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*Review*

# Strategies to Increase the Value of Pomaces with Fermentation

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**Abstract:** The generation of pomaces from juice and olive oil industries is a major environmental issue. This review aims to provide an overview of the strategies to increase the value of pomaces by fermentation/biotransformation and explore the different aspects reported in scientific studies. Fermentation is an interesting solution to improve the value of pomaces (especially from grape, apple, and olive) and produce high-added value compounds. In terms of animal production, a shift in the fermentation process during silage production seems to happen (favoring ethanol production rather than lactic acid), but it can be controlled with starter cultures. The subsequent use of silage with pomace in animal production slightly reduces growth performance but improves animal health status. One of the potential applications in the industrial context is the production of enzymes (current challenges involve purification and scaling up the process) and organic acids. Other emerging applications are the production of odor-active compounds to improve the aroma of foods as well as the release of bound polyphenols and the synthesis of bioactive compounds for functional food production.

**Keywords:** pressing residue; grape; apple; silage; animal production; enzyme production; polyphenols



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## 1. Introduction

Pomace is the main residue (a humid, solid material) generated from the pressing of fruits and olives to obtain juices and olive oil, respectively. This residue is heterogeneous and may contain seeds, pulp, stems, and peels, depending on the source [1,2]. In terms of the global production of juices and olive oil, the amount of pomace produced every year achieves several millions of tons [3,4]. Its high organic matter, nutrients, and moisture content favor the growth of microorganisms to decompose this residue (the generation of greenhouse gases, unpleasant odors, and contamination of groundwater) and can attract pests, which ultimately leads to an important environmental impact [5]. Additionally, the consumption of juices [6,7] and olive oil [8–10] is expected to increase in the upcoming years. In this sense, the residues from these two sectors of the food industry are expected to increase.

Another important aspect related to pomaces is the presence of bioactive compounds that are lost when these residues are discarded. One of the most studied classes of phytochemicals are polyphenols. This class of compounds is characterized by the antioxi-

dant [11–13], antimicrobial [3,12], anti-inflammatory [13], and anti-diabetic [14] activities tested in vitro and in vivo. This scenario can be seen as a relevant opportunity to explore strategies to improve the management of pomace and reduce its environmental impact.

In this sense, the concept of a circular economy is favored to improve the sustainability in this sector of the food industry, i.e., transforming residues into raw materials with high-added value and connecting them with other chains of food processing [15]. Moreover, a circular economy is one of the principles of the European Green Deal that aims to improve the efficiency of resource use and to cut pollution, for instance [16,17]. Recent publications support the potential utilization of this strategy [18–24]. It is also important to mention that the reutilization of residues of the food industry and the consequent development of food products are concepts supported and well-accepted by consumers [25,26].

Among the possible solutions to manage pomaces, fermentation has been suggested to obtain high-added value products and compounds. Moreover, fermentation can be seen as an important and more sustainable strategy to treat food industry residues [2,27,28]. Thus, this review aims to provide an overview of the utilization of fermentation (mainly involving lactic acid bacteria and yeasts) and biotransformation (biotransformation) of pomace in the production of silage and supplement feed for animal, enzymes, polyphenols, bioactive compounds (release of bound polyphenols and the synthesis of fatty acids and carotenoids), odor-active volatile compounds, and organic acid production.

## 2. Utilization in Silage or as a Feed Supplement for Animal Production

The feeding of animals reared for food production is one of the possible applications of fermented pomaces (Figure 1), for which there are two main strategies: adding the pomace in silage production or fermenting/biotransforming the pomace and using it as feed supplement (Table 1). Regarding the first strategy, the production of silage consists of preserving pasture grass for further use (especially during dry periods). The process occurs mainly by fermenting pasture with bacteria that are naturally or strategically added to acidify the material and delay microbial and biochemical spoilage [29]. Considering the importance of silage and the fermentation process, many studies have explored the effect of pomace in the characteristics of silage and its effect in animal health and performance.

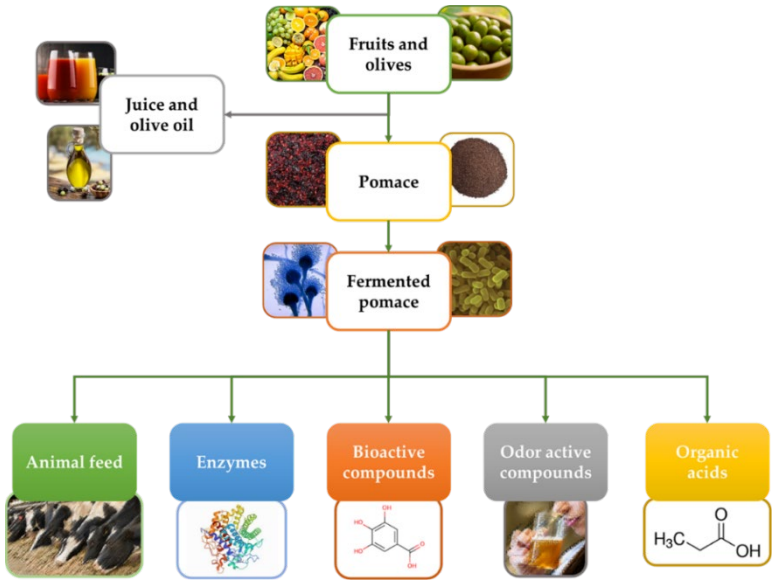


**Figure 1.** Schematic representation of strategies to valorize pomaces with fermentation.

**Table 1.** Effect of fermentation in the characteristics of silage produced from apple, white mulberry, and grape pomace.

| Source | Experimental Conditions | Effect in Silage Characteristics | Ref. |
| --- | --- | --- | --- |
| Apple pomace | Alfalfa hay, timothy hay, soybean meal, and vitamin and mineral supplement, pomace (0, 5, 10, and 20%), 3.4–22.4 °C, and 60 days | Increased pH, ethanol, acetic acid, and ammonia nitrogen levels; reduced lactic acid content | [30] |
| Apple pomace | Maize, wheat bran, soybean meal, timothy hay, alfalfa hay and vitamin-mineral supplement, pomace (20% in silage), 4.4–25.8 °C, and 60 days | Increased pH and ethanol content; reduced lactic and acetic acid, and ammonia nitrogen contents | [31] |
| Grape pomace | *Calotropis procera*, pomace (0, 10, 20, 40% in silage), and 90 days | Increased ethanol, acetic, propionic and butyric acid contents, effluent loss and gas loss; reduced soluble carbohydrate and lactic acid content and digestibility; no effect in pH | [32] |
| Grape pomace | Sweet sorghum silage, pomace (0, 5, 10, and 15% in silage), and 90 days | Increased acetic acid (only 10%) and total polyphenol content; reduced water-soluble carbohydrates, lactic acid (only 15% treatment), and butyric acid contents; no effect in dry matter and protein neutral and detergent fiber contents, pH, and ammonia nitrogen level | [33] |
| White mulberry pomace | Meadow grass, pomace (0, 25, 50, 75, and 100% in silage), and 60 days | Increased gas production, organic matter digestibility, and metabolizable energy | [34] |

Using pomace as a raw material for silage production may shift the characteristics of silage and change its content and composition of organic acids, digestible matter, and pH. These results were reported in studies with apple pomace that also indicated a reduction in the production of lactic acid [30,31]. Along with the increase in ethanol content in silage, the pH was increased, and the accumulation of lactic acid was reduced in relation to silage without pomace. However, these studies also indicated an unclear effect in the accumulation of ammonia nitrogen.

