Recent trends on the valorization of winemaking industry wastes

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Abstract

Wine production is an activity of high global economic impact, and generates considerable amounts of wastes leading to severe problems in ecosystems. Therefore alternative uses need to be proposed. Winery wastes are rich in phytochemicals with great opportunities of utilization given their chemical structure and biological properties. In the last two years an intense research has been carried out aiming to generate new alternatives to characterize, purify or apply phytochemicals. As a result, new technologies have risen in different fields ranging from animal and human nutrition to new nanotechnological applications.

Keywords

Winery wastes, revalorization, fields of application, new technologies

Introduction

In 2018 global production of grape destined for wine reached 44.35 Mton, and 292 MhL of wine were obtained. **[1]**. After wine making, a considerable amount of solid waste containing skins, seeds and stems, regarded as grape marc o grape pomace is generated **[2]**, and represents approximately 20% of the total processed grapes **[3, 4]**. In white wine making, pomace is obtained after pressing before fermenting, while in red wine making, pomace is generated after fermentation **[5, 6]**. Lees are the wastes generated at the bottom of wine containers after fermentation, during storage, after the first or the second racking or after filtration, and are composed mainly by yeast **[7, 8]**. **Figure 1** shows an overview of the main generation flows of wastes in winemaking.

High useful content in phytochemicals, such as phenolic acids, flavonoids, and anthocyanins **[9, 10]**, structural polymers like cellulose and hemicellulose **[11, 12]** and lipids **[13]** have been found in winery wastes. However, in order to know the extent and to focus the field for the revalorization, comprehensive characterization **[13, 14, 6]** and optimization studies **[15]** are necessary. In this regards, besides the structural components, a wide array of phenolic compounds in a highly variable range of concentration have been found. Compounds and their concentrations, are influenced by the cultivar and the extraction method. For example a total phenolic contents ranging from 11.8 to 32.1 g/Kg grape pomace, and from 10.4 to 64.8 g/Kg pomace when studying four white grape cultivars and four red grape cultivars respectively when working with an acetone/water/acetic acid based extraction method was found **[13]**. With regards to phenolic profile, four phenolic acids (gallic acid, ferulic acid, p-coumaric acid, cafeic acid) rangin from 2.01 to 6.59 µg/mg of extract, and four flavon -3-ols (+Catechin, - epicatechin, epicatechin gallate, epigallocatechin gallate) ranging from 7.01 to 16.8 µg/mg of extract, when working with white grape marc, with a water based extraction method were found **[6]**. While when working with red wine and with an ultrasound assisted extraction a quite wider variety of phenolic compounds was found **[2]**, and included 13 anthocyanins, 2 were flavonols, the most representative anthocyanins were Malvidin-3-O-glucoside, Malvidin 3-(acetyl)–glucoside and Malvidin 3-(coumaroyl) –glucoside. Structural carbohydrates found in eight grape cultivars were Glucan, xylan, galactan, arabinan and manan. Glucan where the most abundant, ranging from 8.19 to 15.2 and from 8.04 to 12.2 % dry weight basis in white and red wine grape pomace, respectively. Additionally, grape pomace from eight cultivars showed a high concentration of unsaturated fatty acids, mainly linoleic and oleic acid **[13]**.

On the other hand, in recent decades large research has been conducted in order to take advantage of the biological properties of compounds provided by winery wastes, in different areas of interest, like food sector towards nutraceutical properties, and therefore functional in the prevention of various pathological conditions in humans **[16]**. Additionally, in agreement with industrial ecology concepts, like cradle-to-cradle and circular economy, it is necessary to recycle, reuse and recover valuable chemicals from waste and wastewaters as a major topic in wine processing industry. Besides costumer is increasingly aware of functional and natural and safe sources of food **[7, 11].** Within those principles, several research topics have been identified as trend on revalorization wine industry. In order to fully recover the compounds in winery wastes, several extraction methods have been proposed. **Table 1** shows the main trends analyzed in the last two years regarding extraction methods for wastes valorization. Moreover, the properties of phytochemicals as antioxidants, anti-inflammatory of polyphenols have been highly regarded not only in the field of human and animal health and nutrition but also in the field of microbiology as nutrients or biocides. Additionally, structural cellulose have been led to the production of new materials. **Table 2** shows the main trends of wastes valorization.

