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TESAF

Dipartimento Territorio
e Sistemi Agro-Forestali
Università di Padova



Effect of storage on stilbenes contents in cv. *Pinot Noir* grape canes collected at different times before pruning

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Global grape production has been estimated in 2014 (OIV, 2015)

- ❖ Europe: 41%
- ❖ Asia: 29%
- ❖ America: 21%

80% of the cultivated grape is used for winemaking

Pesticides



Sustainability



Society demand

Generation of by-products



Grape canes

(Muir et al., 2004; Zhu et al., 2015)

Luckily, there is a tendency to replace the:



Grapes canes



Rich source of
resveratrol

(Raynea, Karacabeya and Mazza, 2008)



To the best quality grapes, some practices including irrigation, fertilization and **pruning** should be performed in grapevines, annually.



The main purpose of **pruning** is to keep a balance between



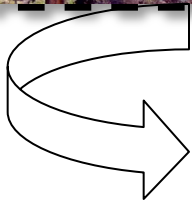
Vegetative
growth



Fruit
production

(Tassie and Freeman, 2001)

Grape canes represent a large source of waste derived from the viticulture, with an estimated volume of 1–5 tons hectare year.



Burnt



Composted





However, **grape canes waste** can be a **source of stilbenes** as proposed by several authors (Karacabey and Mazza, 2008, 2012; Rayne, Karacabey and Mazza, 2008; Vergara et al., 2011; Zhang et al., 2011; Çetin et al., 2011; Vergara et al., 2011; Lambert et al., 2013; Gorena et al., 2014; Houillé et al., 2015).



