17	Phenylethyl acetate	0,48±0,03	0,52±0,04
18	Ethyl butyrate	0,14±0,02	0,12±0,01
19	Ethyl caprylate	0,32±0,02	0,26±0,01
20	Ethyl caproate	0,44±0,03	0,48±0,04
21	Ethyl caprate	0,020±0,001	0,020±0,001
22	∑Volatile esters	28,8±1,3	51,1±2,8

The analysis of the data presented in table 5.2 regarding the influence of yeast strains on the content of volatile substances in dry white wines reveals significant differences between two types of yeast used in the fermentation process. The concentrations of higher alcohols, isopentanol, isobutanol, phenyl-2-ethanol, hexanol and propanol-1, as well as fatty acids and ethyl esters of fatty acids, vary depending on the yeast strain used. The results obtained indicate that both samples of dry white wines fall within the recommended limits of higher alcohol concentrations. Thus, yeast strains significantly influence the aromatic profile and the final composition of dry white wine, contributing to obtaining a product with desired organoleptic characteristics.

5.1.2. Study of the ability of selected yeast strains to form sulfur compounds in the fermentation process in dry white Chardonnay wines

Aroma is one of the main factors contributing to the quality of dry white wines. The aroma of wine is a complicated mixture formed by a very large number of volatile compounds, which contains: alcohols, esters, aldehydes, ketones, fatty acids, terpenes, sulfur compounds, etc. The contribution of each compound to the overall aroma of wines can be estimated both positively and negatively.

Sulfur compounds (thiols, sulfides, etc.) play an important role in the formation of wine aroma, since they have a very low perception threshold and usually affect the organoleptic quality of the wine. Their presence in wines is due to the activity of yeasts, which are able to metabolize sulfites and sulfates added to the must, with the formation of volatile sulfur compounds, responsible for many olfactory defects in wines [12, 37, 38].

In order to evaluate the impact of selected yeast strains on the content of sulfur compounds, they were determined in experimental wines. By analyzing in detail the influence of selected yeast strains on sulfur compounds, the aim is to understand how they influence the organoleptic quality of wine. This research also aims to identify and evaluate the positive or negative influence of selected yeast strains on each volatile compound in the wine aroma, with particular emphasis on volatile sulfur compounds resulting from the fermentation process.

The results obtained can provide valuable information for improving wine production processes and optimizing the quality of dry white wines.

Table 5.3 presents the results of the determination of some sulfur compounds in dry white Chardonnay wines obtained under production conditions in 2011 at SA "Cricova".

The results obtained indicate that the concentrations of all the compounds determined in the experimental dry white Chardonnay wine does not differ significantly, compared to the control wine, fermented with ADY and no sulfur compound exceeds the threshold values of perception, but the wine fermented with Zymaflore yeasts shows higher concentrations for some compounds, such as ethanethiol, dimethyl disulfide and diethyl disulfide. These differences may influence the olfactory characteristics of the wines produced with this strain. Methanethiol shows traces in both experimental wines, and its concentrations are below the threshold of perception. This aspect can be considered beneficial for avoiding unpleasant aromas associated with this compound.

Thus, the indigenous yeast strain No. 2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 contributes to the formation of lower concentrations of sulfur compounds, improving the organoleptic quality of dry white Chardonnay wine.

Table 5.3. Comparative analysis of the content of sulfur compounds in dry white Chardonnay wines, μg/L

№	Sulfur compounds	Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26	Zymaflore yeasts (control)	Specific aroma	Organoleptic perception threshold [37,38]
1	Methanethiol	TRACES	TRACES	Onion, boiled cabbage	0,3 μg/L
2	Ethanethiol	0,48±0,02	0,64±0,02	Onion, rubber	1,1 μg/L
3	Dimethyl sulfide	3,15±0,01	3,21±0,05	Corn, boiled cabbage. Raspberries - low concentrations	27 μg/L
4	Diethyl sulfide	0,41±0,01	0,46±0,08	Boiled vegetables, onion, garlic	15-18 μg/L
5	Dimethyl disulfide	4,70±0,08	5,23±0,09	Boiled cabbage, onion	30-45 μg/L
6	Diethyl disulfide	1,80±0,05	2,11±0,05	Garlic, burnt rubber	25-40 μg/L

5.1.3. Influence of selected yeast strains on the process of biogenic amine formation

Biogenic amines, such as ethanolamine, phenylethylamine, methylamine, agmatine, histamine, putriscine, cadaverine, and tyramine, are low molecular weight organic bases found in wine that are formed primarily by microbial decarboxylation of the respective amino acid. These compounds negatively affect the hygienic condition of wine. [21, 30].

Biogenic amines are compounds of nitrogen metabolism and can cause toxic effects depending on the concentration and individual sensitivity. They are a real problem for people sensitive to histamine. In general, biogenic amines cause headaches and other adverse effects. Moreover, the ethanol present in wine reinforces the toxic effects of biogenic amines. Yeasts actively participate in the generation of biogenic amines, as they are able to produce histamine, an amine known for its significant level of toxicity [24, 32].

Because winemaking is a complicated biotechnological process, related to the activity of yeasts, It was necessary to determine the content of biogenic amines, which is strictly limited by EU regulatory documents. The results of the analyses performed are presented in table 5.4.

Table 5.4. The content of biogenic amines in dry white wines obtained with different yeast strains selected under production conditions at SA "Cricova", (mg/dm^3)

№	Wine name	Yeast strain	Putriscina	Cadaverine	Histamine	tyramine	Ethanolamine
1	Chardonnay (control)	*Zymaflore yeasts*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit
2	Chardonnay	*Nr.2-Cricova-2 - S.cerevisiae CNMN-Y-26*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit

According to the results presented in Table 5.4, it is observed that the use of the indigenous yeast strain in alcoholic fermentation did not contribute to the increase in the content of biogenic amines in dry white wines. In the analyzed wine samples, only traces of histamine were detected, which do not influence the quality of the wines obtained.

5.2. Testing and implementation of yeast strains isolated and selected from the 'Purcari' wine center under production conditions

5.2.1. Determinarea indicilor fizico-chimici ai vinurilor albe și roșii seci obținute în condiții de producere

Based on the research conducted and the results obtained under the laboratory and microvinification conditions of SPIHFT, the yeast strain No. 1-FNFTP-1- *S.cerevisiae* CNMN-Y-32 was selected for the production of dry white wines and the yeast strain No. 30-RN-120-P-5 - *S.cerevisiae* CNMN-Y-31 for the production of dry red wines for the purpose of obtaining industrial batches of wine.

The comparative study of the fermentation process of must from the Chardonnay and Pinot gris grape varieties and must from the Malbec grape variety, harvested from the plantations of the 'Purcari' wine center (h.y.2017), in order to obtain dry white and red wines using the indigenous yeast strains No. 1-FNFTP-1- *S.cerevisiae* CNMN-Y-32 and No. 30-RN-120-P-5 - *S.cerevisiae* CNMN-Y-31, was carried out under production conditions at the ÎM „Vinăria Purcari" SRL.

Industrial Active dry yeast (Anchor Alchemy I, Anchor NT 202, South Africa) were used as controls for the alcoholic fermentation of must and must. The initial physicochemical characteristics of Chardonnay, Pinot gris and Malbec grape must are shown in table 5.5.

The studied yeast strains were subjected to research in order to determine their fermentative activity and their influence on the physicochemical and organoleptic indices of dry white and red wines.

Table 5.5. Physico-chemical indices of grape must used for the comparative assessment of indigenous yeast strains (h.y. 2017)

№	Grape variety name	The volume, decalitre	Mass concentration of sugars, g/dm^3	Mass concentration of titrable acids, g/dm^3	pH	OR potential, mV
1	Chardonnay	1000	218,0±0,5	6,8±0,1	3,20±0,03	214,0±0,1
2	Pinot gris	1000	210,0±0,5	6,1±0,1	3,20±0,01	214,0±0,2
3	Malbec	2000	191,0±0,5	8,7±0,1	3,15±0,04	214,5±0,2

After completion of alcoholic fermentation, the dry white and red wines, obtained using the selected yeast strains, were subjected to physico-chemical analyses, and the results of the evaluation are shown in tables 5.6 and 5.7.

Dry white Chardonnay wines fermented using yeast strains No.1-FNFTP-1- *S.cerevisiae* CNMN-Y-32 and active dry yeast Anchor Alchemy I are characterized by an identical concentration of ethyl alcohol, as are dry white Pinot gris wines fermented using yeast strains No.1-FNFTP-1- *S.cerevisiae* CNMN-Y-32 and active dry yeast Anchor Alchemy I.

Table 5.6. Physico-chemical indices of dry white wines obtained with different yeast strains selected under production conditions (h.y. 2017)

№	Grape variety	Yeast strain	Physico-chemical indices					
			Alcohol concentration, % vol.	Mass concentration of:				pH
				residual sugars, g/dm^3	titrable acids, g/dm^3	volatile acids, g/dm^3	non-reducing dry extract, g/dm^3	
1	Chardonnay (control)	*Anchor Alchemy I*	12,9±0,1	2,0±0,2	6,1±0,1	0,42±0,03	19,5±0,3	3,15±0,02
2	Chardonnay	*Nr.1-FNFTP-1- S.cerevisiae CNMN-Y-32*	13,0±0,1	1,2±0,2	6,1±0,1	0,39±0,03	19,8±0,3	3,17±0,03
3	Pinot gris (control)	*Anchor Alchemy I*	12,4±0,1	1,8±0,2	5,6±0,1	0,42±0,03	20,1±0,3	3,16±0,01
4	Pinot gris	*Nr.1-FNFTP-1- S.cerevisiae CNMN-Y-32*	12,4±0,1	2,0±0,2	5,5±0,1	0,42±0,03	20,0±0,3	3,16±0,02

The data in Table 5.6 indicate that dry white wines fermented using the indigenous yeast strains No. 1-FNFTP-1- *S.cerevisiae* CNMN-Y-32 are characterized by the complete fermentation of sugars in the must.The mass concentration of residual sugars in dry white wines does not exceed the permissible limit of 4 g/dm^3 for this category of wines.

The concentration of titrable acids in Chardonnay wine does not vary and is 6.1 g/dm^3 for wine fermented with the experimental strain and the control strain.

The mass concentration of volatile acids differs insignificantly and constitutes 0.39-0.42 g/dm^3, a phenomenon explained by the diversity of enzymatic reactions and the initial level of sulfurous anhydride, which directly affects the acetic acid content.

The concentration of the non-reducing dry extract does not vary significantly and is within the range of 19.5 – 19.8 g/dm^3 for Chardonnay wines and 20.0 – 20.1 g/dm^3 for Pinot gris wines.

The pH value in the experimental dry white wine samples using different yeast strains is in a narrow range and is 3.15-3.17 depending on the strain used.

The concentration of ethyl alcohol in dry red Malbec wines fermented using yeast strains No.30-RN-120-P-5 - *S.cerevisiae* CNMN-Y-31 is identical to the concentration of ethyl alcohol in dry red Malbec wines fermented using active dry yeast Anchor NT 202 (tab.5.7).

The data in Table 5.7 indicate that dry red wines fermented using indigenous yeast strains No. 30-RN-120-P-5 - *S.cerevisiae* CNMN-Y-31 are characterized by complete fermentation of sugars.

The mass concentration of residual sugars in dry red wines does not exceed the permissible limit of 4 g/dm^3 for this category of wines.

The concentration of titrable acids in dry red wines does not vary significantly and is 8.0 g/dm^3 for wine fermented with the experimental strain and the control strain.

The mass concentration of volatile acids does not differ and is 0.52 g/dm^3 for wine fermented with the experimental strain and the control strain.

