Final Scientific Report

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Project: Innovative approach to develop value-added snack products through extrusion technology

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1. Objectives preconized/achieved

The project focuses on obtaining new snack products by valorization in a high amount of grape pomace for superior health properties with maize flour addition in extrusion technology. Grape pomace will be the component of the new direct expanded product formula which will provide improved nutritional quality, with a low glycemic index.

The **concrete objectives** of the projects are as follows:

O1. Physico-chemical characteristics analysis, molecular characterization, and functional properties determination of grape pomace and maize flour used as raw materials

O2. Feed mixture preparation, physico-chemical characteristics analysis, molecular characterization, and functional properties determination

O3. Testing, establishing the extrusion conditions (processing parameters and system parameters) of the formulated mixtures, and performing extrusion

O4. Evaluation of the new snacks' quality characteristics

O5. Complex evaluation of the new snack product obtained after the optimization process.

The concrete objectives were reached and fulfilled to a degree of 100%. To fulfill each objective, all the activities associated with objectives were carried out according to the implementation plan, without differences between those originally expected and those achieved, obtaining the following results.

2. Presentation of the results obtained, of the achieved result indicators

O1/A1.1. Physico-chemical composition, molecular characterization, and functional properties of grape pomace

The following estimative results were obtained: moisture, ash, acidity, color, protein content, fat, dietary fiber, total sugar, total phenolic content, antioxidant activity, molecular characteristics, and functional properties (water retention capacity and swelling capacity).

The investigations began with acquiring grape pomace and maize flour, representing the raw materials used in the experimental research. Maize flour was supplied from Ruagnessy S.R.L., Satu Mare, and grape pomace from Iasi Research and Development Center for Viticulture and Vinification. To preserve the grape pomace, two gently drying methods were used, drying in a convection oven at 50 ºC and lyophilization which may help us to maintain the biochemical components compactly. The values obtained for *moisture* content depended on the initial moisture of grape pomace and the type of the drying method applied. No regular trend was observed when comparing *ash* among white and red grape pomace samples with the same components and of the same type, concerning the drying method. Grape pomace *acidity* showed variations from a kind of sample to another as a function of components, drying method, and grape variety, but are within the recommended limits. The *colour parameters*, luminosity (*L**), red or green nuance (+*a**/- *a**), and yellow or blue nuance (+*b**/- *b**) of analyzed samples were directly connected to the chemical compounds of grape pomace such as pigments. All lyophilized samples presented a lighter color compared to the oven-dried samples for both white and red grape pomace samples. The content of *lipids* was lower in seedless grape pomace, for both white and red varieties, dried in an oven or lyophilizated, whereas the carbohydrates decreased. A high content of lipids was found in both varieties of grape pomace with seeds. Lipids value is mainly associated with the seeds that are richer in oil, more than peels. There were no significant differences between grape pomace samples dried in an oven or through lyophilization from the point of view of lipids, proteins, and ash. Ash, *protein*, and lipids were in higher amounts in red grape pomace compared to white grape pomace. These findings are in agreement with those observed by other authors (Mironeasa, 2017; Spanghero et al., 2009). The results obtained for *amino acid* content showed that the values depend on the grape pomace type, seedless or with seeds. A high content of essential *amino acids* was found in whole grape pomace compared to seedless grape pomace, which can be due to the essential amino acids present in seeds. There were no significant differences between grape pomace samples dried in an oven or through lyophilization. The results obtained for *total dietary fiber* content showed that the values depended on variety and grape pomace sample type. Seedless grape pomace contained a lower total dietary fiber compared to grape pomace with seeds. Also, some differences weres identified between samples dried in an oven and those lyophilized from the total fiber content point of view. Regarding *total sugar*, grape pomace from the red variety presented a higher content compared to grape pomace from the white variety, probably due to the different winemaking procedures applied. White grape pomace was procured right after pressing juice, whereas red grape pomace was obtained after fermentation for several days to extract colour and polyphenols. The high content of total sugar was obtained in grape pomace lyophilized samples compared to the samples dried in a conventional oven, a fact that revealed the strong influence of the drying methods applied.

The results obtained for the physical-chemical characteristics of grape pomace showed that the difference in winemaking practices resulted in different chemical compositions of grape pomace. The data obtained are included within the usual range reported for white and red grape pomaces from wine production (Saura-Calixto, 1998; Llobera and Canellas, 2007). Also, the characteristics varied depending on the grape variety, the drying method, and the type of grape pomace sample (seedless or with seeds). The values obtained were according to those presented in specialty literature (Iuga and Mironeasa, 2020; Beres et al., 2019; Maurer et al., 2019).

The *molecular characteristics* evaluated by using the FTIR spectroscopy technique allow obtaining spectra that present some characteristic bands of individual components, providing information about the chemical composition, including both primary and secondary metabolites. The functional groups of the bioactive compounds, different phenolic compounds, fatty acids, polysaccharides, lignins, pectins, and organic compounds, such as sugars, alcohols, and organic acids present in grape pomace were identified. These results show that FTIR spectral features are linked to grape variety, methods used to dry, and also on the grape pomace sample type.

The *total phenolics* from grape pomace were influenced by the pomace drying methods, depending also on the grape variety, and by the grape pomace sample type. The higher values for the total phenolic content were found in seedless grape pomace samples. The amount of total phenolic compounds determined in the maize flour is much lower than that obtained in grape pomace. The *antioxidant activity* of grape pomace samples, evaluated through their action in DPPH radical inhibition, showed different trends depending on the sample type. The high antioxidant activity was observed in grape pomace with seeds compared to seedless grape pomace. This data indicates that the antioxidant activity is not only related to the total phenolic content but also to the different phenolic compounds present in each sample extract. Different compounds have different antioxidant activities (Natella et al, 1999). The results obtained highlighted that grape pomace, an underused by-product of the wine-making process, could be an alternative source of natural antioxidants that can be considered completely safe in comparison with synthetic ones.

