

Review

# Citrus and Winery Wastes: Promising Dietary Supplements for Sustainable Ruminant Animal Nutrition, Health, Production, and Meat Quality

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**Abstract:** Citrus and grapes are the most widely grown fruits globally, with one-third of total production used for juice and wine making. The juice and winemaking processes generate large quantities of solid organic wastes including citrus pulp and grape pomace. These fruit wastes pose serious economic, environmental, and social challenges, especially in low-to-middle-income countries due to financial, technological, and infrastructural limitations. They are, however, rich in valuable compounds which can be utilized in the ruminant livestock industry as novel, economical, and natural sources of cellulose, polyunsaturated fatty acids, and phytochemicals, which have nutritional, anthelmintic, antioxidant, and antimicrobial properties. Despite citrus and grape fruit wastes having such potential, they remain underexploited by the livestock industry in low-to-middle-income countries owing to lack of finance, skills, technology, and infrastructure. Inclusion of these fruit wastes in ruminant diets could combine the desirable effects of enhancing animal nutrition, health, welfare, production, and meat quality attributes with the prevention of challenges associated with their disposal into the environment. The current review explores the valorization potential of citrus and winery wastes as dietary supplements to sustainably enhance ruminant animal nutrition, health, welfare, production, and meat quality.

**Keywords:** fruit wastes; sustainability; ruminants; natural bioactive compounds; valorization

## 1. Introduction

Ruminant animals in low-to-middle-income countries far outperform all other domestic animals not only as a high-quality source for the human world food system, but also regarding other issues like being a source of fiber, energy/power, fertilizer, employment, income, capital accumulation, export earnings, and by-products and also having social and cultural significance [1]. In addition to supporting over 600 million smallholder farmers, livestock is an important risk aversion strategy for these resource-poor households in low-to-middle-income countries [1]. The global importance of ruminants and their products is increasing as consumer demand in the low-to-middle-income countries expands with population growth and rising incomes [1–3]. In that regard, it is estimated that at least 60 to 70% more ruminant livestock products should be produced to feed the population which is predicted to grow from 7.3 billion in 2015 to 9.5 billion in 2050, and most of this increase will be from low-to-middle-income countries [1,2]. Overall, there is a global need to sustainably increase ruminant production in dwindling arable areas and rangelands [1]. This must be achieved while minimizing the negative effects of livestock agriculture on the environment and, in the face of climate change, extreme weather events and threats from emerging diseases and parasites [1,2].

Ruminant livestock production in low-to-middle-income countries, particularly in the tropics, suffers greater setbacks compared to in high-income countries, mostly owing to the available feed

resources [1,4]. The growing demand for animal products in most low-to-middle-income countries imposes huge demand on the available feed resources [1,4]. In response to this demand, large amounts of feed resources will be needed, compromising the sustainability of the current feed production systems [1,3]. On one hand, the availability of rangelands for ruminants has become limited, owing to the growing human population, urbanization, industrialization, and rising demand for utilization of agricultural land for crop production [1,2]. On the other hand, climate change is limiting the productivity and grazing capacity of rangelands, which negatively affects animal health, welfare, and production [1]. Besides these preharvest losses, ruminant livestock production in low-to-middle-income countries is further challenged by severe postharvest losses caused by meat discoloration, rancidity, and microbial spoilage [5]. To improve livestock nutrition, health, welfare, and meat shelf life, the livestock industry often relies on extensive usage of synthetic chemicals including nutritional supplements, antiparasitics, antimicrobials, and antioxidants [6,7]. Cases of chemical residues in meat, resistance to antiparasitic and antimicrobial drugs, and negative human health effects from these synthetic products are, however, increasing [7,8]. In addition, the affordability and accessibility of these compounds is a challenge among many smallholder farmers in low-to-middle-income countries due to their low socio-economic status [1]. Correspondingly, there is a paradigm shift towards the search for more “natural” and sustainable ways of managing animal nutrition, health, welfare, production, and meat healthfulness, safety, and shelf life [7].

Low-to-middle-income countries are currently contributing about 51% to global total fruit production [2]. Of this global production, citrus and grapes are economically the most important fruit crops with approximately more than 100 and 80 million metric tons produced annually, respectively [2,9]. As a result, large quantities of citrus pulp (14.4 million metric tons) and grape pomace (9 million metric tons) are generated per annum with little economic interest and are posing severe storage, processing, and disposal problems including landfill gas and leachate generation, which is subjected to stern regulations and presents a cost for the fruit processing industry [3]. The recent characterization of unutilized and discarded fractions of the fruit wastes indicates their potential candidature for processing and value addition [10]. In this regard, wastes from citrus and winery fruits have been shown to be rich sources of natural cellulose, minerals, polyunsaturated fatty acids (PUFAs), and phytochemicals, which have nutritional, anthelmintic, antioxidant, and antimicrobial properties [11,12]. These wastes, henceforth referred to as “by-products”, can therefore be adopted in the citrus- and grape-producing low-to-middle-income countries as a strategy of economic advantage in ruminant diets as they lower feed shortages and costs and enhance animal nutrition, health, welfare, production, meat fatty acid profile, and shelf life. In addition, the utilization of citrus and winery by-products as dietary supplements can produce meat with human-health-promoting properties [6,7] and help mitigate sustainability challenges that would arise from their disposal [7]. The current review therefore explores the potential of utilizing citrus and winery by-products as dietary supplements for sustainable improvement of ruminant animal nutrition, health, welfare, production, and meat quality.

## 2. Nutrient and Bioactive Profiles of Citrus and Winery By-Products

### 2.1. Nutrient Composition of Citrus Pulp and Grape Pomace

Citrus pulp is the solid residue that remains after extraction of juice from fresh fruits. It contributes 50–70% of the fresh weight of the original fruit and is made up of the peel (60–65%), internal tissues (30–35%), and seeds (0–10%) [9]. The genus citrus includes several important fruits, with the most important on a worldwide basis being sweet orange (*C. sinensis*, 67.8% of world citrus production), tangerine (*C. reticulata*, 17.9%), lemon (*C. limon*, 6.3%), and grapefruit (*C. paradise*, 5.0%) [13]. Grape pomace comprises stalks (2%), seeds (47%), skin, and pulp (51%) on a dry matter basis [14]. Globally, Sauvignon Blanc and Shiraz (Syrah) are among the most economically important cultivars of *Vitis vinifera*, which is the predominant species in genus *Vitis*.