The mass concentration of the non-reducing dry extract in dry red wines does not vary significantly and is 21.3 g/dm^3 for the wine fermented using active dry yeast Anchor NT 202, and for the wine fermented using No. 30-RN-120-P-5 - *S.cerevisiae* CNMN-Y-31 the concentration is 21.6 g/dm^3. According to the Regulation on the organization of the wine market, the mass concentration of the non-reducing dry extract, expressed in grams per cubic decimeter, must be at least 18 g/dm^3 for dry red wines.

The pH value in experimental wine samples using different yeast strains does not differ and is 3.15.

Comparative analysis indicates that the wine samples obtained have similar physicochemical indices, with insignificant variations between varieties and yeast strains.

The selected yeast strain No.1-FNFTP-1- *S.cerevisiae* CNMN-Y-32 contributes to the formation of dry white Chardonnay wines with a clean, fine aroma, the taste being full, full-bodied, harmonious. The dry white wine obtained from the Pinot gris grape variety using the strain No.1-FNFTP-1- *S.cerevisiae* CNMN-Y-32 is characterized by a clean, harmonious, full, soft, balanced taste, floral aroma, such as jasmine and acacia flowers. These aromas contribute to obtaining a fragrant character of the wine.The selected yeast strain No.30-RN-120-P-5 - *S.cerevisiae* CNMN-Y-31 contributes to the formation of dry red Malbec wines with a clean aroma, aromas of cherries and plums, the taste being full, full-bodied, tannic, harmonious.

The analysis of the results obtained demonstrated that both in the case of using indigenous yeast strains and in the case of using active dry yeast, clean, complex and typical aromas predominate in the wines.

Table 5.7. Physico-chemical indices of dry red wines obtained with selected yeast strains under production conditions (h.y. 2017)

№	Grape variety	Yeast strain	Physico-chemical indices					
			Alcohol concentration, % vol.	Mass concentration of:				pH
				residual sugars, g/dm^3	titrable acids, g/dm^3	volatile acids, g/dm^3	non-reducing dry extract, g/dm^3	
1	Malbec (control)	*Anchor NT 202*	11,4±0,1	1,9±0,2	8,0±0,1	0,52±0,03	21,3±0,3	3,15± 0,03
2	Malbec	*Nr.30-R-N-120-P-5 - S.cerevisiae CNMN-Y-31*	11,3±0,1	1,5±0,2	8,0±0,1	0,52±0,03	21,6±0,3	3,15± 0,02

5.2.2. Determination of the content of volatile substances in dry white and red wines obtained using yeast strains selected from the ′Purcari′ wine center under production conditions

In order to study the influence of selected yeast strains on the aromatic potential of dry white and red wines, obtained under production conditionsit is, some volatile substances were determined: higher alcohols, fatty acids, esters, aldehydes, etc. The results of the volatile substances content are presented in table 5.8.

Table 5.8. Content of volatile substances in dry white and red wines, (mg/dm^3)

№	Name of the substance	Chardonnay		Pinot gris		Malbec	
		Yeast strain					
		Nr.1-FNFTP-1- *S.cerevisiae* CNMN-Y-32	Anchor Alchemy I (control)	Nr.1-FNFTP-1- *S.cerevisiae* CNMN-Y-32	Anchor Alchemy I (control)	Nr.30-R-N-120-P-5 - *S.cerevisiae* CNMN-Y-31	Anchor NT 202 (control)
Higher alcohols							
1	Propanol-1	18,1±0,9	25,4±2,1	18,7±0,8	28,9±2,7	19,2±0,5	28,7±1,9
2	Hexanol	0,75±0,04	0,86±0,09	0,83±0,03	0,99±0,09	0,89±0,07	1,12±0,05
3	Isobutanol	22,4±1,3	30,5±1,8	21,9±0,9	32,4±1,6	22,1±0,9	23,1±0,7
4	Phenyl-2-ethanol	51,48±3,21	53,08±2,53	52,12±1,15	56,09±3,27	49,80±1,51	52,10±2,12
5	Isopentanol	156,0±5,2	164,4±6,1	151,2±4,9	163,2±3,1	161,1±5,4	166,2±6,3
6	∑ Higher alcohols	248,7±8,9	274,2±7,5	244,7±8,7	281,5±8,1	253,1±9,7	271,2±9,9
Fatty acids							
7	Butyric acid	0,57±0,05	0,61±0,03	0,55±0,03	0,59±0,06	0,61±0,05	0,59±0,03
8	Isovaleric acid	0,55±0,04	0,53±0,03	0,47±0,03	0,51±0,04	0,57±0,04	0,61±0,03
9	Caproic acid	4,37±0,12	4,71±0,15	4,55±0,17	4,98±0,20	4,99±0,20	5,05±0,19
10	Caprylic acid	2,52±0,09	2,76±0,08	2,49±0,08	2,75±0,07	2,63±0,09	2,73±0,08
11	Capric acid	0,040±0,005	0,050±0,004	0,040±0,007	0,050±0,008	0,050±0,006	0,050±0,007
12	∑ Fatty acids	8,05±0,04	8,66±0,09	8,10±0,07	8,88±0,08	8,85±0,07	9,03±0,09
Esters							
13	Ethyl acetate	48,8±1,8	55,8±2,1	31,5±0,9	49,5±1,8	65,1±2,8	69,5±2,5
14	Isobutyl acetate	0,010±0,001	0,010±0,001	0,010±0,001	0,010±0,001	0,020±0,002	0,020±0,002
15	Isoamyl acetate	1,76±0,05	1,72±0,06	1,95±0,09	1,85±0,06	1,99±0,09	1,89±0,08
16	Hexyl acetate	0,030±0,001	0,040±0,002	0,030±0,001	0,040±0,002	0,040±0,001	0,040±0,001
17	Phenylethyl acetate	0,58±0,05	0,62±0,07	0,55±0,04	0,60±0,03	0,62±0,04	0,65±0,05
18	Ethyl butyrate	0,15±0,03	0,13±0,02	0,14±0,01	0,13±0,02	0,17±0,03	0,19±0,02
19	Ethyl caprylate	0,42±0,05	0,46±0,06	0,56±0,06	0,62±0,03	0,66±0,02	0,69±0,05
20	Ethyl caproate	0,54±0,03	0,58±0,02	0,55±0,03	0,62±0,02	0,72±0,04	0,82±0,04
21	Ethyl caprate	0,060±0,001	0,060±0,001	0,060±0,001	0,070±0,002	0,090±0,003	0,100±0,003
22	∑Volatile esters	52,3±1,8	59,2±2,1	35,3±0,9	53,4±2,2	69,4±3,2	73,9±3,7

From the results presented in table 5.8. it can be concluded that the content of volatile substances in dry white and red wines varies depending on the yeast strain used in the fermentation of the must or must.

In all wine samples studied, the sum of higher alcohols does not exceed the permissible limit of 300 mg/dm^3.

The concentration of isopentanol is quite low, but the concentration of this alcohol is higher in wines fermented using active dry yeast and is 163.2-166.2 mg/dm^3.

The concentrations of isobutanol and phenyl-2-ethanol in fermented wines indicate that active dry yeast have a higher potential to produce these alcohols as a result of alcoholic fermentation of must or must.

According to research, dry white and red wines obtained using indigenous yeast strains contain lower concentrations of hexanol compared to wines fermented using industrial active dry yeast.

Propanol-1 was not detected in large quantities in the wines obtained, but its content is quite considerable and constitutes 25.4 mg/dm^3 in the dry white Chardonnay wine fermented with the Anchor Alchemy I yeast strain, which is 7.3 mg/dm^3 more than in the wine fermented with the *No.1-FNFTP-1- S.cerevisiae CNMN-Y-32* yeast strain. In the Pinot gris white wine its concentration constitutes 28.9 mg/dm^3, which is 10.2 mg/dm^3 more than in the wine fermented with the *No.1-FNFTP-1- S.cerevisiae CNMN-Y-32* yeast strain. The concentration in Malbec red wine was 28.7 mg/dm^3, which is 9.5 mg/dm^3 more than in wine fermented using yeast strain *No. 30-RN-120-P-5 - S.cerevisiae CNMN-Y-31*.

The amount of fatty acids in the analyzed wine samples did not differ significantly. However, the synthesis of fatty acids by yeasts is highly variable, and changes that occur during alcoholic fermentation (pH, temperature, presence of nutrients), as well as the growth rate of yeasts, can affect the content of individual components.

A significant difference is observed in the concentration of ethyl acetate, which influences the organoleptic properties, giving the wines a fresh apple aroma. In the case of using the indigenous yeast strains *No. 1-FNFTP-1- S.cerevisiae CNMN-Y-32* and *No. 30-RN-120-P-5 - S.cerevisiae CNMN-Y-31*, the concentration of ethyl acetate is lower than in dry white and red wines, which were fermented using active dry yeast. At the same time, high concentrations of ethyl acetate negatively influence the organoleptic qualities of dry white and red wines.

The analysis of volatile substances in dry white and red wines obtained with yeast strains selected from the 'Purcari' wine center indicates significant variations in the content of these components.

Higher alcohols, such as propanol-1, hexanol, isobutanol, phenyl-2-ethanol and isopentanol, show significant differences between the dry white and red wines obtained and the yeast strains used. The concentrations of these alcohols are generally higher in wines fermented with active dry yeast Anchor Alchemy I.

Fatty acids, including butyric, isovaleric, caproic, caprylic and capric acid, indicate a relative uniformity in concentration between the samples analyzed, suggesting a consistent synthesis of these components in all wines analyzed.

Volatile esters such as ethyl acetate, isobutyl, isoamyl, hexyl, phenylethyl, ethyl butyrate, ethyl caprylate, ethyl caproate and ethyl caprate show significant variations between wines obtained using the selected strains. Ethyl acetate concentrations are higher in wines fermented with active dry yeast Anchor Alchemy I.

The general conclusions show that higher alcohols and fatty acids are within acceptable limits, and ethyl acetate concentrations significantly influence the organoleptic qualities of wines, being lower when using autochthonous strains.

In this way, the choice of yeast strains plays an essential role in the development of the aromatic profile of wines, highlighting significant variations in the content of volatile substances and influencing their organoleptic characteristics.

5.2.3. Study of the ability of selected yeast strains to form sulfur compounds in the fermentation process in dry white and red wines

The determination of sulfur compounds in wines has several key purposes: controlling organoleptic quality, monitoring yeast activity, preventing olfactory defects and ensuring compliance with production standards. Sulfur compounds, such as thiols and sulfides, have a very low perception threshold and can affect the organoleptic quality of wine.

Table 5.9. Comparative analysis of the content of sulfur compounds in dry white wines Chardonnay and Pinot gris, µg/L

№	Sulfur compounds	Chardonnay		Pinot gris		Specific aroma	Organoleptic perception threshold [37,38]
		(Nr.1-FNFTP-1- *S.cerevisiae* CNMN-Y-32)	(Anchor Alchemy I) control	(Nr.1-FNFTP-1- *S.cerevisiae* CNMN-Y-32)	(Anchor Alchemy I) control		
1	Methanethiol	TRACES	TRACES	TRACES	TRACES	Onion, boiled cabbage	0,3 µg/L
2	Ethanethiol	0,47±0,02	0,42±0,02	0,45±0,02	0,48±0,05	Onion, rubber	1,1 µg/L
3	Dimethyl sulfide	3,13±0,01	3,20±0,03	2,95±0,02	3,10±0,01	Corn, boiled cabbage. Raspberries - low concentrations	27 µg/L
4	Diethyl sulfide	0,42±0,01	0,45±0,06	0,38±0,07	0,40±0,03	Boiled vegetables, onion, garlic	15-18 µg/L
5	Dimethyl disulfide	4,71±0,08	4,56±0,03	4,10±0,03	4,10±0,08	Boiled cabbage, onion	30-45 µg/L
6	Diethyl disulfide	1,81±0,05	2,12±0,05	2,13±0,04	2,18±0,03	Garlic, burnt rubber	25-40 µg/L

Their low perception threshold means that even low concentrations can influence the quality of wine. The presence of these compounds in wines is due to the activity of yeasts, which can metabolize sulfites and sulfates added to the must, forming volatile sulfur compounds. Determining the concentrations of these compounds is essential to prevent or minimize potential olfactory defects and to ensure the production of higher quality wines. This analysis provides information on how yeasts influence the aromatic profile of wines and helps in making decisions in the production process [12].