In the case of grape pomace, the depletion in lactic acid content and the increase in the production of other organic acids, polyphenol content, effluent and gas loss were also reported in two recent studies [32,33]. Both studies did not indicate significant differences in the pH of silage. It is relevant to mention that the study carried out by Li et al. [33] also evaluated the combination of grape pomace with the starter culture composed of *Lactobacillus plantarum* and *Lactobacillus buchneri*. These microorganisms led to a better control of fermentation and quality of silage by favoring the accumulation of lactic and acetic acid, water soluble carbohydrates, and crude protein. Moreover, ammonia nitrogen levels were reduced and no effect in the neutral detergent fiber content and the pH of silage were reported. A related experiment evaluated the production of silage with white mulberry pomace with meadow grass [34]. In this case, significant effects in organic matter digestibility and metabolizable energy, as well as in gas production, were reported.

Since silage is an important component for animal production in periods and regions of reduced feed availability, some studies reported the effect of silage with pomace and fermented pomace in animal nutrition, health and the composition and characteristics of foods obtained from animals in these experimental diets (Table 2). For instance, recent experiments reported the effect of silage added with apple pomace in the diet of Suffolk wethers [30,31]. In both cases, significant reductions in digestibility and nitrogen retention, in relation to the control diet, were reported. No effect in feed intake between control and experimental diets were indicated in these studies.

**Table 2.** Effect of fermented apple, grape, pomegranate, olive, and tomato pomaces in animal production and foods obtained from animals fed with these fermented pomaces.

| Source | Experimental Conditions | Animals and Study Characteristics | Effect in Animal Production and Related Food | Ref. |
| --- | --- | --- | --- | --- |
| Apple pomace | Silage: alfalfa hay, timothy hay, soybean meal, and vitamin and mineral supplement, pomace (0, 5, 10, and 20%), 3.4–22.4 °C, and 60 days | Suffolk wethers (4 animals), initial weight 50.3 kg, and 21 days of experiment | Reduced digestibility, gross energy, and nitrogen retention; no effect in feed intake and fiber content | [30] |
| Apple pomace | Silage: maize, wheat bran, soyabean meal, timothy hay, alfalfa hay and vitamin-mineral supplement, pomace (20% in silage), 4.4–25.8 °C, and 60 days | Suffolk wethers (4 animals), initial weight 65.3 kg, and 21 days of experiment | Increased organic acid content (except propionic and butyric); reduced digestibility and nitrogen retention; no effect in feed intake | [31] |
| Apple pomace | Silage: control feed, pomace (14.8% in silage), 9.7–20.1 °C, and 21 days | Male Yorkshire × Duroc × Landrace pigs (10 animals), initial weight 70 kg, and 53 days of experiment | Animals: increased feed efficiency; reduced average daily feed intake; no effect in finished body weight, average, daily gain, carcass weight, back fat thickness or dressing ratio<br>Back fat: increased moisture, linoleic acid (C18:2n6), linolenic acid (C18:3) and arachidic acid; reduced water holding capacity, palmitic acid (C16:0), palmitoleic acid (C16:1) and heptadecenoic acid (C17:1) proportion | [35] |
| Apple pomace | Silage: minced sardine, pomace (15%), *Lactobacillus plantarum* (starter culture), 35 °C for 7 days | Juvenile European sea bass fish (240 animals), initial weight 15 g, and 9 weeks of experiment | Increased feed conversion ratio, relative average daily feed intake, leukocyte count, and carcass composition (moisture, lipid and ash contents); reduced final body weight, weight gain, specific growth rate, protein efficiency, apparent net protein utilization, and microvilli density | [36] |
| Grape pomace | Silage: sorghum, pomace (0, 10, 20, and 30%), and 7 months | Male mixed breed lambs (24 animals), initial weight 21.5 kg, and 35 days of experiment | No effect in performance, carcass composition, and meat quality | [37] |
| Grape pomace | Silage: corn, water, starter culture, and pomace (43.6 g/kg feed) | Landrace × Large White − Duroc − Pietrain piglets (24 animals), and 15 days of experiment | Animals: increased antioxidant defense system response, average daily gain, growth of facultative probiotic bacteria, and LAB; reduced oxidative stress and pathogen<br>Meat: increased omega-3 fatty acids content; reduced n-6/n-3 ratio | [38] |
| Grape pomace | SSF: 1 kg substrate, *Aspergillus niger*, 30 °C, and 7 days; pomace (15 g/kg feed) | Male Ross 308 broiler chicks (140 animals), and 42 days of experiment | Animals: increased body weight and serum CAT level; reduced *Clostridium perfringens* count in cecum; no effect in feed intake, feed conversion ratio, serum GPx and SOD, other microorganism in cecum, and intestinal morphology<br>Liver: no effect in pH and color | [39] |

**Table 2.** *Cont.*

| Source | Experimental Conditions | Animals and Study Characteristics | Effect in Animal Production and Related Food | Ref. |
|---|---|---|---|---|
| Olive pomace | SSF: Two-step fermentation: *Bacillus subtilis* var. natto N21, 37 °C, 2 days; *Lactobacillus casei*, 25–35 °C, and 5 days; pomace (7.5, 15, and 30%) | Male Ross 308 broiler chicks (1400 animals), initial weight 44–47 g, and 42 days of experiment | Animals: increased feed conversion ratio, antioxidant status and defense system response; reduced body weight gain, protein efficiency ratio, nutrient digestibility, serum triglycerides and total cholesterol; no effect in feed intake, serum LDL cholesterol, ALT and AST<br>Breast meat: increased GPx and SOD; reduced fat and cholesterol content, and lipid oxidation status; no effect in moisture and protein | [40] |
| Tomato pomace | SSF: pomace (10% in silage), *Lactobacillus plantarum* (starter culture), and 30 days | Pregnant Holstein dairy cows (50 animals), initial weight 710–715 kg, 7 days of experiment | Animals: increased feed intake and digestibility, blood cholesterol and HDL, IgA, IgG, IgM, and antioxidant defense system response; no effect in feed intake, digestibility, milk yield and composition<br>Milk: increased vitamin A, C, and E contents; no effect milk yield and composition | [41] |
| Pomegranate pomace | SSF: 100 g substrate, *Aspergillus niger*, 30 °C, and 7 days; pomace (5 and 10 g/kg feed) | Male Ross 308 broiler chicks (175 animals), initial weight 39 g, and 42 days of experiment | Animals: increased crypt depth; reduced lipid oxidation, *Clostridium perfringens* in cecum, and villus height; no effect in body weight, feed intake and conversion ratio, carcass characteristics, antioxidant defense system response, and muscularis mucosa thickness<br>Meat and liver: no effect in color and pH | [42] |

ADF: Acid detergent, ALT: alanine aminotransferase, AST: aspartate aminotransferase, CAT: catalase, GPx: glutathione peroxidase, HDL: high-density lipoprotein cholesterol, IgA: immunoglobulin A, IgG: immunoglobulin G, IgM: immunoglobulin M, LAB: lactic acid bacteria, LDL: low-density lipoprotein cholesterol, NDF: neutral detergent fiber, SOD: superoxide dismutase, and SSF: solid-state fermentation.

