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Fast grape cold drying using a heat pump technology as an alternative to traditional postharvest dehydration: Effects on the phenolic composition and extractability

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ABSTRACT

Cold drying technology aims to achieve fast water loss by relying on low relative humidity while avoiding high thermal stress on the product. The study compares its effect to two traditional dehydration conditions used to process grapes destined to special wine production, focusing on the preservation and extraction of anthocyanins and tannins in *Vitis vinifera* L. cv. 'Nebbiolo'. Fast rate dehydration (FD) at 15 °C and 40% relative humidity (RH), medium rate dehydration (MD) at 25 °C and 40% RH, and slow rate dehydration (SD) at 10 °C and 75% RH were tested after 15 days and at 35% weight loss. Grape must composition, potential phenolic content of skin, seeds, and pulp, anthocyanin profile, and mechanical properties were assessed, as well as extracted phenolic content and color traits during 14-day simulated maceration in wine-like solution. FD, while achieving a 35% weight loss in the shortest time (15 days), did not preserve skin polyphenols, mostly decreasing the anthocyanin content, which limited the formation of polymeric pigments during maceration. FD also resulted in highest contents of seed phenolics. This fast dehydration may induce stress responses, skin structural changes or biochemical imbalances in grape tissues, compromising the preservation of phenolic compounds, especially from skins. MD showed better outputs, especially for anthocyanin retention and color preservation during maceration, whereas SD was also effective in preserving skin polyphenols, but longer dehydration promoted anthocyanin degradation. Shorter dehydration treatments remain useful to produce high quality special wines from partially dehydrated grapes, particularly to reduce storage time.

1. Introduction

In a highly competitive global market, the production of wines with distinctive characteristics is of great interest for wineries. Products obtained from grapes that undergo significant water loss are renowned for their complexity and high quality. Well-known examples include sweet wines such as *Passito*, Tokaji, Pedro Ximénez, and Sauternes as well as dry reinforced wines like Amarone and Sforzato di Valtellina (Mencarelli & Tonutti, 2013). Different dehydration techniques are employed around the World. Grape berries dehydration can occur on-vine, through late harvest, noble rot or freezing, and after harvest involving more extreme water loss (typically drying in open-air settings) or withering as a result of slow dehydration (Mencarelli & Tonutti, 2013). Therefore, two key factors govern the grape dehydration process: the extent and the rate of water loss. Water losses of 20%−30% are usually used in the production of dry wines whereas significantly higher levels (40%−50%) are reached for sweet wines (Mencarelli & Tonutti, 2013).

The dehydration process induces significant chemical and physical transformations in the grape berries, depending on both intrinsic and extrinsic factors. Intrinsic factors include varietal characteristics like berry size, bunch compactness, skin surface morphology, and microporosity (Sanmartin et al., 2021). Extrinsic factors encompass environmental conditions such as relative humidity, temperature, air flow, exposure to sunlight, and the overall duration of the process (Matera et al., 2017; Sanmartin et al., 2021).

Postharvest dehydration has a high impact on primary and

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secondary grape metabolites. Regarding the phenolic composition, changes have been attributed to a dynamic interplay between biosynthetic and oxidative mechanisms occurring throughout the dehydration process, beyond the concentration effect. Several studies suggest that physiological and metabolic variations are closely linked to both temperature and water loss rate (Mencarelli et al., 2010), being likewise genotype-dependent (Torchio et al., 2016; Zenoni et al., 2016). On the one hand, the rate at which berry weight loss occurs significantly affects secondary metabolism in the grape skin. Zenoni et al. (2020) observed an inhibited accumulation of phenolic compounds, particularly stilbenes, after accelerated postharvest dehydration compared to the natural process, even under the same temperature conditions. Moreover, the strongest average genes downregulation involved anthocyanin regulators, particularly in the accelerated process. On the other hand, Shmuleviz et al. (2023) studied the single temperature effect and reported a reduction of total anthocyanin content during postharvest dehydration even though the differences were not significant between the two temperatures used (8 and 12 °C). However, lower temperature stimulated the increase of resveratrol whereas higher temperature promoted stilbene oligomeric forms probably due to oxidative polymerization. Other studies have identified a temperature range of approximately 10–20 °C as optimal for preserving anthocyanin integrity during grape dehydration (Mencarelli et al., 2010; Sanmartin et al., 2021). The content of phenolic compounds, such as stilbenes, flavonols, and catechins, increases when dehydration temperature rises from 10 °C to 20 °C for weight losses between 20% and 30%, but decreases strongly at 30 °C (Mencarelli et al., 2010). It is important to highlight that prolonged dehydration under non-optimal or uncontrolled conditions can lead to the degradation or oxidation of phenolic molecules. Such degradation is often associated with the activity of oxidative enzymes such as peroxidase and polyphenol oxidase (PPO) from grapes or laccase from molds (Constantinou et al., 2018; Mencarelli et al., 2010) affecting mainly anthocyanins. Therefore, the management of the dehydration process is of paramount importance in minimizing phenolic oxidation to produce high quality wines (Mencarelli et al., 2010, 2021).

Postharvest dehydration also causes structural modifications of skin cell wall (Río Segade et al., 2019; Zoccatelli et al., 2013). This skin cell wall degradation can enhance the extractability of phenolic compounds, thereby influencing the final chemical profile of the wine (Panceri & Bordignon-Luiz, 2017; Río Segade et al., 2019). Previous studies have underlined that berry skin mechanical properties, such as skin hardness and thickness, usually increase during grape postharvest dehydration but the magnitude of these changes depends on the dehydration conditions and grape variety (Río Segade et al., 2019; Scalzini et al., 2023).

Regarding the phenolic composition of berry seeds, few studies have been published on the impact of the dehydration process, which reported an increase in the content of proanthocyanidins and low molecular weight flavanols (Centioni et al., 2014; Moreno et al., 2008; Rolle et al., 2013). Moreover, it is important to know if these compounds can be easily released during maceration because they influence sensory traits such as astringency and bitterness (Río Segade et al., 2016). ‘Nebbiolo’ is a grape variety native from northwestern Italy, widely recognized for its role in the production of the internationally renowned dry red wines *Barolo* and *Barbaresco* from fresh grapes. Moreover, ‘Nebbiolo’ grapes are used to produce *Sforzato di Valtellina DOCG*, a reinforced dry red wine emblematic of the "heroic viticulture" practiced on the steep slopes of the Valtellina valley in Northern Italy. This wine is produced from partially dehydrated grapes under uncontrolled postharvest conditions in dedicated rooms (Scalzini et al., 2023). ‘Nebbiolo’ grapes present a high share of di-substituted anthocyanins, which are prone to oxidation and lead to scarce and unstable wine color, although they have a high content of tannins, key players in wine astringency.

Based on these considerations, the present study aimed to evaluate the impact of cold drying technology for fast dehydration at low temperature on the phenolic composition and extractability of skins and seeds from ‘Nebbiolo’ grapes, in comparison with two alternative dehydration protocols: a controlled temperature method, representative of modern practices used in the production of reinforced wines and an uncontrolled temperature method, which reflects traditional dehydration techniques. Combining low temperature with an accelerated dehydration rate provides an innovative controlled approach and, to our knowledge, this study represents the first application of cold drying using heat pump technology to wine grape dehydration. Fast dehydration using a cold dryer is a technique already tested in several industries such as meat preparation and officinal herbs preservation, and relies on the use of temperatures below 20 °C and low relative humidity, under different construction approaches (Aykın-Dinçer & Erbaş, 2019). The use of a fast dehydration approach is a challenge for wineries: while it potentially avoids the prolonged storage of grapes and the susceptibility to fungal infections during extended withering periods, at the same time its effect on preserving important grape metabolites needs to be assessed.

2. Materials and methods

2.1. Grape samples and dehydration processes

For this study, approximately 35 kg of *Vitis vinifera* L. cv. ‘Nebbiolo’ grapes were harvested by hand at commercial ripeness (24.8 °Brix) in 2023 from a vineyard located in Barolo (Piemonte, Italy) and then transported to the laboratory. Bunches were visually inspected to remove damaged berries and then manually sectioned into smaller clusters (5–10 berries) to ensure homogeneous air exposure during dehydration. A single layer of small clusters was randomly distributed on perforated boxes (about 2 kg each) and then subjected to three different dehydration treatments. Approximately 10 kg of ‘Nebbiolo’ grapes were used for each dehydration trial. The first dehydration method was based on the use of a cold dryer using heat pump technology (NWT35, Northwest Technologies, Boves, Italy), hereafter referred to as fast rate dehydration (FD). This technique is based on the reduction of water vapor content and subsequent heating at low temperature of the air stream contacting the product. The use of dehumidified air increases the capability to absorb moisture from the product while maintaining relatively low temperatures, with potential benefits in terms of food quality and energy consumption (Bhattacharjee et al., 2024; Salehi, 2021). The process consists in three phases: (i) the airflow entering the drying chamber absorbs the grape moisture; (ii) the outlet wet air flows though the evaporator of a heat pump and it cools below the dew point resulting in water vapor condensation; (iii) the dehumidified air flows through the condenser of the same heat pump increasing its temperature before being reintroduced into the drying chamber (Salehi, 2021). The grape dehydration conditions were set to 15 °C temperature and 40% relative humidity (RH).