Tables 5.9 and 5.10 present the results of the determination of sulfur compounds in dry white and red wines obtained under production conditions in 2017 at the ÎM "Vinăria Purcari" SRL.

The results obtained indicate that the concentrations of all determined compounds do not differ significantly compared to the control wines fermented with ADY and no sulfur compound exceeds the perception threshold values.

Table 5.10. Comparative analysis of the content of sulfur compounds in dry red Malbec wines, µg/L

№	Sulfur compounds	Malbec (Nr.30-R-N-120-P-5 - *S.cerevisiae* CNMN-Y-31)	Malbec (Anchor NT 202) control	Specific aroma	Organoleptic perception threshold [37,38]
1	Methanethiol	TRACES	TRACES	Onion, boiled cabbage	0,3 µg/L
2	Ethanethiol	0,64±0,06	0,66±0,02	Onion, rubber	1,1 µg/L
3	Dimethyl sulfide	3,21±0,03	3,25±0,03	Corn, boiled cabbage. Raspberries - low concentrations	27 µg/L
4	Diethyl sulfide	0,50±0,05	0,46±0,05	Boiled vegetables, onion, garlic	15-18 µg/L
5	Dimethyl disulfide	5,35±0,07	5,23±0,08	Boiled cabbage, onion	30-45 µg/L
6	Diethyl disulfide	2,15±0,05	2,11±0,01	Garlic, burnt rubber	25-40 µg/L

Thus, the selected indigenous yeast strains *No. 1-FNFTP-1- S.cerevisiae CNMN-Y-32* and *No. 30-RN-120-P-5 - S.cerevisiae CNMN-Y-31* contribute to the formation of low concentrations of sulfur compounds, thus improving the organoleptic quality of dry white and red wines.

5.2.4. Influence of selected yeast strains on the process of formation of biogenic amines

Since winemaking is a complicated biological process, related to the activity of yeasts, it was necessary to determine the content of biogenic amines, which is strictly limited by EU regulatory documents. The results of the analyses performed are presented in table 5.11.

According to the results presented in Table 5.11, it was established that the use of different yeast strains for fermentation did not contribute to the increase in the content of biogenic amines in dry white and red wines. Traces of histamine were detected in the analyzed wine samples, which do not influence the quality of the obtained wines.

Table 5.11. The content of biogenic amines in dry white and red wines obtained with different yeast strains selected under production conditions at the ÎM ”Vinăria Purcari”, (mg/L)

№	Wine name	Yeast strain	Putriscina	Cadaverine	Histamine	Tyramine	Ethanolamine
1	Chardonnay (control)	*Anchor Alchemy I*	Below detection limit	Below detection limit	<0,5	Below detection limit	Below detection limit
2	Chardonnay	*FNFTP-1*	Below detection limit	Below detection limit	<0,5	Below detection limit	Below detection limit
3	Pinot gris (control)	*Anchor Alchemy I*	Below detection limit	Below detection limit	<0,5	Below detection limit	Below detection limit
4	Pinot gris	*FNFTP-1*	Below detection limit	Below detection limit	<0,5	Below detection limit	Below detection limit
5	Malbec (control)	*Anchor NT 202*	Below detection limit	Below detection limit	<0,5	Below detection limit	Below detection limit
6	Malbec	*R-N-120-P-5*	Below detection limit	Below detection limit	<0,5	Below detection limit	Below detection limit

5.3. Testing and implementation of yeast strains isolated and selected from the 'Trifești' wine center under production conditions

After completion of alcoholic fermentation, the dry white and red wines, obtained using the selected yeast strains, were subjected to physicochemical analyses, and the results obtained are shown in tables 5.12 and 5.13.

Dry white Chardonnay wines fermented using yeast strains *No.15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34* and active dry yeast *IOC B-2000* are characterized by an identical concentration of ethyl alcohol, as well as dry white Muscat Ottonel wines, fermented using yeast strains *No.22-Atr-2.3 - S.cerevisiae CNMN-Y-35* and active dry yeast *IOC B-2000*.

The data in table 5.12 indicate that dry white wines fermented using the indigenous yeast strains *No. 15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34* and *No. 22-Atr-2.3 - S.cerevisiae CNMN-Y-35* are characterized by the complete fermentation of the sugars in the wine.

The mass concentration of residual sugars in white wines does not exceed the permissible limit of 4 g/dm^3 for this category of wines.

The concentration of titrable acids in dry white wines Chardonnay and Muscat Ottonel does not vary significantly and is in the range of 6.7-6.8 g/dm^3 and 5.8-5.9 g/dm^3, respectively.

The mass concentration of volatile acids does not differ for dry white wines Chardonnay and Muscat Ottonel, respectively.

The concentration of the non-reducing dry extract does not vary significantly and is within the range of 19.6 – 19.7 g/dm^3 for Chardonnay wines and 18.8 – 19.1 g/dm^3 for Muscat Ottonel wines.

The pH value in experimental wine samples using different yeast strains is within a narrow range and is 3.19-3.21 depending on the strain used.

The concentration of ethyl alcohol in dry red Merlot wines fermented using yeast strains *No.32-M100Tr-1- S.cerevisiae CNMN-Y-36* is identical to the concentration of ethyl alcohol in dry red Merlot wines fermented using active dry yeast *IOC R-9008*, as well as for dry red Cabernet Sauvignon wines fermented using yeast strains *No.41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37* and active dry yeast *IOC R-9008*.

Table 5.12. Physico-chemical indices of dry red wines obtained with different yeast strains under production conditions (h.y. 2019)

№	Grape variety	Yeast strain	Physico-chemical indices					
			Alcohol concentration, % vol.	Mass concentration of:				pH
				residual sugars, g/dm^3	titrable acids, g/dm^3	volatile acids, g/dm^3	non-reducing dry extract, g/dm^3	
1	Chardonnay (control)	*IOC B-2000*	12,6±0,1	2,1±0,2	6,7±0,1	0,39±0,03	19,6±0,3	3,19±0,02
2	Chardonnay	*Nr.15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34*	12,5±0,1	2,0±0,2	6,8±0,1	0,39±0,03	19,7±0,3	3,20±0,01
3	Muscat Ottonel (control)	*IOC B-2000*	12,9±0,1	4,0±0,2	5,9±0,1	0,49±0,03	19,1±0,3	3,21±0,01
4	Muscat Ottonel	*Nr.22-Atr-2.3 - S.cerevisiae CNMN-Y-35*	12,9±0,1	3,7±0,2	5,8±0,1	0,49±0,03	18,8±0,3	3,19±0,02

The data in Table 5.13 indicate that dry red wines fermented using the indigenous yeast strains *No. 32-M100Tr-1- S.cerevisiae CNMN-Y-36* and *No. 41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37* are characterized by complete fermentation of the sugars in the wine.

The mass concentration of residual sugars in dry red wines does not exceed the permissible limit of 4 g/dm^3 for this category of wines.

The concentration of titrable acids in dry red wines does not vary significantly and is within the range of 6.6-6.9 g/dm^3.

The mass concentration of volatile acids does not differ significantly and is in the range from 0.45 g/dm^3 to 0.52 g/dm^3.

The mass concentration of non-reducing dry extract in dry red wines does not vary significantly and ranges between 20.1-20.7 g/dm^3.

The pH value in experimental wine samples using different yeast strains does not differ significantly and is 3.27 for dry red wines. Merlot and 3.31 for Cabernet Sauvignon dry red wines.

The analysis of the results obtained demonstrated that when using both indigenous yeast strains and active dry yeast in dry white wines, clean, complex, typical, floral aromas predominate.

Selected yeast strains *No.15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34* contribute to obtaining dry white Chardonnay wines with a clean, fine aroma, the taste being full, full-bodied, harmonious. The dry white wine obtained from the Muscat Ottonel grape variety using the strain *No.22-Atr-2.3 - S.cerevisiae CNMN-Y-35* is characterized by a clean, harmonious, full, soft, balanced taste, floral aroma, as well as fruity notes, such as peach, lemon and grapefruit. These aromas contribute to the complexity and overall freshness of the wine.

Table 5.13. Physico-chemical indices of dry red wines obtained with different yeast strains under production conditions (h.y. 2019)

№	Grape variety	Yeast strain	Physico-chemical indices					
			Alcohol concentration, % vol.	Mass concentration of:				pH
				residual sugars, g/dm^3	titrable acids, g/dm^3	volatile acids, g/dm^3	non-reducing dry extract, g/dm^3	
1	Merlot (martor)	*IOC R-9008*	13,8±0,1	1,9±0,2	6,7±0,1	0,49±0,03	20,7±0,2	3,27±0,03
2	Merlot	*Nr.32-M100Tr-1-S.cerevisiae CNMN-Y-36*	13,7±0,1	2,5±0,2	6,6±0,1	0,45±0,03	20,5±0,4	3,27±0,02
3	Cabernet Sauvignon (martor)	*IOC R-9008*	13,5±0,1	2,9±0,2	6,7±0,1	0,56±0,03	20,1±0,3	3,31±0,03
4	Cabernet-Sauvignon	*Nr.41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37*	13,5±0,1	2,5±0,2	6,9±0,1	0,53±0,03	20,6±0,2	3,31±0,01

Dry red wines were characterized by rich aromas of red fruits,such as currants, blackberries and black cherries. The taste was characterized by black fruits, including plums and black strawberries, and the aromas of cocoa, spices and sometimes dark chocolate contributed to the complexity of the wines obtained. The organoleptic analysis of the dry red wines obtained under production conditions demonstrated that the quality of the wines is high, and the wines fermented with the use of yeast strains *No.32-M100Tr-1- S.cerevisiae CNMN-Y-36* and *No.41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37* were appreciated at the same level with a score of 8.1 points.

The researched yeast strains contribute to the formation of wines with a clean, fine aroma, the taste being full, full-bodied, harmonious.

5.3.1. Determination of the content of volatile substances in dry white and red wines obtained using yeast strains selected from the 'Trifești' wine center under production conditions

In order to study the influence of selected yeast strains on the aromatic potential of dry white and red wines, obtained under production conditions, some volatile substances were determined: higher alcohols, fatty acids, esters, aldehydes, etc. The results of the volatile substances content are presented in table 5.14.

From the results presented in Table 5.14, it can be concluded that the content of volatile substances in dry white and red wines varies depending on the yeast strain used in the fermentation of the must or must.

In all wine samples studied, the sum of higher alcohols does not exceed the permissible limit of 300 mg/dm^3.

The isopentanol concentration is quite low, but the content of this alcohol is higher in wines fermented with the use of active dry yeast and constitutes 169.2-174.4 mg/dm^3 (tab.5.14).