A related experiment with pigs fed with silage containing apple pomace indicated minimal or non-significant effects in the growth performance, except for a reduction in daily feed intake and an increase in feed efficiency in animals fed with apple pomace silage [35]. Additionally, this study also indicated a significant increase in the content of some individual polyunsaturated fatty acids in back fat, whereas the content of few saturated and monounsaturated fatty acids in back fat were reduced. This effect was attributed to the dietary fiber found in apple pomace that favored the growth of probiotic microorganisms in pig intestine and led to the potential changes in back fat fatty acid composition.

Another interesting strategy to use silage with apple pomace was reported for the production of fish. Davies et al. [36] studied the effect of a silage produced with apple pomace, minced sardine, and *Lactobacillus plantarum* as a starter culture in the production of juvenile European sea bass. In these animals, the silage with apple pomace improved the health status of fish, whereas growth performance indicators were reduced in relation to the control diet (without apple pomace).

The effect of feeding animals with silage containing grape pomace was also reported in recent studies but contrasting results have been reported. In the experiment carried out by Massaro Junior et al. [37], increasing levels of silage with grape pomace (up to 30% in feed) did not cause significant changes in indicators of growth performance (initial and final body weight, average daily gain and feed conversion ratio), carcass characteristics (hot and cold carcass yield, for instance), and meat quality (such as pH, shear force, lipid oxidation, and color) in lambs. Conversely, the use of silage produced with grape pomace in piglets induced the antioxidant defense system, reduced the indicators of oxidative stress, and the counts of pathogenic microorganisms (*Campylobacter jejuni*, for instance) in fecal samples [38]. Additionally, the meat produced from animals fed with the experimental diet had more omega-3 fatty acids in comparison to the meat from animals fed with the control diet.

Fermentation in a solid state has also been explored to obtain potential feed additives for animal production. In the case of broiler chicks, the incorporation of fermented grape pomace in animal diets produced heavier animals with increased serum levels of catalase (a component of the antioxidant defense system) [39]. Additionally, no significant reductions in other components of the antioxidant defense system, intestinal morphology, and the pH or color of liver in the animals fed with silage containing grape pomace were reported in this study.

Olive pomace has been indicated as an interesting component to improve the diet of chicks [40]. Adding fermented olive pomace in animal feed enhanced the antioxidant status and the antioxidant defense system as well as reduced serum triglycerides and total cholesterol. Conversely, body weight gain was affected and no major effects in liver enzymes were indicated by the authors. The effects on animal health were also observed in meat in terms of reduced fat, cholesterol contents and lipid oxidation levels in breast meat. Another study indicated a favorable effect of solid-state fermented pomace in animal health [41]. In this case, the consumption of fermented tomato pomace improved health indicators (serum lipids and immune and antioxidant defense systems) in Holstein cows. However, the authors indicated no effects in terms of feed intake and milk production and composition (except for vitamins A, C, and E).

The effect of silage produced with pomegranate pomace in broiler chicks was evaluated by Gungor et al. [42]. The oxidative status was improved and some effects in the internal morphology were reported in animals consuming the experimental silage. No significant effects were reported for carcass characteristics, the antioxidant defense system, and meat and liver characteristics (pH and color).

From these experiments, it seems reasonable to consider that mixing pomace with other components for silage production modifies the microbial activity as well as the characteristics of silage. These effects can be attributed to the composition and content of nutrients (such as water-soluble carbohydrates). It is important to mention that the effect is dependent on the extract composition (apple vs. grape pomace, for instance).

Additionally, the shift in the fermentation process by using pomace as a raw material in silage production (especially for the production of lactic acid to ethanol) may be reduced from the addition of starter cultures. In terms of animal production, the main benefit seems to be related to animal health and the quality of foods obtained from these animals (chicks, cows, fish, lambs, and pigs), regardless of pomace source. In terms of animal production, the use of either pomace as silage raw material or fermented feed supplement seems to have a negative impact, such as in growth performance. It is worth mentioning that the modification of foods obtained from animals fed with fermented pomace fits in the strategy to naturally enrich foods with nutrients and functional compounds [43]. This strategy is supported by studies with apple [35], grape [38,39], olive and tomato pomaces [40,41]. However, additional studies are still necessary to identify relevant sources due to the controversial results such as those reported for pomegranate pomace in chicken meat [42].

## 3. Enzyme Production and Potential Applications

The use of pomace for the production of enzymes obtained from the agro-industrial processing of foods is an interesting strategy for producing high-added value products (Table 3). One of the main pomaces explored in the production of enzymes is obtained from apple processing. Recent studies point out that apple pomace can be used to obtain different enzymes without an additional carbohydrate source [44–48]. For instance, the production of lignin peroxidase and manganese peroxidase were reported from the fermentation of apple pomace with *Phanerochaete chrysosporium* BKM-F-1767 [48]. In this study, apple pomace was indicated as the most versatile residue to produce these enzymes in comparison to brewery residue, pulp and paper residue, and fishery waste.

The production of amylase, cellulose, pectinase, and xylanase was reported for fermentation with *Rhizopus delemar* $F_2$ [44]. Similarly, the production of pectinase was reported in another study carried out with *Aspergillus parvisclerotigenus* KX928754 where the fermentation was optimized in terms of pH, temperature, and the period of fermentation [45]. Similarly, the combination of two *Bacillus* strains, *Bacillus subtilis* and *Bacillus pumilus*, was indicated as a relevant strategy to produce pectinase from apple pomace [46]. In this study, the authors optimized the fermentation by exploring the effect of solid content and the ratio between *B. subtilis* and *B. pumilus* in the production of this enzyme.

**Table 3.** Production of enzymes from the fermentation of apple, grape, olive, tomato, orange, pea, and carrot pomaces.

