



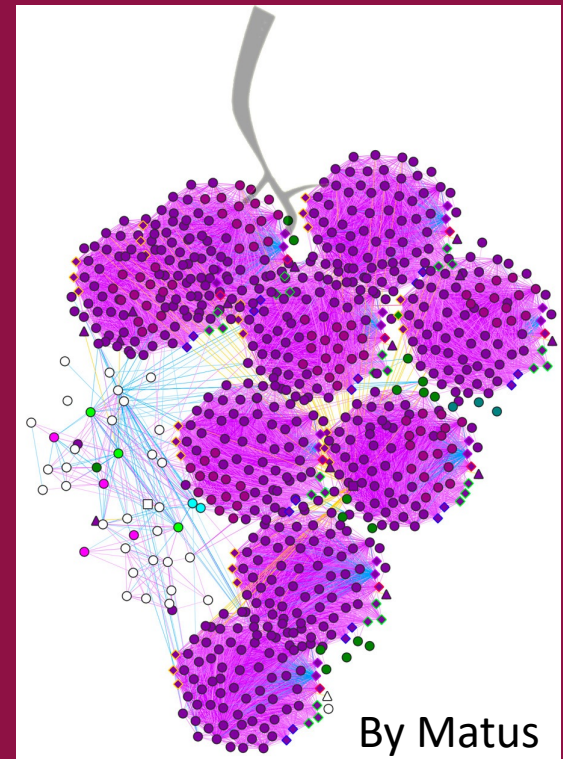
fondazione banfi

SANGUIS JOVIS

ALTA SCUOLA DEL SANGIOVESE

Maturazione delle bacche d'uva :
reti regolative associate a processi
endogeni e fattori ambientali

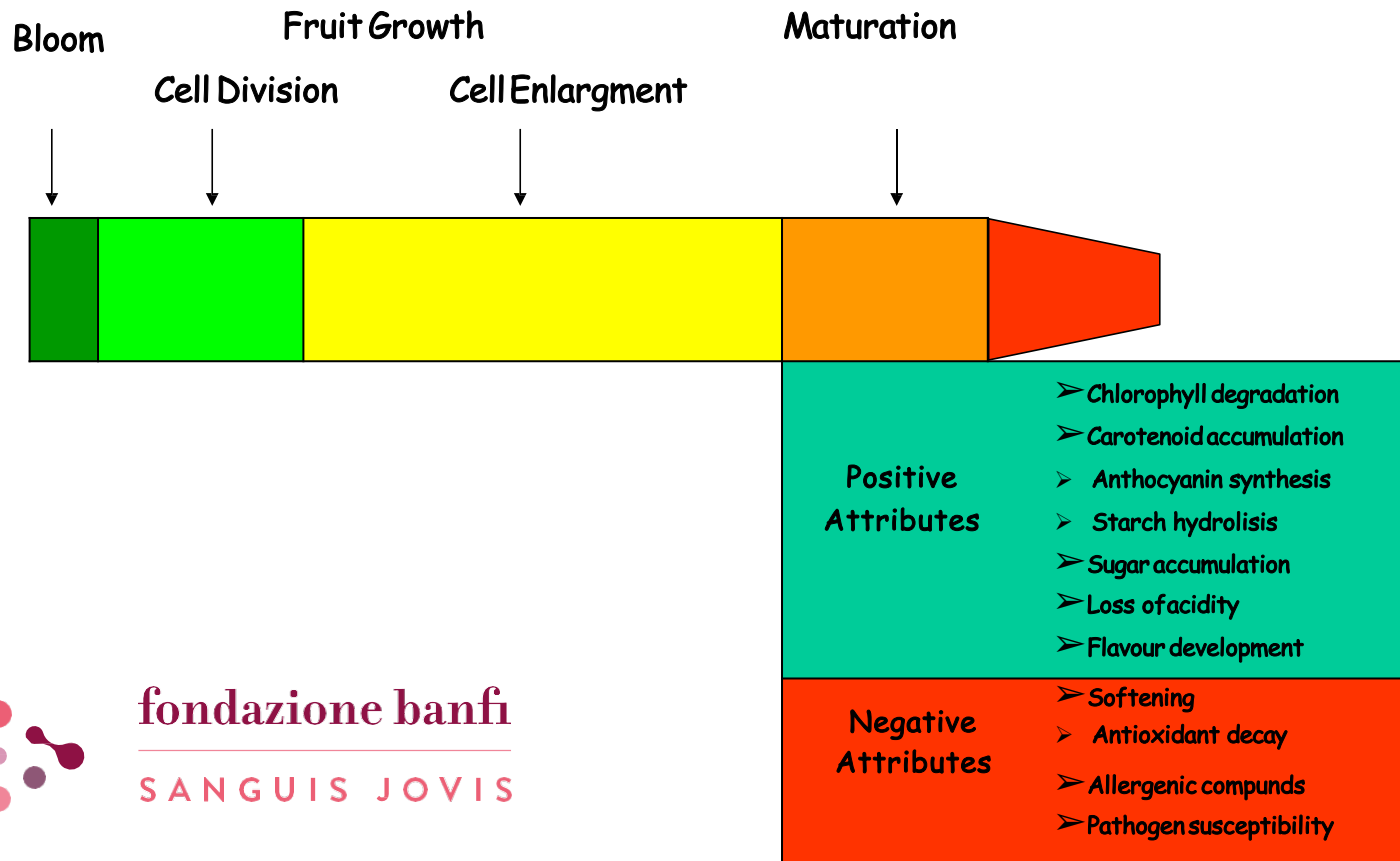
Claudio Bonghi
Dipartimento di Agronomia, Animali,
Alimenti, Risorse Naturali e Ambiente
UNIPD