The concentrations of isobutanol and phenyl-2-ethanol in fermented wines indicate that active dry yeast have a higher potential to produce these alcohols as a result of alcoholic fermentation of must or must, the exception being the dry white wine Muscat Ottonel fermented with the indigenous yeast strain *Nr.22-Atr-2.3 - S.cerevisiae CNMN-Y-35* where the concentration of these compounds is higher than in the control sample. In the research conducted, dry white and red wines obtained using indigenous yeast strains contain lower concentrations of hexanol compared to wines fermented using industrial active dry yeast.

Propanol-1 was not detected in large quantities in the obtained wines, but its content is quite considerable, reaching 27.3 mg/dm^3 in the dry white Chardonnay wine fermented with the IOC B-2000 yeast strain, which is 6.2 mg/dm^3 more than in the wine fermented with the *Nr.15-S75Tr-4.4 yeast strain – S. cerevisiae CNMN-Y-34.* In the Muscat Ottonel white wine, its concentration is 28.5 mg/dm^3, which is 7.8 mg/dm^3 more than in the wine fermented with the *Nr.22-Atr-2.3 yeast strain – S. cerevisiae CNMN-Y-35*.

The concentrations of this compound in red wines do not differ significantly and are 18.2-19.2 mg/dm^3.

The total fatty acid content in the analyzed wine samples does not differ significantly. However, the synthesis of fatty acids by yeasts is highly variable.

A significant difference is observed in the concentration of ethyl acetate, which influences on the organoleptic properties, giving the wines a fresh apple aroma.

In the case of using indigenous yeast strains *No.15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34, No.22-Atr-2.3 - S.cerevisiae CNMN-Y-35, No.32-M100Tr-1- S.cerevisiae CNMN-Y-36* and *No.41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37* the concentration of ethyl acetate is lower than in wines that were fermented using active dry yeast. At the same time, high concentrations of ethyl acetate negatively influence the organoleptic qualities of dry white and red wines.

Table 5.14. Content of volatile substances in dry white and red wines, (mg/dm^3)

№	Name of the substance	Chardonnay		Muscat Ottonel		Merlot		Cabernet-Sauvignon	
		Yeast strain							
		Nr.15-S75Tr-4.4 - *S.cerevisiae* CNMN-Y-34	**IOC B-2000**	**Nr.22-Atr-2.3 - *S.cerevisiae* CNMN-Y-35**	**IOC B-2000**	**Nr.32-M100Tr-1-*S.cerevisiae* CNMN-Y-36**	**IOC R-9008**	**Nr.41-C-S60Tr-2 - *S.cerevisiae* CNMN-Y-37**	**IOC R-9008**
	Higher alcohols								
1	Propanol-1	21,1±0,8	27,3±1,2	20,7±0,8	28,5±1,4	18,2±0,5	18,7±0,4	18,5±0,6	19,2±0,9
2	Hexanol	0,85±0,05	0,96±0,06	0,93±0,04	0,95±0,05	0,83±0,03	1,05±0,06	0,99±0,03	1,07±0,06
3	Isobutanol	21,8±0,9	25,5±1,1	29,9±1,9	26,6±1,2	23,1±1,1	29,1±1,4	24,2±1,3	30,1±2,2
4	Phenyl-2-ethanol	56,32±2,1	52,08±2,2	57,21±2,8	54,19±2,1	51,61±2,1	53,12±2,5	54,21±3,2	53,19±2,9
5	Isopentanol	166,0±5,4	174,4±6,1	159,2±4,9	169,2±7,6	160,1±5,8	171,2±6,8	159,3±4,4	173,4±8,6
6	∑ Higher alcohols	266,07±9,9	280,24±7,8	267,94±5,9	279,44±9,8	253,84±6,7	273,13±7,8	257,20±7,4	276,96±8,2
	Fatty acids								
7	Butyric acid	0,59±0,02	0,63±0,03	0,59±0,02	0,62±0,03	0,60±0,01	0,61±0,02	0,61±0,02	0,62±0,03
8	Isovaleric acid	0,54±0,02	0,55±0,03	0,57±0,02	0,59±0,04	0,67±0,04	0,71±0,04	0,65±0,03	0,72±0,04
9	Caproic acid	4,25±0,08	4,61±0,09	4,35±0,12	4,68±0,11	5,03±0,17	5,15±0,09	5,05±0,11	5,12±0,14
10	Caprylic acid	2,62±0,09	2,78±0,10	2,59±0,08	2,76±0,11	2,73±0,10	2,83±0,12	2,69±0,10	2,78±0,09
11	Capric acid	0,050±0,003	0,050±0,005	0,040±0,002	0,050±0,005	0,060±0,006	0,070±0,006	0,060±0,004	0,070±0,005
12	∑ Fatty acids	8,05±0,12	8,62±0,21	8,14±0,17	8,70±0,21	9,09±0,32	9,37±0,27	9,06±0,21	9,31±0,17
	Esters								
13	Ethyl acetate	28,5±1,5	35,8±1,7	31,5±1,1	39,5±1,4	65,1±2,7	69,5±2,8	65,9±2,2	67,6±2,9
14	Isobutyl acetate	0,030±0,001	0,040±0,002	0,020±0,001	0,030±0,001	0,090±0,003	0,090±0,003	0,100±0,005	0,100±0,005
15	Isoamyl acetate	1,56±0,09	1,62±0,08	1,85±0,08	1,95±0,08	1,69±0,07	1,69±0,09	1,72±0,06	1,75±0,09
16	Hexyl acetate	0,030±0,001	0,040±0,002	0,030±0,001	0,040±0,002	0,040±0,003	0,040±0,001	0,040±0,002	0,050±0,003
17	Phenylethyl acetate	0,58±0,07	0,55±0,06	0,65±0,05	0,61±0,04	0,64±0,08	0,66±0,07	0,65±0,06	0,67±0,07
18	Ethyl butyrate	0,130±0,01	0,130±0,01	0,150±0,02	0,170±0,02	0,190±0,03	0,190±0,03	0,210±0,02	0,190±0,02

19	Ethyl caprylate	0,62±0,09	0,66±0,05	0,76±0,07	0,72± 0,07	0,86±0,09	0,89± 0,08	0,83± 0,06	0,89± 0,04
20	Ethyl caproate	0,74±0,09	0,78±0,07	0,75±0,08	0,72± 0,05	0,82±0,06	0,89± 0,05	0,85± 0,05	0,90± 0,06
21	Ethyl caprate	0,100±0,001	0,160±0,002	0,160±0,002	0,170± 0,001	0,190±0,003	0,190± 0,003	0,180± 0,002	0,210± 0,004
22	∑Volatile esters	32,3±1,2	39,8±2,1	35,9±1,8	43,9± 2,6	69,6±2,1	74,1± 2,8	70,5± 3,1	72,4± 3,5

5.3.2. Study of the ability of yeast strains to form compounds with sulfur in the fermentation process

The determination of sulfur compounds in wines fulfills multiple objectives of scientific and practical relevance in the wine industry. This process serves as an essential tool for controlling and ensuring the organoleptic quality of wine, monitoring yeast activity, preventing olfactory defects and guaranteeing compliance with rigorous production standards.

Sulfur compounds, including thiols and sulfides, have an extremely low threshold of perception, having the ability to substantially influence the organoleptic quality of wine even at minimal concentrations. Their low threshold of perception underlines the high sensitivity of tasters to these compounds, requiring careful and precise analysis.

Table 5.15. Comparative analysis of the content of sulfur compounds in dry white wines Chardonnay and Muscat Ottonel, µg/L

№	Sulfur compounds	Concentration of sulfur compounds				Specific aroma	Organoleptic perception threshold [37,38]
		Chardonnay		Muscat Ottonel			
		(Nr.15-S75Tr-4.4 - *S.cerevisiae* CNMN-Y-34)	(IOC B-2000) control	(Nr.22-Atr-2.3 - *S.cerevisiae* CNMN-Y-35)	(IOC B-2000) control		
1	Methanethiol	TRACES	TRACES	TRACES	TRACES	Onion, boiled cabbage	0,3 µg/L
2	Ethanethiol	0,37±0,02	0,37±0,02	0,55±0,01	0,58±0,03	Onion, rubber	1,1 µg/L
3	Dimethyl sulfide	4,23±0,01	4,55±0,03	4,95±0,02	4,10±0,02	Corn, boiled cabbage. Raspberry-low concentrations	27 µg/L
4	Diethyl sulfide	0,72±0,01	0,75±0,02	0,78±0,01	0,80±0,05	Boiled vegetables, onion, garlic	15-18 µg/L
5	Dimethyl disulfide	5,11±0,08	5,32±0,08	5,10±0,06	5,05±0,05	Boiled cabbage, onion	30-45 µg/L
6	Diethyl disulfide	2,11±0,07	2,22±0,05	2,33±0,05	2,58±0,06	Garlic, burnt rubber	25-40 µg/L

The origin of these compounds in wines is linked to the activity of yeasts, which can metabolize sulfites and sulfates added to the must, generating volatile sulfur compounds. By determining the concentrations of these compounds, potential olfactory defects can be prevented or minimized, thus strengthening the assurance of the quality and gustatory integrity of the wine.

This analysis provides essential scientific information on how yeasts influence the aromatic profile of wines. Moreover, the results obtained contribute to the reasoning of decisions in the production process, optimizing the control and manipulation of factors involved in the formation of sulfur compounds.

Tables 5.15 and 5.16 present the results of the determination of sulfur compounds in dry white and red wines obtained under production conditions in 2019 at "Vierul - Vin" SRL.

The results obtained indicate that the concentrations of all determined compounds do not differ significantly compared to the control wines fermented with active dry yeast and no sulfur compound exceeds the perception threshold values.

In this way, the selected autochthonous yeast strains *No.15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34, No.22-Atr-2.3 - S.cerevisiae CNMN-Y-35, No.32-M100Tr-1- S.cerevisiae CNMN-Y-36 and No.41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37* intended for obtaining dry white and red wines contribute to the formation of low concentrations of sulfur compounds, thus improving the organoleptic quality of dry white and red wines.

Table 5.16. Comparative analysis of the content of sulfur compounds in dry red wines Merlot and Cabernet-Sauvignon, µg/L

№	**Sulfur compounds**	**Concentration of sulfur compounds**				**Specific aroma**	**Organoleptic perception threshold [37,38]**
		Merlot		**Cabernet- Sauvignon**			
		(Nr.32-M100Tr-1-*S.cerevisiae* CNMN-Y-36)	**(IOC R-9008) control**	**(Nr.41-C-S60Tr-2 - *S.cerevisiae* CNMN-Y-37)**	**(IOC R-9008) control**		
1	Methanethiol	TRACES	TRACES	TRACES	TRACES	Onion, boiled cabbage	0,3 µg/L
2	Ethanethiol	0,61±0,02	0,62±0,01	0,65±0,02	0,68±0,02	Onion, rubber	1,1 µg/L
3	Dimethyl sulfide	5,13±0,01	5,21±0,04	5,25±0,01	5,31±0,01	Corn, boiled cabbage. Raspberry-low concentrations	27 µg/L
4	Diethyl sulfide	0,48±0,04	0,51±0,02	0,48±0,02	0,50±0,04	Boiled vegetables, onion, garlic	15-18 µg/L
5	Dimethyl disulfide	5,75±0,07	5,95±0,08	5,62±0,05	5,85±0,05	Boiled cabbage, onion	30-45 µg/L
6	Diethyl disulfide	2,56±0,09	2,78±0,09	2,25±0,07	2,81±0,07	Garlic, burnt rubber	25-40 µg/L

5.3.3. Influence of yeast strains on the process of biogenic amine formation

The purpose of determining biogenic amines in wines is to monitor and evaluate the content of these compounds to ensure the quality and safety of the product. Biogenic amines are nitrogenous organic substances, such as tyramine, histamine and putrescine, which can appear in wines as a result of microbial activity or other fermentation processes. Since the production of wines is a complicated biological process, related to the activity of yeasts, it was necessary to determine the content of biogenic amines, which is strictly limited by EU regulatory documents. The results of the analyses performed are presented in table 5.17.