1. Extraction methods

In order to reach bioactive compounds chemically bonded to different matrix in plant tissues, it is necessary to find suitable processes **[17, 18]**. For a success in extraction, mild conditions of operation are needed **[19, 7]**, and economic considerations should be considered **[13].** In this regard, several approaches have been undertaken. Typical extraction procedures include the use of solvents like, methanol, acetone when solid liquid extraction is performed, or for liquid/liquid extractions hexane, chloroform or ethyl acetate are used [14, 15]. Solutions can be acidified [13]. Glycerol has been proposed as green solvent [9].

As alternatives to the use of solvents, environmentally friendly alternatives have been conducted with promising results. Micronization allowed to extract dietary fiber, and polyphenol extracts improving compound extractability, antioxidant capacity and radical scavenging activity [18]. On the other hand, due to low water solubility of polyohenols, the β-cyclodextrin has been proposed as hydrophobic molecules carrier in aqueous solution, and sonication for collapsing cavitation voids to generate high sheer forces and disrupt cell walls [20]. After sonication, further clarification procedures using ceramic membranes can be applied [2].

Enzymes can also aid polyphenol extraction. Polygalacturonase and pectin lyase depolymerize pectin through hydrolysis of the ester bond between the carboxyl and methyl of pectin groups as a result of de-esterification reactions [19]. Additionally, enzyme - assisted extraction and high hydrostatic pressure are foresighted as promising alternatives as they can render higher concentration of both total phenolic compounds and proanthocyanidins when enzymes where pretreated with high pressure before the extraction of phenolic compounds [17].

Microwave and ultrasound are promising for lees as source of polyphenols, as the extraction yield is doubled and the time for the extraction is reduced tenfold under the microwave, while ultrasound reduced the time three fold [10]. Microwave and ultrasound have also been performed for pomace as a source of phytochemicals [21].

When purification is needed, silica particles grafted with polyethylene glycol is used. Besides, tenability can maximize the adsorption capacity of polyphenols, with a complete recovery of compounds with up to 12-fold antioxidant activity compared to crude extracts [22].

Membrane technology is used for clarifications concentrate the bioactive compounds for lees as the source of phytochemicals [7, 23].

2. Animal nutrition

The use of by products from the winery industry has been motivated by several needs in the research field of animal nutrition like shortages in pasture availability and the need to lower methane emissions without compromising production yields [24]; the effect of feed in milk yield or composition [25]; study of physiological changes induced by polyphenols at cellular and organic level [26]; beef or chicken meat quality regarding the changes in composition in fatty acids and oxidative stress [3, 27, 4, 28]; the expression of genes in dairy cows [29] or changes in their proteome [25, 30], and last but not least, the need of replacement of antibiotics by bioactive phytochemicals [31]. All efforts have led to a noticeable amount of information to take into account when considering winery wastes as replacements for feedstock.

3. Human nutrition

Winery wastes are useful for applications as ingredient replacer in protein based food matrix. Grape pomace showed to work properly as seasoning ingredient for marinated chicken breasts [32]. Besides, several technological advantages were found regarding not only to the physicochemical properties, like firmeness, adhesiveness, but also both the total phenol content the antioxidant activity where increased, and a reduction of glycemic index by the increase in the slowly digestive starch, when using grape pomace as semoline replacer [33]. Additionally, in hamburger meat, grape pomace showed to fulfil at solving problems related to microbial spoilage, low content of unsaturated fatty acids, low antioxidant capacity and low dietary fiber [34]. On the other hand, advances have been done in the use of purified grape pomace as fining agent of wines [35].

4. Medical applications

Anti – inflammatory effects have been tested in animal models, and related with a range of illness. In human mesenchymal stem cell cultures, grape pomace proved to affect cell gene expression, by stimulating differentiation into the osteoblastic lineage. This finding is useful towards the enhancement of tissue response to dental implants and biomaterials [5]. Besides, in a murine collagen – induced arthritis model, treatments with mixtures of herbal origin containing grape pomace alleviated the severity of clinical symptoms. Moreover, an early treatment lead to a reduction on proinflammatory citokynes [36]. In mice models, it was also found that the addition of grape pomace attenuated the proinflammatory state showed in animals fed with atherogenic diets [37]. On the other hand, when combining grape pomace extracts with lactobacilli, it was found a synbiotic effect over several anti-inflammatory properties using an intestine-like in vitro model [38]. When fed with diets supplemented with grape pomace, mice showed an improvement in lifespan; an increased antioxidant activity in plasma; and a significant attenuation in atherosclerosis development [37]. Additional groundbreaking findings using animal models include the increase in the survival rates of *Caenorhabditis elegans* when submitted to thermal stress under treatments with grape pomace [39].