Source of Stilbenes

Phenolic
compounds

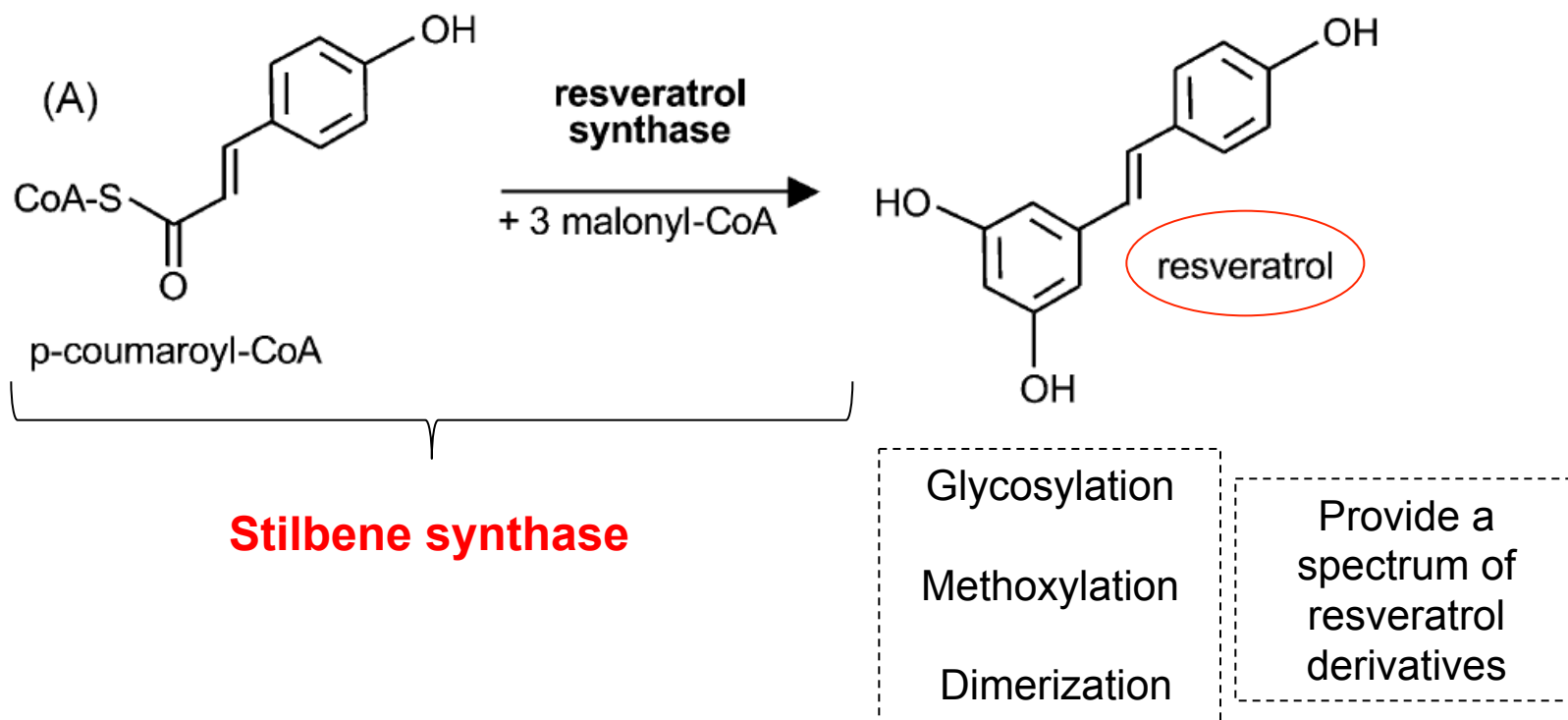
Secondary
metabolism

Accumulated in
response to stress



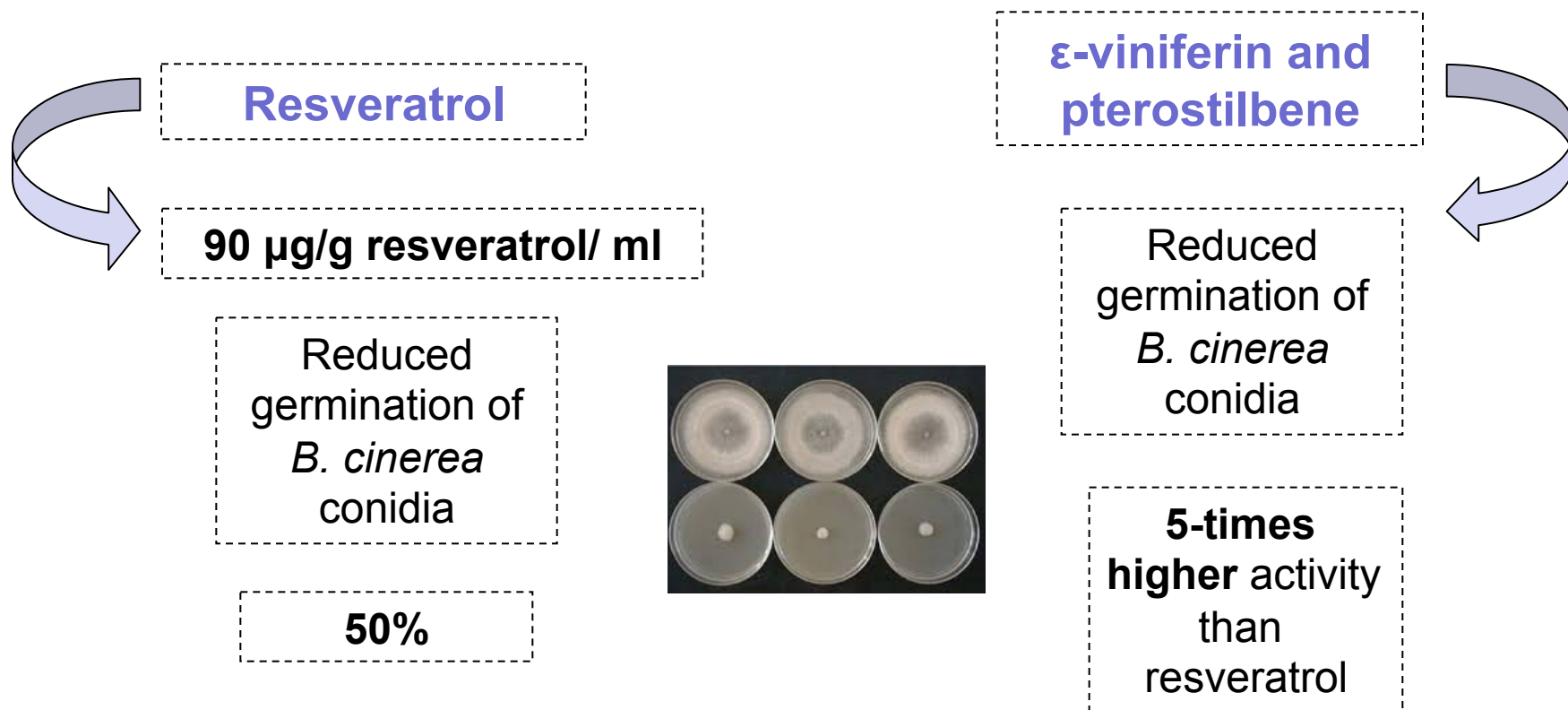
(Nürnberger et al., 2004; Kuc, 1995; Jeandet et al., 2013; Hart and Shrimpton, 1979; Hart, 1981)

Most plant stilbenes are derivatives of the basic unit **trans-resveratrol** (3,5,4'-trihydroxy-transstilbene).