Maturazione dei frutti

La maturazione è una sindrome fisiologica che, anche se presenta alcuni aspetti comuni, si differenzia marcatamente dall'ultima fase di sviluppo rappresentata dalla senescenza.

La maturazione è caratterizzata da un insieme di processi strettamente regolati da un punto di vista genetico ed epigenetico



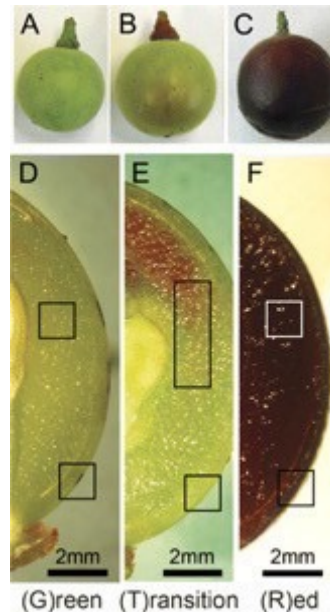
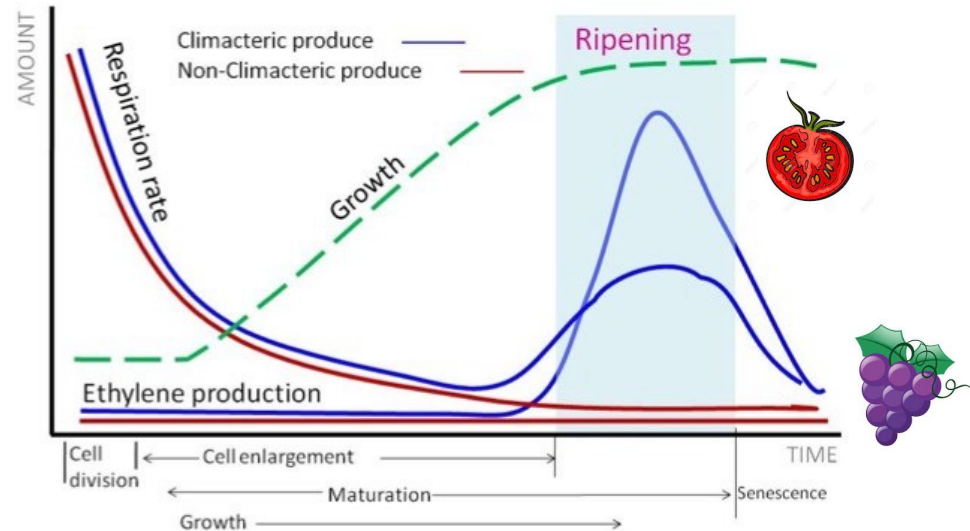
Maturazione della bacca della vite

La bacca d'uva è un frutto non climaterico

È un processo molto lungo che dura circa 2 mesi in cui si verifica una profonda modificazione della composizione della bacca