5. New materials

Revalorization of winery wastes can also be achieved via the generation of new materials. For example grape marc used as bioadsorbent for the removal of lead (II) resulted more effective when encapsulated in calcium alginate [40]. Moreover, calcium alginate entrapped grape marc is able to remove high percentages of methylene blue and methyl in liquid matrix [41]. Furthermore, the removal of reactive black 5 dye from aqueous solution was achieved when using a high active composite photocatalyst made with marc – based carbon and titanium dioxide in the mineral form of Anatase [42]. Moreover, zirconium – loaded grape pomace proved to provide superior defloridation than traditional resins [43]. Additionally, cellulose nanocrystals were successfully obtained from grape pomace, and sent to a process for obtaining self-healing nanocomposite hydrogels with efficient performance [12].

Nanoparticles also represent a field of interest. Phytochemicals from grape pomace can act as both reducing and capping agents to synthesize Ag dendritic nanostructures. The type of growth of structures was associated to the presence of organic components in the pomace, and showed properties similar to those obtained by chemically. Moreover, nanoparticles obtained with pomace showed both antimicrobial activity against *Fusarium graminearum*, and no obstruction on germination of wheat seeds [44]. Nanoparticles from grape pomace can also be obtained using plasma chemical reactor and can reduce the 4-nitrophenol to 4-aminophenol in the presence of sodium borohydride [45].

6. Agriculture applications

The market value of winery wastes is relatively poor, as the remaining phenolic compounds prevents germination [7, 46]. In order to increase the value as fertilizers, several studies have been undertaken with promising results. Water extractions and washes with hydraulic pressure can remove polyphenolic compounds to usable level. Seedling emergence was not affected for *Daucus carota* L. and *Zea mays* when depleted marc or blends with compost were performed [47]. Treatments with *Peniophora albobadia* LPSC # 285 decrease the phenolic compound content in grape pomace [48]. On the other hand, vermicomposting can efficiently decrease the total polyphenols and total antocyanins contents. Additinoally, changes on cellulose and hemicellulose content were observed. Stabilization of the grape marc was reached by growth of *Eisenia andrei* earthworms, increasing the mineral content of vermicompost [46]. Besides, results of vermicomposting experiments carried out using white and red grape marc, and also distilled grape marc showed changes in bacterial community and increases in processes related to cellulose metabolism, and synthesis of antibiotic and hormones [49, 50, 51]. This findings led to a better understanding of the underlying biological process towards the optimization of vermicomposting.

7. Biotechnological applications

Grape marc aqueous extract at low minimum bactericidal concentration showed antibacterial activity against *Staphylococcus aureus* and *Escherichia coli* [6]. Similarly, grape pomace entrapped in poly(lactic-co-glycolic) acid nanoparticles was effective against *Candida albicans* mature biofilm [52] the effectiveness was conferred to the phenolic content. Approaches have also been done regarding a wider array of pathological organisms affecting plants and humans, such as *Bacillus cereus*, *Escherichia coli*, *Salmonella enterica*, *Toxoplasma gondii*, *Staphylococcus aureus*, *Streptococcus uberis*, *Plasmodium falciparum* and *Phytophthora cinnamomi*, with activity against all tested species [16].

Value added products can also be obtained when using winery wastes as substrate in biological processes. By solid state fermentation of a blend containing grape pomace and wheat bran (1:1), an extract with polygalacturonase, CMCase and β-glucosidase activities was obtained [17]. Similarly, biopreservation compounds identified as isoamyl acetate, isoamyl alcohol, 2-phenyl ethylacetate and 2-phenyl ethanol were produced on fermentation media made with grape pomace extracts using the yeasts *Candida pyralidae* Y1117; *Pichia kluyveri* Y1125; *P. kluyveri* Y1164. Moreover inhibition activity against *Z. baillii*, *D. bruxellensis*, *D. anomala* was also proved [53]. In the same way, in order to diversify the revalorization of grape marc, different fermentation modes namely solid state, semiliquid and submerged fermentation were assayed using *Pleurotus* spp. Phenolic compounds were depleted and laccases were obtained. Moreover, mushrooms with enhanced nutritional value were obtained [54]. Lastly, in the tannase production by *Aspergillus niger*, the use of tannic acid can be reduced by adding grape pomace extracts [55].