(Chong, Poutaraud and Hugueney, 2009)

The antifungal activity X stilbenes



In **grape canes**, stilbenes are accumulated at high concentrations in the heartwood.



Different grape canes varieties

Pinot noir



Collected before pruning



Stored at room temperature



Increased concentration of stilbenes

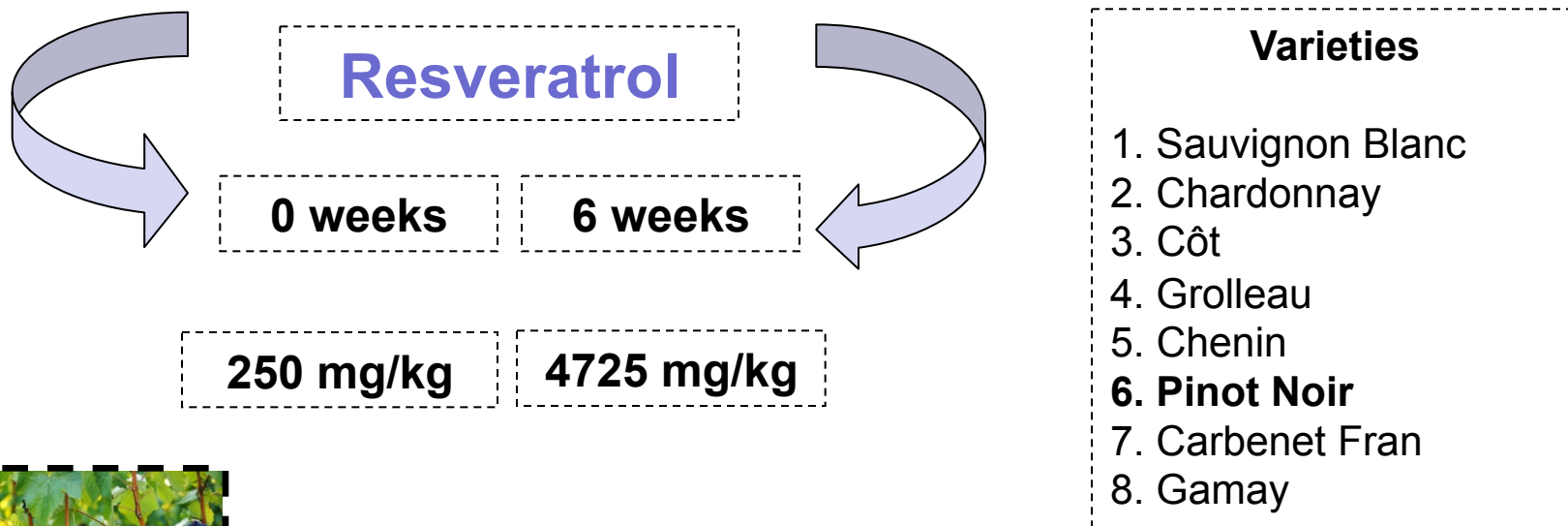


Mechanisms is not known yet

(Hart and Shrimpton, 1979; Hart, 1981; Karacabey and Mazza, 2008; Vergara et al., 2012; Zhang et al., 2011; Houillé et al., 2015; Gorena et al., 2014).



According to the study performed by authors Houillé et al. (2015) was evaluated the resveratrol content in 8 varieties of grape canes after 6 weeks of storage and reported that the resveratrol content in Pinot Noir canes had the highest accumulation of resveratrol.





According to the study performed by authors Rayne, Karacabey and Mazza (2008) was evaluated 16 different reagents for the extraction of *trans*-resveratrol and *trans*-viniferin in *Pinot Noir* canes.