The *functional properties* (water retention capacity and swelling capacity) of grape pomace varied depending on the sample type, and as a function of the drying method used. Remarkable differences were obtained between some lyophilized and dried oven grape pomace samples, depending on grape variety from water retention capacity and swelling capacity points of view. The drying and the milling processes change physico-chemical features, affecting their functional properties.

The results of the research were presented at international conferences or a workshop - see **3.3.** (1, 2, 3, and 4) and **3.4.** (2).

O1/A1.2. Physico-chemical, and molecular characterization and functional properties of maize flour

The estimative results that were obtained consist of moisture, ash, acidity, colour, protein content, fat, dietary fiber, total sugar, total phenolic content, antioxidant activity, molecular characteristics, and functional properties (water retention capacity and swelling capacity). Maize flour *moisture* had a higher value compared to dried grape pomace but ensured appropriate storage stability. Maize flour *acidity* is within the recommended limits. The *colour parameters* of maize are dictated by its pigments, flour luminosity being approximately twice that of the grape pomace samples. Maize flour has a much lower *lipid* content compared to grape pomace. Compared to the essential *amino acids* present in maize flour, grape pomace represents an interesting source of economic amino acids to utilize in food preparation. For maize flour, the *total fiber* content was much lower compared with the grape pomace samples analyzed. Results obtained for physico-chemical characteristics of maize flour revealed values that are within the limits mentioned in national or international standards. The *molecular characteristics* of maize flour revealed the distribution of nutritional components, such as moisture, proteins, lipids, ash, carbohydrates, starchy polysaccharides, and also amide I, amide II, amylose, and amylopectin at typical bands of individual components. Maize flour presented lower contents of *polyphenols* and *antioxidant activity* as compared to grape pomace. The data obtained for maize flour's *functional properties* revealed that it can absorb and retain water similar to grape pomace. The drying and the milling processes change the physicochemical characteristics of materials, affecting the functional properties of studied samples.

The results of the research were presented at an international conference or a at a workshop - see **3.3.** (1, 2, 3, and 4).

O1/A1.3. Data collection, elimination of aberrant results, and evaluation of the degree of validity and fidelity of the obtained results

The data collected from the research activities were assessed from a statistical point of view, the aberrant results were eliminated, and then the degree of validity and fidelity of the obtained data was evaluated. A database with physico-chemical, molecular characteristics, and functional properties of raw materials, grape pomace, and maize flour was achieved. This database with the results obtained will serve as a basis for subsequent objectives that were included in the work plan.

According to the work plan, the main attributes and characteristics of extrusion equipment were established and the necessary steps for the purchase were carried out. In this regard, the specifications were drawn up and uploaded to the platform of the Electronic Public Procurement System (SEAP), the offers were analyzed, the gainer was designated and the purchase contract was concluded with the supplier S.C. NITECH S.R.L. Bucureşti.

O2/A2.1. Mixture preparation for extrusion, physicochemical characterization, and functional properties

Grape pomace was mixed thoroughly with maize flour to the ratio of 0, 10, 20, 30, and 40% (w/w, d.b.), and the moisture, colour, ash, acidity, protein content, amino acids, fat, total dietary fiber, total phenolic content, antioxidant activity, degree of starch gelatinization were determined. The data regarding moisture, ash, acidity, colour, protein content, amino acids, fat, total dietary fiber, total phenolic content, antioxidant activity, starch gelatinization, and functional properties (water retention capacity and swelling capacity) were obtained. The physico-chemical characteristics of mixtures formulated varied depending on the grape variety, the drying method, the type of grape pomace sample, seedless or with seeds, and the amount that substituted maize flour. An increase in *ash*, *lipids*, *protein*, and *fiber* content and a decrease of carbohydrates with grape pomace amount increase was observed in the mixtures. The grape pomace features impacted mixture's characteristics. There were no significant differences between mixtures formulated with the same amount of grape pomace dried in an oven or through lyophilization from the point of view of ash, lipids, and proteins. The mixtures with grape pomace presented up to 3 times more *lipids* and *fiber* compared to maize flour, higher values being obtained for mixture with whole grape pomace compared to those with seedless grape pomace, especially from red grape variety. The lipids and fiber content rise with the increase of the amount of grape pomace in mixtures formulated. A high content of *proteins* was found in all mixtures with grape pomace from the red variety compared to those from the white variety. The *amino acid* content in the mixtures prepared was influenced by the amino acids present in studied grape pomace samples. There was no regular trend regarding amino acid content in mixtures with the increase of the amount of grape pomace which substitutes partial maize flour. An increase in essential amino acids was observed in mixtures including grape pomace with seeds compared to those with seedless grape pomace. Moreover, an enhanced profile in some *essential amino acids* such as valine, methionine, phenylalanine, isoleucine, leucine, and lysine were obtained for mixtures compared to maize flour. The *acidity* values for all mixtures were within the recommended limits, even though there are variations from one type of sample to another as a function of grape pomace type, grape variety, drying method, and the amount of grape pomace added in maize flour. The mixtures' *moisture* content is influenced by the grape pomace and maize flour moisture, but all mixtures formulated ensure suitable storage stability. The *colour* of the mixtures formulated for extrusion ranged with the amount of grape pomace added in maize flour and was impacted by the pigments from grape pomace and maize flour. Low values of luminosity (L^*) were observed in mixtures with whole grape pomace from the red variety compared with those with grape pomace from the white variety, and the lowest L^* value was registered in the mixture with 40% grape pomace. With the increasing grape pomace content, the mixture's colour became darker, indicating that the grape pomace, seedless or with seeds, had a strong effect on the colour of the mixtures. The redness (a^*) parameter increased with the increased amount of grape pomace in mixtures, and higher values were obtained for the samples containing lyophilized seedless red grape pomace. An opposite trend was observed for the yellowness (b^*) parameter in the mixtures with lyophilized seedless red grape pomace. Measurements of *starch gelatinization* by differential scanning calorimetry revealed that the values for onset temperature (T_o), peak temperature (T_p), final temperature (T_f), and endothermic enthalpies (ΔH) for the gelatinization were different from each other. The grape pomace variety, sample type, seedless or with seeds, and the proportion that substituted maize flour impacted the thermal properties of the mixtures formulated. The possible interactions between grape pomace and maize flour constituents may modify the binding forces among the crystallites and amorphous network by linking to the amorphous area of starch grains and thus influenced gelatinization temperatures, T_o, T_p and T_f (Zhu et al., 2009). The increase of grape pomace amount in mixtures determined a decrease of ΔH for all mixtures. This effect can be related to the immobilization of water due to the high fiber content from the mixtures formulated which also determines an increase in gelatinization temperatures. Regarding the *total phenolics*, mixtures prepared with seedless grape pomace samples showed a higher value compared with the mixtures with seeds grape pomace, the values depending on the addition level of grape pomace in maize flour. Different trends for the antioxidant activity of the mixtures formulated, depending on the mixture type, were observed. The amount of grape pomace which substituted maize flour influenced more or less the antioxidant activity, depending on the phenolic compounds that are present in each sample extract.