According to the results presented in table 5.17, it was established that the use of different yeast strains for fermentation did not contribute to the increase in the content of biogenic amines in dry white and red wines. Traces of histamine were detected in the analyzed samples, which do not influence the quality of the wines obtained.

Table 5.17. The content of biogenic amines in dry white and red wines obtained using different yeast strains selected under production conditions at "Vierul-Vin" SRL, (mg/L)

№	Wine name	Yeast strain	Putriscina	Cadaverine	Histamine	Tyramine	Ethanolamine
1	Chardonnay	*Nr.15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit
2	Chardonnay (control)	*IOC B-2000*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit
3	Muscat Ottonel	*Nr.22-Atr-2.3 - S.cerevisiae CNMN-Y-35*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit
4	Muscat Ottonel (witness)	*IOC B-2000*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit
5	Merlot	*Nr.32-M100Tr-1-S.cerevisiae CNMN-Y-36*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit
6	Merlot (control)	*IOC R-9008*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit
7	Cabernet Sauvignon	*Nr.41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit
8	Cabernet Sauvignon (witness)	*IOC R-9008*	Below detection limit	Below detection limit	<0.5	Below detection limit	Below detection limit

5.4. Improvement of technological regimes for the production of dry white and red wines using selected indigenous yeasts

Based on studies conducted during the years 2009-2019, indigenous yeast strains isolated and selected from the wine centers "Chișinău," "Purcari," and "Trifești" with advanced technological properties were tested under production conditions at S.A. "Cricova," ÎM "Vinăria Purcari" SRL, and "Vierul-Vin" SRL, where experimental batches of dry white and red wines were produced.

Industrial-scale trials of the selected indigenous yeast strains for alcoholic fermentation of must and must slurry for the production of dry white and red wines demonstrated their positive influence on the fermentation process and the quality of the final product.

Commissions consisting of specialists from S.A. "Cricova," ÎM "Vinăria Purcari" SRL, and "Vierul-Vin" SRL, along with IȘPHTA, recommended the indigenous yeast strains obtained from various wine centers for use in the production of dry white and red wines.

Based on the results obtained under industrial conditions, the technological regimes for the production of dry white and red wines using indigenous yeasts were improved.

VI. THE INFLUENCE OF NON-*SACCHAROMYCES* YEAST ON THE QUALITY OF DRY WHITE WINES

In recent years, the use of non-Saccharomyces yeasts in must fermentation and the formation of dry white wines has increasingly concerned the interests of winemakers both in countries with a tradition in winemaking (France, Spain, Italy, Portugal, Greece) and more novice ones (Australia, Chile, Argentina, South Africa) [23, 36].

Some researchers consider that the impact of non-*Saccharomyces* yeasts on the quality of dry white wines is negative, others discover some favorable technological and organoleptic capacities of this group of yeasts. Their capacity to render complexity to the final product, to produce fruity varietal aromas, to exercise an enzymatic activity of potential interest it is reported in numerous scientific publications [1, 22, 23, 36].

6.1. Study of the influence of non-Saccharomyces yeast strains on the must fermentation process

For the comparative assessment of the influence of non-Saccharomyces yeast strains Torulaspora delbrueckii (Enartis FERM, Italy) on the physicochemical and organoleptic indices of white wines, the fermentation process was studied under laboratory conditions.

Non-Saccharomyces yeast strains can contribute to the development of a broader spectrum of aromatic compounds in wines. *Torulaspora delbrueckii*, in particular, is known for its ability to produce enzymes that can positively influence the aromatic profile of wine. This can lead to wines with more complex and varied aromas. Aligote grape must with an initial sugar concentration of 218 g/dm^3 was used as raw material.The fermentation of the must (3 L) was carried out at a temperature of 18±2°C and monitored for 23 days. The indigenous yeast strain *No. 2-Cricova-2 - S.cerevisiae CNMN-Y-26* was used as a control culture in the research.

Two fermentation schemes were used: co-inoculation, where non-Saccharomyces yeasts (10^5 CFU/mL) and *Saccharomyces* yeasts (10^6 CFU/mL) were inoculated simultaneously, and sequential fermentation, with inoculation of non-*Saccharomyces* yeasts (10^5 CFU/mL) and Saccharomyces yeasts (10^6 CFU/mL) after reaching an alcohol concentration of 3% vol. In the study process, the fermentative activity of mixed-combined and mixed-successive wort fermentation processes was determined under laboratory conditions. The results obtained are presented in figure 6.1.

According to the results obtained, it was established that the cultures and their combinations have an increased fermentation dynamics, the exception being the pure non-Saccharomyces strain. This is explained by the fact that the yeast strain *Torulaspora delbrueckii*, being a culture poorly resistant to high alcohol concentrations, demonstrates a reduced ability to ferment carbohydrates.

According to the results obtained, which are shown in figure 6.1, a more active fermentation is observed when using the indigenous yeast *strain No. 2-Cricova-2 - S.cerevisiae CNMN-Y-26*, which consumes all the sugars in the must within 20 days of inoculation, compared to the non-*Saccharomyces* yeast strain studied.

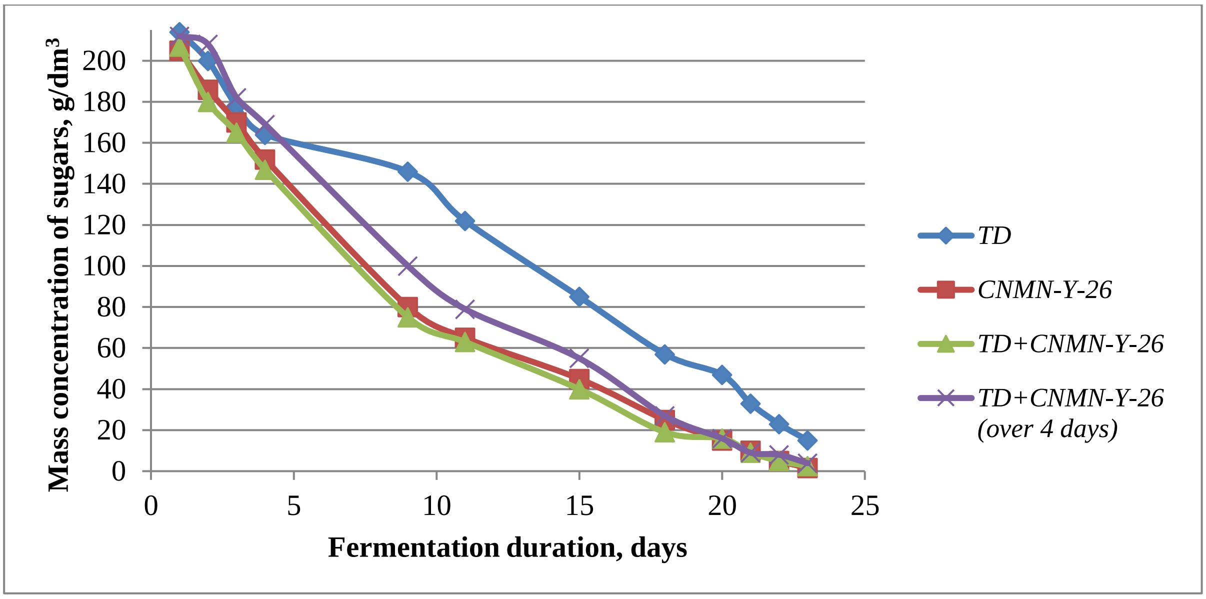

Fig. 6.1. The dynamics of sugar fermentation in grape must Aligotes using yeast strains *Torulaspora delbrueckii* and *Nr.2-Cricova-2 - S.cerevisiae CNMN-Y-26*

6.2. Study of the influence of non-*Saccharomyces* yeast strains on the physicochemical indices of dry white wines

After completing the alcoholic fermentation process, the dry white wines obtained using different fermentation schemes were subjected to physicochemical analyses, and the results obtained are presented in table 6.1.

According to the results in Table 6.1, it can be mentioned that in wines fermented with the use of yeast strains *No. 2-Cricova-2 - S.cerevisiae CNMN-Y-26* (control) the highest concentration of ethyl alcohol of 12.9% vol. was determined. In the Aligote white wine fermented with the use of non-*Saccharomyces Torulaspora Delbrueckii* yeast strains the lowest concentration of ethyl alcohol was determined, which constitutes 11.8% vol.

Dry white Aligote wine fermented using *Torulaspora delbrueckii* yeast strains in combination with selected yeasts is characterized by a high alcohol concentration: 12.5% vol. (*Torulaspora delbrueckii+ No.2-Cricova-2 - S.cerevisiae CNMN-Y-26*), and the sequential inoculation of indigenous yeasts contributes to obtaining a dry white wine with a lower alcoholic strength (12.1% vol).

The results obtained confirm that *Torulaspora delbrueckii* transforms part of the sugars in the must into other secondary compounds, which lead to an improvement in the quality of the wine, which was proven by organoleptic analysis.

The concentration of titrable acids in white wines, obtained under laboratory conditions, varies insignificantly depending on the fermentation scheme used and is 7.0 - 7.5 g/dm^3.

The mass concentration of volatile acids varies in Aligote white wines in a wider range from 0.33 g/dm^3 to 0.50 g/dm^3.

The variation of the pH index value in Aligote white wine samples is also within a narrow range and ranges from 3.13 to 3.18.

The residual sugar concentration values in white wines do not exceed the permissible limits for this category of wines, with the exception of dry white wine obtained using the non-*Saccharomyces* yeast strain *Torulaspora delbrueckii* where it constitutes 15.0 g/dm^3 (table 6.1).

Table 6.1. Physico-chemical and organoleptic indices of dry white Aligote wines, obtained according to different fermentation schemes (SPIHFT, 2016 h.y.)

№	Yeast strain, name	Alcohol concentration, % vol.	Mass concentration of:			pH	Organoleptic rating, points
			titrable acids, g/dm^3	volatile acids, g/dm^3	residual sugars, g/dm^3		
1	Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (control)	12,90±0,45	7,00±0,10	0,40±0,02	1,60±0,10	3,13±0,01	7,95
2	*Torulaspora Delbrueckii*	11,80±0,30	7,50±0,20	0,50±0,03	15,0±1,10	3,15±0,1	7,60
3	*Torulaspora delbrueckii+ Nr.2-Cricova-2 - S.cerevisiae CNMN-Y-26*	12,50±0,35	7,20±0,10	0,40±0,03	2,5±0,20	3,18±0,02	7,90
4	*Torulaspora delbrueckii+ Nr.2-Cricova-2 - S.cerevisiae CNMN-Y-26 (over 4 days)*	12,10±0,20	7,20±0,10	0,33±0,03	3,90±0,35	3,15±0,01	8,10

Sensory analysis of dry white Aligote wines demonstrated that the sequential fermentation method with inoculation of *Saccharomyces* yeasts on the 4th day of fermentation (average alcohol 3% vol.) contributes to improving the quality of the wine. Dry white Aligote wine is characterized by a complex aroma and balanced taste, which is confirmed by the high organoleptic score of 8.1 points (*Torulaspora delbrueckii+ No.2-Cricova-2 - S.cerevisiae CNMN-Y-26* (over 4 days).