80% Ethanol: 20% Water

***trans*-
Resveratrol**

3450 mg/kg

***trans*-
Viniferin**

1300 mg/kg

Effective in the
extraction of both
compounds

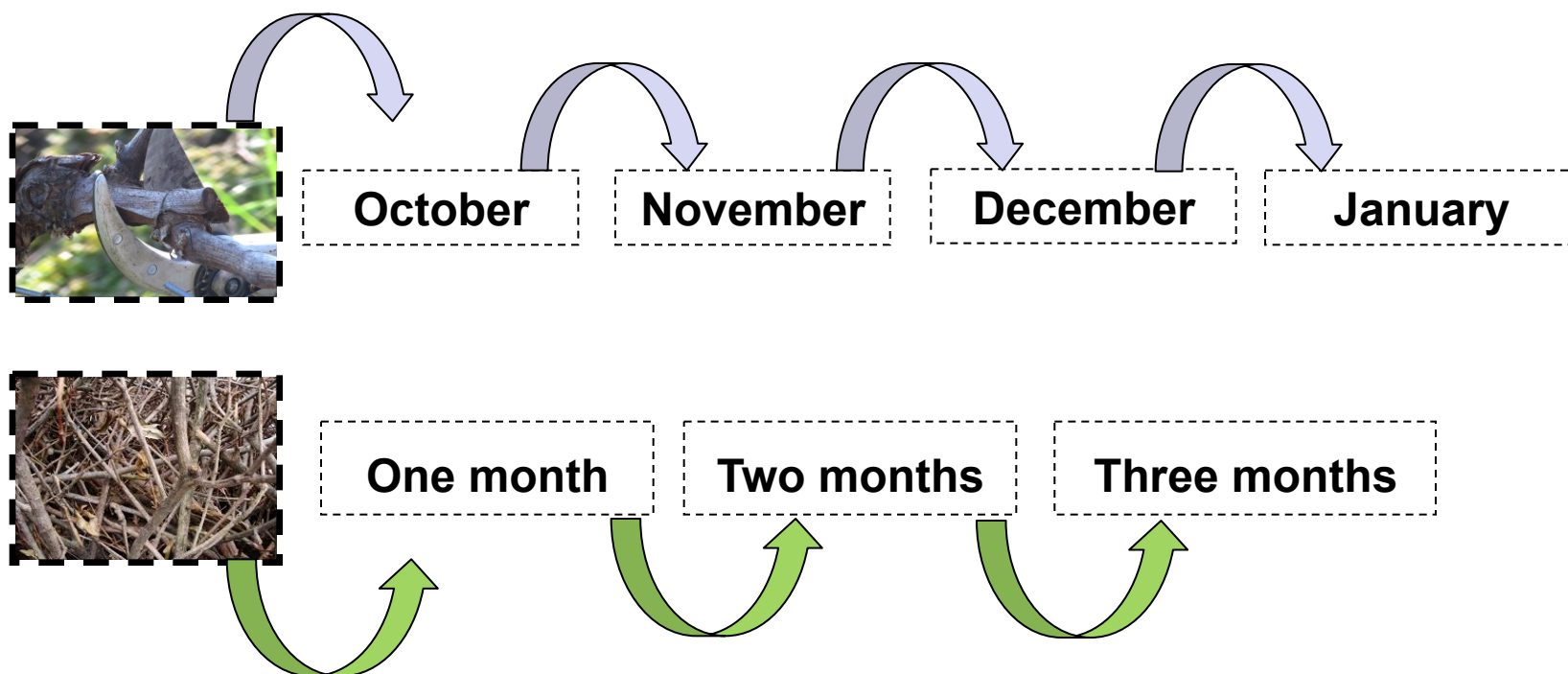
Reagents

1. Diethyl ether
2. 2-propanol
3. Acetone
4. Ethanol
5. 95% ethanol:5% water
6. Methanol
7. **80% ethanol:20% water**
8. Acetonitrile
9. Dimethylformamide
10. 70% ethanol:30% water
11. 54% dimethylsulfoxide:46% acetone
12. Dimethylsulfoxide
13. 60% ethanol:40% water
14. 50% ethanol:50% water
15. 50% methanol:50% water
16. 30% ethanol:70% water

(Rayne, Karacabey and Mazza 2008)



The objective of the study was to evaluate the content of stilbenes in *Pinot noir* canes collected at different times before pruning, and stored for one, two or three months at room temperature.