E' fortemente condizionato dall'ambiente

Inizia nella polpa in prossimità della fine dello stilo



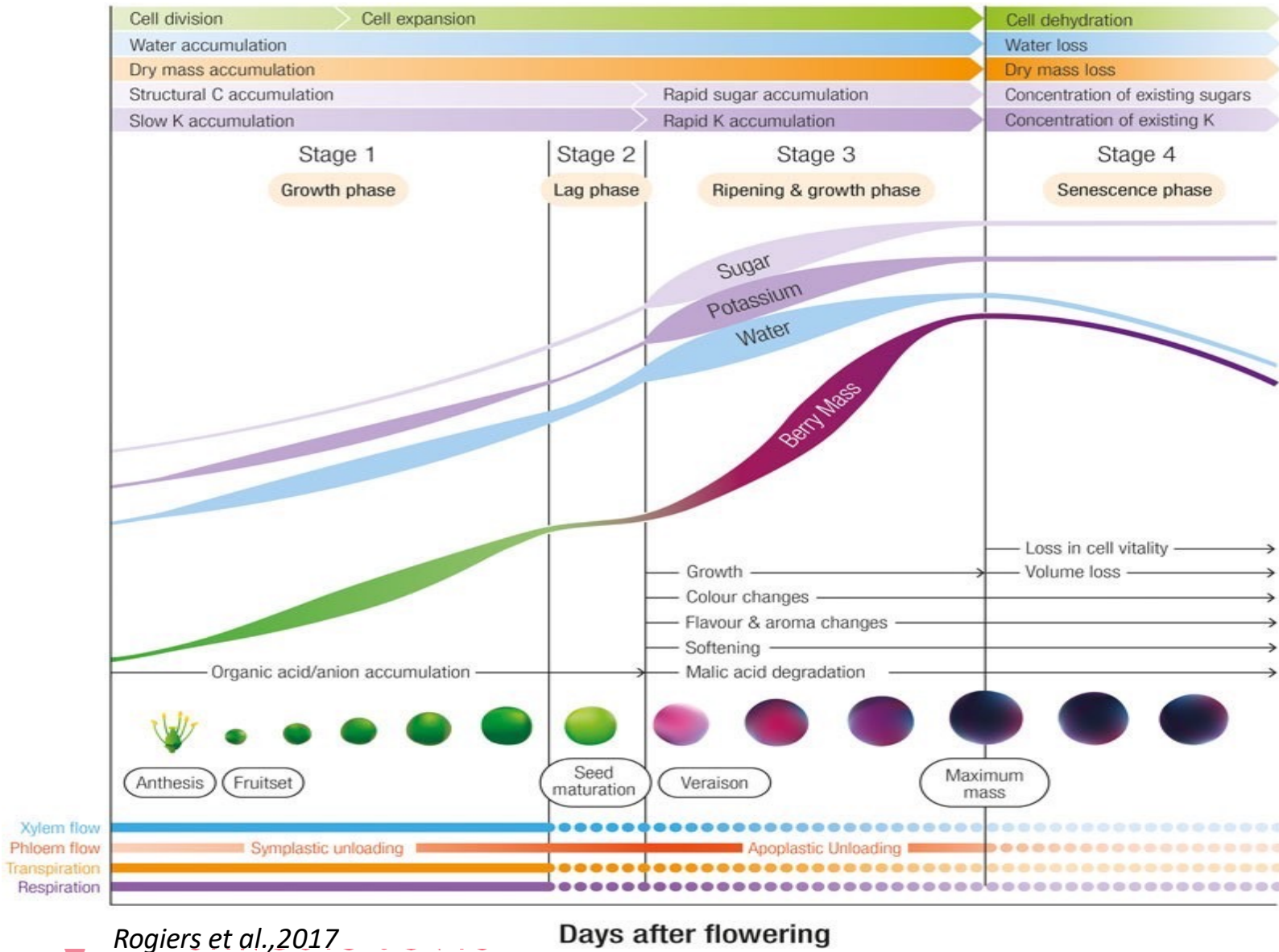
Castellarin et al., 2011



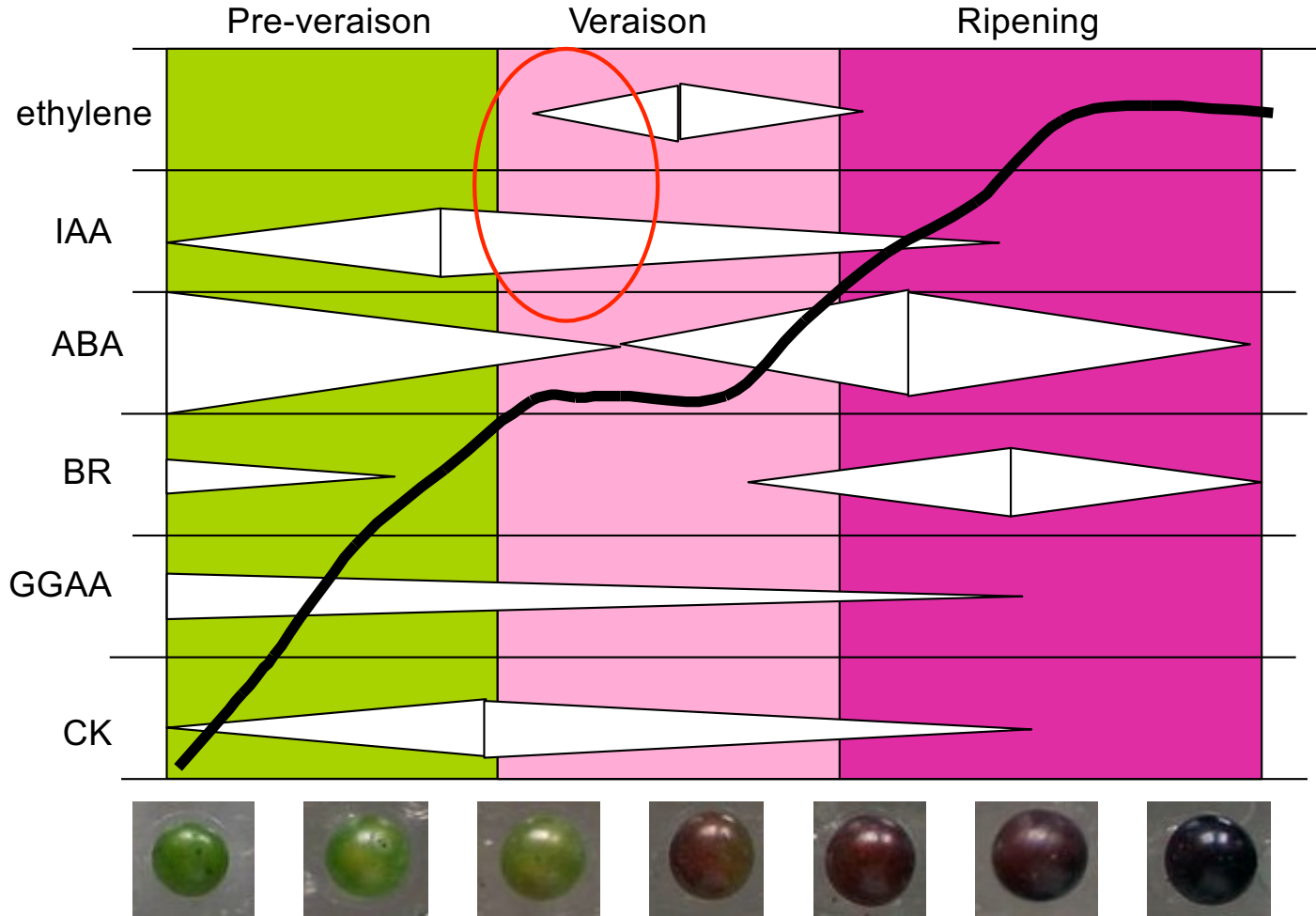
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Cambiamenti chimico-fisiologico che avvengono durante la maturazione della bacca d'uva 1



Cambiamenti chimico-fisiologico che avvengono durante la maturazione della bacca d'uva 2



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Trattamenti con etilene (1000 ppm for 36h) su bacche di Sangiovese

Due giorni dopo pigiatura



C₂H₄

Controllo



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Botton et al., 2017

Trattamenti con etilene (1000 ppm for 36h) su bacche di Sangiovese

Analisi del profilo polifenolico

	Controllo	C ₂ H ₄	
Flavanol vanillin (mg/L)	939	1224	→ + 30%
Polifenoli totali (mg/L)	1984	2395	→ + 20 %
Antocianine totali (mg/L)	228	319	→ + 40 %
Monomeri antocianine (mg/L)	126	185	→ + 45%
Proantocinanidine (mg/L)	2802	3441	→ + 20 %
Flavonoidi totali (mg/L)	1413	1636	→ + 15 %