The mixture's *functional properties*, in terms of water retention capacity (WRC) and swelling capacity (SC), showed different values depending on the amount of grape pomace added in maize flour, grape pomace type, seedless or with seeds, grape variety, white or red, and also by the type of drying used. Functional properties of the mixtures prepared highlighted considerable differences between samples due to the milling processes of grape pomace which produce physicochemical changes in the features of the material. Different particle-size fractions of grape pomace have different chemical compositions and properties, and these influence the functionality of the mixtures to which they are added. The high values obtained for WRC and SC may be due to the high soluble dietary fiber from mixtures, since these functional properties are determined by the content in water-soluble-fiber-components of the material (Sosulski & Caden, 1982), or due to the other components, such as lignin which has water affinity (Lopez et al., 1996). The structural properties of the fiber have a stronger effect on the water-binding capacity of fiber than its chemical composition (Robertson & Eastwood, 1981). The results obtained suggest that the functional properties of mixtures formulated for extrusion are dependent on various factors and grape pomace properties.

Some of the obtained results regarding physicochemical characteristics of mixes formulated and functional properties were disseminated by their presentation at international conferences and scientific articles publication – see **3.1.** (3, 4, 5, 6, and 7), **3.3.** (1,2, 5, 7, 8, 9, 10, and 11) and **3.4.** (2).

O2/A2.2. Molecular analysis of grape pomace-maize flour mixtures formulated

Study of the molecular characteristics by using Fourier-transform infrared spectroscopy with attenuated transmission (ATR-FT-IR) for mixtures formulated was performed. The *molecular characteristics* of the mixtures allow the acquisition of spectra that present some characteristic bands of individual components. Valuable information about the chemical composition, including both primary and secondary metabolites, of the investigated samples was provided by these bands. FTIR spectra for *maize flour* revealed the distribution of nutritional components, such as moisture, proteins, lipids, ash, carbohydrates, starchy polysaccharides, and also amide I, amide II, amylose, and amylopectin at typical bands of individual components. Compared to maize flour, functional groups of the bioactive compounds, different phenolic compounds, fatty acids, polysaccharides, lignins, pectins, and organic compounds, such as sugars, alcohols, and organic acids were identified in the mixtures due to the presence of these constituents in grape pomace. Overall, all of the mixtures show similar spectra with peaks occurring in similar locations. Compared to maize flour, there is a variation in peak intensities that is approximately proportional to the grape pomace concentration added in mixtures. Moreover, the molecular characteristics are linked to grape pomace type, seedless or with seeds, and grape variety, white or red, but no differences were observed between the samples with grape pomace dried in an oven and those with grape pomace lyophilized.

The results regarding molecular characteristics of grape pomace were presented at the Elsevier conference, 3rd Food Chemistry Conference: Shaping a Healthy and Sustainable Food

Chain through Knowledge, Dresden, Germany, 10-12 October 2023; 19th Food Colloids Conference: Using colloids science to find new sustainable solutions in food, Thessaloníki, Greece, 14-18 April 2024 – see **3.3.** (4, 8, 9, and 10) and in scientific papers – see **3.1.** (3, 4, 5, 6, and 7).

O2/A2.3. Data collection, elimination of aberrant results, and evaluation of the degree of validity and fidelity of the obtained results

The data collected from the research activities was assessed from a statistical point of view, the aberrant results were eliminated, and then the degree of validity and fidelity of the obtained data was evaluated. For maize-based mixtures formulated with 10, 20, 30, and 40% whole grape pomace and seedless grape pomace, from white and red grape varieties, dehydrated by using a convective drying oven and lyophilization, respectively, a database with their physicochemical, and molecular characteristics, and functional properties was achieved. This database with the results obtained will serve to evaluate the extrudability of the mixture formulated or as the basis for subsequent objectives, such as setting up the extrusion process parameters or conditions.

The research was continued only for mixtures containing oven-dried grape pomace, because overall there were no significant differences between oven-dried and lyophilized samples regarding the characteristics evaluated. This solution was also decided due to the high costs involved in drying grape pomace by lyophilization. Also, because no remarkable differences were registered between the snack features obtained with different die diameters, and due to high material consumption, we decided to continue the experiment only with the die with 2 mm in diameter.

According to the work plan, *laboratory extrusion equipment was purchased*, it was put in service and training was made by the C. W. Brabender Instruments, Inc. representant, and S.C. NITECH S.R.L. representant.

O3/A3.1. Preliminary investigations for exploring and selecting the processing parameters and system parameters on mixtures formulated

Different parameter combinations of extrusion processing will be investigated by varying some parameters e.g., the levels of water added to the system, the feed rate, the temperature, the screw speeds, and specific mechanical energy. Due to the complexity, the parameters involved in the extrusion process will be tested to achieve the desired results, e.g., adequate expansion index, and density of the extrudates, by manipulating properties of feed mixtures and extrusion processing parameters. As deliverables associated with this activity, a database with variable levels tested in different kinds of extrusion processing (e.g., feed moisture, feed rate, die temperature, die dimensions, screw speed, and specific mechanical energy) was achieved.