The second method involved dehydration in a room at 25 °C without RH control (average value of 40% RH), referred to as medium rate dehydration (MD), to simulate the natural dehydration process occurring in a closed room. The third treatment used a cooled incubator (FOC200E, Velp Scientifica Srl, Usmate Velate, Italy), in which the temperature was set to 10 °C without RH control (average value of 75% RH); this condition is referred to as slow rate dehydration (SD). This last treatment aimed to reach optimal temperatures for preserving secondary metabolites (Mencarelli et al., 2010). Under each condition, the dehydration process was stopped when the grape samples reached a weight loss (WL) of 35%. Sampling was done at three experimental stages: on fresh grapes (T0), for all conditions when FD-treated grapes reached 35% WL (T1, time-matched comparison point), and when the SD- and MD-treated grapes also achieved 35% WL (T2, weight loss-matched comparison point), corresponding to 15, 27, and 60 days for FD, MD, and SD, respectively. Therefore, T2 involves different dehydration time to reach same target WL for each of the three conditions studied. Three biological replicates of each sample were analyzed just after dehydration.

2.2. Mechanical properties

Grape texture analysis was carried out using a TA.XTplus texture analyzer (Stable Micro Systems, Godalming, Surrey, UK), equipped with an HDP/90 platform and a 5 kg load cell, according to Rolle, Torchio et al. (2012). The mechanical properties of whole berries and berry skins were determined on fresh and dehydrated grapes to evaluate the skin wall degradation. For each sample, 30 berries were individually measured. The berry skin hardness was evaluated by a puncture test using a P/2 N needle probe through skin break force (N, as F_{sk}), skin break energy (mJ, as W_{sk}), and skin stiffness (N/mm, as E_{sk}). Skin thickness (μm, as Sp_{sk}) was determined on a piece of skin (~0.25 cm^2) using a P/2 flat probe. The softening of whole berries was assessed using a double compression test under 25% deformation with a P/35 flat probe. The parameters determined were hardness (N, as BH), cohesiveness (adimensional, as BCo), gumminess (N, as BG), springiness ratio (adimensional, as BS-*r*), and resilience (adimensional, as BR).

2.3. Grape must analysis

For fresh and dehydrated grapes, three independent samples consisting of 100 g of berries were randomly selected and manually crushed. The resulting must was centrifuged at 4000 rpm for 15 min at 20 °C using a Heittch 32R centrifuge (Tuttlingen, Germany). Quantification of organic acids (malic, tartaric, and citric), glycerol, and reducing sugars (glucose and fructose) was performed using high-performance liquid chromatography (HPLC) equipped with a refractive index detector and a diode array detector (DAD) set to 210 nm (Agilent Technologies, Santa Clara, CA, USA), in accordance with the procedure described by Giordano et al. (2009). The pH value was determined using an InoLab 730 pH meter (WTW, Weilheim, Germany). Total acidity, expressed as g/L of tartaric acid, was assessed following the protocol outlined by the International Organization of Vine and Wine (method OIV-MA-AS313–01; OIV, 2022).

2.4. Grape skin, pulp, and seed polyphenol extraction

For each sampling point and treatment, a total of ten berries were manually peeled, and their components (skin, seeds, and pulp) were carefully separated. Skins and seeds were quickly and individually immersed into 40 mL of an extraction solution at pH 3.20 consisting of 5 g/L tartaric acid, 12% v/v ethanol, and 2 g/L sodium metabisulfite (Di Stefano & Cravero, 1991; Di Stefano & Flamini, 2008). The samples were subsequently frozen at −20 °C for later analysis. Prior to analysis, the samples were thawed at ambient temperature. Skins were homogenized at 8000 rpm for 1 min using an immersion blender (Ultra-Turrax T25, IKA Labortechnik, Staufen, Germany) and centrifuged at 4000 rpm for 15 min at 20 °C. Seeds were macerated in the extracting solution at 25 °C for 7 days and then centrifuged. The supernatant was then collected, diluted to a final volume of 50 mL with the same extraction solution, and used for the determination of potential content of phenolic compounds. The pulp was introduced into a tube containing sodium metabisulfite (100 mg), diluted (9:1, m/m) with 5 mol/L sulfuric acid, and frozen at −20 °C for later analysis (Di Stefano & Cravero, 1991; Río Segade et al., 2014). Once thawed, it was homogenized at 9500 rpm for 30 s with an Ultraturrax T10 high-speed homogenizer and centrifuged for phenolic compounds determination in the supernatant.

Simulated maceration trials were conducted using a wine-like solution at pH 3.40 composed of 5 g/L tartaric acid and 100 mg/L sodium metabisulfite. For each experimental condition, 80 g of berries were randomly selected. Then, skins and seeds were manually separated from the pulp and individually immersed into 100 mL of the model solution. Maceration was carried out over 14 days at 24 °C. To monitor the extraction kinetics, 3 mL aliquots were collected at 8, 24, 48, 72, 96, 120, 168, and 240 h. In accordance with the method described by Paissoni et al. (2020), the sampled volume was replaced with a mixture of absolute ethanol and model solution to simulate the progressive increase in ethanol concentration during fermentation, corresponding to 0%, 1%, 3%, 6%, 9%, 12%, 13.5%, and 15% v/v, respectively. After this stepwise addition, a final ethanol concentration of 15% v/v was kept from 240 h to 336 h (14 days).

2.5. Physical-chemical analysis of the extracts

Spectrophotometric analysis of phenolic composition. Phenolic compounds were quantified using a UV-1800 spectrophotometer (Shimadzu Corp., Kyoto, Japan), following spectrophotometric methods adapted from Torchio et al. (2010) and Di Stefano and Cravero (1991). The total phenol index (TPI), expressed as mg (−)-epicatechin per kg of grape and per 100 berries, was determined by measuring the absorbance at 280 nm. Total anthocyanin content (TA), expressed as mg malvidin-3-*O*-glucoside chloride per kg of grape and per 100 berries, and total flavonoids (TF), expressed as mg (+)-catechin per kg of grape and per 100 berries, were assessed in skin extracts by diluting samples in a mixture of absolute ethanol, water, and 37% hydrochloric acid (70:30:1, v/v/v). Absorbance was measured at 536–540 nm for anthocyanins whereas at 280 nm for flavonoids. Condensed tannins were quantified by precipitation with 0.04% w/v methyl cellulose (MCP) and reported as mg (−)-epicatechin per kg of grape and per 100 berries, after measuring absorbance at 280 nm (Sarneckis et al., 2006). Flavanols reactive to vanillin (FRV), expressed as mg (+)-catechin per kg of grape and per 100 berries, were determined by the reaction with a methanolic solution of 4% w/v vanillin and concentrated hydrochloric acid by measuring the absorbance at 500 nm.

At the end of skin maceration, monomeric pigments (MON), small polymeric pigments (SPP), and large polymeric pigments (LPP) were evaluated by LPP precipitation with bovine serum albumin and MON bleaching using sulfur dioxide (Harbertson et al., 2003), and subsequent absorbance measurement at 520 nm.

Color characteristics. The color parameters of the macerating skin extracts were determined through color intensity and hue, following the OIV-MA-AS2–07B method, and CIELab coordinates including lightness (L^*), red/green component (a^*), and yellow/blue component (b^*), as described in the guidelines provided by the OIV-MA-AS2–11 method (OIV, 2022). The ΔE^* parameter, representing the color difference between differently treated samples, was calculated as follows (Equation 1; OIV, 2022):

$$\Delta E^* = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2} \quad (1)$$

HPLC anthocyanin profile. The anthocyanin profile was analyzed following the procedure reported by Río Segade et al. (2014). Each skin extract was diluted 1:1 with a 0.3 mol/L hydrochloric acid solution at pH 0.5 and filtered through a 0.45 μm PTFE membrane filter. A 50 μL aliquot was injected into an Agilent 1260 HPLC system (Agilent Technologies) equipped with a LiChrospher® 100 RP-18 analytical column (25 cm × 0.4 cm, 5 μm particles; Merck, Darmstadt, Germany). The mobile phase consisted of solvent A (formic acid:water, 10:90 v/v) and solvent B (formic acid:methanol:water, 10:50:40 v/v/v). Anthocyanins were detected at 520 nm with a diode array detector. Data acquisition and processing were performed using the Agilent ChemStation software (Agilent Technologies). Individual monomeric forms were identified according to their retention time and spectral characteristics. The quantification was performed using an external standard calibration and results were expressed as mg of malvidin-3-*O*-glucoside chloride per kg of grape.

2.6. Statistical analysis

Statistical analysis was carried out using R software (R Foundation for Statistical Computing, Vienna, Austria). The assumptions of normal data distribution and homogeneity of variances were verified using the

Shapiro–Wilk test and Levene's test, respectively. For each measured variable, a one-way analysis of variance (ANOVA) was applied to assess differences among treatments, followed by Tukey's HSD test for post hoc comparisons. A significance threshold of $p < 0.05$ was adopted. When the assumptions of normality or homoscedasticity were not satisfied, a Welch's ANOVA was performed and the Games–Howell test was used for subsequent pairwise comparisons.