| Source | Microorganism | Fermentation Conditions | Enzyme and Enzymatic Activity | Ref. |
|---|---|---|---|---|
| Apple pomace | *Phanerochaete chrysosporium* BKM-F-1767 | 40 g substrate, 60% moisture, pH 4.5, 37 °C, and 14 days | Lignin peroxidase: 141.4 U/gds<br>Manganese peroxidase: 631.2 U/gds<br>Laccase: 719.9 U/gds | [48] |
| Apple pomace | *Rhizopus delemar* $F_2$ | 5 g apple pomace, 10 mL moistening agent (6.0 g $Na_2HPO4$, 3.0 g $KH_2PO4$, 0.5 g NaCl, 1.0 g $NH_2Cl$, 2 mL of 1 M $MgSO_4$ and 0.1 mL of 1 M $CaCl_2$) | Amylase: 21.0 U/g<br>Cellulase: 18.2 U/g<br>Pectinase: 61.5 U/g<br>Xylanase: 158.3 U/g | [44] |
| Apple pomace | *Aspergillus parvisclerotigenus* KX928754 | 5 g substrate, pH 7.0, 168 h, 30 °C, 2% sucrose and 3% peptone, 30 °C, and 168 h | Pectinase: 1366.3 U/mL | [45] |
| Apple pomace | *Bacillus subtilis* and *Bacillus pumilus* (20 and 80% in inoculum, respectively) | 15 g substrate/L, 0.2 g/L pectin, 0.2 g/L $MgSO_4$ $7H_2O$, and 0.2 g/L $K_2HPO_4$, pH 9.0, 130 rpm, 30 °C, and 24 h | Pectinase: 11.25 U/mL | [46] |
| Apple pomace and dahlia tubers | *Mucor circinelloides* | 10 g substrate, apple pomace: dahlia tubers (9:1), 83.5% moisture, 0.3% $NH_4H_2PO_4$, 0.2% $KH_2PO_4$ and 0.1% KCl, pH 6.4, 30 °C, 5.8 days | Inulinase: 411.3 U/gds | [49] |
| Apple pomace | *Cellulosimicrobium* sp. CKMX1 (wild) and its mutant $E_5$ | 10 g substrate, 20 mL basal salt medium, pH 8.0, 35 °C, and 72 h | Xylanase: 418 (wild) and 568 (mutant $E_5$) U/g | [47] |
| Grape pomace | *Aspergillus niger* NRRL3 | 100 mL modified Czapex minimal medium with grape pomace, 4% tannic acid, pH 5.50, 120 rpm, 30 °C | Tannase: 3.0–4.5 U/mL | [50] |
| Grape pomace | *Bacillus subtilis* natto DSM 17766 | 15 g/100 mL, 3% $H_2SO_4$, pH 6.0, and 7 days | Cellulase: 0.2 U/mL | [51] |
| Grape pomace | *Pleurotus ostreatus* and *Pleurotus pulmonarius* | 4 g, 26 °C, 140 rpm, and 15 days | Laccase: 26.2 and 15,273.0 U/g for *Pleurotus ostreatus* and *Pleurotus pulmonarius*, respectively | [52] |
| Grape pomace and wheat bran | *Aspergillus niger* 3T5B8 | Grape pomace: wheat bran (50 and 50%), 60% moisture, 0.91% ammonium sulfate solution, 37 °C, and 96 h | Tannase: 0.30 U/g | [53] |
| White grape pomace, olive mill wastewater, red grape pomace and wheat bran | *Aspergillus niger* B60 | 50 g substrate, white grape pomace and olive mill wastewater, red grape pomace, and wheat bran (15, 15 and 70% of total substrate, respectively), 30 °C, and 120 h | CMCase: 668 U/g<br>Polygalacturonase: 3151 U/g<br>Amylase: 1099 U/g<br>Xylanase: 579 U/g<br>Protease: 204 U/g | [54] |
| Olive pomace | *Kluyveromyces marxianus* | 5 g substrate, 45 °C, and 48 h | Tannase: 42.4 U/mg | [55] |
| Exhausted olive pomace | *Aspergillus niger* CECT 2915 | 10 g substrate, 30 °C, and 6 days | Xylanase: 28 U/g<br>Cellulase: 38 U/g | [56] |

**Table 3.** *Cont.*

| Source | Microorganism | Fermentation Conditions | Enzyme and Enzymatic Activity | Ref. |
|---|---|---|---|---|
| Olive pomace and wheat bran | *Aspergillus ibericus* MUM 03.49, *Aspergillus niger* MUM 03.58, and *Aspergillus tubingensis* MUM 06.152 | 30 g olive pomace: wheat bran (50 and 50%), 75% moisture, 30 °C, and 7 days | Lipase: 223, 53.6 and 7.6 U/g for *A. ibericus*, *A. niger* and *A. tubingensis*, respectively | [57] |
| Exhausted olive pomace | *Crypthecodinium cohnii* ATCC 30772 | 5 and 8 g substrate/L, 27 °C, 160 rpm, and 7 days | Pectinase: 37 and 33 U/mL for 8 and 5 g/L olive pomace, respectively | [58] |
| Tomato pomace | *Aspergillus oryzae* NRRL 2220 in SSF or SmF | 10 g substrate, 19.8 g/L casein, 0.92 g/L NaCl, 30 °C, and 72 h | Protease: 21,309 and 2343.5 U/g for SSF and SmF, respectively | [59] |
| Tomato pomace | *Aspergillus oryzae* 2220 | 20 g, 50% initial moisture content, pH 6 and 1 mL of 5-day-old inoculum, 30 °C, and 72 h | Protease: 12 U/gds after 42 h | [60] |
| Tomato pomace | *Aspergillus oryzae* 2220 (static bioreactor) | 5 kg, 10 cm bed, 30 °C, and 44 h | Protease: 13.6 U/gds | [60] |
| Tomato pomace and sorghum stalks | *Pleurotus ostreatus* and *Trametes versicolor* | 500 g tomato pomace, 100 g sorghum stalks, and 28 °C | Laccase: 15 and 35 U/g for *P. ostreatus* (4 days) and *T. versicolor* (18 days), respectively<br>Protease: 13,000 and 34,000 U/g for *P. ostreatus* (4 days) and *T. versicolor* (13 days), respectively<br>Xylanase: 9 and 50 U/g for *P. ostreatus* (4 days) and *T. versicolor* (13 days), respectively | [61] |
| Tomato pomace, wheat bran, and canola meal | *Bacillus subtilis* T4b | Wheat bran 30 g/L, canola meal 40 g/L, and tomato pomace 15 g/L, 180 rpm, 28 °C, and 48 h | Xylanase: 315 U/mL | [62] |
| Orange pomace | *Aspergillus niger* | 5 g substrate, 30 °C, and 96 h | Pectinase: around 17 U/g (endo+exo enzyme activities) | [63] |
| Orange pomace | *Aspergillus niger* (tray bioreactor) | 285 g substrate/tray, 30 °C, and 96 h | Pectinase: around 60 U/g (endo+exo enzyme activities) | [64] |
| Orange pomace with sugarcane bagasse | *Aspergillus niger* (tray bioreactor) | 285 g substrate/tray, 30 °C, and 96 h | Pectinase: around 75 U/g (endo+exo enzyme activities) | [64] |
| Orange pomace | *Aspergillus niger* (rotating-drum bioreactor) | 285 g substrate/batch, 30 °C, and 96 h | Pectinase: around 40 U/g (endo+exo enzyme activities) | [64] |
| Carrot pomace | *Penicillium oxalicum* BGPUP-4 | 10 g substrate, 90% moisture, 0.5% inulin, 0.2% $NaNO_3$, 0.2 g/mL $KH_2PO_4$, 0.1% KCl, 0.05% $MgSO_4 \cdot 7H_2O$, 0.001% $FeSO_4$ $7H_2O$ and 0.2% $NH_4H_2PO_4$, pH 7.0, 30 °C, and 4 days | Inulinase: 322.10 U/g | [65] |

CMCase: carboxymethyl cellulase, SmF: submerged fermentation, and SSF: solid-state fermentation.

Combining apple pomace with other sources of nutrients can improve enzyme production yields. This factor was considered in the experiment carried out by Singh et al. [49], who used dahlia tuber powder (source of inulin) to produce inulinase with apple pomace. These authors optimized the fermentation in terms of moisture, fermentation period, and pH. Another interesting strategy to obtain extracts rich in enzymes from apple pomace consist in generating mutant strains such as those indicated by Guleria et al. [47]. In this case, the new mutant of *Cellulosimicrobium* sp. CKMX1 $E_5$ increased the production of xylanase in relation to its parent strain.

