Effect of fining with new plant proteins on the aroma composition, phenolic compounds, and color of a Monastrell wine

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Abstract. The wine industry has increased its interest in finding new fining agents, and vegetable proteins have received increasing attention. Quinoa and kiwicha are native pseudocereals to the Andean highlands with high protein content and are considered gluten-free products. This work aimed to determine the effect of fining with quinoa and kiwicha protein extracts (QP and KP respectively) at different doses (30 and 50 g/hL) on the aroma composition, color, and phenolics of a Monastrell wine compared to pea proteins and gelatin. Fining treatments produced no significant reduction of TPI. Except for QP at 30 g/hL and the lowest dose of pea proteins, the fining treatments decreased the content of total anthocyanins. The doses of 50 g/hL of QP and KP were particularly efficient, producing the largest decrease in total tannins. Analyses by size exclusion chromatography showed that treatments with QP and KP were capable of selectively removing high molecular weight phenolics. All fining agents slightly decreased the color intensity. All fining treatments showed a significant decline in total ester concentration (26-45%). Instead, total alcohols remained constant upon the fining treatments. Five terpenes were identified in all wines, and their content was not affected by the fining treatments.

1 Introduction

The growing demand for foods and beverages that do not include the use of inputs of animal origin as part of their manufacturing process has led researchers to search for new alternative food additives. Faced with this scenario, the wine industry has increased its interest in finding new methods for the phenolic clarification of wine, and plant-based proteins from cereals, potatoes and legumes have received increasing attention. Proteins extracted from various plant sources have been proposed as effective phenolic fining agents (1-4), but due to their allergenic potential, only those obtained from wheat, peas and potatoes have been approved for use in wines (OIV-OENO 495 -2013).

Quinoa (Chenopodium quinoa Willd.) and kiwicha (Amaranthus caudatus L.) are native pseudocereals to the Andean highlands with higher protein content than traditional cereals (5,6) and are considered gluten-free products with low allergenic potential (7,8).

This work aimed to determine the effect of fining with protein extracts from quinoa and kiwicha (QP and KP respectively) at different doses (30 and 50 g/hL) on the aroma composition, color, and phenolics of a Monastrell wine compared to the commercial proteinaceous fining agents Proveget 100 (pea proteins at the doses 30 and 50 g/hL) and Vinigel cristal (liquid gelatin at 125 mL/hL) after 48 h of contact time.

2 Materials and methods

2.1 Fining experiments

For the fining experiments, a Monastrell wine was used. The fining capacity of KP at doses of 30 and 50 g/hL was evaluated in comparison with QP at 30 and 50 g/hL, and two commercial fining agents based on gelatin and pea proteins (i.e., the liquid gelatin Vinigel cristal, Agrovin, at the recommended dose of 125 mL/hL, and the pea protein concentrate Proveget 100, Agrovin, at 30 and 50 g/hL). For the fining trial, 100 mL of wine was transferred into 125 mL glass bottles. The clarifying agents were dispersed into 500 µL of water and then added to the wine samples. The control treatment consisted of 100 mL of wine treated with 500 µL of water.

Fining experiments were performed at room temperature, with 48 h of contact time. After clarification, the wines were centrifuged at 14000 rpm for 10 min. All treatments were conducted in triplicate.

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2.2 Physicochemical analyses

Condensed tannins were determined by the methylcellulose precipitable tannin assay (9). Color intensity was determined by the sum of absorbance at 420, 520, and 620 nm. The total polyphenol index (TPI) was calculated from the absorbance at 280 nm of the 1:100 diluted wine samples. Total and polymeric anthocyanins were determined according to Ho et al. 2001(10).

2.3 Volatile compounds

For the analysis of wine volatile compounds, 8 mL of wine were added to a 20 mL headspace vial. 2 g of sodium chloride and 200 µL of the internal standard (125 µL/L of 2-octanol in absolute ethanol) were added to the same vial. The vial was tightly sealed and then loaded onto a Gerstel autosampling device (Gerstel GmbH & Co.KG, Mellinghofen, Germany). The program of the autosampling device consisted of swirling the vial at 500 rpm for 15 min at 40 ºC, then inserting the fibre into the headspace for 30 min at 40 ºC as the solution was swirled again, then transferring the fibre to the injector for desorption at 240 ºC for 5 min. A divinylbenzene-carboxen-polydimethylsiloxane 50/30 micras (DVB/CAR / PDMS) fiber was used.

Gas chromatographic analyses were carried out on a HP 5890 GC system coupled to a HP 5972 quadrupole mass spectrometer (Agilent Technologies, CA, USA). A HP InnoWax 30M capillary column (50 m x 0.32 mm, 0.25 µm film thickness, Agilent Technologies, CA, USA) was used for the analysis, using helium as carrier gas y with a temperature programe starting at 40 ºC for 5 min then raising to 225 ºC at 3 ºC/min, and then holding at that temperature for 5 min. The mass spectrometer was operated in electron ionisation mode at 70 eV. The MS detector was operated in scan mode (mass range 50-200 amu).