3. Results and discussion

3.1. Dehydration kinetics and technological ripeness parameters

Daily berry WL varied markedly among the three dehydration treatments. Grapes subjected to the FD method achieved 35% WL within 15 days, corresponding to an average daily WL of 2.34%. The MD treatment required 27 days to reach the same WL, with an average daily reduction of 1.37%. Conversely, the SD treatment extended over 60 days to attain 35% WL, resulting in an average daily WL of 0.57%. These results clearly demonstrate the significant role of temperature and relative humidity (RH) in modulating the rate of water loss (Mencarelli et al., 2010), which in turn affects the berry metabolism during the dehydration process (Zenoni et al., 2020). At T1 sampling point, defined as the moment in which the grapes subjected to the FD process already reached 35% WL, those from MD and SD processes achieved a WL of 18% and 6%, respectively. As previously mentioned, at T2 the grapes from the different dehydration treatments achieved 35% WL.

In relation to sugar content, all three dehydration treatments resulted in grapes with higher sugar concentrations compared to fresh grapes. Among the treatments, the FD condition produced the highest final sugar concentration, followed by the MD and SD treatments at T1 and T2 (Table 1). The increase in sugar content corresponds to the concentration effect due to the berry water loss in the SD-treated grapes, exceeding the concentration factor in MD and FD. These results support that temperature and relative humidity influence the water loss dynamics and may induce processes such as the release of sugars from the grape cell wall matrix into the must, being glucose the main sugar of

Table 1
Standard parameters of grape must and potential concentration of phenolic compounds in berry skins, pulp, and seeds from fresh grapes and dehydrated grapes under different environmental conditions.

Sample (weight loss)	Fresh grapes (-)	Dehydrated grapes T1				Dehydrated grapes T2			
		FD (35%)	MD (18%)	SD (6%)	Sign	FD (35%)	MD (35%)	SD (35%)	Sign
Grape must									
Reducing sugars (g/L)	260±1	380±5a	332±44b	274±0c	***	380±5a	363±3b	349±8c	**
pH	3.29±0.01	3.63±0.05a	3.39±0.01b	3.33±0.02b	***	3.63±0.05a	3.40±0.02c	3.50±0.02b	***
Total acidity (g/L as tartaric acid)	5.31±0.09	4.78±0.14a	4.80±0.24a	4.99±0.04a	ns	4.78±0.14c	5.20±0.07b	5.59±0.04a	***
Citric acid (g/L)	0.06±0.01	0.08±0.01a	0.06±0.03a	0.05±0.01a	ns	0.08±0.01b	0.04±0.01b	0.24±0.08a	*
Tartaric acid (g/L)	7.59±0.07	5.17±0.10b	6.35 ±1.14ab	7.11±0.08a	**	5.17±0.10a	5.43±0.17a	4.40±0.04b	***
Malic acid (g/L)	1.11±0.01	1.14±0.04a	0.96 ±0.14ab	0.86±0.06b	*	1.14±0.04a	0.77±0.08b	0.86±0.04b	***
Glycerol (g/L)	-	0.37±0.18a	0.10 ±0.05ab	0.07±0.04b	*	0.37±0.18c	0.89±0.09b	3.06±0.90a	***
Grape skins									
TPI (mg (−)-epicatechin/kg grape)	2623±92	2418±60ab	2721±79a	2314±249b	*	2418±60b	3159±121a	2617±181b	**
TF (mg (+)-catechin/kg grape)	3531±53	3924±190a	4079±87a	3552±93b	**	3924±190b	4883±140a	3849±36b	***
TA (mg malvidin-3-*O*-glucoside chloride/kg grape)	495±20	335±28b	529±29a	455±60a	**	335±28c	612±25a	463±31b	**
FRV (mg (+)-catechin/kg grape)	986±65	466±52c	947±65a	749±46b	***	466±52b	838±106a	765±56a	**
MCP (mg (−)-epicatechin/kg grape)	914±76	859±117b	1167±87a	794±28b	**	859±117b	1244±67a	825±167b	*
Delphinidin-3-*O*-glucoside (mg/kg grape)	26.7 ± 3.9	11.6 ± 1.9b	27.9 ± 0.8a	22.7 ± 5.2a	***	11.6 ± 1.9b	26.1 ± 3.3a	20.8 ± 2.4a	***
Cyanidin-3-*O*-glucoside (mg/kg grape)	48.1 ± 4.0	27.6 ± 3.3b	54.1 ± 2.5a	44.1 ± 10.6a	**	27.6 ± 3.3b	48.5 ± 5.5a	38.3 ± 3.9a	***
Petunidin-3-*O*-glucoside (mg/kg grape)	22.5 ± 3.4	13.9 ± 1.5b	24.0 ± 0.9a	19.3 ± 3.4a	**	13.9 ± 1.5b	24.7 ± 3.0a	19.2 ± 2.0a	***
Peonidin-3-*O*-glucoside (mg/kg grape)	157.1 ± 10.6	94.9 ± 7.1b	170.0 ± 8.9a	152.5 ± 13.9a	***	94.9 ± 7.1c	187.1 ± 5.5a	149.4 ± 12.1b	***
Malvidin-3-*O*-glucoside (mg/kg grape)	111.0 ± 15.5	52.3 ± 3.6b	109.5 ± 11.8a	96.0 ± 6.9a	***	52.3 ± 3.6b	122.6 ± 15.2a	93.7 ± 11.0a	***
Sum acetyl-glucosides (mg/kg grape)	8.5 ± 0.9	5.4 ± 0.4b	8.4 ± 1.9a	8.5 ± 0.5a	*	5.4 ± 0.4c	8.2 ± 0.4b	11.1 ± 1.5a	***
Sum cinnamoyl-glucosides (mg/kg grape)	44.6 ± 4.5	31.7 ± 2.1b	50.6 ± 8.4a	46.5 ± 2.4a	**	31.7 ± 2.1c	66.9 ± 4.2a	55.3 ± 2.8b	***
Grape pulp									
TF (mg (+)-catechin/kg grape)	221.4 ± 99.0	310.7 ± 244.6a	187.3 ± 31.4a	100.6 ± 14.8a	ns	310.7 ± 244.6a	282.6 ± 52.7a	237.7 ± 42.7a	ns
TA (mg malvidin-3-*O*-glucoside chloride/kg grape)	17.0 ± 9.0	40.3 ± 35.1a	16.0 ± 4.0a	7.9 ± 2.1a	ns	40.3 ± 35.1a	22.7 ± 5.9a	19.8 ± 3.7a	ns
FRV (mg (+)-catechin/kg grape)	104.1 ± 53.5	105.5 ± 35.6a	123.6 ± 4.4a	44.4 ± 8.2b	**	105.5 ± 35.6a	191.9 ± 38.6a	147.6 ± 31.4a	ns
MCP (mg (−)-epicatechin/kg grape)	44.4 ± 23.3	127.2 ± 82.6a	51.4 ± 6.5a	19.9 ± 2.0a	ns	127.2 ± 82.6a	100.7 ± 22.9a	64.3 ± 14.6a	ns
Grape seeds									
TPI (mg (−)-epicatechin/kg grape)	1644±301	4236±360a	3005±522b	2641±79b	**	4236±360a	3131 ±604ab	2730±430b	*
FRV (mg (+)-catechin/kg grape)	1653±195	4068±313a	3399±118b	2566±91c	***	4068±313a	2536±375b	2773±431b	**
MCP (mg (−)-epicatechin/kg grape)	853±27	2335±100a	1602±255b	1440±83b	**	2335±100a	1502±363b	1304±201b	**

All data are expressed as average value ± standard deviation (n = 3). Sign: *, **, ***, and ns indicate significance at $p < 0.05$, 0.01, 0.001, and not significant differences, respectively. Different Latin letters within the same row indicate significant differences among treatments inside each time point according to Tukey test ($p < 0.05$). Anthocyanin compounds are expressed as mg malvidin-3-*O*-glucoside chloride/kg grape. TPI, total phenol index; TF, total flavonoids; TA, total anthocyanins; FRV, flavanols reactive to vanillin; MCP, methyl cellulose precipitable tannins; FD, fast rate dehydration; MD, medium rate dehydration; SD, slow rate dehydration; T1, time at which FD-treated grapes reached 35% weight loss (15 days; time-matched comparison point); T2, 35% weight loss for all treatments (weight loss-matched comparison point).

structural polysaccharides from both pulp and skin cell walls (Nunan et al., 1998; Tomasi et al., 2021; Vidal et al., 2001). The lower concentration of sugars in SD samples may be due to their consumption by *Botrytis cinerea* in agreement with an increased glycerol concentration, a marker of noble rot development (Modesti et al., 2024; Rolle, Giordano et al., 2012).

Regarding total acidity, the concentration effect induced by dehydration partially offset the metabolic degradation of organic acids typically observed in withered grapes in relation to fresh samples. Consequently, FD-treated grapes exhibited the lowest total acidity and the highest pH values at the end of the dehydration process (35% WL, T2 sampling point), followed by the MD and SD treatments (Table 1). Changes were also observed in the individual concentrations of organic acids. Tartaric content decreased in all treatments compared to fresh grapes, with the greatest reduction corresponding to higher WL mainly under SD conditions. Rösti et al. (2018) reported that the reduction in tartaric acid content is associated with precipitation phenomena occurring within the berry tissues, likely due to the disruption of cellular compartmentalization during dehydration. The observed decrease in malic acid levels in MD and SD-treated grapes may be attributed to a negative balance between catabolic activity and the concentration effect resulting from water loss (Scalzini et al., 2023). It is important to highlight that SD treatment significantly increased citric and glycerol concentrations in the grape must. An increased concentration of glycerol, consistent with a decrease in tartaric acid content, may be related to the presence of *Botrytis cinerea* (Modesti et al., 2024; Rolle, Giordano et al., 2012).