Conclusions

The technologies afore discussed represent the up to date trends on the revalorization of winery wastes for obtaining high value products, plenty of them with promising results towards the utilization in practical applications. Better yet, when undertaken under frames proposed by the circular economy and the cradle to cradle concepts. More research is needed for optimization procedures have to be done in order to meet industrial criteria and validation trials, particularly those related to extraction methods and those pointed to improvements of human health, as the composition and concentration of compounds vary in crude extracts according to the extraction method and grape variety under study. Due to the amount of research generated, the diversification of applications, and the increasing awareness of consumer for natural products, the linkage of the winery industry to new industrial processes via revalorization of winery wastes generated can be foresighted as a reality in the years to come.

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Figure

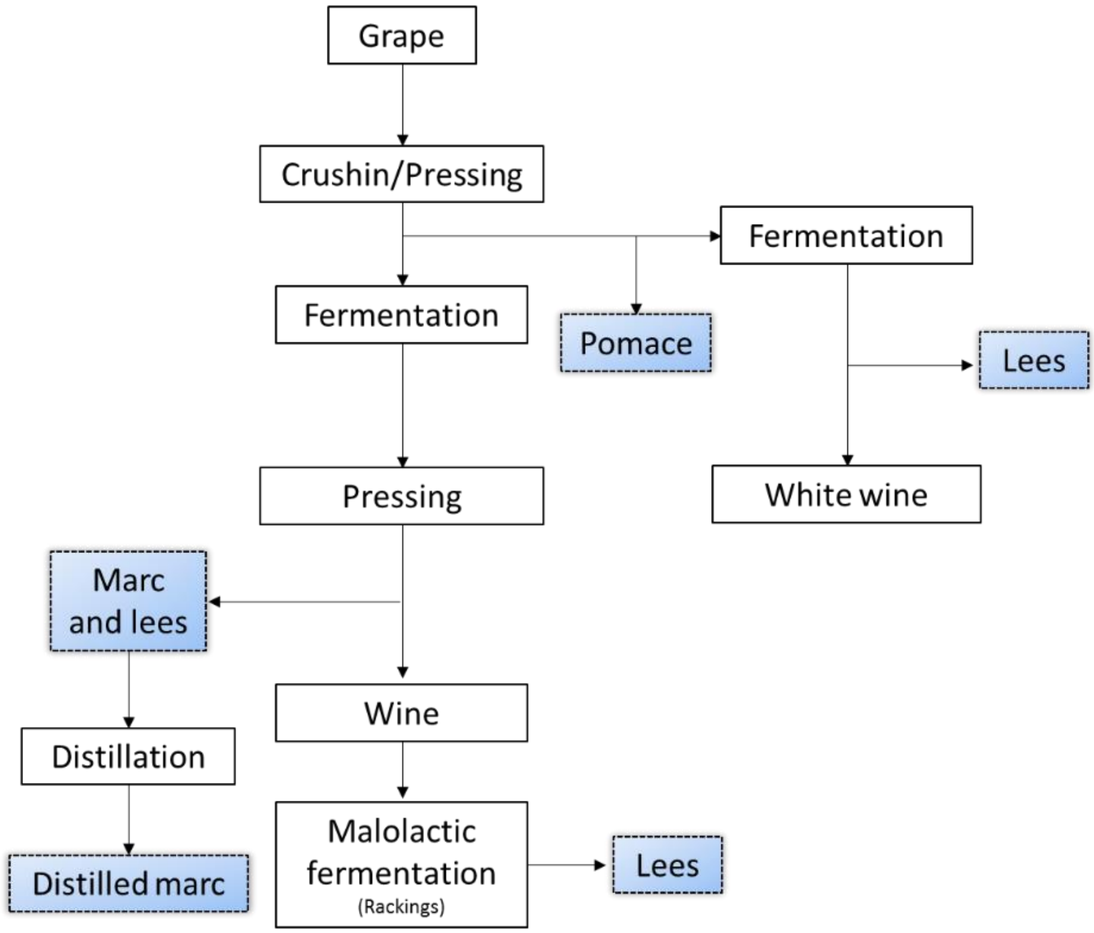

Figure 1. Main generation flows of wastes. Shaded rectangles show the wastes covered in this review.

Tables

Table 1. Main trends on winery wastes valorization regarding the extraction methods.