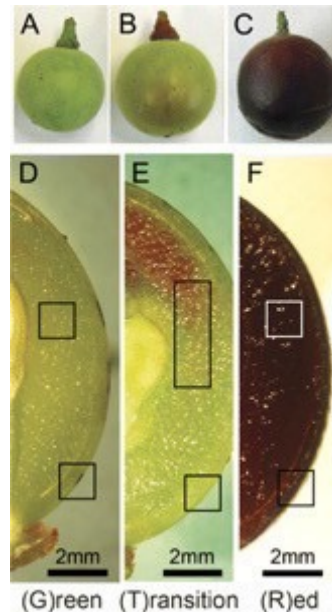
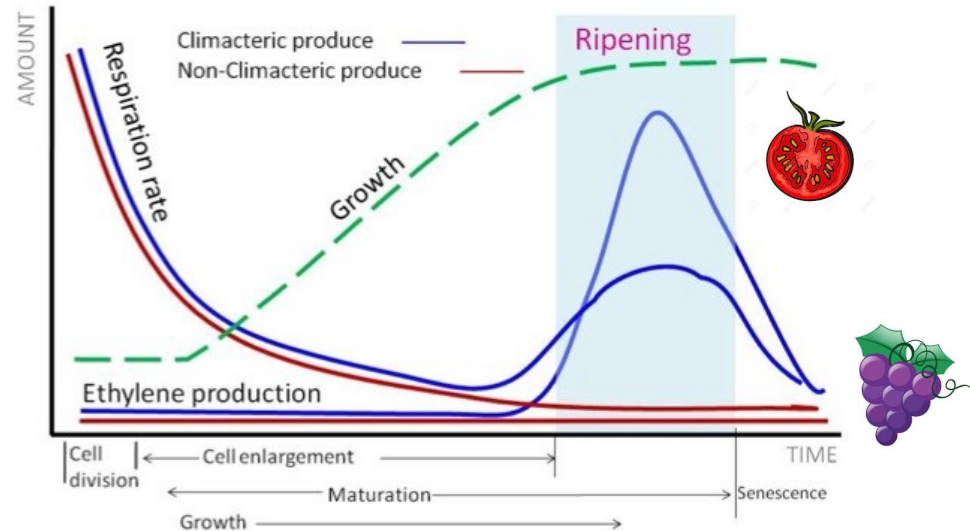
Maturazione della bacca della vite

La bacca d'uva è un frutto non climaterico

È un processo molto lungo che dura circa 3 mesi in cui si verifica una profonda modificazione della composizione della bacca