3.2. *Effect of postharvest dehydration on the potential content of phenolic compounds*

At T1 sampling point, the FD-treated grapes exhibited significantly lower values across almost all analyzed skin-related parameters when compared to those from MD and SD treatments, particularly for TA and FRV (Table 1). In berry skins, MD-treated grapes showed the higher contents of all determined compounds, although the differences were not always statistically significant. However, when data were expressed per 100 berries with the aim of minimizing the impact of berry WL associated with dehydration (Table S1), all parameters resulted to be significantly lower in FD-treated grapes compared to MD and SD, which showed similar results. Notably, the FD treatment yielded lower values of all skin-related parameters than those observed in fresh, non-dehydrated grapes (Table 1 and Table S1). At T2 sampling point (35% WL for all trials), no statistically significant differences were observed between FD- and SD-treated grapes for TPI, TF, and MCP in berry skins when data were expressed per kg of grape, with MD standing out for the highest values. However, FD-treated grapes consistently showed lower values than MD and SD among all skin-related parameters when data were expressed per 100 berries while this trend was observed only for TA and FRV considering the expression by kg of grape.

A plausible explanation for this pattern is that the accelerated water loss in FD-treated grapes induces more severe physiological stress during withering. Rapid dehydration likely causes extensive wrinkling and microfractures in the epidermal layer, leading to increased oxygen exposure and, consequently, greater non-enzymatic and enzymatic oxidation mediated by polyphenol oxidases (PPO) (Zheng et al., 2021). This oxidative degradation, particularly of anthocyanins and other phenolic compounds, may surpass the concentration effect typically observed during dehydration, as well as the biosynthetic activity stimulated by stress responses (Tomasi et al., 2021). These findings align with several studies reporting enhanced polyphenolic content in response to prolonged dehydration. This may be attributed to an inhibited expression of anthocyanin-regulation genes when the postharvest dehydration process is shortened (Zenoni et al., 2020). For instance, Bellincontro et al. (2016) observed higher concentrations of anthocyanins and polyphenols in the skins of 'Corvina','Corvinone', and 'Rondinella' following extended withering periods. Panceri et al. (2013) also reported significant increases in the polyphenol and anthocyanin content in 'Cabernet Sauvignon' and 'Merlot' grapes subjected to controlled slow dehydration. Conversely, other studies have highlighted cultivar-dependent variability in the effect of dehydration kinetics on the skin phenolic composition. Bonghi et al. (2012) found that rapid dehydration in 'Raboso Piave' grapes was effective in delaying the reduction of skin flavan-3-ol content, with (+)-catechin and (−)-epicatechin concentrations at 10% berry WL remaining comparable to or slightly exceeding those observed at harvest but they decreased at 30% WL. Additionally, Torchio et al. (2016) reported no significant differences in TA, FRV, TPI, or TF content in 'Nebbiolo' grape skins at 14% berry WL between fast and slow dehydration protocols.

On the other hand, the postharvest dehydration process degrades the skin cell wall facilitating the diffusion of phenolic compounds to the pulp but the dehydration rate can influence this degradation. In the present study, the berry pulps from the FD treatment showed the highest TF, TA, and MCP contents even though the differences were not significant due to high variability among replicates, probably associated with dissimilar degradation of skin cell wall influencing the release of phenolic compounds from the berry skin to pulp during the dehydration process (Río Segade et al., 2019). These results are in agreement with those reported by Torchio et al. (2016) who observed higher concentrations of total phenolic compounds in pulps from fast dehydration of 'Nebbiolo' grapes.

Regarding the anthocyanin profile (Table 1), the five main free anthocyanin forms, along with their corresponding acetylated and cinnamoylated (caffeoylated + *p*-coumaroylated) derivatives, showed a similar trend to the total anthocyanin content (TA) at T1 and T2, with significantly lower values for FD-treated grapes compared to MD and SD. At T2, other treatment-related differences were evident. Specifically, MD-treated grapes exhibited significantly higher concentrations of peonidin-3-*O*-glucoside and cinnamoylated derivatives compared to FD and SD, whereas SD showed the highest levels of acetylated anthocyanins. It is important to highlight that the percentage of acylated forms increased from 12.7% to 15.5%−17.1% after the dehydration process, while the total anthocyanin content decreased for FD and SD. Therefore, changes in the chemical structure of anthocyanins can also occur during postharvest dehydration (Bonghi et al., 2012).

In contrast to the trends observed in the skins, seed-related parameters exhibited an opposite pattern. At 15 days of dehydration (T1, corresponding to the point at which the FD treatment reached 35% berry WL), FD samples displayed significantly higher values of TPI, MCP, and FRV compared to those subjected to the MD and SD treatments, when expressed by kg of grape (Table 1). This trend persisted when MD and SD treatments reached the level of 35% WL (T2 sampling point). At T2, the differences were statistically significant for all parameters when the results were expressed by kg of grape or 100 berries, with the sole exception of TPI as mg/100 berries (Table 1 and Table S1). As observed for skin data, the seed-related parameters obtained for MD and SD treatments were generally similar one to another and, in most cases, not statistically different. Furthermore, the differently dehydrated grapes showed higher values across all measured parameters when compared to fresh grape samples. This increase exceeding the WL% can be largely attributable to the concentration effect resulting from water loss but also to enhanced extraction and potential biosynthesis or structural changes (Centioni et al., 2014). The consistently higher TPI, MCP, and FRV values in the FD treatment compared to MD and SD suggest that rapid dehydration conditions may enhance the potential concentration of seed polyphenols.

These findings are consistent with previous studies. Rolle et al. (2013) reported increased concentrations of seed proanthocyanidins, vanillin-reactive flavanols, and total flavonoids in Corvina grapes under the faster dehydration conditions. Similarly, Torchio et al. (2016) noted comparable trends in 'Nebbiolo' seeds under fast dehydration conditions, with high contents of the same compounds compared to slow

dehydration. Additionally, Rolle et al. (2009) found increasing contents of both proanthocyanidins and flavanols in seeds during the on-vine withering process of 'Mondeuse' grapes. In seeds, leucocyanidin reductase (LAR) expression peaked at ripening, inducing the catechin monomer formation from leucocyanidins (Rinaldi et al., 2017). Likewise, during the late seed maturation and dehydration, phenylpropanoid metabolic pathway involved in proanthocyanidin accumulation and cell wall modifying enzymes are up-regulated as