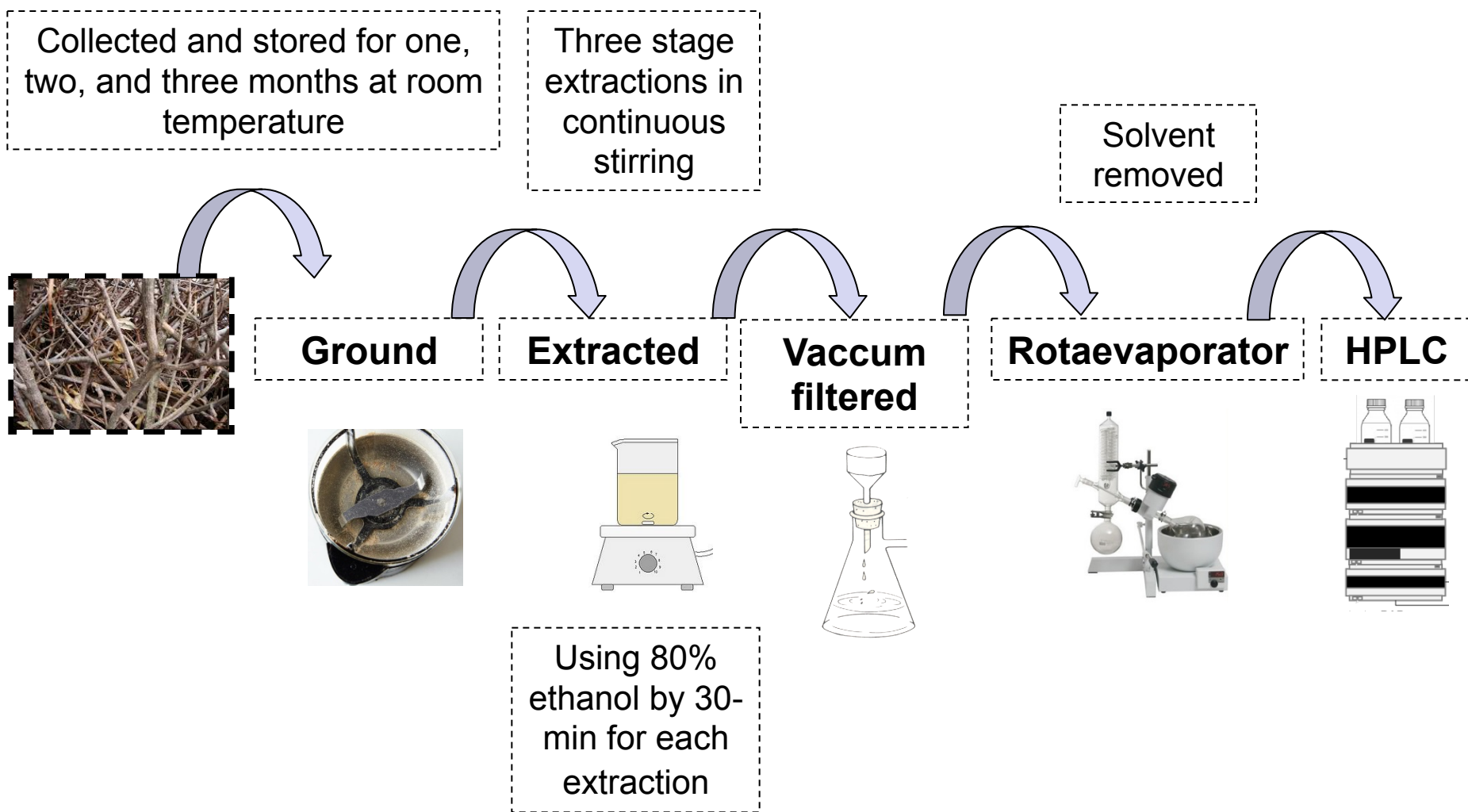




Table 1. Content of stilbenes in the Pinot Noir grape canes collected in different times before pruning.

Months	October			November		
	Control sample	*RT1	*RT2	Control sample	*RT1	*RT2
Resveratrol (mg/Kg)	65,84cB	640,92bB	1431,25aB	80,99cA	1228,81bA	3124,19aA
Viniferin (mg/Kg)	1511,69cA	1761,13aA	1626,68bB	1260,28bB	1122,79cB	3520,41aA
Piceatannol (mg/Kg)	31,51cB	324,92bB	557,64aB	34,07cA	528,22bA	1369,67aA

Means followed by the same letter on the line and capitalized in the column do not differ statistically by Tukey test at the 5% level of significance.