E' fortemente condizionato dall'ambiente

Inizia nella polpa in prossimità della fine dello stilo



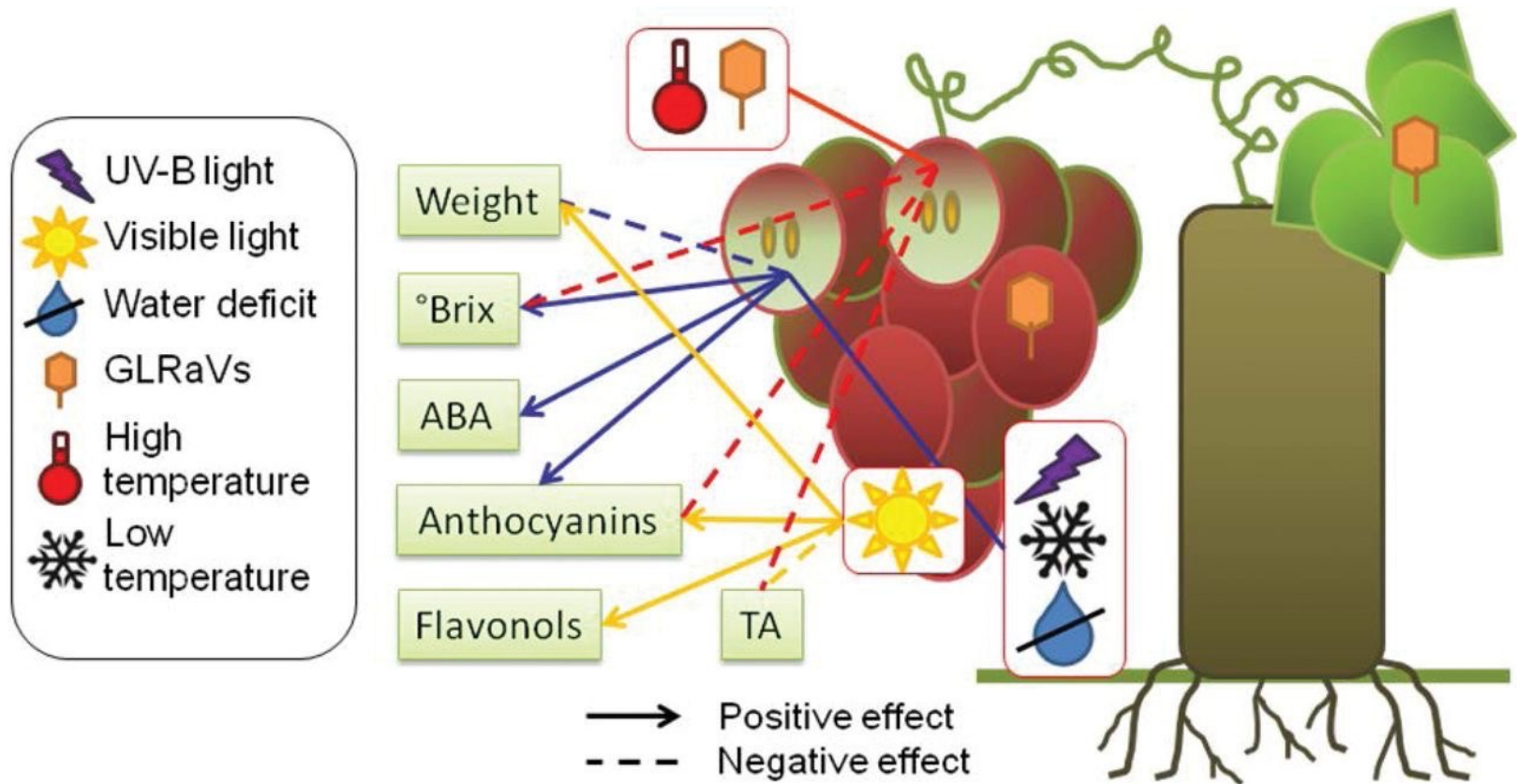
Castellarin et al., 2011



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Fattori ambientali in grado di modificare parametri biochimici associati alla maturazione di bacche pigmentate



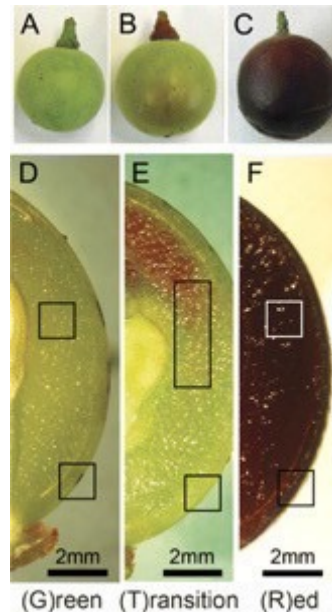
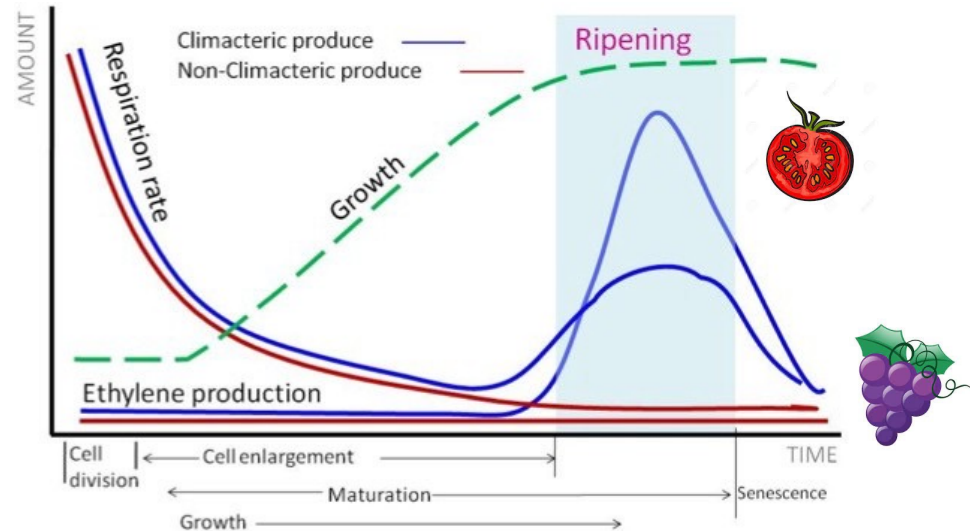
Maturazione della bacca della vite