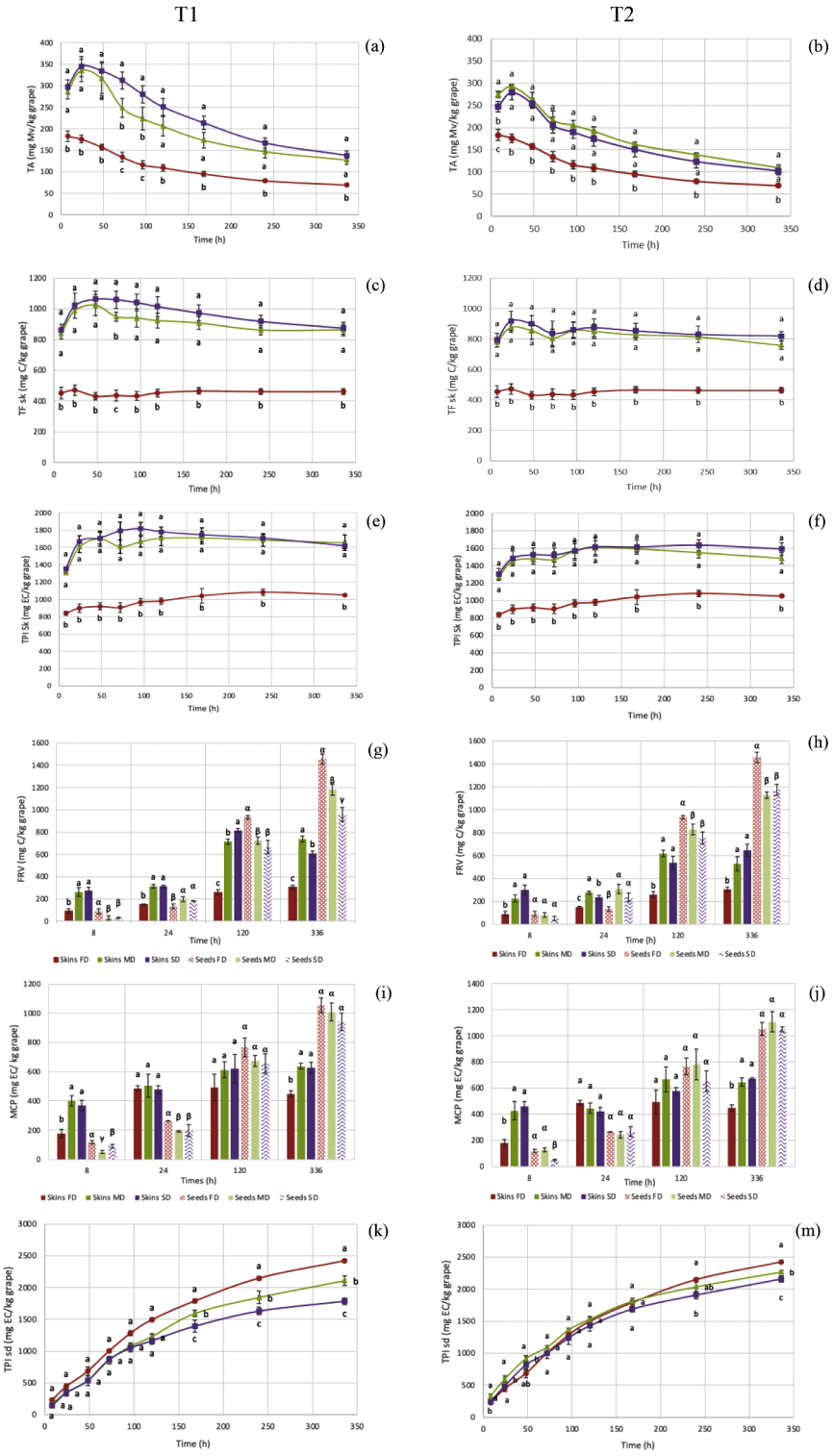

Fig. 1. Extraction curves of phenolic compounds (mg/kg grape) from berry skins and seeds of dehydrated grapes during simulated maceration. All data are expressed as average value ± standard deviation (n = 3). Different Latin letters within the same row indicate significant differences among treatments for each time according to Tukey test ($p < 0.05$). TPI, total phenol index; TF, total flavonoids; TA, total anthocyanins; FRV, flavanols reactive to vanillin; MCP, methyl cellulose precipitable tannins; sk, skins; sd, seeds; Mv, malvidin-3-*O*-glucoside chloride; C, (+)-catechin; EC, (−)-epicatechin; FD (red), fast rate dehydration; MD (green), medium rate dehydration; SD (purple), slow rate dehydration; T1, time at which FD-treated grapes reached 35% weight loss (15 days; time-matched comparison point); T2, 35% weight loss for all treatments (weight loss-matched comparison point).

protection mechanism, as observed for other orthodox seeds (Mazlan et al., 2018). These changes occurring during postharvest dehydration seem to be influenced by the different environmental conditions used.

3.3. Polyphenol extraction during simulated maceration

To investigate the extraction kinetics of phenolic compounds from grape skins and seeds, a 14-day maceration was conducted using a model wine solution. Ethanol concentration was progressively increased to simulate the alcoholic fermentation process. This simulated maceration was applied to both fresh and dehydrated grapes, allowing for a comparative assessment of the effects of different dehydration treatments on polyphenol release from berry skins and seeds under fermentation-like conditions.

According to the extraction kinetics for anthocyanins from fresh grape skins, TA concentration showed a clear peak at 24 h of maceration, followed by a gradual decline over time (Figure S1a). A similar pattern was observed across all dehydration treatments at T1 and T2 sampling points as shown in Fig. 1a-b and Figure S2a-b. This trend is consistent with previous studies (Giacosa et al., 2023; Paissoni et al., 2017; Río Segade et al., 2019). As maceration progresses, the concentration of anthocyanins declines due to concurrent oxidative reactions, polymerization and condensation processes, as well as adsorption on solid parts, often surpassing the rate of continued release (Setford et al., 2017). Among treatments, FD-treated grapes consistently exhibited the lowest TA concentrations, whereas SD yielded the highest ones. Nevertheless, the differences between SD and MD were not statistically significant at most sampling points. For T2 samples after 14 days of maceration, the TA concentration in FD samples was 70±2 mg malvidin-3-*O*-glucoside chloride/kg grapes, compared to 110±6 mg malvidin-3-*O*-glucoside chloride/kg grapes in MD and 102±7 mg malvidin-3-*O*-glucoside chloride/kg grapes in SD. This represents a reduction of 31%−36% in FD samples relative to the other conditions, even though the anthocyanin losses during maceration were lower. At the end of maceration, the extraction percentages for T2 samples (35% berry WL) in relation to potential content were 21±8%, 18±4%, and 22±7% for FD-, MD-, and SD-treated grapes, respectively.

On the other hand, the extraction kinetics for TF from skins of both fresh and dehydrated grapes showed an initial increase up to 48 h or 24 h of maceration for WL less than or equal to 35%, respectively, followed by a slight decline that stabilized into a plateau for FD samples (Figure S1b, Fig. 1c-d, and Figure S2c-d). As observed for anthocyanins, the FD treatment consistently resulted in lower TF concentrations compared to MD and SD samples at T1 and T2 sampling points. When the results were expressed per 100 berries, TF concentrations in SD samples were significantly higher than those in MD (Figure S2c-d). At the end of maceration for T2 samples, corresponding to 35% berry WL across all treatments, the TF content in FD samples was 462±18 mg (+)-catechin/kg grapes, whereas MD and SD samples reached 759±28 and 820±30 mg (+)-catechin /kg grapes, respectively, indicating a reduction of 39%−44% in the first samples. A similar trend was observed for TPI values in dehydrated samples, increasing up to 120 h of maceration (Fig. 1e-f and Figure S2e-f), while in fresh grapes the peak was reached around 50 h (Figure S1c), after which concentrations remained stable. These maceration extraction kinetics are consistent with previous studies (Ferrero, Paissoni et al., 2025). Again, FD samples at T1 and T2 showed statistically lower TPI values during simulated maceration whereas the differences between MD and SD were only significant when the data were expressed per 100 berries (Figure S2e-f), highlighting the role of dehydration level in modulating polyphenol extractability.

MCP and FRV, representing respectively condensed tannins and oligomeric flavanols, also reached their maximum concentrations at 120 h of maceration for skins from fresh and dehydrated grapes, except for MCP for FD samples that reached a plateau in the early 24 h of maceration (Figure S1d-e, Fig. 1g-j, and Figure S2g-j). Similar extraction kinetics have been previously observed for 'Nebbiolo' grapes (Paissoni et al., 2017). As grape ripening progresses, cell wall porosity increases, hindering the release of flavanols from berry skins. A slower and reduced extraction of polymerized flavanols occurs due to their greater retention into the pores. However, changes in skin cell wall composition of overripe grapes favor the diffusion of these compounds into the wine because of structural degradation. The FD conditions may have promoted these changes (Miller et al., 2019; Quijada-Morín et al., 2015). The subsequent decline or plateau observed in the extraction curves may be the result of re-adsorption processes, where tannins and polymeric pigments bind to cell wall materials (Beaver et al., 2020). Once again, FD treatments consistently resulted in significantly lower MCP and FRV values than MD and SD at T1 and T2, except for MCP at 24 and 120 h of maceration at T2 (Fig. 1g-j, and Figure S2g-j). At 336 h of maceration for T2 dehydrated grapes, the concentration of condensed tannins was markedly lower by 30%−33% in FD grapes compared to MD and SD. Specifically, MCP values were 449±23 mg (−)-epicatechin/kg grapes in FD, 645±32 mg (−)-epicatechin/kg grapes in MD, and 671±9 mg (−)-epicatechin/kg grapes in SD. FRV contents followed a similar pattern, with FD yielding 305±19 mg (+)-catechin/kg grapes, while MD and SD reached 529±61 and 647±51 mg (+)-catechin/kg grapes, respectively. These results represent a 42%−53% reduction in FRV content in FD samples compared to the other treatments. The SD-treated grapes consistently showed higher MCP and FRV values when the results were expressed per 100 berries, although the differences were not always significant compared to MD. Therefore, the extractability at the end of maceration at T2 for FD, MD, and SD samples was, respectively, 66±12%, 63±15%, and 85±10% for FRV whereas 52±14%, 52±6%, and 81±20% for MCP.

These results demonstrated that FD grapes consistently exhibited significantly lower contents of extracted phenolic compounds. This outcome is most likely due to the greater oxidative degradation of phenolics induced by the harsher dehydration conditions. In addition, the dehydration process also influenced the release of oligomeric flavanols and condensed tannins from skins during simulated maceration, with the slow process increasing the extractability probably as a result of the higher degradation of skin cell wall during longer withering process (Zoccatelli et al., 2013). Given that polyphenols play a crucial role in determining wine quality, contributing to attributes such as color stability, flavor complexity, structure, astringency, and bitterness (Gutiérrez-Escobar et al., 2021), wines produced from fast dehydrated grapes may have lower sensory quality compared to those derived from a longer grape dehydration period.