3 Results

3.1 Phenolic composition and color

The effect of the fining treatments with quinoa and kiwicha proteins on the phenolic profile of a Monastrell wine was evaluated and compared against the commercial gelatin and pea proteins (Table 1).

The fining treatments tested in this study produced no significant reduction of total phenolics (TPI). Except for Q30 and P30, the rest of the treatments affected the content of total anthocyanins, causing decreases ranging from 9 to 12%, with gelatin being the clarifying agent that produced the greatest absolute decrease. All treatments reduced the concentration of polymeric anthocyanins, varying from 3 to 11%. In this case, gelatin, and the highest doses of quinoa and kiwicha proteins caused the greater decreases. The highest doses of quinoa, kiwicha proteins, and gelatin were the treatments that caused the greatest decrease. According to the results of the MCPT method, the treatments with quinoa proteins, with the highest dose of kiwicha proteins (50 g/hL) and gelatin caused a significant decrease in tannins, specially the two plant proteins, ranging between 4 and 11%, whilst the treatments with pea and kiwicha proteins at 30 g/hL did not show differences against the control. In this trial, all the evaluated treatments caused decreases in color intensity varying from 3 to 12%.

Table 1. Spectrophotometric measurements of Monastrell wine phenolics after treatments with proteinaceous fining agents.

	TPI	MCP tannins (mg/L)	Total anthocyanins (mg/L)
Control	60.6 ±1.8 a	2456.7 ± 42.9 a	172.0 ± 1.2 a
QP_30	59.4 ± 2.1 a	2317.6 ± 4.8 c	162.7 ± 2.3 ab
QP_50	57.7 ± 24 a	2236.0 ± 40.5 d	155.2 ± 4.0 bc
KP_30	59.5 ± 1.4 a	2416 ± 18.5 ab	156.8 ± 2.5 bc
KP_50	58.0 ± 2.6 a	2288.2 ± 7.4 cd	153.7 ± 4.5 bc
P_30	60.5 ± 3.5 a	2453.3 ± 29.0 a	162.4 ± 5.3 abc
P_50	60.8 ± 1,0 a	2432.5 ± 5.9 a	156.8 ± 2.1 bc
V_125	59.8 ± 1.7 a	2347.8 ± 1.5 bc	151.6 ± 4.5 c

3.2 Aroma composition

The most abundant volatile compounds detected in wine samples correspond to fermentation-derived esters and alcohols. Ethyl octanoate was the major ester identified in the control wine (37.5 µg/L), followed by ethyl succinate (35,7 µg/L), ethyl acetate (18,7 µg/L), and ethyl hexanoate (8.9 µg/L) respectively. All fining treatments showed a significant decline in total ester concentration varying from 26 to 45%, with gelatin being the agent causing the largest decrease. The major volatile loss corresponds to ethyl octanoate, which decreased by about 89%, with all fining agents showing a similar affinity to this substance. All treatments diminished the ethyl hexanoate concentration between 35 and 54%. The ethyl succinate concentration was not affected by the fining treatments.

Instead, the concentration of acetates was not affected by the fining treatments, just like total alcohols that remained constant upon the fining treatments. 3-methyl-1-butanol (91.7 µg/L) and 2-phenylphenol (37 µg/L) were the most abundant alcohols in wine samples. Only Q50 decreased the concentration of 3-methyl-1-butanol by 13%, while none of the fining treatments modified the content of 2-phenylphenol. Regarding C6-alcohols, hexanol was the only volatile identified, being in similar concentrations in all wines (1.8 µg/L). The concentration of hexanol in wines was below its olfactory perception thresholds. Besides, five terpenes were identified in all wines (i.e., linalool, α-terpinene, α-terpineol, nerolidol, and geraniol), but none of them exceeded their odor threshold. Terpenes were not affected by the fining treatments.

4 Conclusions

Quinoa and kiwicha protein extracts were as effective as gelatin reducing the total tannin content of Monastrell wines. Treatments with QP and KP reduced the total anthocyanins content and slightly decreased the color intensity similar to other proteinaceous fining agents. Regarding the aroma composition, QP and KP reduced the total ester concentration similar to the other clarifiers, but total alcohols remained constant upon the fining treatments. Results showed that QP and KP could be used as effective fining agents alternatives to animal proteins such as gelatin, and their effectiveness depends on the chemical composition or variety of the wines tested. The use of QP and KP as fining agents has the advantage that both pseudocereals are recognized as non-allergenic products.

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