Winery waste	Asisted extraction method	Solvent	Reference
Grape Marc	Ultrasound	Grape marc, 23.85% w/w; ethanol, 40% w/w; water, 36.15% w/w	2
Grape pomace	Homogenizer	Glycerol (50 % v/v)	9
	Ultrasonic bath	70 % Acetone/28 water/2%Acitic acid	13
	Agitation	Metanol HCl 1% v/v, hexane, chloroform, ethyl acetate	14
	Bath water from 40 to 60 °C	Ethanol or Acetone 0.02 M sodium acetate	15
	Enzyme and High Hydrostatic Pressure	Buffer (pH 5) with a solid:liquid ratio of 1:8	17
	Micronization	Metanol/water solution	18
	Enzymatic extraction	Buffer acetate pH 4	19
	Probe and bath sonication	1.5 % aqueus solution of β-Cyclodextrin	20
	Microwave and ultrasound (probe)	Acidified aqueous solution with citric acid 2%	21
	Polyethylene glycol - Grafted silica particles	Polyethylene glycol, ethanol, sorbitol, citric acid, 200mM NaCl, water	22
Lees	Microwave and membrane (Microfiltration, Ultrafiltration, and nanofiltration)	Ethanol, hydrocloric acid and water.	7
	Vortex	Methanol/water/formic acid,	8
	Microwave and ultrasound	Water etanol mixture	10
	Poly(vinylidenefluoride) Hollow fibers and Ultra filtration membranes	Water (For extraction)	23

Table 2. Main trends on winery wastes valorization regarding the field of application.

Field of application	Winery waste	Model	Reference
Animal nutrition	Grape pomace	Beef	3
		Poultry	4
		Dairy cows	11
		Lambs	26
		Broilers	27
		Broilers	28
		Dairy cow	29
		Piglets	31
	Grape Marc	Dairy cows	24
		Dairy cows	25
Human nutrition	Grape pomace	Marinated chicken breast	32
		Durum wheat pasta	33
		Hamburger meat	34
		Fining agent for wine	35
Medical applications	Grape pomace	Human mesenchimal stem cells	5
		Murine	36
		Mice	37
		Human adenocarcinoma colon cell line Caco-2	38
		Caenorhabditis elegans	39
New materials	Grape Pomace	Cellulose nanocrystals Deep Eutectic Solvents	12
		Biosorbent with tetravalent zirconium ions	43
		Dendritic nanoparticles	44
		Ag nanoparticles	45
	Grape Marc	Alginate entrapped	40
		Alginate entrapped	41
		Activated carbon	42

Agriculture applications	Grape pomace	Phenolic compounds elimination by WRF	48
	Grape Marc	Vermicomposting	46
		Composting, growth carrots and corn	47
		Vermicomposting bacterial comunity changes white wine	49
		Vermicomposting bacterial comunity changes red wine	50
		Vermicomposting becterial comunity changes distilled grape marc	51
Biotechnological applications	Grape pomace	Production of enzymes	17
		In vitro	52
		Production of compounds inhibiting the growth of beberage spoling organisms.	53
		Production of enzymes and fruiting bodies using *Pleurotus* species.	54
		Tannase induced by grape pomace and tannic acid mixtures.	55
	Grape Marc	Antimicrobial activity against *Staphylococcus aureus* and *Escherichia coli*	6
		Antimicrobial activity angainst several organisms of economical interest.	16

Highlights

- Functional properties of winery wastes phytochemicals towards practical applications.
- Recent advances show the potential uses of winery wastes in a wide range of fields.
- New technologies for winery waste utilization, including nanotechnology applications.
- Outstanding results in medical applications of winery waste phytochemicals.

Annotated references

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* Results are of potentially high impact as represent a step forward on the utilization of grape pomace for on the improvement of nutritional quality of beef by dietary supplementation with a cheap raw material.

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* A self-sufficient approach is presented, as enzymes where produced and then used to treat the grape pomace.

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**Results obtained with grape pomace showed higher adsorption capacity than some other resins, and represent a high potential towards practical applications regarding water defluoridation.

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** Results presented are the cornerstone for understanding the biological processes underlying in the biotranformation of grape marc, resulting in a potential material to be used as a bulk in agriculture. Besides, the biological processes, as studied, can be useful for designing strategies at industrial scale.

Credit author statement

Dr. Oscar Manuel Portilla Rivera. Conceptualization, methotodlogy, formal analysis, writing original paper, resources

Dr. Maria Dolores Saavedra. Conceptualization, formal analysis, writing-review & Editing, visualization

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