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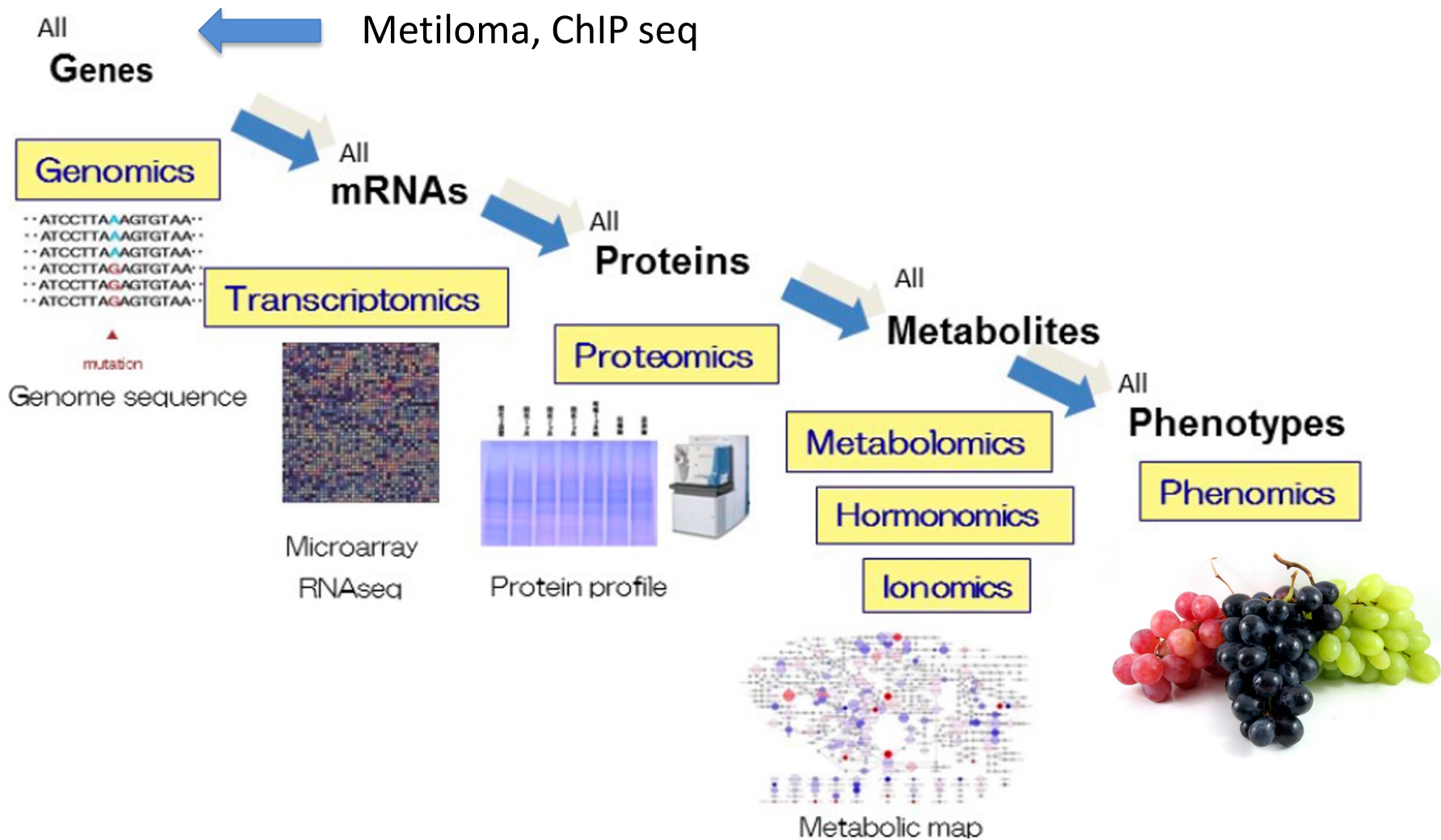
Castellarin et al., 2011



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Approcci omici per lo studio della fisiologia della pianta