The results regarding *preliminary investigations* for exploring and selecting the *processing parameters* and *system parameters on mixtures formulated* revealed the influence of extrusion processing parameters, such as feed moisture, feed rate, screw speed, temperature, and die dimension, and also of system parameters, pressure, torque, and specific mechanical energy on finite products. In this sense, these findings were disseminated at international conferences, workshops, and scientific articles - see **3.1.** (2), **3.3.** (6), and **3.4.** (1).

By manipulating the *extrusion processing parameters*, feed moisture (12 - 18%), feed rate (15 - 26 rpm), screw speed (130 - 170 rpm), barrel temperatures (25 – 180 °C), die dimension (ø 2, ø 3 mm), die temperature (160 – 180 °C) and extrusion system parameters, pressure and torque, a wide range of tests was performed to achieve expanded products like snacks with desired characteristics. Some of the resulting products can be seen in Figure 1. Also, the feed mixture characteristics were taken into account. The results of these tests allowed us to establish the processing conditions. Additionally, the changes produced by extrusion cooking were taken into account, findings which were reported in the article from the Q1 quartile – see **3.1.** (1). As well, a synthesis of the results was presented at the workshop: Current Trends in Research and Dissemination of Results in the Field Food – see **3.4.** (1).

Figure 1. Extrudates snack product results from different tests

O3/A3.2. Extrusion of formulated samples at establishing processing conditions, drying of the collected snacks extrudates and storage until analysis

The extrusion of formulated samples with different levels of grape pomace (10-40%) performed at established processing conditions, highlighted that expanded products have various characteristics, depending on temperatures along the barrel section and die temperature, screw speed, feeder speed, pressure, and specific mechanical energy. The collected expanded products were dried and stored until analysis.

O3/A3.3. Data collection, elimination of aberrant results, and evaluation of the degree of validity and fidelity of the obtained results

The data collected from the research activities was assessed from a statistical point of view, the aberrant results were eliminated, and then the degree of validity and fidelity of the obtained data was evaluated. A database with variable levels tested in different kinds of extrusion processing, e.g., feed moisture, grape pomace level in mixtures, feed rate, screw speed, and die temperature, was achieved. Additionally, a database on variable levels established and used in the experimental design to obtain expanded products was created.

O4/A4.1. Evaluation of the expansion ratio, water absorption index, water solubility index, colour, and textural properties

The data obtained for *expansion ratio* (ER), an indicator of puffy property, showed that the values depended on the grape pomace type, seedless or with seeds, variety of grape pomace, and addition level. For the snacks with the same level of grape pomace, remarkable differences for ER between seedless or whole grape pomace were observed. The magnitude of differences is depending on the grape variety. At high concentrations of seedless or whole grape pomace, a significant diminution in ER was observed. These changes can be associated with increasing fiber content in the extrudates which disrupted the expanded starch structure, affecting the expansion phenomenon. It was reported that the high insoluble fiber content from grape pomace reduced ER and created denser products (Fleischman et al., 2016). The *porosity* of expanded snacks is given by the air cells created during extrusion, products presenting lower bulk density. The degree of expansion is closely related to the size, number, and distribution of air cells surrounded by the cooked material (Hashimoto and Grossmann, 2003). The porosity was influenced by the grape variety, white and red, grape types, seedlsess and whole, and also by the maize to grape pomace ratio. Compared to the maize snacks, a slight increase in porosity was observed, especially for products with white grape pomace where those with seedless grape pomace showed high porosity compared to whole pomace ones. This might be due to their different lipid and fiber content and to the maize to grape pomace ratio. The use of white grape pomace in mixtures for the production of snacks leads to products with greater porosity than the use of red grape pomace.

Some data obtained regarding snacks ER and porosity has been presented at international conferences, and presented and discussed to a large extent in the articles – see **3.3.** (1) and **3.1.** (5, 6).

The snack's *functional properties* in terms of water absorption index (WAI) and water solubility index (WSI) showed variation in function of addition level and grape pomace variety. With the addition level rise, WAI and WSI decreased. These results are consistent with those reported by Raleng et al. (2016) and Poliszko et al. (2019) when pineapple pomace and pumpkin flour, respectively, were used to obtain extruded snacks. Some of the obtained results have been presented at international conferences, and presented and discussed to a large extent in the articles – see **3.3.** (11), and **3.1.** (5, 6).

The addition of seedless or whole grape pomace changes the extruded snacks' *colour*, the lowest luminosity being registered for snacks with a high percentage of pomace, especially for those from the red variety. When increasing the grape pomace concentration, whiteness (L*) and yellowness (b*) values decreased, and redness (a*) value increased, the colour parameters being remarkably affected by the grape pomace content and variety. The increase of a* value might be the consequence of grape pomace colour and thermal oxidation of grape pomace polyphenols during extrusion. Similar results were reported by Yu and Smith (2018). Incorporating whole and seedless grape pomace in snack formulations offers interesting *texture* characteristics. The changes in the textural parameters depend on the addition level and type of grape pomace, seedless or whole. Compared to the texture parameters of maize snacks, there is a reduction or a rise of the maximum cutting force, the crunchiness, the crispness, the compression force, and the fracturability that is approximately proportional to the grape pomace amount incorporated in new snacks. Some of the obtained results have been presented at the Elsevier international conference 11th Conference on Sensory and Consumer Research (EuroSense 2024: A Sense of Global Culture), Dublin, Ireland, 8-11 September 2024 – see **3.3.** (16, 17), and presented and discussed to a large extent in the accepted articles for publication in WoS – see **3.1.** (4, 5).

From the optimization process, results that the optimal doses for whole white and red grape pomace were found to be 21.50 and 18.28% respectively, giving snacks with ER, WSI, crispness at maximum, and WAI, cutting force, and compression force at minimum. These products presented a diminished ER compared to the control, but the porosity increased. More results and discussions to a large extent have been presented in a scientific article – see **3.1.** (6).

When investigating the possibility of using seedless grape pomace from white and red varieties in considerable percentages in the production of snacks maize to obtain products with maximum EI, porosity, and minimum WAI, from optimization process results that the best formation included 30% white seedless grape pomace and 20% red seedless grape pomace. A considerably improved porosity was obtained for snacks with white compared to red seedless grape pomace ones, but no significant difference was found between control and snacks with red seedless grape pomace. More results and discussions have been presented in the accepted article for publication in WoS – see **3.1.** (4).