Grape is another relevant substrate for the production of enzymes. In this case, the production of tannase was obtained from the fermentation with *Aspergillus niger* NRRL3 [50]. Similarly, the production of cellulose using *Bacillus subtilis* was also obtained from the fermentation of grape pomace [51]. Another recent experiment indicated that the production of laccase from grape pomace was dependent on the starter culture [52]. In this case, *Pleurotus pulmonarius* was more efficient for producing this enzyme than *Pleurotus ostreatus*. Moreover, the authors also indicated that solid-state fermentation was more appropriate than semiliquid and submerged fermentations.

The effect of adding wheat bran in grape pomace for the production of different enzymes was studied in a recent experiment [53]. The fermentation with *Aspergillus niger* successfully produced more tannase by combining wheat bran with grape pomace than using only wheat bran. However, the presence of grape pomace limited the production of xylanase and β-glucosidase and slowed the production of polygalacturonase. Additionally, the authors also reported a dependency on time for the production of polygalacturonase and carboxymethyl cellulase (higher enzymatic yields were obtained after 96 h of fermentation). Additionally, Papadaki et al. [54] reported the production of amylase, carboxymethyl cellulase, polygalacturonase, protease, and xylanase from a substrate composed of white grape pomace, olive mill wastewater, red grape pomace and wheat bran. *Aspergillus niger* was used to obtain these enzymes.

Olive processing for oil extraction also generates a valuable substrate for microbial enzyme production. For instance, a recent experiment with olive pomace indicated that tannase could be obtained from the fermentation with *Kluyveromyces marxianus* [55]. Another relevant example that supports the use of this pomace in the production of enzymes is the study carried out by Leite et al. [56]. In this case, the authors fermented the exhausted olive pomace with *Aspergillus niger* and reported the production of xylanase and cellulose. In the case of lipase production from grape pomace, the effect of *Aspergillus* species was evaluated in a recent study [57]. *Aspergillus ibericus* was a more efficient species in relation to *Aspergillus niger* and *Aspergillus tubingensis*. Interestingly, a related experiment with exhausted olive pomace reported the production of pectinase from the growth of the microalgae *Crypthecodinium cohnii* [58]. Additionally, no significant differences in terms of substrate concentration (5 vs. 8 g/L) in the production of this enzyme were reported.

Tomato is another relevant source of pomace that can be utilized in the production of enzymes. Proteases could be obtained from tomato pomace using *Aspergillus oryzae* according to recent studies [59,60]. Moreover, the study carried out by Belmessikh et al. [59] indicated that the production of protease from tomato pomace was more efficient in solid-state rather than submerged fermentation. The optimization also indicated that casein and NaCl levels are significant factors in improving the production of protease.

The combination of tomato pomace with other sources of nutrients for enzymatic production has also been explored [61]. Particularly, for the combination with sorghum stalks, the production in a laccase, protease, and xylanase were dependent on the starter culture [61]. In this case, *Pleurotus ostreatus* was associated with a faster but less intense production of these enzymes. Conversely, *Trametes versicolor* had higher production yields but after longer fermentation periods. Another more recent experiment with tomato pomace, wheat bran, and canola meal indicated that the fermentation with *Bacillus subtilis* was associated with high xylanase content [62].

Another relevant pomace for the production of enzymes is obtained from orange processing. In this case, recent experiments explored the generation of pectinase from the fermentation with *Aspergillus niger* [63,64]. It is also relevant to comment that a recent experiment indicated that the use of sugarcane bagasse is a relevant strategy to reduce moisture loss during fermentation and improve the production yield of pectinase from orange pomace [64]. In a similar way, carrot pomace was indicated as an interesting substrate for fermentation, which can be utilized in the production of inulinase [65]. The production of inulinase was affected by moisture content, fermentation period, and pH.

The production of enzymes from pomaces can also be improved by the use of emerging technologies such as microwave heating and ultrasound. This aspect was reported in the production of carbohydrases from apple pomace by Pathania et al. [44]. According to these authors, the intensity of microwaves (as a pre-treatment) had a significant effect on the production yield. The maximum values for amylase, pectinase, and xylanase were reported for the 450 W treatment. Additional power (up to 600 W) caused a reduction in the production of enzymes. In the same line of thought, the use of ultrasound can improve the production of enzymes. Leite et al. [56] indicated that using 750 W and 20 kHz and optimizing the time and liquid/solid ratio (12.4 min and 7.3) maximized the production yields of xylanase (75 U/g) and cellulase (35 U/g).

It is also important to highlight that some experiments to scale up the production of enzymes from pomaces have been carried out in the last decade. One relevant example that explored this aspect was performed by Boukhalfa-lezzar et al. [60] with tomato pomace fermented with *Aspergillus oryzae*. In this study, similar production yields were reported between lab scale and a bioreactor for protease production (12 U/gds after 42 h with 20 g of substrate vs. 13.6 U/gds after 44 h with 5 kg of substrate). Another relevant experiment supporting the increase in the production scale of enzymes was carried out with orange pomace in a tray reactor and a rotating-drum reactor [64]. In this case, differences in production yield were reported between these two reactors wherein the bioreactor with trays had the highest yield. Moreover, both reactors increased the production of pectinase in relation to a previous experiment from the same research group [63].

The purification of enzymes obtained from fermentation is another relevant aspect considered in recent studies. In order to explore potential solutions to improve the separation of enzymes, an experiment with lignin peroxidase and manganese peroxidase explored the use of centrifugation and filtration after the fermentation of apple pomace [48]. The results revealed that centrifugation was more efficient for separating both enzymes than filtration. A recent study compared the use of fractionation with ammonium sulfate and chromatography filtration in the purification of tannase from fermented olive pomace [55]. Both methods led to extracts with increased enzymatic activity wherein the chromatography filtration was more efficient than fractionation with ammonium sulfate (1026.1 vs. 664 U/mg, respectively). A similar outcome was obtained in another study with pectinase from apple pomace (1081.7 vs. 860.6 U/mg for chromatography filtration and ammonium sulfate fractionation, respectively) [45].

Potential applications can also be considered in the context of enzyme production. Since pomace is a by-product from food processing, the use of these enzymes in food production can be suggested. A relevant example is the experiment carried out by Mahmoodi et al. [63]. In this study, cubic pieces of fresh apple were treated with pectinase to produce apple juice. The main effects were the reduction in juice viscosity and increased juice yield, soluble sugar content, and pectate content. A similar experiment with polygalacturonase obtained from apple pomace was efficient for clarifying apple juice [66].

An interesting application for enzymes obtained from pomace fermentation is the detoxification of food. This approach was evaluated by Cuprys et al. [67] who applied laccase from apple fermentation with *Trametes versicolor* to decompose ciprofloxacin (an antibiotic). However, the presence of a reducing agent (syringaldehyde in this study) was necessary to favor the enzymatic degradation of this antibiotic in water. Although the scientific information about the application of microbial enzymes from pomace fermentation

in food processing is limited, the use of these enzymes could be considered to improve the processing of beer, bread, cheese, syrup, and wine [68] in further experiments. Moreover, potential applications in other research areas are in pharmaceutical, chemical, fuel, and paper production [69].