In the 2016 wine campaign, research was conducted on the comparative assessment of the use of non-*Saccharomyces* and *Saccharomyces* yeasts in the alcoholic fermentation of must under microvinification conditions within the PI SPIHFT. Active dry yeasts (Oenologia LB8, Germany), the indigenous yeast strain *No. 2-Cricova-2 - S.cerevisiae CNMN-Y-26* and the *Torulaspora delbrueckii* strain, Enartis FERM, were used as controls. Previous research (under laboratory conditions) demonstrated the positive effect of successive alcoholic fermentation of must on the quality of dry white wines. Therefore, in order to establish the optimal moment for inoculation of *Saccharomyces* yeasts, the successive alcoholic fermentation scheme was used, and the inoculation of *Saccharomyces* yeasts was carried out when the alcoholic concentration in the must was reached at 3%

vol. and 6% vol. The initial concentration of sugars in the must was 220 g/dm^3, and the mass concentration of titrable acids was 8.2 g/dm^3.

After completion of alcoholic fermentation, dry white Chardonnay wines obtained using different must fermentation schemes were subjected to physicochemical and organoleptic analyses, and the results are presented in Table 6.2.

Table 6.2. Physico-chemical and organoleptic indices of dry white Chardonnay wines obtained using Saccharomyces and non-*Saccharomyces* yeasts under microvinification conditions (SPIHFT)

№	Yeast strain, name	Alcohol concentration, % vol.	Mass concentration of:			pH	Organoleptic rating, points
			titrable acids, g/dm^3	volatile acids, g/dm^3	residual sugars, g/dm^3		
1	ADY- (control)	13,00±0,50	7,70±0,10	0,36±0,04	1,60±0,04	3,12± 0,01	7,90
2	Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (control)	12,90±0,40	7,90±0,10	0,36±0,04	2,40±0,10	3,15± 0,02	7,90
3	*Torulaspora delbrueckii* (control)	12,60±0,30	8,00±0,20	0,30±0,03	7,60±0,40	3,13± 0,01	7,85
4	TD+ Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (3%vol.alcohol)	12,90±0,40	8,00±0,20	0,30±0,03	3,30±0,10	3,12± 0,01	7,95
5	TD+ Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (6%vol.alcohol)	12,90±0,40	8,00±0,10	0,30±0,02	3,40±0,20	3,15± 0,02	7,85
6	TD+ ADY- (3%vol.alcohol)	12,90±0,35	7,90±0,10	0,30±0,03	3,60±0,10	3,14± 0,02	7,95
7	TD+ ADY- (6%vol.alcohol)	12,80±0,45	8,00±0,20	0,30±0,02	3,90±0,20	3,13± 0,01	7,85

Legend: TD- Torulaspora delbrueckii;
ADY- Active dry yeast.

According to the results in table 6.2, it can be mentioned that dry white wines fermented with the use of *Torulaspora delbrueckii* yeast strains are characterized by a lower alcohol concentration: 12.6% vol., and the successive inoculation of yeasts *No. 2-Cricova-2 - S.cerevisiae CNMN-Y-26* contributes to obtaining wines with a higher alcohol content - 12.9% vol., which confirms the fact that *Torulaspora delbrueckii* is a culture poorly resistant to high alcohol concentrations and demonstrates a reduced ability to ferment carbohydrates.

The concentration of titrable acids in dry white Chardonnay wines obtained under microvinification conditions varies insignificantly depending on the fermentation scheme used and ranges from 7.7 g/dm3 to 8.0 g/dm^3.

The mass concentration of volatile acids varies in all Chardonnay white wines in the range of 0.30-0.36 g/dm^3. It is known that the use of *Saccharomyces* and *Torulaspora* *delbrueckii* yeasts in successive fermentation contributes to the reduction of acetic acid, which also had a positive effect on the mass concentration of volatile acids.

The variation of the pH index value in the dry white wine samples obtained is also within a narrow range and is 3.13-3.15.

The residual sugar concentration values in dry white Chardonnay wines do not exceed the permissible limits for this category of wines, with the exception of wine obtained using the non-*Saccharomyces* yeast strain *Torulaspora delbrueckii* and are 7.6 g/dm^3.

Sensory analysis of dry white Chardonnay wines demonstrated that successive fermentation with inoculation of *Saccharomyces* yeasts when reaching an average alcoholic concentration of 3% vol. contributes to improving the quality and gives the wine a complex aroma and balanced taste, which is confirmed by high organoleptic scores of 7.95 points (tab.6.2.). Successive alcoholic fermentation with inoculation of *Saccharomyces* yeasts when reaching an average alcoholic concentration of 6% vol. does not significantly influence the organoleptic properties of the wines compared to the control samples.

6.3. Study of the influence of non-*Saccharomyces* yeast strains on the physicochemical indices of dry white Chardonnay wines obtained under production conditions

To confirm the results obtained under microvinification conditions at SPIHFT, experiments were conducted with the use of non-*Saccharomyces* yeasts in successive alcoholic fermentation under the production conditions of the SA "Cricova" wine complex. The clarified must of the Chardonnay variety with an initial concentration of sugars of 215 g/dm^3 and titrable acids of 6.2 g/dm^3 was used as the research object. Active dry yeast Aroma White (Italy) was used as a control. The results of the physicochemical analysis and organoleptic assessment of dry white Chardonnay wines are presented in table 6.3.

According to the results in table 6.3, it can be mentioned that the dry white wine Chardonnay obtained using the *Torulaspora delbrueckii* yeast strain is characterized by a lower alcohol concentration: 12.1% vol., and the successive inoculation of the *Nr.2-Cricova-2 - S.cerevisiae CNMN-Y-26* yeasts contributes to obtaining wines with a higher alcoholic strength - 12.8% vol., which confirms the results obtained previously.

The concentration of titrable acids in dry white wines obtained under the conditions of SA "Cricova", varies insignificantly within the limits of 5.7 g/dm^3 to 6.0 g/dm^3.

The mass concentration of volatile acids varies in all white wines obtained in the range of 0.33-0.36 g/dm^3.

The variation of the pH index value in samples of dry white Chardonnay wines is within a fairly narrow range and is 3.27-3.32.

The residual sugars in dry white wines do not exceed the permissible limits for this category of wines, with the exception of wine obtained using the non-*Saccharomyces* yeast strain *Torulaspora delbrueckii* (9.0 g/dm^3).

Sensory analysis of dry white Chardonnay wines confirmed that the successive fermentation of the must with inoculation of *Saccharomyces* yeasts upon reaching an average alcoholic concentration of 3% vol. contributes to improving the quality of the wine obtained, which is confirmed by the high organoleptic score of 7.95 points (tab.6.3).

Table 6.3. Physico-chemical and organoleptic indices of dry white Chardonnay wines obtained using *Saccharomyces* and non-*Saccharomyces* yeasts under production conditions at SA "Cricova", h.y.2016

№	Yeast strain, name	Alcohol concentration, % vol.	Mass concentration of: titrable acids, g/dm^3	volatile acids, g/dm^3	residual sugars, g/dm^3	pH	Organoleptic rating, points
1	*Torulaspora delbrueckii* (control)	12,10±0,25	5,7±0,10	0,38±0,06	9,00±0,45	3,28± 0,01	7,80
2	ADY "Aroma White"(control)	12,80±0,25	5,8±0,15	0,36±0,03	4,0±0,50	3,27± 0,02	7,90
3	TD+ Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (3%vol.alcohol)	12,80±0,25	6,0±0,20	0,33±0,03	3,0±0,35	3,30± 0,02	7,95
4	TD+ Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (6%vol.alcohol)	12,70±0,35	6,0±0,10	0,36±0,03	3,3±0,10	3,32± 0,03	7,90

Legend: TD- Torulaspora delbrueckii; ADY- Active dry yeast.

The successive alcoholic fermentation of the must with inoculation of *Saccharomyces* yeasts upon reaching an alcoholic concentration of 6% vol. does not significantly influence the organoleptic properties, compared to the control samples, but the organoleptic score is lower compared to the wine fermented according to the scheme TD+ No.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (3%vol.alcohol).

6.4. Determination of the content of volatile substances in dry white Chardonnay wines obtained under production conditions

Recent studies have shown that the use of non-*Saccharomyces* yeasts can increase the complexity and intensity of aromas in wine. Various species of non-*Saccharomyces* yeasts, including *Torulaspora delbrueckii,* can produce unique aroma compounds during wine fermentation. The interaction between non-*Saccharomyces* yeasts and *Saccharomyces* yeasts can have a positive effect on the volatile complex of the wine, contributing to the development of a more complex and balanced aroma profile.

In this context, a comparative analysis of the content of volatile substances in dry white Chardonnay wines, which were obtained using different fermentation schemes, was carried out. The results obtained are presented in table 6.4.

From the results presented in table 6.4 it can be concluded that different fermentation schemes have an impact on the final content of volatile compounds in dry white Chardonnay wines.

The concentration of acetaldehyde was low in all wines, the one fermented with *No.2-Cricova-2 - S.cerevisiae CNMN-Y-26* having a lower concentration than the other white wines.

The ethyl acetate concentrations for the three wines fermented with the control yeasts did not have significant differences, but In the wine obtained with the yeast combination, a lower concentration (almost half) of ethyl acetate was recorded.

Table 6.4. Content of volatile substances in Chardonnay white wines, mg/L

Name of the substance	Yeast strain						
	ADY (control)	Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (control)	*Torulaspora delbrueckii* (control)	TD+ Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (3%vol.alcohol)	TD+ Nr.2-Cricova-2 - *S.cerevisiae* CNMN-Y-26 (6%vol.alcohol)	TD+ADY (3% vol.alcohol)	TD+ ADY (6% vol.alcohol)
Acetaldehyde	10,40±0,69	6,80±0,38	10,50±0,73	7,70±0,45	8,30±0,51	8,50±0,56	8,90±0,61
ethylacetate	35,90±3,45	37,50±3,75	35,50±3,56	18,90±1,25	19,10±1,56	17,80±1,16	18,10±1,63
Propanol-1	30,50±4,11	29,40±3,62	45,10±4,25	66,40±4,89	59,40±4,23	71,60±4,89	63,90±4,29
Isobutanol	23,90±3,23	20,50±3,13	21,60±3,42	21,50±3,44	20,70±3,23	22,70±3,77	23,50±3,78
Σ Amyl alcohols	95,60±5,36	100,10±5,47	63,50±4,23	75,80±4,56	85,60±5,42	77,90±5,42	82,50±5,78
Phenyl-2-ethanol	49,50±4,31	54,60±4,54	70,20±4,41	62,30±3,21	59,20±4,09	60,10±3,89	55,80±3,44

The sum of amyl alcohols was lower in the wine fermented with the use of the control yeast *Torulaspora delbrueckii*.The concentration of propanol-1 was higher in wines fermented with the TD + No.2-Cricova-2 - S.cerevisiae CNMN-Y-26 (3% vol. alcohol) yeast combination and TD+ADY (3% vol. alcohol), while the isobutanol concentration was the same for all white wines obtained using the control strains and their combinations. However, the concentration of phenyl-2-ethanol with its distinctive rose aroma was significantly higher for the wine fermented with the control strain *Torulaspora delbrueckii* and its combination with the control yeasts, than for the wines fermented using ADY (control) and *No.2-Cricova-2 - S.cerevisiae CNMN-Y-26* (control).

CONCLUSIONS AND RECOMMENDATIONS

The problems addressed in the thesis are devoted to the isolation, identification and selection of yeast strains from different wine-growing centers of the Republic of Moldova, the study of their influence on the quality of dry white and red wines, by elucidating the physicochemical and biochemical transformations that take place in the alcoholic fermentation process. The main results of the research are formulated in the following conclusions:

1. 34 yeast strains were isolated from the ′Chișinău′ wine center, 64 yeast strains (34 from white varieties, 30 from red varieties) from the ′Purcari′ wine center and 61 yeast strains (33 from white varieties, 28 from red varieties) from the ′Trifești′ wine center, chapter 3 (Soldatenco, O., 2019).