In relation to seeds, all fresh grapes and dehydrated grapes under different treatments showed a consistent trend of increasing phenolic concentration (FRV, MCP, and TPI) during simulated maceration (Figure S1d-f, Fig. 1g-m, and Figure S2g-m). This finding corroborates the pattern observed by Ferrrero, Paissoni et al. (2025), reporting a constant increase of TPI during seed maceration for 'Nebbiolo', among other red varieties. Likewise, Canals et al. (2005) observed that while phenolic extraction from grape skins tends to stabilize after an initial increase, the extraction of phenolic compounds from seeds is initially marked by a low diffusion rate that intensifies approximately halfway through alcoholic fermentation. This late-stage acceleration is likely driven by the synergistic effects of seed hydration and the increasing ethanol concentration in the fermenting medium, which promotes the dissolution of the seed cuticle and facilitates the release of phenolic compounds (Bautista-Ortín et al., 2016; Rousserie et al., 2020). Hernández-Jiménez et al. (2012) reported that adequate seed hydration is essential for the effective release of phenolic substances. It is noteworthy that differences among the three treatments (FD, MD, and SD) were less significant for seed-derived compounds than for those extracted from skins. Moreover, statistical significance varied depending on whether the results were expressed per kg of grapes or 100 berries. For instance, at T1 sampling point, the FD sample exhibited a significantly higher TPI value when expressed per kg of grapes from 168 h of

maceration (Fig. 1k). However, when data were normalized to 100 berries, the SD and MD treatments showed higher TPI values also from 168 h of maceration (Figure S2k). A similar trend was observed at T2 sampling point, where FD exhibited a statistically significant higher TPI value at the end of maceration (336 h), with 2425±19 mg (−)-epicatechin/kg grapes, compared to 2267±33 mg (−)-epicatechin/kg grapes for MD and 2158±50 mg (−)-epicatechin/kg grapes for SD (Fig. 1m). Conversely, when the results were expressed per 100 berries, SD displayed the statistically significant higher IPT value from 120 h of maceration (Figure S2m). In the case of MCP content, the differences among treatments were generally not statistically significant at either T1 or T2, irrespective of how the results were expressed. Nonetheless, at the end of the maceration period for T2 samples, the SD treatment yielded the highest MCP content when expressed per 100 berries (Figure S2j). For the FRV content, the SD and MD treatments showed significantly higher values during the early stages of maceration (24 h), whereas the FD treatment exhibited the statistically significant higher values at 120 h and 336 h at T1, when expressed in mg (+)-catechin/kg grapes (Fig. 1g). This pattern was confirmed at T2, where FD again displayed the highest FRV values at 120 and 336 h (Fig. 1h). It is important to highlight that at the end of maceration for T2 samples, while the extractability was similar (around 40%) for low molecular flavanols, it differed for polymeric flavanols reporting 45±5%, 74±25%, and 80±16% for FD, MD, and SD, respectively.

Despite the higher potential content of polymeric tannins in FD samples, the extracted content at the end of the maceration process was similar to SD and MD as a consequence of reduced extractability. Dehydration causes notable physiological and structural alterations within the seed coat and internal tissues, reducing the release of phenolic compounds (Cadot et al., 2006; Rousserie et al., 2020). Lignification of the inner layers and outer integument may occur in different extension depending on the dehydration conditions, in agreement with an increase in the acoustic energy produced during a compression test of berry seeds through grape dehydration (Río Segade et al., 2016). During fermentative maceration, seeds are hydrated and ethanol concentration increases, reducing the physical barriers that typically limit the release of polyphenols, particularly condensed tannins, increasing their extractability. Interestingly, this extractability seems to be conditioned by the physiological and structural characteristics of seeds dehydrated under different environmental conditions, influencing both the rehydration ability and/or the re-adsorption of released polymeric tannins on seed cell wall materials (Giacosa et al., 2023; Rousserie et al., 2020). The lower extractability of these compounds is advantageous since they are directly involved in the bitterness and astringency of wines.

3.4. Anthocyanin profile during skin simulated maceration

At two critical points during simulated maceration, specifically at 24 h (representing the peak of anthocyanin extraction) and at 336 h (representing the endpoint of extraction), the anthocyanin profile of grape skin extracts was determined. These results are summarized in Table 2 for the T1 and T2 sampling stages. The specific anthocyanin forms identified through chromatographic separation exhibited trends consistent with the total anthocyanin content (TA) determined by spectrophotometry. 'Nebbiolo' grapes are known to have a distinctive anthocyanin profile dominated by di-substituted forms, particularly peonidin-3-*O*-glucoside, which is especially prone to oxidative degradation (Locatelli et al., 2016). These structural characteristics can hinder the formation of stable pigments and compromise long-term color retention and polymeric pigment development (Ferrero, Beria D'Argentina et al., 2025).

At 24 h for T1, the grapes subjected to SD treatment retained levels of the major anthocyanins, including delphinidin-3-*O*-glucoside, cyanidin-3-*O*-glucoside, peonidin-3-*O*-glucoside, petunidin-3-*O*-glucoside, malvidin-3-*O*-glucoside and their acetylated and cinnamoylated derivatives that were comparable to those found in fresh grapes whereas a concentration effect was observed for MD, particularly for non-acylated forms (Table 2). At 24 h for T2, only MD treatment preserved the concentration of individual anthocyanin forms. After 336 h of maceration, the SD samples showed the higher concentrations of all anthocyanin forms but they were lower than those found in fresh grapes at T2. Conversely, FD treatment consistently yielded significantly lower concentrations of nearly all anthocyanin forms, both at the early extraction stage (24 h) and after extended maceration (336 h) for T1 (Table 2). At T2, this pattern remained evident, with FD showing significantly reduced anthocyanin content compared to MD at 24 h, although differences between FD and SD at 24 h were not statistically significant. At 24 h and 336 h, FD-treated samples had the lowest concentrations for all anthocyanin forms, with a reduction exceeding 50% for total free anthocyanins compared to MD and SD (Table 2). These results agree with the effect of dehydration treatments on the anthocyanin profile of dehydrated grapes (Table 1), even though long maceration has produced a significant decrease in all forms, particularly peonidin-3-*O*-glucoside and cyanidin-3-*O*-glucoside that are more prone to oxidation. This

Table 2
Anthocyanin profile during simulated maceration of berry skins from fresh grapes and dehydrated grapes under different environmental conditions.

	Fresh grapes (-)	Dehydrated grapes T1				Dehydrated grapes T2			
Sample (weight loss)		FD (35%)	MD (18%)	SD (6%)	Sign	FD (35%)	MD (35%)	SD (35%)	Sign
24 h maceration									
Delphinidin-3-*O*-glucoside	34.1 ± 1.5	16.1 ± 1.6b	39.7 ± 3.7a	34.5 ± 1.6a	***	16.1 ± 1.6b	29.5 ± 4.6a	16.8 ± 0.8b	***
Cyanidin-3-*O*-glucoside	64.6 ± 7.7	37.4 ± 4.8b	78.7 ± 12.1a	62.9 ± 1.4a	**	37.5 ± 4.7ab	53.1 ± 8.3a	33.9 ± 3.0b	**
Petunidin-3-*O*-glucoside	29.2 ± 1.1	14.8 ± 1.7b	34.7 ± 3.0a	29.9 ± 1.2a	***	14.8 ± 1.7b	27.5 ± 4.4a	15.3 ± 0.5b	***
Peonidin-3-*O*-glucoside	202.5 ± 8.3	114.4 ± 21.3b	254.0 ± 29.6a	214.8 ± 11.6a	***	114.7 ± 21.2b	205.1 ± 36.4a	121.5 ± 4.0b	**
Malvidin-3-*O*-glucoside	137.5 ± 10.6	63.2 ± 9.0b	162.7 ± 14.8a	139.5 ± 11.3a	***	63.4 ± 9.0b	133.4 ± 21.5a	74.7 ± 2.9b	***
Sum acetyl-glucosides	30.0 ± 1.6	14.8 ± 4.2b	32.7 ± 3.4a	26.6 ± 2.1a	***	14.8 ± 4.2ab	21.5 ± 5.2a	9.7 ± 1.5b	***
Sum cinnamoyl-glucosides	40.7 ± 2.5	19.4 ± 3.2b	44.6 ± 3.0a	41.0 ± 3.4a	***	19.5 ± 3.2b	33.5 ± 6.9a	18.6 ± 1.6b	***
336 h maceration									
Delphinidin-*O*-3-glucoside	2.8 ± 0.7	0.5 ± 0.1b	1.9 ± 0.1a	2.6 ± 0.1a	***	0.5 ± 0.1b	1.4 ± 0.1a	1.8 ± 0.4a	***
Cyanidin-3-*O*-glucoside	3.5 ± 1.6	0.7 ± 0.2b	1.5 ± 0.2ab	2.8 ± 0.7ab	*	0.7 ± 0.2b	1.6 ± 0.1ab	1.8 ± 1.2ab	*
Petunidin-3-*O*-glucoside	5.0 ± 0.7	1.2 ± 0.1b	3.8 ± 0.4a	4.7 ± 0.4a	***	1.2 ± 0.1b	2.8 ± 0.2a	3.4 ± 0.4a	***
Peonidin-3-*O*-glucoside	30.4 ± 7.7	7.3 ± 0.8b	18.7 ± 1.3a	28.1 ± 1.7a	***	7.3 ± 0.7b	18.2 ± 0.9a	20.4 ± 6.2a	**
Malvidin-3-*O*-glucoside	46.0 ± 3.1	13.6 ± 1.1b	36.3 ± 5.7a	40.6 ± 1.5a	***	13.6 ± 1.1c	31.3 ± 2.6b	38.1 ± 2.2a	***
Sum acetyl-glucosides	4.4 ± 1.3	1.1 ± 0.3b	4.7 ± 0.9a	4.8 ± 2.1a	*	1.1 ± 0.3b	2.3 ± 1.4ab	1.9 ± 0.2ab	**
Sum cinnamoyl-glucosides	4.8 ± 0.5	1.6 ± 0.6b	4.2 ± 0.5a	5.3 ± 0.4a	***	1.6 ± 0.6b	3.0 ± 1.0ab	3.7 ± 0.1a	***

All data are expressed as average value ± standard deviation (n = 3). Sign: *, **, and *** indicate significance at $p < 0.05$, 0.01, and 0.001, respectively. Different Latin letters within the same row indicate significant differences among treatments inside each time point according to Tukey test ($p < 0.05$). Anthocyanin compounds are expressed as mg malvidin-3-*O*-glucoside chloride/kg grape. FD, fast rate dehydration; MD, medium rate dehydration; SD, slow rate dehydration; T1, time at which FD-treated grapes reached 35% weight loss (15 days; time-matched comparison point); T2, 35% weight loss for all treatments (weight loss-matched comparison point).

decrease, particularly significant for di-substituted forms of anthocyanins, is consistent with previous studies carried out on the 'Nebbiolo' variety during fermentative macerations on lab-scale winemaking (Ferrero, Beria D'Argentina et al., 2025). The pronounced decline in anthocyanin content over time, especially in FD samples, underscores the impact of oxidation reactions and polymerization processes occurring throughout maceration. In the early stages of dehydration, water loss prevents oxygen intake. When dehydration progresses, the stress induced by rapid water loss appears to accelerate skin collapse, microcracking, and tissue rupture. These alterations likely enhance oxygen permeability and disrupt cellular compartmentalization, thereby triggering phenolic oxidation pathways (Zheng et al., 2021). Regarding the faster water loss relative to FD treatment, the changes in the skin microstructure could have promoted the faster extraction of anthocyanins, particularly di-substituted forms, which are more easily oxidized, and therefore their concentration significantly decreased during maceration.