O4/A4.2. Evaluation of starch gelatinization degree and *in vitro* starch digestibility of snacks from grape pomace-maize flour

Regarding *starch gelatinization degree*, DSC was characterized by evaluating the thermal properties of snacks. As no peak was observed in all snacks obtained, no gelatinization onset temperature (T_o), peak temperature (T_p), final temperature (T_f), or enthalpy (ΔH) values were detected for these samples. This indicated that during the extrusion process, the starch was completely gelatinized. Similar findings were reported by Han et al. (2021).

The data obtained revealed that extrusion cooking changed the *in vitro starch digestibility* of snacks from grape pomace-maize flour. By increasing the level of grape pomace from both varieties, white and red, the diminution of *in vitro* starch digestibility of snacks occurred. This trend may be due to the increase in fiber, and also in protein and lipid content of the extruded snacks. The reduction of digestibility can be attributed to starch, retrogradation of gelatinized starch, or formation of some complexes, such as amylose-lipid and starch-protein that may reduce the susceptibility of starch to enzymatic attack. Also, the high temperature during the extrusion process is another possible factor that can influence the *in vitro* starch digestibility of snacks. The obtained database with starch digestibility of snacks from grape pomace-maize flour was used to disseminate results in scientific articles and to propose patent applications – see **3.1.** (5, 6), and **3.2.** (1, 2).

Some of the obtained data were disseminated by presenting them at Elsevier international conferences or other international conferences, in scientific papers or patent proposals – see **3.3.** (12, 16, 17), **3.2.** (1, 2).

From the optimization process results that the optimal doses for white grape pomace, seedless and with seeds, were found to be 29.66% and 29.22% respectively, giving snacks with enhanced nutritional profile and a good acceptability from the sensory point of view due to the score more than 7. Additional results were presented and discussed to a large extent in the accepted article for publication in WoS – see **3.1.** (4)

When investigating the development of new expanded products with enhanced nutritional profiles by valorization of red grape pomace in maize snacks, from optimization process results that 30% seedless and 30% with seeds were found to be the adequate doses to improve samples from the nutritional point of view compared with the maize snacks. More results have been reported and discussed to an extent in the article – see **3.1.** (3).

O4/A4.3. The sensory evaluation of the snacks from GP-maize flour

Snack sensory characteristics were evaluated with a hedonic test, following of guidelines of ISO 8589:2007. Although having good acceptability, the *sensory characteristics* of the new snacks were different in appearance and form, color, smell, taste, flavor, and texture point of view. The snacks with the highest quantity of seedless grape pomace were described by darker colour and grape pomace aroma. A bitter taste was revealed for the snacks with the highest quantity of seedless grape pomace from the red variety, but the snacks were not considerably different in terms of flavor. The obtained results revealed that the acceptability of the products was within good limits up to an optimal amount of grape pomace added in maize flour. The database obtained with opinions regarding sensory characteristics of snacks from grape pomace-maize flour was used to disseminate results in scientific articles and to propose patent applications – see **3.1.** (4), and **3.2.** (1, 2).

O4/A4.4. The nutritional evaluation of snacks from GP-maize flour obtained at optimum extrusion processing conditions

The *nutritional profile* of snacks obtained at optimum extrusion processing conditions (24 rpm feed rate, 15% moisture, 150 rpm screw speed, and processing temperature of 50°C, 95°C, 175°C, and 180°) varied depending on the grape variety, the type of grape pomace sample, seedless or whole, and the amount that substituted maize flour. The *moisture* content was decreased with the addition level. The *acidity* of new snacks showed values within the acceptable range, not exceeding the maximum acid value of 4.0 mg KOH/g lipids, according to the Codex Alimentarius (CODEX-STAN 210 - 1999). An increase in *ash*, *lipids*, *protein*, and *fiber* content and a decrease of carbohydrates proportionally with the amount of grape pomace incorporated, was observed. The snack's nutritional profile was impacted also by the grape variety and nutritional profile of grape pomace components, peels, and seeds. Grape peels are interesting for their content of fiber and phenolic compounds, protein, ash, and soluble sugars, whereas grape seeds have a typically high content of fiber, oils, proteins, phenolic compounds, minerals and vitamins, sugars, organic acids, etc. (Iuga and Mironeasa, 2020; Deng et al., 2011). The snacks with grape pomace presented up to 11 times more lipids and more than 54 times fiber compared to maize snacks, depending on the grape pomace type, seedless or with seeds, and grape variety, white and red. Snacks with whole grape pomace from the red variety presented a high content of fibers and protein compared to from white grape pomace ones. The protein content of maize-grape pomace snacks was up to 2 times more than that of maize snacks, a higher content being obtained for samples with red grape pomace compared to with white grape pomace ones. In respect to the quality of protein from new snacks, an improved profile in the *amino acids essential* such as lysine, phenylalanine, leucine, valine, threonine, and methionine was found, still their content depending on grape pomace variety and type, seedless or whole. The total essential amino acids content increased considerably in grape pomace-maize snacks, approximatively proportionally with grape pomace level added, compared with maize snacks. As was expected, the most abundant *non-essential amino acids* observed in the expanded snacks were aspartic acid and glutamic acid, amino acids found in high proportion in grape pomace. Also, arginine, glycine, alanine, cysteine, tyrosine, and norvaline were found in snacks. The snack's amino acid content was impacted by the amino acid profile of grape pomace. Regarding starch digestibility, the results revealed that the snacks with grape pomace-maize showed a high content of *resistant starch* compared with maize snacks. Overall, there was an increasing trend with the rise of the amount of grape pomace in expanded products. A high content of resistant starch was obtained for the snacks with red grape pomace compared with those with white grape pomace. Starch structure, and consequently its digestibility can be changed by the polyphenols present in grape pomace (Chi et al., 2017).

Some of the obtained data was disseminated by presenting at international conferences and in scientific articles – see **3.3.** (16, 17), **3.1.** (3, 4, 5, 6, and 7).