Citrus pulp contains up to 105 g/kg dry matter (DM) crude protein (CP), while grape pomace contains up to 123 g/kg DM CP (Table 1). The CP content of these fruit by-products is within the recommended metabolizable protein requirements for maintenance (60–80 g/kg DM) of ruminants [15]. Furthermore, grape pomace has high ether extract (EE) in g/kg DM compared with citrus pulp (Table 1). The ether extract levels of these fruit by-products are within the recommended range (<80 g/kg DM) for ruminants [15]. In terms of neutral detergent fiber (NDF), citrus pulp has lower values than grape pomace (Table 1), but values for both fruit by-products lie within the recommended range of 170–330 g/kg DM for ruminants [15]. In contrast, citrus pulp has higher content of water-soluble sugars than does grape pomace (Table 1). Recent studies done by Gobindram [16] and Winkler [17] reported high metabolizable energy values for grape pomace compared to citrus pulp. However, both values were within the recommended range of 361–556.5 Kilojoules (KJ) metabolizable energy (ME)/kg of metabolic live-weight required for maintenance of ruminants [18]. The lignin content in grape pomace is higher than that in citrus pulp (Table 1) and is above the 40 g/kg DM suggested as the threshold level at which DM intake and digestibility in ruminants could be depressed [19]. The calcium, phosphorus, and magnesium content of citrus pulp and grape pomace are within the recommended maintenance requirements for ruminants of 15.4 mg Ca/kg, 16 mg P/kg, and 12–16 mg Mg/kg body weight, respectively [18]. The levels of the most limiting amino acids for ruminants, methionine and cysteine, in citrus pulp and grape pomace do not meet the required levels for maintenance [18]. Use of citrus pulp and grape pomace in ruminant diets therefore requires supplementation of the diet with amino acids or use of amino-acid-rich ingredients to complement these fruit by-products. Overall, the observed variation in chemical composition reported in Table 1 can be explained by fruit origin, environmental factors, production techniques and cultivation conditions, dehydration methods, extraction procedures, and fruit cultivar. The nutritional composition variation of these fruit by-products implies that recommendations for inclusion level in ruminant diets can only be made after chemical characterization.

**Table 1.** Chemical composition of citrus and winery by-products.

Chemical Composition (g/kg)	Citrus Pulp <sup>a</sup>			Grape Pomace <sup>b</sup>		
	Min	Max	Mean ± SD	Min	Max	Mean ± SD
Dry matter	858	955	910 ± 5.0	351	955	918 ± 0.1
Organic matter	934	955	928 ± 3.1	866	938	910 ± 0.3
Ash	46.3	134	69.0 ± 1.40	33.0	134	60 ± 0.68
Crude protein	64.9	105	70.0 ± 1.50	54.0	123	115 ± 0.5
Ether extract	27.0	58.0	37.0 ± 2.00	52.0	71.0	68.0 ± 0.65
Neutral detergent fiber	155	387	242 ± 10.6	376	630	322 ± 2.5
Neutral detergent solubles	613	845	729 ± 10.2	373	744	678 ± 3.16
Acid detergent fiber	100	307	222 ± 10.4	317	550	326 ± 2.9
Acid detergent lignin	21.0	25.0	22.0 ± 4.90	161	446	197 ± 2.8
Hemicellulose	55.0	287	183 ± 4.2	59.0	313	208 ± 2.1
Cellulose	128	128	128 ± 3.1	540	540	540 ± 3.4
Metabolizable energy (MJ/kg)	105	128	119 ± 2.4	58.0	130	88.6 ± 5.31
<b>Mineral composition (g/kg)</b>						
Calcium	4.90	22.4	17.0 ± 2.30	2.29	6.10	3.0 ± 0.05
Phosphorus	0.70	1.50	1.0 ± 0.20	0.60	3.10	2.6 ± 0.05
Potassium	6.60	11.6	9.3 ± 1.30	15.0	24.1	20.4 ± 0.41
Magnesium	1.00	2.10	1.3 ± 0.40	1.00	1.20	1.1 ± 0.03
Sulphur	0.80	1.20	0.8 ± 0.20	1.02	1.35	1.2 ± 0.02
Sodium	0.30	4.00	1.2 ± 1.30	0.10	0.90	0.7 ± 2.42
Zinc (mg/kg)	6.00	57.0	14 ± 14.0	7.30	14.6	11.3 ± 0.34
Manganese (mg/kg)	5.00	14.0	8.0 ± 3.00	14.2	21.0	16.8 ± 0.41
Iron (mg/kg)	46.0	170	80 ± 32.0	115	147	126 ± 2.9
Copper (mg/kg)	3.00	6.30	4.5 ± 1.00	7.30	13.4	9.8 ± 0.31

<sup>a</sup> Reported mean values were for *C. Sinensis* and *C. Reticulate Blanco* cultivars in Egypt, Brazil, Morocco; USA, Turkey, Pakistan, and South Africa; <sup>b</sup> Reported mean values were for *Pinotage*, *Shiraz*, and *Blanc* cultivars in South Africa, Spain, Italy, Iran, and Chile; SD means standard deviation, Organic matter = 100 – Total Ash; Neutral Detergent Solubles = 100 – Neutral detergent fiber; Hemicellulose = Neutral detergent fiber – Acid detergent fiber. Sources: [3,8,13,16,19–22].

## 2.2. Bioactive Compounds in Citrus and Winery By-Products

### 2.2.1. Citrus Pulp

Citrus pulp has high proportions of PUFAs, especially linoleic acid (15–45% of total fatty acids) [23]. There is, however, scant data on the fatty acid composition of citrus pulp; hence, further research is recommended. Citrus fruits and their by-products are rich sources of bioactive compounds such as phenolics, flavonoids, and ascorbic acid. The total phenolic and flavonoid contents for citrus pulp range from 8.25 to 397 mg gallic acid equivalent (GAE)/g of extract [24,25] and 0.3 to 31.1 mg quercetin acid equivalent/g of powder [24], respectively. Ascorbic acid levels in citrus pulp range from 18.2 to 46.2 mg/100 mL [26]. Overall, ascorbic acid and carotenoid contents of citrus fruits meet the recommended requirements for ruminants [12].

### 2.2.2. Grape Pomace

Grape pomace has elevated proportions of PUFAs, particularly linoleic acid (55–75%) [27]. Further studies are, however, recommended since there is limited information on the fatty acid composition of grape pomace. Grapes contain high levels of polyphenols with as much as 70% retained in the pomace after the extraction of winery juice [11]. The main polyphenols in grape pomace are phenolic acids, flavan-3-ols, flavonols, anthocyanins, and proanthocyanidins. Total phenolic and flavonoid contents among cultivars range from 55.5 to 153.8 mg GAE/g and 38.9 to 91.7 mg rutin equivalent/g, respectively [11]. Total tannin, monomeric anthocyanin, and proanthocyanidin contents in grape pomace extracts range from 54 to 152.2 mg GAE/g, 0.02 to 11.2 mg cyanidin-3-glucoside equivalent/g, and 21 to 51.7%, respectively [11].