3.5. Color evolution during skin simulated maceration

Monomeric anthocyanins are the principal pigments responsible for the red to purple hues observed in red grape varieties and in the resulting wines. These water-soluble flavonoid compounds are predominantly located in the grape skins and are gradually extracted into the must during the maceration phase of red winemaking. This extraction occurs via passive diffusion and is influenced by factors such as skin integrity, temperature, ethanol concentration, and time (Ferrero, Beria D'Argentina et al., 2025). The extraction curves of anthocyanins in model wine solution, shown in Fig. 1a-b and Figure S2a-b, affected the color evolution of skin extracts during maceration as can be observed in Fig. 2. The visual colors were obtained by the conversion of the CIELab coordinates in the corresponding RGB scale. The color differences among treatments were visually perceptible during simulated maceration at T1 and T2, achieving ΔE^* values (total color difference) higher than 7 for MD and SD treatments with respect to FD at the end of maceration (336 h). At T1, after 336 h of maceration, the impact of different dehydration treatments on CIELab coordinates was already significant, with lower values of L^* (lightness), a^* (redness), and b^* (yellowness) for FD-treated grapes (Table 3). Therefore, skin extracts derived from FD grapes appear darker with lower red and yellow hues (Fig. 2). At T2, higher differences in color parameters emerged among treatments with MD-treated grapes displaying significantly higher values of a^*, chroma (C^*), and color intensity compared to those treated with either FD or SD. Interestingly, the FD treatment outperformed SD in color traits (ΔE^* value of 9.7), as evidenced by a higher value of a^* and lower ones of L^*, b^*, H^*, and hue, indicating a shift toward redness rather than yellowish (Table 3, Fig. 2). It could be due to the higher percentage of monomeric anthocyanins (MP%) for the skin extracts from FD-treated grapes, while the MD treatment led to a greater formation of large polymeric pigments (LPP%). FD samples displayed significantly lower anthocyanin and flavanol concentrations overall (Table 3). In addition, a lower initial free anthocyanin concentration in skin extracts would have limited the formation of anthocyanin–flavanol derivatives. Monomeric anthocyanins are highly unstable and are particularly vulnerable to oxidation (Tindal et al., 2021). FD may have preserved the color at the end of extended maceration, but the absence of stable pigments likely compromises color stability over time. The higher LPP% value for MD samples could enhance pigment stability through the formation of anthocyanin-tannin condensation products. Anthocyanin–tannin and anthocyanin–anthocyanin adducts play a key role in color stabilization by shielding the chromophore from nucleophilic attack at the C4 position. This protection reduces vulnerability to bleaching by bisulfite ion and hydrolytic reactions, thereby improving pigment persistence throughout aging (Boulton, 2001; He et al., 2012). A joint skins and seeds maceration probably would have a similar impact on the formation of polymeric pigments for the FD, MD, and SD treatments since the concentration of tannins (MCP value) in the seed extracts at the end of maceration was not significantly different among treatments (Table 3).

3.6. Berry mechanical properties

Finally, the mechanical properties of the skin and whole berries were studied to assess whether the differences in the diffusion of phenolic compounds during skin maceration could be due to changes in structural characteristics after grape dehydration. The berry skin texture parameters, such as skin break force (F_{sk}), skin break energy (W_{sk}), and skin thickness (Sp_{sk}), increased with the fast dehydration (FD) whereas skin stiffness (E_{sk}) decreased (Table 4). When compared with other dehydration techniques at the same time (T1), the first three skin mechanical properties were significantly higher for the FD treatment compared to MD and SD as a consequence of the higher water loss, resulting in harder

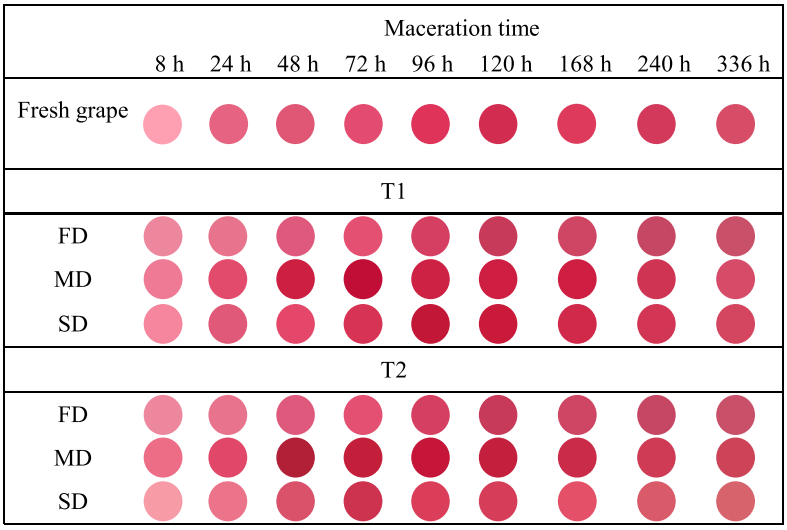

Fig. 2. Color evolution during skin simulated maceration for fresh and dehydrated grapes under different environmental conditions. Each color was obtained by conversion of CIELab coordinates to RGB values. FD, fast rate dehydration; MD, medium rate dehydration; SD, slow rate dehydration; T1, time at which FD-treated grapes reached 35% weight loss (15 days; time-matched comparison point); T2, 35% weight loss for all treatments (weight loss-matched comparison point).

Table 3
Phenolic composition of skin and seed extracts and color traits of skin extracts at the end of simulated maceration (336 h) from fresh grapes and dehydrated grapes under different environmental conditions.

Sample (weight loss)	Fresh grapes (-)	Dehydrated grapes T1				Dehydrated grapes T2			
		FD (35%)	MD (18%)	SD (6%)	Sign	FD (35%)	MD (35%)	SD (35%)	Sign
Grape skins									
TPI (mg (−)-epicatechin/kg grape)	1673±60	1051±9b	1655±90a	1619±32a	***	1051±9b	1484±63a	1591±71a	***
TF (mg (+)-catechin/kg grape)	839±38	462±18b	862±39a	874±40a	***	462±18c	759±28b	820±29a	***
TA (mg malvidin-3-*O*-glucoside chloride/kg grape)	141±7	70±2b	128±11a	138±10a	***	70±2b	110±6a	102±9a	***
FRV (mg (+)-catechin/kg grape)	673±12	305±19c	739±25a	609±23b	***	305±19b	529±61a	647±51a	***
MCP (mg (−)-epicatechin/kg grape)	599±19	449±23b	637±20a	628±37a	***	449±23b	646±32a	671±8a	***
MP%	74.05±0.76	67.99 ±2.01b	70.90 ±0.34ab	72.39 ±1.18a	*	67.99 ±2.01a	62.10 ±0.62b	63.90 ±0.92b	**
SPP%	13.96±0.59	11.77 ±1.04a	14.42±2.40a	12.92 ±1.36a	ns	11.77 ±1.04a	9.43±0.66b	12.51 ±0.73a	**
LPP%	11.99±0.83	20.23 ±1.97a	14.68±2.62a	14.69 ±2.17a	ns	20.23 ±1.97b	28.47 ±0.85a	23.59 ±1.65b	**
SPP+LPP%	25.95±0.76	32.01 ±2.01a	29.10 ±0.34ab	27.61 ±1.18b	*	32.01 ±2.01b	37.90 ±0.62a	36.10 ±0.92a	**
*L**	52.5 ± 0.1	50.8 ± 2.4a	52.2 ± 2.1a	50.8 ± 1.6a	ns	50.8 ± 2.4b	49.3 ± 1.6b	56.5 ± 2.2a	*
*a**	56.53±0.45	50.28 ±0.46b	56.70±2.27a	57.97 ±2.10a	**	50.28 ±0.46b	55.17 ±1.04a	46.53 ±1.74c	***
*b**	14.86±2.83	10.52 ±0.65b	13.50 ±0.93ab	16.39 ±2.66a	*	10.52 ±0.65b	18.78 ±1.37a	17.39 ±3.86a	*
*C**	58.50±0.29	51.37 ±0.47b	58.29±2.27a	60.29 ±1.71a	**	51.37 ±0.47b	58.30 ±0.68a	49.74 ±2.90b	**
*H**	14.72±2.79	11.82 ±0.72a	13.40±0.93a	15.81 ±2.80a	ns	11.82 ±0.72b	18.81 ±1.55a	20.38 ±3.51a	**
Color intensity	2.38±0.06	2.19±0.16a	2.36±0.17a	2.53±0.16a	ns	2.19±0.16b	2.58±0.11a	2.07±0.18b	*
Hue	0.63±0.02	0.65±0.01a	0.62±0.02a	0.59±0.03a	ns	0.65±0.01b	0.69±0.02b	0.84±0.01a	***
Grape seeds									
TPI (mg (−)-epicatechin/kg grape)	1407±167	2425±19a	2112±76b	1786±50c	***	2425±19a	2267±23b	2158±50c	***
FRV (mg (+)-catechin/kg grape)	797±84	1457±46a	1182±49b	959±61c	***	1457±46a	1128±30b	1176±47b	***
MCP (mg (−)-epicatechin/kg grape)	677±118	1053±50a	1008±62a	942±59a	ns	1053±50a	1108±78a	1049±19a	ns