O4/A4.5. Determining the total polyphenolic content, antioxidant activity, and lipid oxidation index of snacks from grape pomace-maize flour obtained at optimum processing conditions

The total polyphenolic content, antioxidant activity, and lipid oxidation index of snacks from grape pomace-maize flour obtained at optimum processing conditions were determined. The *total polyphenol content* and *antioxidant activity* of snack products were influenced by the type of grape pomace, addition level, and grape variety. The grape pomace-maize snacks showed up to 3 times more total polyphenol content and up to 2.5% more antioxidant activity compared to maize snacks. The presence of phenols like gallic acid and catechin from grape pomace can determine the enhancement of antioxidant activity due to their great antioxidant power (Mildner-Szkudlarz et al., 2012). Snack products obtained have a relatively low content of vegetable oils, up to 3.5%, depending on grape pomace type, and *peroxide index* values for all obtained snacks were within the recommended limits, even though there were variations from one type sample to another as a function of grape pomace type, grape variety and the amount of grape from snacks. The addition of grape pomace in the extruded product has the potential to reduce *lipid oxidation* (Yu et al., 2018). The snacks that include grape pomace with seeds presented high values for peroxide index compared to seedless grape pomace ones. This trend can be due to the oil content from seeds. Some of the obtained data were disseminated by presenting at international conferences and in scientific articles – see **3.3.** (13), **3.1.** (3, 4, 5, and 7).

O4/A4.6. Data collection, elimination of aberrant results, and evaluation of the degree of validity and fidelity of the obtained results

The data collected from the research activities was assessed from a statistical point of view, the aberrant results were eliminated, and then the degree of validity and fidelity of the obtained data was evaluated. As deliverables, a database with snack product characteristics, such as expansion ratio, porosity, water absorption index, water solubility index, colour and textural properties, degree of starch gelatinization and starch digestibility, and with opinions regarding sensory characteristics of snacks from grape pomace-maize flour was achieved. Additionally, a database with the nutritional profile of snacks from GP-maize flour in terms of moisture, ash, acidity, proteins, amino acids, fat content, dietary fiber, total polyphenolic content, antioxidant activity, and lipid oxidation index of snacks was achieved. Also, the scientific report on characteristics of snacks (expansion ratio, porosity, water absorption index, water solubility index, colour, and textural properties); degree of starch gelatinization and *in vitro* starch digestibility of snacks; nutritional characteristics; polyphenolic content, antioxidant activity, lipid oxidation index; opinions regarding sensory characteristics, was made. These characteristics of snack products were used to propose patent applications, in scientific papers published in ISI and BDI journals, and also disseminated at international conferences, as was shown above.

O5/A5.1. Microstructure analysis of the new snack product

Information on the microstructure of the new extruded product was obtained based on microscopic analysis by using a scanning electron microscope. In the extrusion process, starch granules are destroyed and a porous honeycomb structure is formed as the extrudate exits the extruder (Ye et al., 2018; Yan and Zhengbiao, 2010).

O5/A5.2. Evaluation of the microstructure

The microstructure images were evaluated to understand the changes that occur during the extrusion processing, the effects on the raw materials, and processing parameters on the snack features. Incorporating different levels of grape pomace in maize flour resulted in snack products with a wide range of *microstructure* which presents different sizes for the cells and their wall thickness. At high levels of seedless grape pomace, the amount of fiber is more than what the starch matrix can handle, and the walls of the cells collapse as the fiber particles pierce through them. Some of the obtained results regarding snacks microstructure were disseminated by presenting them at Elsevier international conferences 6th Food Structure and Functionality Symposium: Meeting the sustainability challenge, Bruges, Belgium, 6-9 October 2024 – see **3.3.** (16, 17), and in the scientific article – see **3.1.** (7).

O5/A5.3. Thermal properties of the new snack product

Thermal properties of snacks in terms of onset melt temperature, peak or gelatinization temperature, and enthalpy were determined through Differential Scanning Calorimetry (DSC). The *thermal properties* of snacks measured by using DSC showed that the extrusion process caused a complete gelatinization of starch granules. During extrusion, the interaction between temperature and moisture content affected starch transformation in extruded products. The high temperature during processing can enhance the extent of transformation while decreasing the die nozzle size has combined effects on shear rate and residence time (Chinnaswamy & Hanna, 1987).

O5/A5.4. Study of the molecular characteristics of the new snack product

Fourier-Transform Infrared Spectroscopy with Attenuated Transmission (FTIR-AT) was used to study the molecular characteristics of the new extruded product. The *molecular characteristics* of snacks obtained showed different spectra that present characteristic bands of particular components from product composition. The addition of grape pomace determined a rise of FT-IR peaks intensities compared to the control, highlighting peaks for different phenolic compounds, fatty acids, protein, polysaccharides, pectin, and organic compounds, depending on the grape variety and addition level. Overall, expanded snacks presented similar spectra with peaks occurring in similar regions. The molecular characteristics identified are related to seedless or whole grape pomace, and grape variety, white or red. The results obtained were presented at the Elsevier conference, 6th Food Structure and Functionality Symposium: Meeting the sustainability challenge, Bruges, Belgium, 6-9 October 2024 – see 3.3. (16, 17). The snacks with optimal doses of grape pomace white and red presented similar FT-IR spectra and intensities of absorbances, with a considerable difference being observed compared to the control. The presence of biologically active compounds from grape pomace in new snacks was indicated by the molecular characteristics which show an enrichment nutritional profile of these products. The addition of optimal doses of seedless grape pomace increased FT-IR peaks intensities compared to the control, indicating the presence of some components from grape peels, such as fiber, protein, pectin, etc. More results and discussions to a large extent have been presented in the scientific articles – see **3.1.** (3, 4, 5, 6, and 7).

O5/A5.5. Data collection, elimination of aberrant results, and evaluation of the degree of validity and fidelity of the obtained results

The data collected from the research activities was assessed from a statistical point of view, the aberrant results were eliminated, and then the degree of validity and fidelity of the obtained data was evaluated. The obtained database with the microstructure of snack products, and their molecular characteristics was used to disseminate results at international conferences and in scientific papers – see **3.3.** (16, 17), **3.1.** (7).