2. The morphological and cultural characteristics of the isolated yeast strains were determined. It was demonstrated that the cells of the studied yeasts are well emphasized and differ in shape and size, and all yeast strains are in a state of budding. All yeast strains isolated from the wine centers ′Chișinău′, ′Purcari′ and ′Trifești′ are of the eukaryotic type, in which the cellular components are well distinguished. Of the 34 cultures isolated from the wine center ′Chișinău′, 64 cultures isolated from the wine center ′Purcari′, 61 cultures isolated from the wine center ′Trifești′, 16, 31 and, respectively, 47 yeast strains belong to the genus Saccharomyces. The comparison of the nucleotide sequences obtained with those placed in the NCBI gene bank (ncbi.nlm.nih.gov) and the identification of the yeasts by the FT-IR method was performed. It was demonstrated that all the yeast strains investigated are from three species: *Saccaromyces cerevisiae* (~75%), *Saccharomyces pastorianus* (~20%), *Saccharomyces bayanus* (~5%), chapter 3 (Soldatenco, O., 2019).

3. The biochemical and technological indices of the isolated yeast strains were evaluated. The following yeast strains were selected: from the wine center ′Chișinău′ - No. 2, No. 8, No. 9, No. 10, No. 14, No. 16 – for the production of dry white wines; from the wine center ′Purcari′ - No. 1, No. 3, No. 7, No. 12 – for the production of dry white wines and No. 21, No. 24, No. 29, No. 30 – for the production of dry red wines; from the wine center ′Trifești′ - No. 1, No. 10, No. 15, No. 19, No. 22 – for the production of dry white wines and No. 27, No. 32, No. 35, No. 41, No. 43 – for the production of dry red wines, chapter 3 (Soldatenco, O., 2021).

4. It has been demonstrated that the use of yeast strains selected from the winemaking centers ′Chișinău′ No. 2 (Cricova-2/CNMN-Y-26), ′Purcari′ No. 1 (FNFTP-1/CNMN-Y-33), No. 12 (Ch75P-3ÎF/CNMN-Y-32), No. 30 (RN-120-P-5/CNMN-Y-31) and ′Trifești′ No. 15 (S75Tr-4.4/CNMN-Y-35), No. 22 (Atr-2.3/CNMN-Y-34), No. 32 (M100Tr-1/CNMN-Y-36), No. 41 (C-S60Tr-2/CNMN-Y-37) allows the production of quality dry white and red wines, both according to physicochemical indices and organoleptic notes, and does not compromise the quality of the wines obtained. with the use of imported Active dry yeasts. It has been demonstrated that the dynamics of must and must fermentation when using selected yeast strains does not differ from the dynamics of must and must fermentation when using ADY, consuming practically all the sugars in the must within 14-15 days from inoculation, chapter 4 (Soldatenco, O., 2020; Taran, N.; Soldatenco, O., 2021).

5. It was demonstrated that the physicochemical composition and quality of dry white and red wines depend largely on the yeast strain used in the must fermentation process. It was found that higher alcohols, esters, aldehydes, volatile acids and other substances formed during the fermentation of the obtained wines contribute to the formation of a very complex aroma, and the mentioned yeast strains have a positive influence on the physicochemical indices and organoleptic note of the finished product. It was demonstrated that all experimental wines have a glycerol concentration higher than 6 g/dm^3, and the highest values are in the samples when using yeast strains No. 2 (′Chisinau′ wine center), No. 1, No. 12 and No. 30 (′Purcari′ wine center), No. 15, No. 22, No. 32 and No. 41 (′Trifești wine center). Thus, it can be concluded that the influence of yeast strains on the formation of glycerol in dry white and red wines is significant. It has been established that the studied yeast strains greatly influence the concentration of some organic acids, especially the content of succinic acid, chapter 4 (Taran, N.; Soldatenco, O., 2020; Soldatenco, O. and others, 2022).

6. The indigenous yeast strains No. 2-Cricova-2, No. 1-FNFTP-1, No. 12-Ch75P-3ÎF, No. 30-RN-120-P-5, No. 15-S75Tr-4.4, No. 22-Atr-2.3, No. 32-M100Tr-1, No. 41-C-S60Tr-2, were deposited in the The National Collection of Non-Pathogenic Microorganisms of the Institute of Microbiology and Biotechnology. Deposit certificates and a passport were obtained for each yeast strain with the number assigned by the National Collection of Nonpathogenic Microorganisms. The yeast strains were registered in the NCBI (National Center for Biotechnology Information, USA, Maryland) global database.

7. Based on the scientific results obtained, the yeast strain No. 2-Cricova-2 - S.cerevisiae CNMN-Y-26 for the production of dry white wines was recommended for implementation in production conditions at JSV "Cricova", at JV "Vinăria Purcari" LTD the yeast strains No. 1-FNFTP-1- S.cerevisiae CNMN-Y-32 and No. 12-Ch75P-3ÎF - S.cerevisiae CNMN-Y-33, for the production of dry white wines and the yeast strain No. 30-RN-120-P-5 - S.cerevisiae CNMN-Y-31 for the production of dry red wines, at "Vierul-Vin" LTD the yeast strains No. 15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34 and No. 22-Atr-2.3 - S.cerevisiae CNMN-Y-35, for the production of dry white wines and No.32-M100Tr-1- S.cerevisiae CNMN-Y-36 and No.41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37, for the production of dry red wines in order to obtain experimental batches. The indigenous yeast strains contributed to the production of dry white and red wines with a clean, fine aroma, full, full-bodied, harmonious taste. Based on the studies conducted during the aa. 2009-2019, the indigenous yeast strains with advanced technological properties, isolated and selected from the winemaking centers ′Chișinău′, ′Purcari′ and ′Trifești′ were tested under production conditions at SA ”Crocova”, ÎM "Vinăria Purcari" SRL and "Vierul-Vin" SRL, where experimental batches of dry white wines in a total volume of 100,000 L and dry red wines in a total volume of 60,000 L were obtained.

8. Based on the results obtained, the technological regimes for the production of dry white and red wines using selected local yeasts were improved. The improved technologies were optimized based on the following criteria: low fermentation temperature under industrial conditions (minimum 13 °C for white wines); lower sulfite content of must and grape pomace (for white wines: 50-75 mg/L total SO_2, when the grapes are healthy and 120-150 mg/L total SO_2 if they have been attacked by gray rot; for red wines: 40-60 mg/L total SO_2 if healthy grapes are used and 120-150 mg/L total SO_2 if they have been attacked by gray rot), chapter 5.

9. 8 patents were obtained for yeast strains isolated and selected from the winemaking centers of ′Chișinău′, ′Purcari′, ′Trifești′.

10. The study of the influence of non-*Saccharomyces* yeast *Torulaspora delbrueckii* has demonstrated that the successive fermentation of the must with the inoculation of Saccharomyces yeasts upon reaching an average alcoholic concentration of 3% vol. contributes to the improvement of the quality and gives the wine a complex aroma and balanced taste. Based on the results obtained, technological recommendations were developed regarding the use of *Saccharomyces* and non-*Saccharomyces Torulaspora delbrueckii* yeasts for the production of dry white wines, chapter 6 (Soldatenco., O. 2024)

PROPOSALS FOR THE USE OF THE RESULTS OBTAINED IN ECONOMIC FIELDS

Within the thesis, the yeast strains *No. 2-Cricova-2 - S.cerevisiae CNMN-Y-26* were isolated, identified and selected for the production of dry white wines in the ′Chișinău′ wine center, the yeast strains *No. 1-FNFTP-1- S.cerevisiae CNMN-Y-32, No. 12-Ch75P-3ÎF - S.cerevisiae CNMN-Y-33* for the production of dry white wines and the yeast strain *No. 30-RN-120-P-5 - S.cerevisiae CNMN-Y-31* for the production of dry red wines in the ′Purcari′ wine center, the yeast strains *No. 15-S75Tr-4.4 - S.cerevisiae CNMN-Y-34, No. 22-Atr-2.3 - S.cerevisiae CNMN-Y-35* for the production of dry white wines and yeast strains *No. 32-M100Tr-1- S.cerevisiae CNMN-Y-36, No. 41-C-S60Tr-2 - S.cerevisiae CNMN-Y-37* for the production of dry red wines, which are recommended for their implementation in the oenological industry, according to the patents: **MD 4210 C1 2013.10.31, MD 4678 C1 2020.11.30, MD 4679 C1 2020.11.30, MD 4680 C1 2020.11.30, MD 4727**

C1 2021.06.30, MD 4728 C1 2021.06.30, MD 4729 C1 2021.06.30, MD 4730 C1 2021.06.30.

The technological regimes for the production of dry white and red wines using selected local yeasts have been improved. The improved technologies have been optimized based on the following criteria: low fermentation temperature under industrial conditions (minimum 13 °C for white wines); lower sulfite content of must and grape pomace (for white wines: 50-75 mg/L total SO_2 when grapes are healthy and 120-150 mg/L total SO_2 if they have been attacked by gray rot; for red wines: 40-60 mg/L total SO_2 when using healthy grapes and 120-150 mg/L total SO_2 if they have been attacked by gray rot.

Technological recommendations have been developed on the use of selected indigenous and non-*Saccharomyces* yeasts in the production of dry white wines:

- It is recommended to use non-*Saccharomyces* yeast *Torulaspora delbrueckii* in the successive fermentation of the must with the inoculation of Saccharomyces yeasts upon reaching an average alcoholic concentration of 3% vol. for the production of dry white wines.

SUGGESTIONS ON POTENTIAL FUTURE RESEARCH DIRECTIONS RELATED TO THE TOPIC ADDRESSED

1. Identification and genetic characterization of indigenous yeasts: Genetic evaluation of indigenous yeasts can help identify the genes responsible for specific aromatic compounds, as well as develop new technologies to manipulate these genes and control the production of the aromatic complex.
2. Assessing native yeast diversity: Studying native yeast diversity can help identify different types of yeast and assess their role in producing unique aromas and flavors. This can also help identify regions with unique native yeasts and harness them to produce distinctive wines.
3. Development of techniques for selecting and improving indigenous yeasts: Development of techniques for selecting and improving indigenous yeasts can help create indigenous yeast strains that produce higher amounts of aromatic compounds or that are more resistant to difficult winemaking conditions, such as high temperatures or low pH conditions.
4. Assessing the impact of terroir on indigenous yeasts: Studying the impact of terroir on indigenous yeasts can help understand the influence of soil, climate, and other environmental factors on the microbial composition of wine. This research can help identify factors that contribute to the unique taste and aroma of wine from a particular region.
5. Evaluating the effects of native yeast combinations: Evaluating the effects of native yeast combinations can help identify synergies and interactions between different native yeast strains and develop technologies to control these interactions in wine production.