*RT1 (1 month at room temperature)

*RT2 (2 months at room temperature)



Table 2. Content of stilbenes in the Pinot Noir grape canes collected in different times before pruning

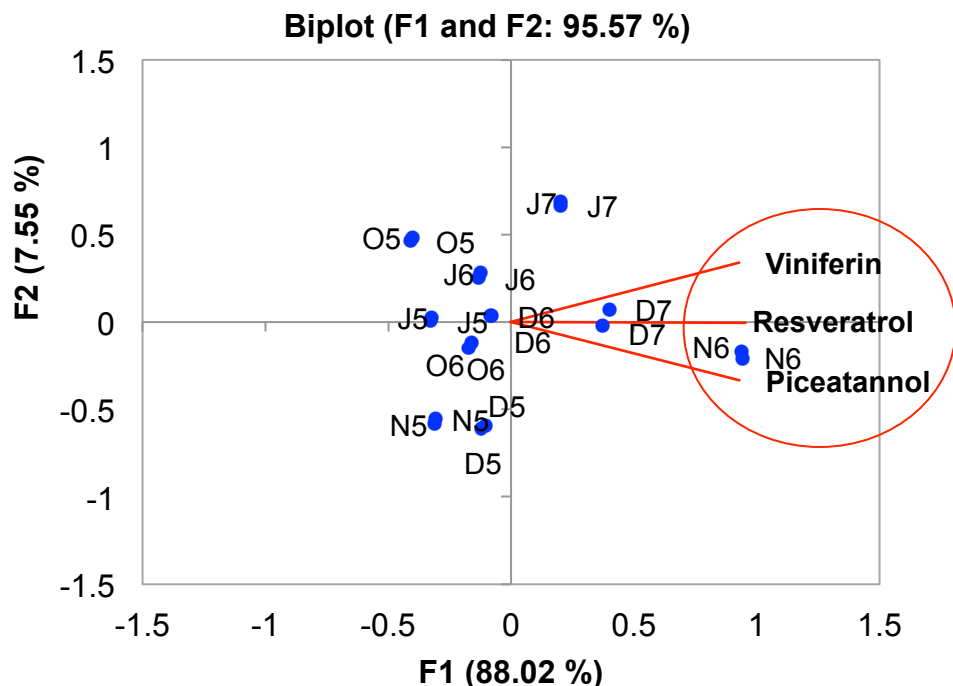
Months	December				January			
Treatment	Control sample	*RT1	*RT2	*RT3	Control sample	*RT1	*RT2	*RT3
Resveratrol (mg/Kg)	45,62 dB	1516,14 cA	1780,04 bA	2850,21 aA	92,31 dA	1173,47 cA	1430,62 bB	2268,14 aB
Viniferin (mg/Kg)	1449,30 cA	1446,61 cA	1778,24 bB	2424,15 aB	1161,68 dB	1446,83 cA	1951,85 bA	2616,91 aA
Piceatannol (mg/Kg)	26,32 dB	708,61 bA	536,54 cA	804,81 aA	38,82 dA	410,89 cB	499,94 bB	574,75 aB

Means followed by the same letter on the line and capitalized in the column do not differ statistically by Tukey test at the 5% level of significance.

*RT1 (1 month at room temperature)

*RT2 (2 months at room temperature)

*RT3 (3 months at room temperature)



Our results about resveratrol

2 months stored

81 mg/Kg --- 3200 mg/Kg

Increased 40 times

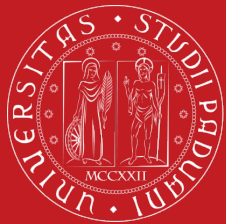
Houillé et al., 2015 resveratrol

1.5 months stored

250 mg/Kg --- 4725 mg/Kg

Increased 19 times

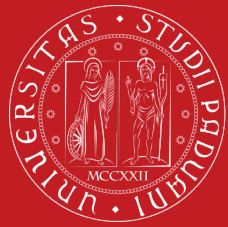
Figure 1. Principal component analysis of the stilbenes content in Pinot Noir canes collected in October (O), **November (N)**, December (D) 2015 and January (J) 2016 being that: (5) = 1 month at room temperature (RT); **(6) = 2 month at RT**; (7) = 3 month at RT.



To study the mechanisms which increase the content of stilbenes in the grape canes collected before pruning and stored at room temperature.

Stilbenes synthase





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Thank you for your attention!

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