3. The estimated impact of the results obtained, with the underlining of the most significant result obtained

The expected result indicators: at least 6 articles in WoS and BDI, 3 international conference participation, and 1 EPO proposed patent.

The achieved result indicators: 6 articles published/accepted for publication in WoS and BDI, 1 article (under review) uploaded in a WoS journal, 17 scientific papers presented at the international conference, 1 OSIM proposed patent application, and 1 EPO proposed patent applications, and 2 workshop participations.

1. Articles published in indexed/quoted journals in Web of Science and in journals indexed in other international databases

1. Mironeasa, S., Coţovanu, I., Mironeasa, C., & Ungureanu-Iuga, M.* (2023). A Review of the Changes Produced by Extrusion Cooking on the Bioactive Compounds from Vegetal Sources. *Antioxidants*, *12*(7), 1453. Impact factor: 6.0 (Q1). DOI: 10.3390/antiox12071453. https://www.mdpi.com/2076-3921/12/7/1453
2. Coţovanu, I., Mironeasa S.*, Ungureanu-Iuga, M., Mironeasa, C. (2023). Effect of extrusion parameters on the extruded products' features. International Multidisciplinary Scientific GeoConference-SGEM, *Proceedings of 23rd International Multidisciplinary Scientific GeoConference SGEM 2023*, 23, 133-140. ISSN: 1314-2704. ISI Proceeding. DOI: 10.5593/sgem2023V/6.2/s25.17 https://www.sgem.org/index.php/component/jresearch/?view=publication&task=show&id=9593&Itemid=
3. Ungureanu-Iuga, M., Mironeasa, S.*, Batariuc, A., Mironeasa, C., Oroian, M.-A. (2024). Extruded snacks from maize flour with red grape pomace. *Ukrainian Food Journal*, 13(3), 557–575. ISSN: 2313-5891. Impact factor: 0.79 (Q4). DOI: 10.24263/2304-974X-2024-13-3-9. https://doi.org/10.24263/2304-974X-2024-13-3-9
4. Mironeasa S., Ungureanu-Iuga, M.*, Mironeasa, C. (2024). Assessing maize-based snacks formulated with whole and seedless white grape pomace. *Carpathian Journal of Food Science and Technology*, 16(4), 5-14. ISSN: 2066-6845. Impact factor: 0.50 (Q4). https://chimie-biologie.ubm.ro/carpathian_journal/
5. Mironeasa S., Ungureanu-Iuga M.*, Ursachi V.-F., Mironeasa C. (2024). Seedless grape pomace in expanded snacks for fiber improvement. *Ukrainian Food Journal*, 13(4), 681–695. ISSN: 2313-5891. Impact factor: 0.79 (Q4). https://doi.org/10.24263/2304-974X-2024-13-4-4.
6. Ungureanu-Iuga, M., Mironeasa S.*, Mironeasa, C. (2024). Evaluation of crisps from maize-integral grape pomace flour mix manufactured by extrusion cooking. *Journal of Agroalimentary Processes and Technologies*, 30(3), 264-273. ISSN (online): 2068-9551. Indexed: Google Scholar, EBSCO, CAS, CAB Abstract, IFIS. https://journal-of-agroalimentary.ro/journal-of-agroalimentary-processes-and-technologies-2024-30-3/evaluation-of-crisps-from-maize-integral-grape-pomace-flour-mix-manufactured-by-extrusion-cooking

7. Ungureanu-Iuga, M., Mironeasa S.*, Mironeasa, C. Oroian M.-A., Rotaru G.-M. (2024). Assessing changes in grape pomace-maize flour mixtures and extruded products. *Food Hydrocolloids*, FOODHYD-D-24-05062 – Under Review.

2. Patents applications submitted for evaluation to OSIM and EPO

1. Mironeasa S., Ungureanu-Iuga M., Mironeasa C. (2024). *Procedeu de obţinere a unui produs aglutenic extrudat, direct-expandat şi produs astfel obţinut*, nr. A/00678 din 07.11.2024 – OSIM.

2. Mironeasa S., Ungureanu-Iuga M., Mironeasa C. (2024). *Procedure for obtaining an extruded non-gluten product, direct-expanded and product so obtained*, No. 24464010.8/EP24464010 from 14.11.2024 – EPO.

3.3. International Conference Participations – scientific papers presented

4th International Conference on Food Bioactives and Health, Prague, Czech Republic, 18-21 September, 2023

1. Mironeasa, S., Ungureanu-Iuga, M., Batariuc, A., Coţovanu, I., Mironeasa, C. (2023). *The impact of two different drying methods on the polyphenolic content and antioxidant activity of grape pomace.* Abstract Book at: https://fbhc2023.com/wp-content/uploads/2023/09/fbh2023-abstract-book-a4-r49-FINAL-25-09-2023.pdf

2. Mironeasa, C., Batariuc, A., Ungureanu-Iuga, M., Coţovanu, I., Mironeasa, S. (2023). *Functional properties of grape pomace used in the extrusion process* Abstract Book at: https://fbhc2023.com/wp-content/uploads/2023/09/fbh2023-abstract-book-a4-r49-FINAL-25-09-2023.pdf

3rd Food Chemistry Conference: Shaping a Healthy and Sustainable Food Chain through Knowledge, Dresden, Germany, 10-12 October 2023

3. Mironeasa, S., Batariuc, A., Coţovanu, I., Codină, G.G., Mironeasa, C. (2023). *Grape pomace characterization for future valorization in extruded snacks.* https://virtual.oxfordabstracts.com/#/event/public/1683/people

4. Mironeasa, C., Ungureanu-Iuga, M., Oroian, M.-A., Mironeasa, S. (2023). *Use of ATR-FTIR spectroscopy technique for the estimation of grape pomace compounds* https://virtual.oxfordabstracts.com/#/event/public/1683/people