## 3. Influence of Feeding Citrus and Winery By-Products on Ruminant Animal Nutrition, Health, and Production

### 3.1. Influence on Dry Matter Intake

Feeding high levels (>150 g/kg DM) of citrus pulp and grape pomace has been reported to reduce dry matter intake (DMI) in ruminants [28–30]. The low DMI is attributed to the high fiber content and reduced palatability due to the sensation of astringency that proanthocyanidins confer on feed by binding with salivary proteins, giving an unpleasant feeling of dryness and harshness [31,32]. Recent studies have, however, shown increased DMI when the inclusion level of citrus and winery by-product is less than 150 g/kg DM in ruminant diets [30,33,34]. This increased intake for citrus and winery by-products might be due to their distinctive scent and taste and better palatability at lower inclusion levels [35].

### 3.2. Influence on Rumen Digestibility

Rumen digestibility of DM, organic matter (OM), CP, and NDF decreases with increasing levels of citrus pulp and grape pomace above 150 g/kg DM in ruminant diets (Table 2). This might be connected to a theory that polyphenols from these fruit by-products form complexes with natural polymers such as proteins and carbohydrates [36] and, therefore, may reduce their digestibility in the digestive tract of ruminants. The binding property of bioactive compounds from citrus and winery by-products such as proanthocyanidins, eugenol, and limonene results from a large number of free phenolic groups that form strong hydrogen bonds at numerous sites with proteins [37]. In addition, proanthocyanidins also form complexes with proteins through hydrophobic binding between the aromatic ring structure of tannins and hydrophobic regions of proteins [37].

In vivo results show that CP digestibility decreased with increasing levels of citrus and winery by-products in ruminant diets and the relationship is stronger than that of OM digestibility [20,38]. This suggests that polyphenols from citrus and winery by-products have a stronger interaction with proteins than do the other organic components in the diet, particularly fiber fractions. The greater

negative effects of dietary polyphenols such as proanthocyanidins on CP digestibility relative to fiber suggest that the effect of tannins on fiber digestion is a secondary effect [36]. Protein appears to have more possible binding sites with tannins than with fiber. This is because fiber appears to interact with tannins through only hydrogen bonds while protein may also form complexes with tannins through hydrophobic binding and covalent bonds [37]. It may be possible that proteolytic bacteria are more tannin sensitive than fibrolytic bacteria [37]. Only few studies have reported that dietary inclusion of 50–100 g/kg DM of citrus and winery by-products in ruminant diets enhances DM, OM, CP, NDF, and ADF digestibility [39,40]. In general, utilization of citrus and winery by-products at inclusion levels of up to 150 g/kg DM in ruminant diets could be useful in finishing ruminants without negative effects on nutrient digestibility.

In vitro and in vivo studies have reported that bioactive compounds from citrus and winery by-products such as polyphenols and essential oils protect the dietary PUFAs from biohydrogenation in the rumen, and/or suppress the growth and metabolism of rumen microbes responsible for biohydrogenation, particularly those involved in the last step, which is the conversion of vaccenic acid to stearic acid [29,41]. In this regard, selective inhibition of *Clostridium proteoclasticum* without influencing *Butyrivibrio fibriosolvens* results in more PUFAs and their biohydrogenation products, such as vaccenic and rumenic acids, bypassing rumen biohydrogenation and being subsequently incorporated into animal tissues [42]. High levels of citrus and winery by-products ( $\geq 200$  g/kg DM) in the diet seem to be more effective in modulating the fatty acid profile of ruminant meat [29,43]. Utilization of such high levels, however, has detrimental effects on DMI and nutrient digestibility.

### 3.3. Influence on Rumen Fermentation Parameters

Overall, rumen pH is not significantly affected by feeding citrus and winery by-products as sources of natural bioactive compounds (Table 3). Proanthocyanidins and essential oils lower the volume of ammonia nitrogen produced in the rumen, which improves assimilation of dietary amino acid by ruminants [44–46]. The decline in ammonia nitrogen concentration is usually accompanied by a decrease in the production of isoacids because of the reduction in degradation of dietary proteins [47]. Furthermore, the effects of bioactive compounds on rumen ammonia concentrations are probably related to a reduction in the protozoal population, which plays a major role in ruminal feed protein degradation [47]. Several studies have shown that ammonia nitrogen emanating from microbial protein degradation can be bound by polyphenols in a balanced chemical reaction regulated by the ammonia concentration to create a continuous supply of sufficient quantities of ammonia for microbial growth in the rumen [48].

**Table 2.** Effects of dietary fruit by-products on the nutrient digestibility of ruminants.

Fruit By-Product	Inclusion (g/kg)	Animal	DM <sup>a</sup> (g/kg)	OM <sup>b</sup> (g/kg)	CP <sup>c</sup> (g/kg)	NDF <sup>d</sup> (g/kg)	ADF <sup>e</sup> (g/kg)	References
Citrus pulp	25	Lambs	800	655	483	580	-	[40]
	50	Lambs	767	733	560	653	-	[40]
	75	Lambs	747	761	521	705	-	[40]
Citrus pulp	90	Cows	741	-	759	574	-	[49]
	180	Cows	754	-	765	576	-	[49]
Citrus pulp	1.25	Steers	530	540		480	-	[28]
	2.5	Steers	600	620		510	-	[28]
Citrus pulp	50	Calves	667	-	698	546	476	[38]
	100	Calves	654	-	696	541	462	[38]
	150	Calves	653	-	691	531	459	[38]
	200	Calves	652	-	690	525	451	[38]
Citrus pulp	60	Lambs	651	662	717	544	-	[50]
	143	Lambs	658	669	725	554	-	[50]
	218	Lambs	639	648	718	530	-	[50]
	265	Lambs	658	666	731	554	-	[50]
Citrus pulp	86.5	Steers	609	-	-	521	537	[51]
	72.8	Steers	673	-	-	574	607	[51]
	72.5	Steers	670	-	-	560	600	[51]
	82.5	Steers	636	-	-	539	561	[51]
Citrus pulp	100	Lambs	695	716	714	501	472	[20]
	200	Lambs	691	713	706	495	470	[20]
Citrus pulp	300	Lambs	681	705	703	488	465	[20]
	400	Lambs	678	704	692	471	461	[20]
Grape pomace	762	Lambs	453	510	345		343	[45]
	300	Wethers	680	690	750	320	500	[52]
Grape pomace	100	Steers	62.5	66.5	72.5	62.2	53.3	[53]

<sup>a</sup> DM—Dry matter, <sup>b</sup> OM—Organic matter, <sup>c</sup> CP—Crude protein, <sup>d</sup> NDF—Neutral detergent fiber, <sup>e</sup> ADF—Acid detergent fiber.