All data are expressed as average value ± standard deviation (n = 3). Sign: *, **, ***, and ns indicate significance at $p < 0.05$, 0.01, 0.001, and not significant differences, respectively. Different Latin letters within the same row indicate significant differences among treatments inside each time point according to Tukey test ($p < 0.05$). TPI, total phenol index; TF, total flavonoids; TA, total anthocyanins; FRV, flavanols reactive to vanillin; MCP, methyl cellulose precipitable tannins; MP, monomeric pigments; SPP, small polymeric pigments; LPP, long polymeric pigments; *L**, lightness; *a**, red/green color coordinate; *b**, yellow/blue color coordinate; *C**, chrome; *H**, hue angle; FD, fast rate dehydration; MD, medium rate dehydration; SD, slow rate dehydration; T1, time at which FD-treated grapes reached 35% weight loss (15 days; time-matched comparison point); T2, 35% weight loss for all treatments (weight loss-matched comparison point).

Table 4
Grape texture parameters of berry skins and whole berries from fresh grapes and dehydrated grapes under different environmental conditions.

Sample (weight loss)	Fresh grapes (-)	Dehydrated grapes T1				Dehydrated grapes T2			
		FD (35%)	MD (18%)	SD (6%)	Sign	FD (35%)	MD (35%)	SD (35%)	Sign
Berry skin parameters									
F_{sk} (N)	0.548±0.102	0.670±0.135a	0.534±0.138b	0.523±0.095b	***	0.670±0.135a	0.591±0.193a	0.613±0.175a	ns
W_{sk} (mJ)	0.495±0.127	0.902±0.264a	0.516±0.167b	0.496±0.121b	***	0.902±0.264a	0.671±0.311b	0.731±0.252b	***
E_{sk} (N/mm)	0.267±0.035	0.184±0.036b	0.223±0.040a	0.236±0.032a	***	0.184±0.036a	0.189±0.060a	0.185±0.050a	ns
Sp_{sk} (μm)	189±34	206±42a	167±42b	190±38ab	*	206±42a	207±44a	201±28a	ns
Whole berry parameters									
BH (N)	3.214±0.398	1.608±0.661c	2.509±0.654b	3.209±0.626a	***	1.608±0.661a	1.576±0.714a	1.488±0.730a	ns
BCo (-)	0.728±0.025	0.617±0.047c	0.667±0.082b	0.738±0.027a	***	0.617±0.047a	0.575±0.075b	0.583±0.079b	*
BG (N)	2.333±0.244	1.000±0.441c	1.690±0.503b	2.368±0.467a	***	1.000±0.441a	0.942±0.528a	0.902±0.521a	ns
BR (-)	0.376±0.027	0.211±0.040c	0.291±0.054b	0.358±0.022a	***	0.211±0.040ab	0.222±0.056a	0.195±0.060b	*
BS-*r* (-)	0.870±0.016	0.710±0.065b	0.727±0.077b	0.846±0.035a	***	0.710±0.065a	0.555±0.097b	0.659±0.122a	***

All data are expressed as average value ± standard deviation (n = 30). Sign: *, ***, and ns indicate significance at $p < 0.05$, 0.001, and not significant differences, respectively. Different Latin letters within the same row indicate significant differences among treatments inside each time point according to Tukey test ($p < 0.05$). F_{sk}, berry skin break force; W_{sk}, berry skin break energy; E_{sk}, berry skin stiffness; Sp_{sk}, berry skin thickness; BH, berry hardness; BCo, berry cohesiveness; BG, berry gumminess; BR, berry resilience; BS-*r*, berry springiness ratio; FD, fast rate dehydration; MD, medium rate dehydration; SD, slow rate dehydration; T1, time at which FD-treated grapes reached 35% weight loss (15 days; time-matched comparison point); T2, 35% weight loss for all treatments (weight loss-matched comparison point).

and thicker skins but, at the same time, more elastic ones according to the lower values of skin stiffness. These differences could be due to gradual skin cell reorganization and to the modifications of cell wall polysaccharides (Scalzini et al., 2024). The effects were not as evident at T2 which considers the same weight loss for all treatments, but significantly higher W_{sk} values were still observed for FD suggesting that factors like temperature, humidity, and time influence how dehydration impacts skin mechanical properties (Zsófi et al., 2014). These structural changes are consistent with differences observed in phenolic extraction during maceration. The increase in skin thickness for MD and SD treatments from T1 to T2 has led to a less anthocyanin extraction during maceration (Fig. 1a-b and Figure S2a-b). It is known that higher values of Sp_{sk} are linked to a lower diffusion of anthocyanins from skins during the maceration phase (Río Segade et al., 2011).

For whole berry measurements (Table 4), at T1, the SD treatment better preserved hardness (BH), cohesiveness (BCo), gumminess (BG), resilience (BR), and springiness ratio (BS-*r*), maintaining values closer to those of fresh grapes. On the other hand, FD caused a notable decrease in these parameters, probably because rapid water loss led to cell collapse and structural integrity losses. In addition, water loss reduces cell turgor and, therefore, berry hardness decreases due to a lower rupture force required (De Belie et al., 1999). As for skin texture parameters, few statistical differences were observed at T2, but the FD treatment usually reported the highest values indicating that environmental conditions also mediate the dehydration impact on berry texture properties. The changes in cell wall polysaccharides occurring during withering lead to grape softness and improved diffusion of anthocyanins from grape skin. Zouid et al. (2013) have reported high negative correlation coefficients with anthocyanin extractability for berry hardness, gumminess, and springiness in grapes with different total soluble sugar content.

4. Conclusions

Grape postharvest dehydration using cold drying technology resulted in the fastest dehydration process among the three treatments considered. FD-treated grapes reached 35% weight loss in 15 days whereas MD in 27 days and SD in 60 days. The results showed that dehydration rate strongly influences the polyphenol content. Under these conditions, the FD treatment did not preserve skin polyphenols, especially anthocyanins, whose contents are already naturally low and they are easily degradable in 'Nebbiolo'. Given that anthocyanins play an essential role in wine color preservation, the higher percentage of monomeric forms at the end of maceration produced darker and redder skin extracts than the SD treatment. However, these forms are more susceptible to oxidation during aging. In contrast, seeds from FD-treated grapes showed higher flavanol concentrations compared to SD and MD treatments, suggesting that their extraction must be monitored during maceration to avoid excessive astringency and bitterness. Overall, the MD treatment provided the most favorable outcomes. Although it did not result in the highest values for all phenolic parameters, MD showed better anthocyanin retention in grapes and extracts, as well as improved color preservation at the end of simulated maceration through the formation of polymeric pigments. The SD treatment also preserved skin polyphenols, but the resulting skin extracts were less dark and more yellowish. These findings suggest that the dehydration rate, rather than temperature alone, plays a key role in preserving secondary metabolites and in maintaining the phenolic integrity of grapes, especially for varieties like 'Nebbiolo' with an important share of anthocyanin forms prone to oxidation.

A modified fast dehydration approach by slightly extending the dehydration time could improve polyphenol preservation without requiring excessively long drying times. Shorter dehydration processes remain advantageous because prolonged withering under high humidity conditions increases the risk of mold development. Further studies on other cultivars and dehydration rates could help confirm and extrapolate these findings. This study contributes to a deeper understanding of how postharvest handling affects the composition of grapes destined to the production of special wines.

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Ethical statement - studies in humans and animals

This study does not involve human or animal subjects.

Data availability

Data will be made available on request.

Ethical statement - studies in humans and animals

The authors declare that this study does not involve human or animal subjects.

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Beatrice Cordero: Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Negin Seif Zadeh:** Writing – review & editing, Investigation, Formal analysis, Data curation. **Alessandro Biglia:** Writing – review & editing, Formal analysis, Data curation. **Alberto Caudana:** Writing – review & editing, Resources. **Maria Alessandra Paissoni:** Writing – review & editing, Validation, Methodology. **Davide Ricauda Aimonino:** Writing – review & editing, Methodology, Conceptualization. **Simone Giacosa:** Writing – review & editing, Validation, Methodology, Formal analysis. **Luca Rolle:** Writing – review & editing, Resources, Methodology, Funding acquisition, Conceptualization. **Susana Río Segade:** Writing – review & editing, Visualization, Supervision, Methodology, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Supplementary materials

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