The use of pomace from different sources can be seen as a relevant strategy to favor the production of enzymes. Current scientific evidence indicates that the production of enzymes can be improved by adding complementary sources of nutrients (such as ingredients rich in carbohydrates for pomaces with reduced levels of this nutrient), applying emerging technologies to favor the exposure of substrates, and increasing the scale of production (minimal effect in production yield, to some extent). The purification with different techniques can also be applied and support the progression towards application in other industrial processes.

## 4. Release and Production of Bioactive Compounds

Improving the biological activity of pomace from food processing is one of the potential and emerging applications of fermentation. This strategy has been applied to obtain carotenoids, fatty acids, γ-linolenic acid, and polyphenols (Table 4). Polyphenols are an important class of bioactive compounds that are found in pomaces. From a broad perspective, polyphenols can be found either in free or bound forms. Polyphenols in free form are those present in the cytosol of vegetable cells, whereas the bound polyphenols are those bound to cell wall constituents [70]. For bound polyphenols in particular, their extraction is complex and conventional extraction methods have low efficiency to separate these compounds from structural components of food. In this context, the use of fermentation (by means of the action of microbial enzymes) has been indicated as a relevant strategy to recovery this compound [70,71].

**Table 4.** Bioactive compounds obtained from pomace fermentation.

| Source | Fermentation Conditions | Bioactive Compounds | Outcome | Ref. |
|---|---|---|---|---|
| Grape pomace | 2 g substrate, *Rhizomucor miehei* NRRL 5282, 37 °C, and 18 days | Polyphenols | Oven dried: reduction in TPC and FRAP, no effect in DPPH<br>Lyophilized: maximum TPC and FRAP values at day 7, no effect in DPPH | [72] |
| Grape pomace | 10 g substrate (grape pomace:wheat bran; 1:1), *Aspergillus niger* 3T5B8, 37 °C, and 96 h | Polyphenols | Increased TPC, ABTS, and ORAC | [53] |
| Grape pomace | 50 g substrate, *Trametes versicolor* TV-6, 5 mycelial plugs, 27 °C, and 15 days | Polyphenols | Reduced 5-lipoxygenase and hyaluronidase activities (up to 4 days of fermentation), and polyphenol content throughout fermentation period | [74] |
| Grape pomace | 60 g substrate, *Actinomucor elegans* ATCC-22963 or *Umbelopsis isabellina* ATCC-36671, 30 °C, and 12 days | γ-Linolenic acid and carotenoids | γ-Linolenic acid: maximum at 4 days for *Umbelopsis isabellina* and 6 days for *Actinomucor elegans*<br>Carotenoids: carotene increased throughout fermentation and maximum at 8 days for lutein | [73] |
| Apple pomace | 2 g substrate, *Rhizomucor miehei* NRRL 5282, 37 °C, and 18 days | Polyphenols | Oven dried: reduced TPC, maximum FRAP value at day 3, no effect in DPPH<br>Lyophilized: slight increase in TPC and DPPH up to day 10, maximum FRAP value at day 10 | [72] |
| Apple pomace | 12.5 g, natural fermentation, 30 °C, and 72 h | Polyphenols | Reduced throughout the fermentation period | [75] |
| Apple pomace | 250 g substrate, *Saccharomyces cerevisiae* ref: 32, *Saccharomycodes bayanus* ref: C6, and *Hanseniaspora uvarum* ref: 62, 25 °C, and 7 days | Fatty acids and polyphenols | Increased fatty acids<br>Slight reduction in polyphenols | [76] |
| Apple pomace | 40, 60 and 80 g substrate/L, *Yarrowia lipolytica*, 28 °C, and 6 days | Fatty acids | Maximum production after day 3 | [77] |
| Elderberry and dwarf elderberry pomace | 50 g substrate, *Aspergillus niger* ATCC-6275, 30 °C, and 6 days | Polyphenols and fatty acids | TPC: maximum release up to 3–4 days of fermentation<br>DPPH: maximum after 3–4 days of fermentation<br>Lipids: slight increase in linoleic and oleic acids up to 4 days of fermentation | [78] |
| Olive pomace | 5 g substrate, *Kluyveromyces marxianus* NRRL Y-8281, 45 °C, and 48 h | Tannic and gallic acids | Reduced tannic acid and increased gallic acid content | [79] |
| Exhausted olive pomace | 5 and 8 g substrate/L, *Crypthecodinium cohnii* ATCC 30772, 27 °C, 160 rpm, and 7 days | Fatty acids | Increased total lipid and DHA content in dry cells | [58] |

**Table 4.** *Cont.*

| Source | Fermentation Conditions | Bioactive Compounds | Outcome | Ref. |
|---|---|---|---|---|
| Exhausted olive pomace | 25 g substrate/L, *Crypthecodinium cohnii* ATCC 30772, 27 °C, 160 rpm, and 5 days | Fatty acids | High production yield; negative effect of detoxification prior to fermentation | [80] |
| Chokeberry pomace | 40 g substrate, *Aspergillus niger* ATCC-6275 or *Rhizopus oligosporus* ATCC-22959, 30 °C, and 12 days | Polyphenols | TPC: maximum at 6 days for or *Rhizopus oligosporus* and 9 days for *Aspergillus niger*; DPPH and TEAC: maximum at 6 days for *Aspergillus niger* and 9 days for *Rhizopus oligosporus* | [81] |
| Plum pomace | 15 g substrate, *Aspergillus niger* ATCC-6275 or *Rhizopus oligosporus* ATCC-22959, 30 °C, and 14 days | Polyphenols | TPC: maximum after 9 days of fermentation; DPPH: maximum at 6 days of fermentation | [82] |
| Apricot pomace | 15 g substrate, *Aspergillus niger* ATCC-6275 or *Rhizopus oligosporus* ATCC-22959, 30 °C, and 14 days | Polyphenols | TPC: maximum at 9 days for *Rhizopus oligosporus*; reduced after 6 days for *Aspergillus niger*; DPPH: maximum at 2 days for both | [83] |
| Pitahaya pomace | 2 g substrate, *Rhizomucor miehei* NRRL 5282, 37 °C, and 18 days | Polyphenols | Oven dried: slight decrease in TPC, decreased FRAP, and no effect in DPPH Lyophilized: slight increase in TPC, maximum FRAP value at day 10 and DPPH value at day 15 | [72] |
| Red bayberry pomace | 0.02% live yeast, 25 °C, 16 h followed by 0.1% probiotic mix, 28 °C, 24 h, and let for up to 7 days | Polyphenols | Increased TPC and TFC values; reduced DPPH value | [84] |

DHA: Docosahexanoic acid, DPPH: (2,2-diphenyl-1-picrylhydrazyl) free radical, FRAP: ferric reducing antioxidant power, ORAC: oxygen radical absorbance capacity, TEAC: trolox equivalent antioxidant capacity, TFC: total flavonoid content, and TPC: total polyphenol content.