**Table 3.** The effects of citrus and winery by-products on rumen fermentation parameters.

Fruit By-Product	Inclusion (g/kg)	Animal	pH	VFA (mmol/L)	Acetic Acid <sup>a</sup>	Propionic Acid <sup>a</sup>	Butyric Acid <sup>a</sup>	NH <sub>3</sub> N (mg/dl)	References
Citrus pulp	477	Steers	6.40	131.0	0.64	0.14	0.15	135	[54]
	300	Steers	6.30	157	0.73	0.13	0.12	109	[54]
Citrus pulp	130	Lambs	6.53	75.4	0.64	0.18	0.14	5.69	[55]
	260	Lambs	6.57	72.8	0.64	0.20	0.12	6.09	[55]
Citrus pulp	390	Ewes	6.63	71.5	0.66	0.18	0.11	4.42	[55]
	300	Ewes	5.90	154	0.65	0.23	0.08	40.0	[46]
Grape pomace	100	Steers	6.37	154	0.59	0.21	0.11	124	[51]
	50	Steers	6.67	116	0.63	0.26	0.11	10.2	[53]
Grape pomace	76.2	Lambs	6.03	-	-	-	-	15.9	[45]
	20	Buffaloes	6.71	57.1	0.66	0.23	0.11	11.4	[56]
Grape pomace	40	Buffaloes	6.71	56.8	0.66	0.23	0.12	13.9	[56]
	60	Buffaloes	6.72	58.5	0.66	0.23	0.11	14.3	[56]
Grape pomace	150	Lambs	6.22	102	-	-	-	103	[57]
	300	Lambs	6.14	97.2	-	-	-	77.5	[57]
	450	Lambs	5.84	72.2	-	-	-	63.6	[57]

VFA means volatile fatty acid; <sup>a</sup> measured in mmol/L.

The inclusion of citrus and winery by-products in ruminant diets up to 150 kg DM improves the individual and total volatile fatty acid VFA profile (Table 3) by providing more lipogenic metabolizable nutrients [46,53,55]. This could be ascribed to a faster rate of feed intake and increased digestibility, since ruminal VFA production depends on the availability and utilization of substrates of rumen fermentation [47]. Therefore, addition of these by-products to concentrate diets will supply a greater amount of highly fermentable substrate to rumen microbes with a resulting increase in VFA concentration [43]. These fruit by-products have been reported to be rapidly and extensively degraded in the rumen due to their high concentration of pectins [13], which have high rumen degradability. However, reduced ruminal VFA concentrations have been reported in ruminants fed citrus and winery by-products [58]. This was attributed to low microbial activity and substrate degradation as a result of decreases in cellulolytic and total bacteria numbers in the rumen in response to proanthocyanidins in citrus and winery by-products [43]. The decline in VFA production may occur with changes in the quantities of main VFAs, such as an increase in acetate and a decrease in propionate [43].

### 3.4. Effects on Methane Production

Polyphenolic compounds from citrus and winery by-products have received attention for their capacity to decrease methane production when added to ruminant diets by suppressing the growth and activity of methanogens, such as *Methanobrevibacter* or *Methanomicrobium*, responsible for methanogenesis [59,60]. Dietary supplementation of essential oils from citrus and winery by-products such as eugenol and limonene in ruminant diets has been reported to directly inhibit methanogenic archaea and/or indirectly reduce methane production by directly suppressing some microbial metabolic processes contributing to methanogenesis [61]. Essential oils may also cause changes in the archaeal community structure and/or in the activity of the methanogenesis pathway, consequently diminishing methanogen abundance and methane production [62]. They may also reduce methanogenesis by lowering the abundance of some protozoa that are symbiotically associated with archaea and can contribute up to 37% to rumen methane production. Essential oils have been confirmed to be effective in reducing methane production in the rumen, with a reduction of up to 94% [48,62]. This raises the opportunity for supplementing ruminant diets with feed ingredients rich in phytochemical–nutrient complexes as a mechanism to provide high feeding value while reducing methane production.

### 3.5. Effects on Nitrogen Emissions

As previously reported, feeding citrus and winery by-products regularly results in a shift in nitrogen excretion from the urine to the feces [43,59]. Urinary nitrogen is deemed more detrimental to the environment than fecal nitrogen, particularly concerning volatilization of ammonia nitrogen and leaching to groundwater reserves [43]. As a result, lowering the proportion of urinary nitrogen and raising the fecal nitrogen with the use of bioactive compounds from citrus and winery by-products would allow for a decrease in ammonia nitrogen volatilization and would lessen environmental pollution. In that regard, further studies aimed at evaluating the effects of feeding citrus and winery by-products on nitrogen emission and the associated mechanisms could be important.

### 3.6. Effects on Nutritional Disorders

#### 3.6.1. Bloat

Based on in vitro and in vivo studies, the beneficial role of bioactive compounds from citrus and winery by-products on ruminant health and welfare is becoming a central issue in livestock production [8,43]. Studies have reported that citrus and winery by-products could prevent bloat in ruminants because of bioactive compounds [43]. The reduction in gas production and prevention of bloat can be explained by the ability of proanthocyanidins to precipitate proteins during chewing and rumination and reduce protein solubility in the rumen, thereby decreasing bloat occurrence [43].

This precipitation also decreases the incidence of bloat by lowering microbial activities, biofilm production, and ruminal gas production [37,43]. In addition, the interaction between polyphenols and protein can improve the flow of nitrogen leaving the rumen to the gut, enhancing protein utilization. The mechanism and ability of bioactive compounds from citrus and winery by-products such as polyphenols and essential oils, eugenol and limonene in particular, to decrease gas production and prevent bloat is not clear [37,43] and merits investigation.

### 3.6.2. Ruminal Parakeratosis and Acidosis

Inclusion of citrus and winery by-products below 200 g/kg DM in ruminant diets has been reported to reduce the occurrence of parakeratosis and acidosis in ruminants [13,63]. However, when elevated levels of citrus pulp were fed, along with low levels of forage, rumen parakeratosis and acidosis occurred in lambs [13]. Dietary inclusion of flavonoids from citrus and winery by-products such as quercetin in ruminant diets reduced the level of parakeratosis, which is an indicator of subacute ruminal acidosis [64]. Feeding either  $\alpha$ -tocopherol from citrus pulp or carsonic acids in ruminant diets corrected the metabolic acidosis in growing lambs [65]. Overall, there is dearth of information on how citrus and winery by-products influence rumen parakeratosis and acidosis, and further research is recommended.