XXIIIrd SGEM GeoConference – "Green Science for Green Life", International Scientific Conference on Earth and Planetary Sciences SGEM, Schönbrunn Palace, Vienna Austria, 28 Nov - 1 Dec 2023

5. Batariuc, A., Mironeasa, S., Ungureanu-Iuga, M., Mironeasa, C. (2023). *Characterization of maize flour-grape pomace mixtures for snack production.* https://www.sgemviennagreen.org/program1/my_halls_day2_list.php?qs=Batariuc&tab=HALL1

6. Coţovanu, I., Mironeasa, S., Ungureanu-Iuga, M., Mironeasa, C. (2023). *Effect of extrusion parameters on the extruded products' features* https://www.sgemviennagreen.org/program1/my_halls_day3_list.php?qs=Cotovanu&tab=HALL1

The 9th Edition of the International Conference BIOTECHNOLOGIES, PRESENT AND PERSPECTIVES, 15th of December 2023

7. Mironeasa, S., Batariuc, A., Ungureanu-Iuga, M.*, (2023). *Assessment of the fiber content of grape pomace-maize flour mixtures used to produce high-fiber snacks.* https://fiajournal.usv.ro/conference2023/#

19th Food Colloids Conference: Using colloids science to find new sustainable solutions in food, Thessaloníki, Greece, 14-18 April 2024

8. Mironeasa, S., Ungureanu-Iuga, M., *Mironeasa C., Oroian M.-A. (2024). Effect of partial substituting maize flour with seedless grape pomace from white and red variety on the degree of starch gelatinization and molecular characteristics.* https://foodcolloids2024.org/abstract-book/

9. Mironeasa C., Ungureanu-Iuga M., Mironeasa S., Oroian M.-A. (2024). *Thermal properties and molecular features of enriched maize flour with grape pomace used in extrusion process.* https://foodcolloids2024.org/abstract-book/

10. Ungureanu-Iuga M., Mironeasa S., Mironeasa C., Oroian M.-A. (2024). *Assessing changes in maize-based mixtures formulated with whole and seedless white grape pomace by means of DSC and FTIR analysis.* https://foodcolloids2024.org/abstract-book/

10th International Conference on Agricultural and Biological Sciences (ABS 2024), Győr, Hungary, July 29th-August 1st 2024

11. Mironeasa, C., Batariuc, A., Mironeasa S., Ungureanu-Iuga M. (2024). *The impact of seedless grape pomace addition on some quality parameters of maize-based snacks.* https://opensz.oss-cn-beijing.aliyuncs.com/web2024/abs2024/file/ABS%20ABB2024-Conference%20Program-August%205th.pdf

12. Mironeasa, S., Batariuc, A., Mironeasa C., Ungureanu-Iuga M. (2024). *In vitro, starch digestibility assessing of maize-grape pomace extrudates.* https://opensz.oss-cn-beijing.aliyuncs.com/web2024/abs2024/file/ABS%20ABB2024-Conference%20Program-August%205th.pdf

3rd European Sample Preparation Conference (EuSP2024) and the 2nd Green and Sustainable Analytical Chemistry Conference (GSAC2024), Chania, Crete, Greece, 15th - 18th September 2024

13. Ungureanu-Iuga M., Mironeasa S., Batariuc A., Mironeasa C. (2024). *Influence of grape pomace addition on the nutritional and functional value of maize extrudates.* https://www.eusp-gsac2024.tuc.gr/fileadmin/users_data/eusp-gsac-2024/_uploads/BoA_EuSP2024-GSAC2024.pdf

11th Conference on Sensory and Consumer Research (EuroSense 2024: A Sense of Global Culture), Dublin, Ireland, 8-11 September 2024

14. Mironeasa S., Codină G.-G., Mironeasa C., Ungureanu-Iuga M. *Colour, texture and sensory acceptability of snacks made with maize and seedless grape pomace.* https://www.eurosense.elsevier.com/conference-programme.html

15. Mironeasa C., Codină G.-G., Mironeasa S., Ungureanu-Iuga M. *Assessing colour, texture and sensory features of snacks from extruded maize-whole grape pomace mix.* https://www.eurosense.elsevier.com/conference-programme.html

6th Food Structure and Functionality Symposium: Meeting the Sustainability Challenge, Bruges, Belgium, 6-9 October 2024

16. Mironeasa S., Batariuc A., Mironeasa C., Ungureanu-Iuga M. (2024). *Nutritional features, structure, texture and molecular characteristics of new high-fibre gluten-free snacks.* https://virtual.oxfordabstracts.com/event/8154/program
17. Mironeasa C., Batariuc A., Mironeasa S., Ungureanu-Iuga M. (2024). *Microstructural, physico-chemical and sensory texture characteristics of maize-base expanded snacks affected by the addition of winemaking by-products.* https://virtual.oxfordabstracts.com/event/8154/program

Workshop Participations

1. Mironeasa S. (2023). *Extrusion technology in food processing*. Current Trends in Research and Dissemination of Results in the Field Food, "Ştefan cel Mare" University of Suceava,

Faculty of Food Engineering, Suceava, Romania. https://fia.usv.ro/wp-content/uploads/sites/7/2023/03/Workshop-25-martie-2023-Program.pdf

2. Coţovanu (Verniceanu) I., Mironeasa S. (2023). *Grape pomace as a fiber source in extruded products*. Sustainable Food: Trends and Opportunities, "Ştefan cel Mare" University of Suceava, Faculty of Food Engineering, Suceava, Romania. https://fia.usv.ro/wp-content/uploads/sites/7/2023/10/Program-Workshop-Sustainable.pdf

The most significant results obtained:

- the established the optimal processing conditions to obtain expanded snacks;
- the patent proposal applications – revealed the expanded snacks with enhanced nutritional profiles by incorporating adequate levels of grape pomace in maize flour obtained;
- the dissemination of research results by participating in a much greater number of conferences than estimated ones.

The website of the project PN-III-P4-PCE-2021-0718, *Innovative approach to develop value-added snack products through extrusion technology* is actualized as can be seen at: (https://fia.usv.ro/cercetare/inadext).

Project director,
Prof. Ph.D. Eng. Silvia MIRONEASA