For instance, studies carried out with grape pomace indicate that polyphenols [53,72], γ-linolenic acid and carotenoids [73] can be obtained from fermentation. In addition to the characterization of the content of these bioactive compounds, these studies also revealed aspects related to the preparation of samples, fermentation period, and the effect of the starter culture.

Regarding the effects of sample preparation and fermentation period in the release of polyphenols, a recent experiment indicated that lyophilization is a better pre-treatment than oven-drying to improve the extraction of polyphenols from grape pomace [72]. Moreover, this study also indicated that long fermentation periods do not favor the accumulation of polyphenols. Additionally, this effect could be explained by the instability of free polyphenols during fermentation. The gradual decomposition of free polyphenols can occur, which may be compensated by the release of bound polyphenols from microbial activity. Another related study with pomace supports this consideration and the necessity to define the optimum fermentation period. The high polyphenol and bioactivity in the beginning of the fermentation period were followed by the reduction in both indicators (polyphenol content and biological activity) as fermentation progressed up to 15 days [74]. Additionally, Teles et al. [53] reported increasing polyphenol content and antioxidant activity during the fermentation of grape pomace with *Aspergillus niger* during a shorter period (96 h) in relation to these aforementioned studies. This study also indicated that polyphenol content was positively correlated with antioxidant potential.

The production of γ-linolenic acid and carotenoids by solid-state fermentation also displayed the same dependency on fermentation time, wherein maximum yields were obtained after 6 days of fermentation [73]. In the case of carotenoids, the synthesis of lutein had a maximum yield after 8 days, whereas the production of carotene increased throughout the fermentation period (18 days).

Apple pomace has also been explored as a relevant source of polyphenols and fatty acids. For instance, the effect of pre-treatment and fermentation on polyphenol accumulation during fermentation was studied by Zambrano et al. [72]. The maximum polyphenol content was not affected by the pretreatment (lyophilization vs. over-drying), but significant changes were reported during the fermentation period. The maximum polyphenol yield and antioxidant potential were obtained at day 10. Conversely, Lohani and Muthukumarappan [75] reported a gradual reduction in the polyphenol content of naturally fermented apple pomace. Madrera et al. [76] reported a slight reduction in the polyphenol content of fermented apple pomace with different yeasts. Additionally, this study also indicated that the production of fatty acids can be obtained from the fermentation of apple

pomace with yeasts. An interesting experiment with apple pomace explored the production of fatty acids in a 5 L bioreactor [77]. In this case, different concentrations of apple pomace were used as a carbon source for lipid biosynthesis. A concentration-dependent effect (40, 60 and 80 g substrate/L) in the production of fatty acids was reported. Moreover, the maximum yield for each tested apple pomace concentration was achieved in a short period (3 days).

In the case of olive pomace, the fermentation with *Kluyveromyces marxianus* led to a reduction in tannic acid content and an increase in the concentration of its depolymerized form, gallic acid [79]. Another interesting application of exhausted olive pomace (residue obtained after the removal of residual oil from olive pomace) is the production of microbial fatty acids, especially docosahexaenoic acid (DHA). A recent experiment indicated that the concentration of exhausted olive pomace had a concentration-dependent effect in the production of DHA by the microalgae *Crypthecodinium cohnii* [58]. Interestingly, another study with the same microalga revealed that detoxification with activated carbon reduced the production of fatty acids [80].

The simultaneous production of polyphenols and fatty acids from fruit pomace was also explored in a recent study with two *Sambucus* species [78]. In these fruits, optimum polyphenol production yield and antioxidant activity were obtained at day 3 and 4 (regardless of species), respectively. A similar effect was observed for the accumulation of linoleic and oleic fatty acids, which had maximum values at day 4. Similarly, the accumulation of polyphenols and antioxidant activity during the fermentation of chokeberry pomace were dependent on the time and starter culture [81]. Maximum values for total phenolic content were obtained between day 6 and 9 of fermentation for *Rhizopus oligosporus* and 9 days for *Aspergillus niger*.

Studies carried out with plum [82] and apricot [83] pomaces indicated that optimum fermentation periods for polyphenol accumulation and antioxidant activity from *Aspergillus niger* fermentation were 9 and 6 days, respectively. Another recent experiment indicated that the accumulation of polyphenols in pitahaya pomace from the activity of *Rhizomucor miehei* was improved by lyophilizing samples before fermentation [72]. A related experiment evaluated the accumulation of polyphenols and antioxidant activity in red bayberry pomace during 7 days during the sequential fermentation with *Saccharomyces cerevisiae* and a mix of lactic acid bacteria (*Lactobacillus bulgaricus*, *Bifidobacterium lactis*, and other lactic acid bacteria) [84]. A gradual increase in the polyphenol content was reported throughout the 7 days of fermentation. Moreover, the antioxidant activity of fermented pomace after this period was improved in relation to non-fermented pomace.

Since the fermentation of pomaces can lead to high polyphenol content and antioxidant activity (Table 4), the biological response to the consumption of fermented pomace was also explored in recent studies. Improvements in the antioxidant defense system and a reduction in the oxidative status of liver and ilium in mice fed with fermented blueberry pomace were reported [85]. The intestine inflammatory response (tumor necrosis factor-alpha and interleukin-10) was also improved in animals that consumed the diet supplemented with fermented blueberry pomace. Concentration-dependent effects were observed in the antioxidant and anti-inflammatory activities. Moreover, these effects ameliorated the modifications induced by a high-fat diet in terms of antioxidant and anti-inflammatory responses.

A further experiment carried out by the same research group explored the functional effect of fermented blueberry pomace in indicators of gut health of mice [86]. The consumption of supplemented diets improved the gut immunological response (secretory immunoglobulin A), affected the gut microbiota and also favored the production of butyric acid (a short fatty acid associated with health benefits). Again, the supplemented diet ameliorated the modifications induced by a high-fat diet in the gut immunological response and gut health. Another experiment in vivo that supports the health benefits associated with the fermentation of pomaces was carried out by Yan et al. [87]. In this case, the consumption of fermented blueberry pomace (rich in polyphenols) improved the resistance to fatigue in relation to control animals that ingested sterile water.

Along with the production of fermented pomaces with increased biological activity, it is also important to develop strategies to isolate active components from the bulk of fermented pomace. This aspect was recently explored by Espinosa-Pardo et al. [88] who optimized the extraction of polyphenols with super-critical $CO_2$ and co-solvents. The authors indicated that the extraction with $CO_2$ (25 MPa at 60 °C) and 90% ethanol as co-solvent was the most efficient extraction condition to obtain the highest polyphenol content and antioxidant activity. Another important aspect to consider is the effect of digestion in the stability of active compounds. Yan et al. [87] evaluated the impact of simulated digestion and indicated significant reduction in the polyphenol content and antioxidant activity of blueberry pomace fermented by *Lactobacillus rhamnosus* GG and *Lactobacillus plantarum*-1 (1:1).

The fermentation of pomaces can be seen as a relevant strategy to produce functional supplements with interesting biological effects, especially from berries. However, additional advances, especially in the application of extraction technologies and the characterization of biological effects in vivo, are still necessary.