## 3.7. Effects of Feeding Citrus and Winery By-Products on Animal Health and Welfare

### 3.7.1. Parasites

Polyphenols and essential oils from citrus and winery by-products exhibit anthelmintic activity both in vitro and in vivo [8,49,66]. An in vivo study showed a decline in the fecal egg counts (FEC) in goats and sheep offered elevated doses of essential oils rich in limonene and eugenol [67]. Furthermore, Squires [68] reported a decline of 97.4% in the FEC when inclusion levels of 95% of limonene and eugenol were administered at a dose of 600 mg/kg live weight. This decline in FEC can be attributed to the fact that limonene is toxic to bacteria and exhibits antioxidant effects in host animals. Limonene and eugenol tend to suppress cell growth and differentiation and, subsequently, embryogenesis of helminth eggs [67]. Intake of dehydrated citrus by-products by animals infected with *Haemonchus contortus* resulted in low hatching rates after 42 days of consumption, signifying a trend to shed less eggs to the environment [8].

The direct effects of citrus and winery by-products rich in proanthocyanidins and essential oils on nematodes include inhibition of eggs and infective larvae and reduction of larvae mobility [8,43]. Indirect effects include increasing protein availability, which strengthens the immune system, thus increasing the resistance to parasitic infections [8,43]. Furthermore, inclusion of grape pomace extract (30 and 60 g on kg fresh weight) in the diet of sheep with an intestinal parasitic infection was able to lessen the development of the infection, indicating a direct anthelmintic effect of the proanthocyanidins [43]. It can be assumed that polyphenols and essential oils (limonene and eugenol) from citrus and winery by-products have the ability to control nematodes and are potential sources of natural anthelmintic agents in ruminants. Further research to clarify the efficacy and mechanism of the action and toxicity to the host parasites and the potential to develop formulations with isolated compounds is warranted.

### 3.7.2. Effects on Oxidative Stress

Inclusion of citrus and winery by-products in ruminant diets has been reported to reduce the occurrence of oxidative stress [69,70]. This may be attributed to the fact that polyphenols act as antioxidants by scavenging free radicals and disrupting oxidative reactions, protecting cells from oxidative damage and decreasing the danger of diseases associated with oxidative stress [71,72]. They also suppress molecular signaling pathways that are stimulated by oxidative stress [72]. Furthermore, polyphenols from citrus and winery by-products are involved in the

suppression of reactive oxygen species (ROS) formation by either the inhibition of enzymes involved in their production, scavenging of ROS, or upregulation or protection of antioxidant defenses [72]. Polyphenolic antioxidants also reduce the catalytic activity of enzymes involved in ROS generation [69]. The elucidation of the molecular mechanisms involved in the improvement of animal redox status after administration of feeds supplemented with citrus and winery by products would help in the development of low-cost interventions for pathological conditions associated with oxidative stress. There is limited information on the effects of feeding citrus and winery by-products on oxidative stress in ruminants; thus, it merits investigation.

### 3.7.3. Effects on Immune System

Bioactive compounds such as flavonoids and proanthocyanidins from citrus and winery by-products have received attention for their ability to enhance and modulate the immune system of ruminants. The main effects of flavonoids on immune responses may be derived from their different mechanisms of action such as protein binding, active site interference, or antioxidant effects [73]. Several flavonoids specifically affect the function of enzymes involved in generating inflammatory responses [74]. Dietary flavonoids appear to moderate the inflammatory response and have primarily inhibitory effects on T-lymphocytes [43,73]. They modulate the cellular immune response in vitro, inhibiting the production of ROS by lymphocytes and granulocytes [43,73]. Phenolic compounds from citrus and winery by-products reduce the proliferation of lymphocytes and production of immunoglobins, and moderate the secretion of pro-inflammatory cytokines by myeloid cells [73]. Some flavonoids also alter immune response, which could be involved in immune surveillance of tumors. Quercetin suppresses antigenic stimulation of cytotoxic T-lymphocytes and inhibits natural-killer-cell-mediated cytotoxicity [73]. Proanthocyanidins have the ability to bind protein in the rumen, thereby making the protein unavailable for digestion and absorption until it reaches the more acidic abomasum. This high-quality protein bypass effect has the potential to enhance the immune response and increase resistance to internal parasites [73]. Bypassing amino acids like arginine, glutamine, and cysteine can boost immune responses as these amino acids regulate activation of T- and B-lymphocytes, natural killer cells and macrophages, gene expression and lymphocyte proliferation, and the production of antibodies, cytokines, and other cytotoxic substances [73]. Further research is required to elucidate the effects of feeding citrus and winery by-products on the immune system of ruminants.

### 3.8. Effects on Growth Performance and Carcass Traits

Recently, citrus and winery by-products have been widely used as an alternative feed and energy source in ruminant diets with no detrimental effects on animal growth [3]. Incorporation of 100 g/kg DM of citrus and winery by-products into ruminant diets was reported to enhance average daily gain and feed conversion efficiency (Table 4). This can be attributed to polyphenols and essential oils in citrus and winery by-products which protect proteins from rumen degradation, thereby increasing the amount of dietary protein reaching the small intestine for absorption [75]. These bioactive compounds improve the efficiency of conversion of dietary protein to animal protein, which subsequently improves the growth performance of ruminants [75]. This is supported by the studies by Zhao [34] and Ioannis [76], who reported that supplementation of ruminant diets with 100 g/kg DM of citrus and winery by-products increased body weight and average daily gain and reduced the feed-to-gain ratio (Table 4). At a high inclusion level (>150 g/kg DM), however, proanthocyanidins, eugenol, and limonene from these fruit by-products would impede feed intake due to their astringent nature. This will, in turn, reduce the digestion of protein and other nutrients by overprotecting dietary protein, decrease ruminal microbial activity, and reduce endogenous digestive enzyme activities, thereby adversely influencing the growth performance of ruminants [77]. It can be concluded that supplementing ruminant diets with up to 100 g/kg DM of citrus and winery by-products improves growth performance and reduces the feed-to-gain ratio of ruminants [78].

At present, there is limited information on the effects of feeding citrus and winery by-products on carcass traits of ruminant animals. In one study, Macias [79] reported an increase in carcass pH up to 45 min postmortem with the dietary inclusion of Ferulic acid from winery by-products in ruminant diets. Several studies have reported that carcass traits were not affected by dietary inclusion of citrus and winery by-products in ruminant diets, except for dressing percentage, which was reduced with the addition of these fruit by-products in ruminant diets [50,80–83]. In another trial, Kafantaris [76] observed that an inclusion level of up to 100 g/kg DM of citrus and winery by-products in ruminant diets increased slaughter, warm, and cold carcass weights and longissimus muscle area. This suggests that the inclusion of less than 100 g/kg DM of citrus and winery by-products in ruminant diets may have neutral to positive effects on ruminant livestock carcass attributes.