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LIST OF SCIENTIFIC WORKS ON THE THESIS TOPIC

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1. **SOLDATENCO, O.** *Bazele științifice și practice ale utilizării levurilor în oenologie.* Chișinău: Tipografia ”PrintCaro”, 2021. 184 pagini. ISBN 978-9975-56-862-3; https://isphta.md/ro/biblioteca/

1.2. collective monographs

1. TARAN, N., **SOLDATENCO, O.** *Evidenţierea şi selectarea suşelor de levuri pentru producerea vinurilor albe seci în plaiul vitivinicol "Cricova".* În: Biotehnologii în vinificație. Red. N. Taran. Chișinău: Tipografia "PrintCaro", 2021. p. 17-74. ISBN 978-9975-56-890-6. https://isphta.md/ro/biblioteca/

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7. TARAN, N., **SOLDATENCO, O.** Aprecierea comparativă de utilizare la fermentarea alcoolică a levurilor Saccharomyces și non-Saccharomyces în condiții de microvinificație. In: *Conferința*

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4. TARAN, N., SOLDATENCO, E., **SOLDATENCO, O.,** VASIUCOVICI, S., BOSTAN, V., CHIOSA, N., CICHIR, L. *Tulpină de levuri Saccharomyces cerevisiae pentru producerea*

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ADNOTARE

Soldatenco Olga: Bazele științifice ale izolării și selectării tulpinilor de levuri autohtone pentru producerea vinurilor albe și roșii în condițiile R. Moldova, teză de doctor habilitat în științe inginerești, Chișinău, 2025.

Structura tezei: constă din introducere, 6 capitole, concluzii și recomandări, bibliografia cu 310 titluri, 7 anexe. Textul de bază conține 204 pagini, inclusiv 65 de figuri și 71 tabele.

Cuvinte cheie: tulpini de levuri, *Saccharomyces,* izolarea, selectarea, autohtone, vinuri albe și roșii seci, fermentație alcoolică, caractere morfologice, fiziologice, biochimice, PCR.

Scopul lucrării: Izolarea, identificarea și selectarea tulpinilor de levuri autohtone din diferite centre vitivinicole pentru producerea vinurilor albe și roșii seci în condițiile R. Moldova.

Obiectivele lucrării: Izolarea levurilor autohtone din diferite centre vitivinicole; identificarea tulpinilor de levuri izolate din centrele vitivinicole ′Chișinău′, ′Purcari′ și ′Trifești′ prin metode tradiționale și moderne (PCR, spectroscopie FT-IR); determinarea indicilor biochimici și tehnologici ai tulpinilor de levuri izolate; studiul influenței tulpinilor de levuri selectate asupra procesului de fermentație a mustului și mustuielii; studiul influenței tulpinilor de levuri selectate asupra indicilor fizico-chimici ai vinurilor albe și roșii seci; testarea și implementarea tulpinilor de levuri selectate și selectate din centrele vitivinicole ′Chișinău′, ′Purcari′ și ′Trifești′ în condiții de producere; perfecționarea regimurilor tehnologice de producere a vinurilor albe și roșii seci cu utilizarea levurilor autohtone; studiul influenței levurilor non-*Saccharomyces* asupra calității vinurilor albe seci.

Noutatea și originalitatea științifică: Pentru prima dată au fost izolate și selectate tulpini de levuri autohtone din diferite centre vitivinicole, și argumentată științific perspectiva utilizării acestor tulpini pentru producerea vinurilor albe și roșii seci.

Rezultatele principale: Au fost izolate și selectate tulpini de levuri autohtoni din diferite centre vitivinicole pentru producerea vinurilor albe și roșii seci; a fost studiată influența levurilor autohtone selectate asupra calității vinurilor albe și roșii seci; a fost stabilit efectul pozitiv a utilizării levurilor selectate asupra notei organoleptice a vinurilor albe și roșii seci; s-a demonstrat că fermentația succesivă a mustului cu inocularea levurilor *Saccharomyces* la atingerea concentrației alcoolice în mediu de 3 % vol. contribuie la ameliorarea calității vinului.

Semnificația teoretică: S-au acumulat date și cunoștințe noi despre caracterele morfologice, fiziologice și biochimice ale tulpinilor de levuri autohtone și influența lor asupra parametrilor de calitate ai vinurilor albe și roșii seci.

Valoarea aplicativă a lucrării: În baza rezultatelor obținute au fost perfecționate regimurile tehnologice de producere a vinurilor albe și roșii seci cu utilizarea levurilor autohtone și elaborate rcomandări tehnologice privind utilizarea levurilor autohtone selectate și non-*Saccharomyces* la producerea vinurilor albe seci. Au fost obținute 8 brevete de invenții.

Implementarea rezultatelor științifice: Rezultatele cercetărilor au fost implementate la S.A.„Cricova” și au fost obținute vinuri albe seci în volum de 40000 L. În baza studiilor efectuate tulpinile de levuri autohtone selectate din centrul vitivinicol ′Purcari′ au fost testate în condițiile de producere la ÎM „Vinăria Purcari”, unde au fost obținute loturil experimentale de vinuri albe (20000 L) și roșii (20000 L) seci. Tulpinile de levuri izolate din centrul vitivinicol ′Trifești′ au fost implementate la SRL ”Vierul-Vin”, unde au fost obținute loturil experimentale de vinuri albe (40000 L) și roșii (40000 L) seci.

ANNOTATION

Soldatenco Olga: Scientific foundations of isolation and selection of indigenous yeast strains for the production of white and red wines in the conditions of the Republic of Moldova, Doctoral Thesis in Engineering Sciences, Chisinau, 2025.

Thesis Structure: It consists of an introduction, 6 chapters, conclusions and recommendations, bibliography with 310 titles, 7 appendices. The main text comprises 204 pages, including 65 figures and 71 tables.

Keywords: yeast strains, *Saccharomyces,* isolation, selection, indigenous, dry white and red wines, alcoholic fermentation, morphological, physiological, biochemical characteristics, PCR.

Thesis Aim: Isolation, identification, and selection of indigenous yeast strains from different viticultural centers for the production of dry white and red wines in the Republic of Moldova.

Thesis Objectives: Isolation of indigenous yeasts from various viticultural centers. Identification of isolated yeast strains from the viticultural centers 'Chișinău,' 'Purcari,' and 'Trifești' using traditional and modern methods (PCR, FT-IR spectroscopy). Determination of the biochemical and technological indices of the isolated yeast strains. Study of the influence of selected yeast strains on the fermentation process of must and grape juice. Study of the influence of selected yeast strains on the physico-chemical indices of dry white and red wines. Testing and implementing the selected yeast strains from the viticultural centers 'Chișinău,' 'Purcari,' and 'Trifești' in production conditions. Refinement of technological regimes for the production of dry white and red wines using indigenous yeasts. Study of the influence of non-Saccharomyces yeasts on the quality of dry white wines.

Scientific Novelty and Originality: For the first time, indigenous yeast strains were isolated and selected from different viticultural centers, and the scientific perspective of using these strains for the production of dry white and red wines was scientifically substantiated.

Main Results: Indigenous yeast strains were isolated and selected from various viticultural centers for the production of dry white and red wines. The influence of selected indigenous yeasts on the quality of dry white and red wines was studied. The positive effect of using selected yeasts on the organoleptic characteristics of dry white and red wines was established. It was demonstrated that successive fermentation of must with the inoculation of Saccharomyces yeasts when reaching an alcohol concentration of 3% vol. contributes to the improvement of wine quality.

Theoretical Significance: New data and knowledge have been accumulated about the morphological, physiological, and biochemical characteristics of indigenous yeast strains and their influence on the quality parameters of dry white and red wines.

Applicative Value of the Work: Based on the obtained results, technological regimes for the production of dry white and red wines using indigenous yeasts were perfected, and technological recommendations for the use of selected indigenous and non-*Saccharomyces* yeasts in the production of dry white wines were developed. Eight invention patents were obtained.

Implementation of Scientific Results: The research results were implemented at S.A. "Cricova" where dry white wines were obtained in a volume of 40000 L. Based on the conducted studies, the selected indigenous yeast strains from the 'Purcari' viticultural center were tested in production conditions at the "Vinaria Purcari", where experimental batches of dry white (20000 L) and red (20000 L) wines were obtained. Yeast strains isolated from the 'Trifești' viticultural center were implemented at SRL "Vierul-Vin," where experimental batches of dry white (40000 L) and red (40000 L) wines were obtained.

АННОТАЦИЯ

Солдатенко Ольга: Научные основы выделения и отбора местных штаммов дрожжей для производства белых и красных вин в условиях Республики Молдова, докторская диссертация в области инженерных наук, Кишинев, 2025 г.

Структура диссертации: Включает введение, 6 глав, заключение и рекомендации, библиографию с 310 названиями, 7 приложений. Основной текст включает в себя 204 страниц, включая 65 рисунков и 71 таблицу.

Ключевые слова: штаммы дрожжей, *Saccharomyces*, выделение, отбор, местные, сухие белые и красные вина, спиртовое брожение, морфологические, физиологические, биохимические характеристики, ПЦР.

Цель диссертации: Выделенте, идентификация и отбор местных штаммов дрожжей из различных винодельческих центров для производства белых и красных сухих вин в Республике Молдова.

Задачи диссертации: Выделение местных дрожжей из различных винодельческих центров. Идентификация выделенных штаммов дрожжей из винодельческих центров 'Кишинев', 'Пуркарь' и 'Трифешты' с использованием традиционных и современных методов (ПЦР, Фурье-спектроскопия). Определение биохимических и технологических показателей выделенных штаммов дрожжей. Изучение влияния отобранных штаммов дрожжей на процесс брожения виноградного сусла. Изучение влияния отобранных штаммов дрожжей на физико-химические показатели сухих белых и красных вин. Тестирование и внедрение отобранных штаммов дрожжей из винодельческих центров 'Кишинев', 'Пуркарь' и 'Трифешты' в условиях производства. Усовершенствование технологических режимов производства сухих белых и красных вин с использованием местных дрожжей. Изучение влияния дрожжей non-*Saccharomyces* на качество белых сухих вин.

Научная новизна и оригинальность: Впервые были выделены и отобраны местные штаммы дрожжей из различных винодельческих центров, и научно обоснованы перспективы использования этих штаммов для производства сухих белых и красных вин.

Основные результаты: Из винодельческих центров различных регионов были выделены и отобраны местные штаммы дрожжей для производства сухих белых и красных вин.

Изучено влияние отобранных местных штаммов дрожжей на качество белых и красных сухих вин. Установлен положительный эффект использования отобранных штаммов на органолептические характеристики сухих белых и красных вин. Показано, что последовательное брожение сусла с внесением инокуляции дрожжей *Saccharomyces* при достижении концентрации алкоголя в среде 3% объемных долей способствует улучшению качества вина.

Теоретическое значение: Получены новые данные и знания о морфологических, физиологических и биохимических характеристиках местных штаммов дрожжей и их влиянии на параметры качества белых и красных сухих вин.

Практическая ценность работы: На основе полученных результатов были усовершенствованы технологические режимы производства белых и красных сухих вин с использованием местных дрожжей, разработаны технологические рекомендации по использованию отобранных местных и non-*Saccharomyces* дрожжей при производстве белых сухих вин. Получены 8 патентов на изобретение.

Реализация научных результатов: Результаты исследований были внедрены в S.A. "Cricova", где были получены сухие белые вина объемом 40000 литров. На основе проведенных исследований отобранные местные штаммы дрожжей из винодельческого центра 'Purcari' были протестированы в условиях производства в ÎM „Vinăria Purcari”, где были получены экспериментальные партии сухих белых (20000 литров) и красных (20000 литров) вин. Штаммы дрожжей, выделенные из винодельческого центра 'Trifeşti', были внедрены в SRL ”Vierul-Vin”, где были получены экспериментальные партии сухих белых (40000 литров) и красных (40000 литров) вин.

SOLDATENCO OLGA

SCIENTIFIC FOUNDATIONS OF ISOLATION AND SELECTION OF INDIGENOUS YEAST STRAINS FOR THE PRODUCTI-ON OF WHITE AND RED WINES IN THE CONDITIONS OF THE REPUBLIC OF MOLDOVA

253.03 TECHNOLOGY OF ALCOHOLIC AND NON-ALCOHOLIC BEVERAGES

Summary of the doctor habilitat thesis in engineering sciences

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