## 5. Production of Organic Acids

Organic acids are multipurpose compounds that have been applied in animal production [89], food processing [90], cosmetic preservation [91] and battery recycling [92], for instance. Due to their importance and the current trends to improve sustainability within the organic acid production sector, several studies have been carried out to explore the use of pomaces in the production of high-added value compounds (Table 5). One relevant example of this strategy is the study performed by Vashisht et al. [93] who evaluated the production of acetic acid using *Acetobacter pasteurianus* SKYAA25 from apple pomace. These authors indicated that the production of acetic acid was affected by the temperature (37 °C), concentration of bioethanol (8%, produced from the same strain), and apple pomace (2%) in fermentation media. Similarly, the production of acetic acid from the fermentation of apple pomace was reported in another study using *Acetobacter aceti* [94].

**Table 5.** Organic acids produced from pomace fermentation.

| Source | Fermentation Conditions | Organic Acid and Yield | Ref. |
|---|---|---|---|
| Apple pomace | 120 g substrate/L, *Acetobacter pasteurianus*, 37 °C, 180 rpm, and 24 h | Acetic acid: 52.4 g/100 g DM | [93] |
| Apple pomace | 1.5 L of substrate, *Acetobacter aceti*, pH 7.0, 28 °C, and 7 days | Acetic acid: 61.4 g/100 g DM | [94] |
| Apple pomace | 14 g substrate/100 g, *Propionibacterium freudenreichii*, 37 °C, and 120 h | Propionic acid: 38 g/100 g DM | [95] |
| Apple pomace | 250 mL substrate, *Propionibacterium freudenreichii*, 37 °C, and 120 h | Acetic acid: 5.01 g/L<br>Propionic acid: 14.54 g/L | [96] |
| Apple pomace | 25 g substrate, *Aspergillus ornatus* and *Alternaria alternate*, pH 5.0, 30 °C, and 48 h | Citric acid: 0.5 g/L | [97] |
| Apple pomace | 25 g substrate, *Rhizopus oryzae*, 30 °C, and 14 days | Fumaric acid: 52 g/kg | [98] |
| Apple pomace | 3–4 L working volume, 50% moisture, *Rhizopus oryzae*, 1.97 atm, and 14 days | Fumaric acid: 138 g/kg | [99] |

Piwowarek et al. [95] studied the optimization of the production of propionic acid from apple pomace fermentation with *Propionibacterium freudenreichii* T82. According to these authors, the accumulation of propionic acid was increased due to a better control of the fermentation process, i.e., adding biotin to fermentation media, carrying out the pH control at 24 and 48 h of fermentation, and increasing the nitrogen level (supplementing the apple pomace with peptone). However, no significant effects were obtained for the variations in temperatures from 30 to 37 °C. In another study from the same research

group with apple pomace, the effect of supplementation (potato wastewater, yeast extract, and peptone) to increase the production yield of propionic acid was evaluated [96]. The use of yeast extract and peptone in apple pomace in the fermentation medium improved the propionic acid yield to a maximum of 14.54 g/L after 120 h of fermentation with *Propionibacterium freudenreichii*. Additionally, the production of acetic acid was also evaluated in this study. A continuous increase in the accumulation of this acid was reported until the end of the fermentation period (120 h) and the most efficient supplement for apple pomace was potato wastewater (maximum yield of 5.01 g/L).

Apple pomace can also be fermented to produce citric acid [97]. In this case, a recent experiment explored the effect of temperature, pH, and substrate amount in the fermentation batch with the combination of *Aspergillus ornatus* and *Alternaria alternate*. The pH and temperature had optimum values of 5 and 30 °C, respectively. Increasing the substrate caused a significant increase in the production of citric acid, which led to choosing the maximum substrate amount tested in this study (25 g). Additionally, supplementing the apple pomace with arginine favored the production of citric acid (maximum yield of 2.7 g/L).

Another relevant acid produced from pomaces is the fumaric acid. The production of this acid with *Rhizopus oryzae* was dependent on the fermentation time [98]. The maximum yield was reported after 14 days (52 g/kg) and no additional increase was observed at up to 21 days of fermentation. The production of fumaric acid using the same microorganism and pomace was also explored in a bench scale fermenter [99]. The system comprised by a rotary drum increased the production of fumaric acid to 138 g/kg within the same fermentation period (14 days).

## 6. Production of Bioflavors

The production of high-added value compounds from pomaces has also been shown to produce bioflavors. The production of aromas from apple pomace fermentation was explored in a recent experiment with yeasts (*Hanseniaspora uvarum*, *Hanseniaspora valbyensis*, and *Saccharomyces cerevisiae*) [100]. This study indicated a strain-dependent effect in the formation of volatile compounds wherein the use of *Saccharomyces cerevisiae* led to a bigger accumulation of volatile fatty acids and their respective ethyl esters, whereas the fermentation with *Hanseniaspora* strains favored the generation of volatile acetic acid esters. A related study evaluated the effect of fermented pomace in a volatile composition of beer [101]. In this case, apple pomace was fermented with lactic acid bacteria (*Lactobacillus rhamnosus* 1473 and 1019, and *Lactobacillus casei* 2246) and significant differences were reported among volatile compositions of apple pomace. However, the fermented pomace (*Lactobacillus rhamnosus* 1473) led to slight modifications in the volatile composition (particularly for ketones and alcohols) of beer. The production of bioflavors was also explored using *Lacticaseibacillus rhamnosus* to ferment orange pomace [102]. This study revealed that fermented pomace had floral (citronellyl formate, 1-nonanol, and β-linalool), citrus (citral and limonene), fruity (β-cyclocitral and benzaldehyde), herbaceous (1-hexanol), bready and caramelly (furfural), and spice (eugenol and carveol) notes.

Finally, another aspect to be considered in the context of the utilization of high-added value compounds obtained from the fermentation/biotransformation of pomaces is their safety. Mycotoxins and pesticides are relevant contaminants in the peels of fruits that may persist in pomaces [103–105]. The effect of fermentation to decontaminate fruits and pomaces is still poorly studied.

## 7. Conclusions

The use of fermentation/biotransformation to obtain high-added value compounds is a valuable solution to improve the reutilization of pomaces from the food industry. The advances in incorporating and optimizing the use of pomaces in animal feed by generating silages and feeds that improve animal health is a relevant alternative to using fermented pomaces. Growth performance can be affected, whereas animal health status can be

improved. The absence of negative effects and the improvement in the nutritional quality of the foods obtained from animals fed with fermented pomaces is another favorable characteristic to support this strategy.

In terms of industrial processes, the production of high-added value products (especially from grape, apple, and olive) such as enzymes and organic acids for application in food processing as well as in other areas is a relevant application. The release of bound phenolics for the development of functional foods (supported by studies in vitro and in vivo), the synthesis of carotenes and fatty acids, and the production of volatile compounds to improve the aroma of food products are potential applications.

One of the main limitations in terms of industrial application consists of its current poor incorporation into other processing chains. Extraction and purification technologies can be seen as current bottlenecks to strengthening the connections between the pomace generation in food industries and their incorporation into other productions chains. In this sense, further studies could aim to explore strategies to improve the isolation of high-added value compounds. Additional studies are still necessary to define strategies to apply the high-added value compounds obtained from pomaces from fermentation/biotransformation in the development of food products. Studies about the detoxification and reduction of potential health risks associated with mycotoxins and pesticides are necessary.

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