**Table 4.** Effects of citrus and winery by-products on the growth performance of ruminants.

Fruit By-Product	Inclusion (g/kg)	Animal	DMI (g/d)	ADG (g/d)	FCR	Reference
Citrus pulp	1.25	Steers	90.3	-	-	[28]
	2.5	Steers	87.3	-	-	[28]
Citrus pulp	240	Lambs	790	178	-	[29]
	350	Lambs	756	179	-	[29]
Citrus pulp	50	Calves	800	517	0.12	[38]
	100	Calves	814	528	0.12	[38]
	150	Calves	829	533	0.11	[38]
	200	Calves	300	539	0.11	[38]
Citrus pulp	300	Lambs	858	188	4.59	[83]
	450	Lambs	880	165	5.37	[83]
Citrus pulp	300	Lambs	928.9	197	4.70	[46]
Citrus pulp	150	Lambs	1230	289	4.31	[44]
	100	Steers	6130	1270	0.21	[33]
Citrus pulp	200	Steers	5960	1000	0.19	[33]
	100	Lambs	1350	67.5	0.05	[20]
	200	Lambs	1370	76.3	0.06	[20]
	300	Lambs	1390	71.7	0.05	[20]
	400	Lambs	1410	75.8	0.05	[20]
	50	Lambs	1.19	208	6.09	[39]
Grape pomace	100	Lambs	1.22	237	5.55	[39]
	150	Lambs	1.14	171	7.99	[39]
	200	Lambs	1.04	140	8.08	[39]
Grape pomace	50	Lambs	2512	283	-	[30]
Grape pomace	50	Lambs	2512	283	-	[30]
Grape pomace	100	Lambs	2495	258	-	[30]
Grape pomace	50	Rams	1379	1789	8.10	[34]
	100	Rams	1482	215	6.90	[34]
Grape pomace	100	Lambs	1142	120	0.10	[81]
	200	Lambs	1120	104	0.09	[81]
Grape pomace	300	Lambs	1240	107	0.08	[81]

DMI, dry matter intake; ADG, average daily gain; FCR, feed conversion ratio.

#### 4. Effects of Feeding Citrus and Winery By-Products on Meat Quality

##### 4.1. Physico-Chemical Meat Quality

Dietary supplementation of citrus and winery by-products in ruminant diets was reported to have no effect on pH, drip loss, cooking loss, or proximate composition of ruminant meat [44,82]. Moran [83] and Gomez [41] found higher water holding capacity when natural polyphenols from winery by-products were included in ruminant finishing diets. This was attributed to the effects of bioactive compounds from winery by-products which have the ability to avoid the loss of membrane integrity and protein cross-links by inhibiting and/or reducing the rate of oxidation in meat [41,84]. Dietary supplementation of citrus and winery by-products in ruminant diets increases meat redness ( $a^*$ ) in ruminants [34,82,85]. Overall, the mechanisms of how natural bioactive compounds in these fruit by-products affect meat redness are not clear but could be related to their ability to

chelate iron in myoglobin, making it non-bioavailable for oxidation [30], and/or the resistance of myoglobin to oxidation [6]. Supplementation of 100 g/kg DM of citrus and winery by-products in ruminant diets improved the instrumental tenderness of ruminant meat by lowering shear force values [34,44]. This reduction in shear force may be associated with the protection exerted by natural polyphenols from citrus and winery by-products against the oxidation of proteolytic enzymes during the ageing process [34,44].

#### 4.2. Effects on Fatty Acid Composition

Feeding citrus and winery by-products as dietary supplements was reported to increase the proportion of omega-3 and omega 6 PUFAs and their biohydrogenation products (Table 5). This could be ascribed to the selective inhibition by bioactive compounds of *Clostridium proteoclasticum* without influencing *Butyrivibrio fibriosolvens*, which results in greater bypass of PUFAs and their biohydrogenation products reaching the small intestines for subsequent absorption and deposition in the muscle [5,42]. The increase in percentages of PUFAs and their biohydrogenation products in ruminant meat varies with the level of citrus and winery by-products fed [29,41]. Overall, the proportion of PUFAs in ruminant meat has been reported to increase linearly with dietary levels of citrus and winery by-products up to 200 g/kg DMI, beyond which rumen function is impaired [42]. Besides increasing the proportion of PUFAs, especially those purported to have human-health-promoting properties such as *n*-3 and *n*-6 PUFAs, as well as rumenic and vaccenic acids [29,86], Table 5 shows that the addition of citrus and winery by-products below <150 kg tends to lower monounsaturated fatty acids (MUFAs) and saturated fatty acids (SFAs) in ruminants. Contrary to the above findings, other researchers [31,43,86] reported that dietary supplementation of citrus and winery by-products at levels lower than 150 g/kg DM in ruminant diets did not modify the fatty acid profile in longissimus muscle. Characterizing the individual and combined effects of specific phytochemicals in citrus and winery by-products on different biohydrogenation products could help to improve the proportions of human-health-promoting PUFAs in ruminant meat.

**Table 5.** Effects of citrus and winery by-products on the meat fatty acid profile of ruminants.

Fruit By-Product	Inclusion Level (g/kg)	SFA	VA	RA	(g/100 g) Fatty Acid				Reference
					MUFA	<i>n</i> -3	<i>n</i> -6	PUFA	
Grape pomace	100	40.45			37.5	2.17	13.7	16.4	[76]
Citrus pulp	250	36.5	1.39	1.08	30.9	5.57	20.3	26.9	[29]
Citrus pulp	350	35.9	1.56	1.02	32.5	5.48	20.4	26.9	[29]
Grape pomace	100	47.8	-	-	39.4	2.34	10.4	12.8	[86]
Grape seed	50	49.7	-	-	37.8	2.19	10.3	12.4	[86]
Grape seed	25	43.3	3.42	-	32.4	5.26	9.90	20.1	[32]
Grape pomace	50	67.8	4.76	0.78	17.1	1.01	5.09	8.94	[43]
Grape pomace	100	64.8	8.18	0.47	22.3	1.08	3.64	8.64	[43]
Citrus pulp	150	3.90	6.47	1.71	31.6	16.1	10.2	19.9	[44]
Grape pomace	50	36.6	2.10	1.00	34.9	5.3	6.61	28.4	[41]
Grape pomace	100	38.0	2.21	1.20	35.4	4.68	5.83	26.6	[41]

PUFA, polyunsaturated fatty acid; VA, vaccenic acid; RA, rumenic acid; MUFA, monounsaturated fatty acid; SFA, saturated fatty acid; *n*-6/*n*-3 is the ratio of omega 6 and omega 3 fatty acids.

#### 4.3. Effects on Sensory Quality

Regarding the use of citrus and winery by-products in ruminant diets, there are little if any studies that have investigated their effect on meat sensory attributes and volatile compounds. However, some bioactive compounds found in winery by-products, such as ferulic acid, have been reported to improve meat tenderness, juiciness, and flavor [87]. In contrast, Chaves [88] reported high off-flavor appreciation values in ovine meat supplemented with cinnamaldehyde and hesperidin, bioactive compounds present in citrus by-products. It is also important to note that citrus and winery by-products contain high levels of dietary polyphenols which are known to alter meat fatty acid composition [6,7]. The fatty acid composition of meat influences sensory quality attributes such as flavor, aroma, and juiciness [89]. In this regard, it is important to investigate the effects of citrus and winery by-products on ruminant meat sensory attributes and volatile compounds.

### 5. Effects of Feeding Citrus and Winery By-Products on Retail Meat Shelf Stability

#### 5.1. Myoglobin Oxidation

Feeding tanniferous-rich fruit by-products such as citrus and winery by-products is known to delay or inhibit meat discoloration during storage, thereby extending its shelf life [30,34,90]. This could be attributed to the positive effects of dietary bioactive compounds on heme pigment concentration and metmyoglobin formation during storage. Changes in redness ( $a^*$ ) values over time describe meat color deterioration from red to brown, and reflect the myoglobin concentration and its redox state in meat [85]. The oxidation of myoglobin and the subsequent buildup of metmyoglobin at the meat surface are the key causes of meat color deterioration [90]. The actual mechanism by which these bioactive compounds influence meat color stability is not clear. However, it has been hypothesized that citrus and winery polyphenolic compounds may affect meat color shelf life by augmenting muscle overall antioxidant capacity and myoglobin resistance to oxidation [6,90].

#### 5.2. Lipid Oxidation

Earlier reports show that dietary supplementation of tanniferous feeds in ruminant diets may protect oxidation of PUFAs in meat during retail display [6,30,34,90]. Thus, dietary supplementation of citrus and winery by-products in ruminant diets may reduce oxidative deterioration [34,90] and the formation of rancid odors, off-flavors, and toxic compounds harmful to human health. A decline in the production of meat lipid oxidation products can also decrease meat discoloration and nutrient losses [6]. In addition, citrus and winery by-products may moderate the formation of lipid oxidation products that influence protein solubility, emulsification, water binding capacity, texture, and other rheological properties via the interactions between lipid and protein products [90,91]. Overall, the mechanisms of the effect of natural bioactive compounds in citrus and winery by-products on lipid oxidation have not been fully explained. Nevertheless, phytochemicals are generally known to prevent lipid oxidation through numerous mechanisms, which include free radical scavengers, reducing agents, metal chelators, and single oxygen quenchers [6,34,90].

#### 5.3. Protein Oxidation

The rate and extent of protein oxidation in meat can be delayed, reduced, or prevented by feeding natural dietary antioxidants in ruminant diets, including citrus and winery by-products [91,92]. Phytochemicals present in these by-products contribute to the antioxidative power protecting proteins from oxidation [11,12]. Despite few studies devoted to the effects of dietary supplementation of citrus and winery by-products in ruminant diets on meat protein oxidation, animal models indicate the potential of natural bioactive compounds as efficient inhibitors of protein oxidation. These bioactive compounds reduce the destruction of amino acids to carbonyl compounds, which are known to affect protein functionality, solubility, and viscosity in meat [91–93]. The reduction of carbonyl formation

may positively improve some meat quality attributes, such as color, aroma, flavor, tenderness, water holding capacity, nutritional value, juiciness, and biological functionality.

#### 5.4. Microbial Growth

Several studies have shown that citrus and winery by-products have the ability to retard the growth of many foodborne pathogens including *Escherichia coli*, *Salmonella typhimurium*, *Enterobacteria*, *Pseudomonas*, *Listeria monocytogenes*, *Staphylococcus aureus* and *Brochothrix thermosphacta* [30,43,94]. Overall, the antimicrobial mechanisms of phenolic plant extracts manifest through attacking the phospholipid bilayer of the cell membrane, disrupting enzyme systems and compromising the genetic material of bacteria [95]. The chelating ability of phenolic compounds can also deprive microbes of essential iron required for growth [96]. Overall, there is little information on the effects of feeding citrus and winery by-products on the microbial shelf life of ruminant meat. Further studies to determine the effects of feeding fruit by-products to ruminants on meat microbiological quality are therefore warranted.

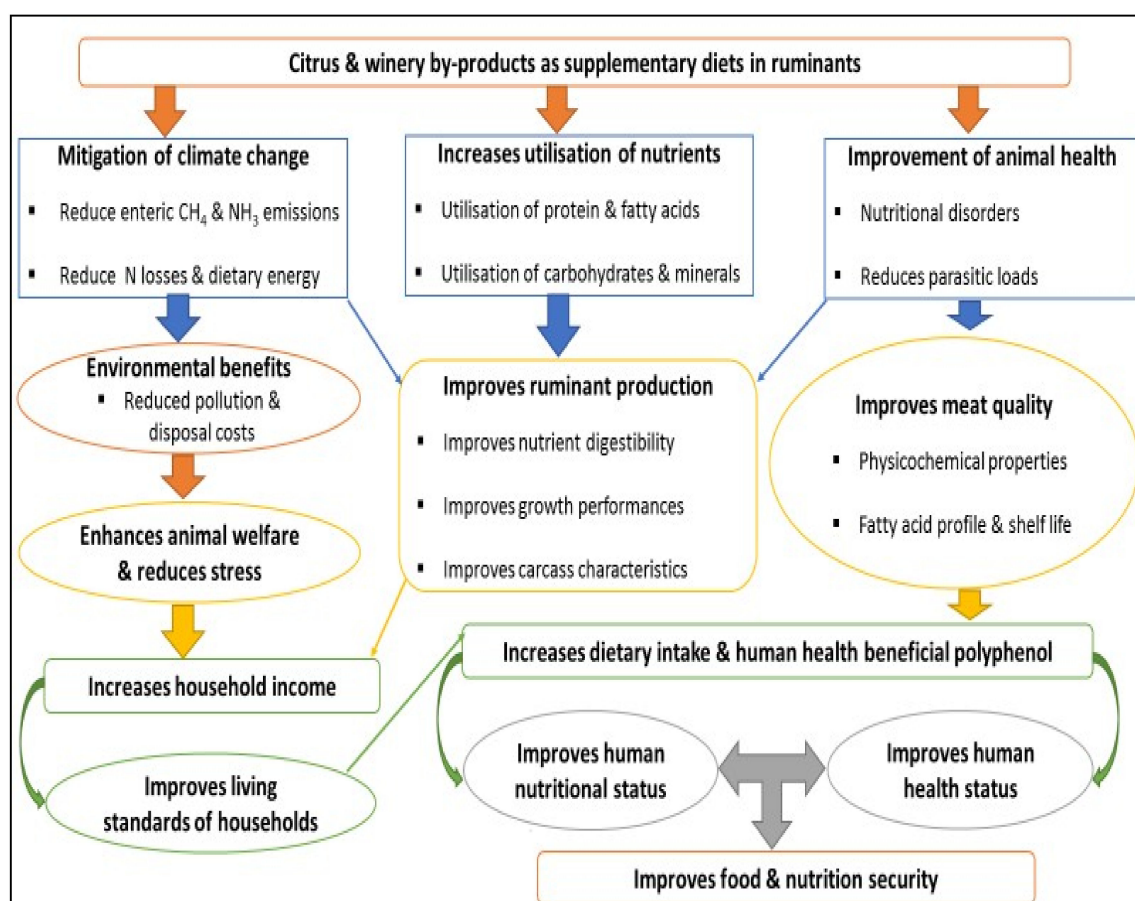
### 6. Potential Utilization of Citrus and Winery By-Products in Low-To-Middle-Income Countries and Future Perspectives

Valorization of citrus and winery by-products as supplementary feed ingredients for ruminant production holds great potential for smallholder farmers in low-to-middle-income countries producing these by-products. This is because the escalating demand for animal products, propelled by rising income, population, and urbanization in low-to-middle-income countries, imposes a huge demand on feed resources [3]. Sustainability of feed production systems is facing several challenges including land, soil, and water scarcity; food–feed–fiber–fuel competition; ongoing global warming and frequent and drastic climatic vagaries; and growing competition for arable land and nonrenewable resources such as fossil carbon sources, water, and phosphorus [3]. A key to sustainable ruminant production in low-to-middle-income countries is, therefore, the efficient utilization of locally available feed resources. These include reduction in wastage of organic content lost through the disposal of fruit by-products and broadening of the feed resource base through a quest for novel feed resources, especially fruit by-products which do not compete with human food.

Citrus and winery by-products have been shown to be rich sources of natural cellulose, minerals, vitamins, PUFAs, and phytochemicals, which have nutritional, anthelmintic, antioxidant, and antimicrobial properties [11,12]. In that regard, these fruit by-products can be adopted as a plan of economic advantage in ruminant diets since they reduce feeding costs and feed shortages, and improve animal nutrition, health, growth, and carcass characteristics, physico-chemical meat quality attributes, and fatty acid composition (Figure 1), thereby generating income which can improve the living standards of smallholder farmers in low-to-middle-income countries. However, utilization of citrus and winery by-products can be limited by the presence of pesticides; thus, it is important to regularly monitor such contaminants before incorporating them into ruminant diets [3]. Owing to their seasonality, bulkiness, and high moisture content (600 g/kg), which expedites microbial spoilage, oxidation of organic macromolecules, and degradation of bioactive compounds [97], these by-products also require preservation prior to utilization as animal feed. In that regard, simple and low-cost methods of practical handling such as dehydration and ensiling should be adopted to conserve these fruit by-products so that they can be fed to the livestock throughout the year or during periods of feed scarcity [3]. Given that ensiling losses are high and can have a negative impact on the farm environment, most studies recommend sun-drying because it is simple, inexpensive, and can be easily performed during the harvest season [27].

For the sustainable use of citrus and winery by-products, it is crucial to seek low-technology-based agro-processing and value-adding opportunities for the utilization of citrus and winery by-products, especially in low-to-middle-income countries. This is because of limited financial resources, lack of storage infrastructure and transport facilities, unstable or weak institutions, lack of information and

skills, and inequitable empowerment and access to resources required for high-technology-based agro-processing and value addition. Collaboration of large companies producing citrus juice and wine in low-to-middle-income countries with smallholder farmers, which could see them establishing small feedlots to utilize citrus and winery by-products as feed ingredients in ruminant diets, is recommended. In that context, it could be important to create a proper fruit by-product supply chain system to connect fruit processors, ruminant livestock farmers, abattoirs, and retailers and ensure the availability of quality meat at a competitive price to consumers. The use of citrus and winery by-products can be a catalyst for organizations to realize that valorization of wastes not only lowers costs and increases processing efficiencies but also decreases the environmental pollution load. In addition, these fruit by-products may significantly contribute towards food and nutrition security by minimizing or preventing discoloration, rancidity, and microbial spoilage; maintain or enhancing nutritional and sensory quality; and extending the shelf life of animal-based foods (Figure 1). This may subsequently improve the distribution of animal foods over long distances [5], which could help smallholder farmers to export to distant niche markets, thereby increasing their revenue. Supplementing citrus and winery by-products in ruminant diets may also reduce economic losses and food-borne illnesses associated with discoloration and with chemical and microbial spoilage. Moreover, the use of citrus and winery by-products could reduce costs for food additives, specialized processing, and packaging often used in the food industry to improve the appearance and maintain the quality of meat following long periods of storage and distribution [5].



**Figure 1.** A summary of the potential benefits of incorporating citrus and winery by-products into ruminant diets.

Research to determine the optimum inclusion levels of these fruit by-products from specific varieties grown in different environments extracted and processed using distinct methods, among

other variables, could be important. A readily accessible facility or analytical method should be available for the complete quantification and classification of micronutrients and phytochemicals from citrus and winery by-products. Future research on the bioactivity, bioavailability, and toxicology of these phytochemicals and their mode(s) of action, stability, and interactions with other food ingredients should be conducted with cautious assessment of in vitro and in vivo studies. If all of these components, including technical and economic feasibility challenges, product quality, and health safety, are in place, valorization of citrus and winery by-products in low-to-middle-income countries producing these by-products could be completed.

## 7. Conclusions

Sustainable utilization of citrus and winery by-products as dietary supplements in ruminant production (up to 150 g/kg DM) holds greatest potential in the low-to-middle-income countries producing these by-products. These by-products have the potential to improve the DMI, nutrient digestibility, rumen fermentation parameters, health and welfare, growth performance, and carcass and meat quality attributes of ruminant animals. In that context, a sustainable food production system can be attained through the utilization of citrus and winery by-products to improve food, nutrition, and income security for resource-poor, vulnerable, and marginal populations who reside in low-to-middle-income countries. Future studies are recommended to determine the effects of feeding citrus and winery by-products on meat fatty acid profiles, shelf life, microbiological quality, and volatile and flavor compounds in ruminants.

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