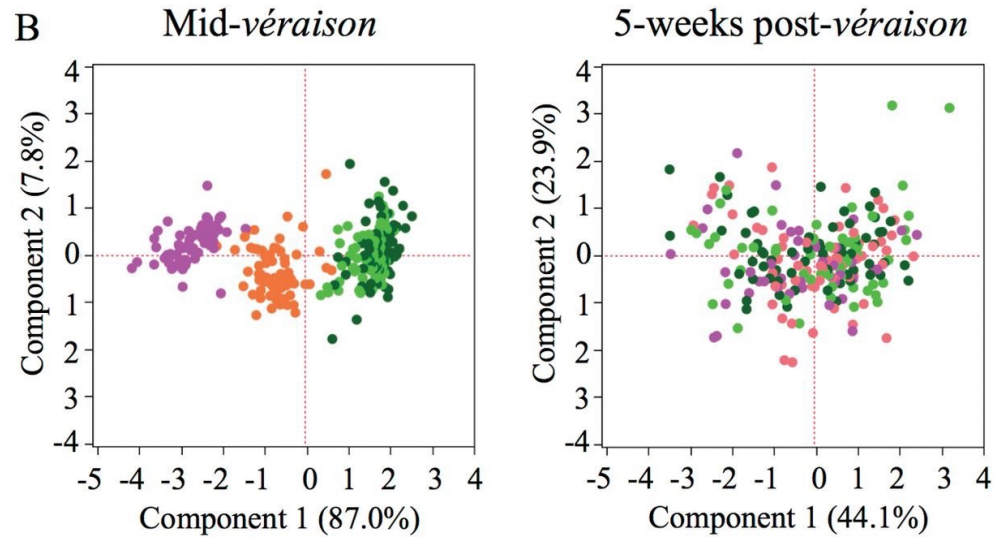
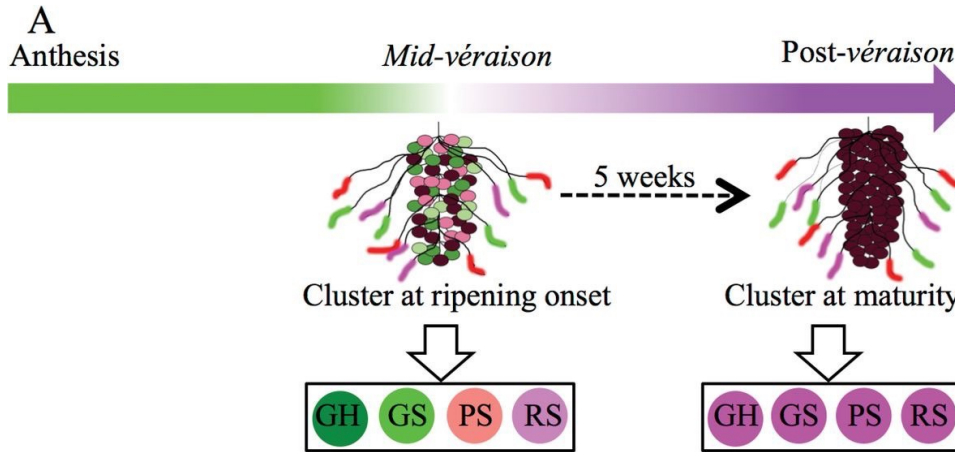




August 10

Dinamica maturazione in un grappolo

Ripening state variation among berries within a cluster at ripening onset and uniform ripeness upon maturity.



Cambiamenti chimico-fisiologico che avvengono durante la maturazione della bacca d'uva:

- a) livelli di espressione di hub genici coinvolti nei principali biochimismi**
- b) diverso comportamento della buccia e della polpa**
- c) il controllo dell'inizio del processo di maturazione: il concetto «switch genes»**
- d) la plasticità della bacca: il controllo dell'ambiente sulla maturazione della bacca**
- e) il controllo genico nella fase di post-raccolta: la disidratazione della bacca.**



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Rogiers et al., 2017 JOVIS



In qualsiasi momento

- Dal 2021
- Dal 2020
- Dal 2017
- Intervallo specifico...

Ordina per pertinenza

Ordina per data

Qualsiasi lingua

Pagine in Italiano

- includi brevetti
- includi citazioni

Crea avviso

Bioinformatics analysis of WRKY transcription factors in grape and their potential roles prediction in sugar and abscisic acid signaling pathway

T Huang, J Yang, D Yu, X Han, X Wang - *Journal of Plant Biochemistry* ..., 2021 - Springer
 ... **berry** ripening, more sugar accumulation and ABA appear in colored **grapes** as compared to ... **berry** of different growth and developmental stages was analyzed by **transcriptomic analysis**, and the co ... Total RNA of **grape berry** was extracted with a RNAprep Pure Plant kit (TIANGEN ...
 ☆ 57 Citato da 2 Articoli correlati Tutte e 3 le versioni

[ACNP Full Text](#)

Transcriptome analysis reveals light-induced anthocyanin synthesis candidate genes in rabbiteye blueberry (*Vaccinium ashei*: Reade)

X Guo, D Wang, M Shakeel - *Biotechnology & Biotechnological* ..., 2021 - Taylor & Francis
 ... These findings validate the results of the **transcriptomic analysis** ... significantly induced by light [51], whereas the induction of anthocyanin accumulation in the **grape** variety 'Shiraz ... these are major factors influencing the production of high-value crops such as **grapes**, apples and ...
 ☆ 57 Articoli correlati 2

[\[PDF\] tandfonline.com Full View](#)

Research progress of proteomics in grapes during grape berry development and under abiotic stress

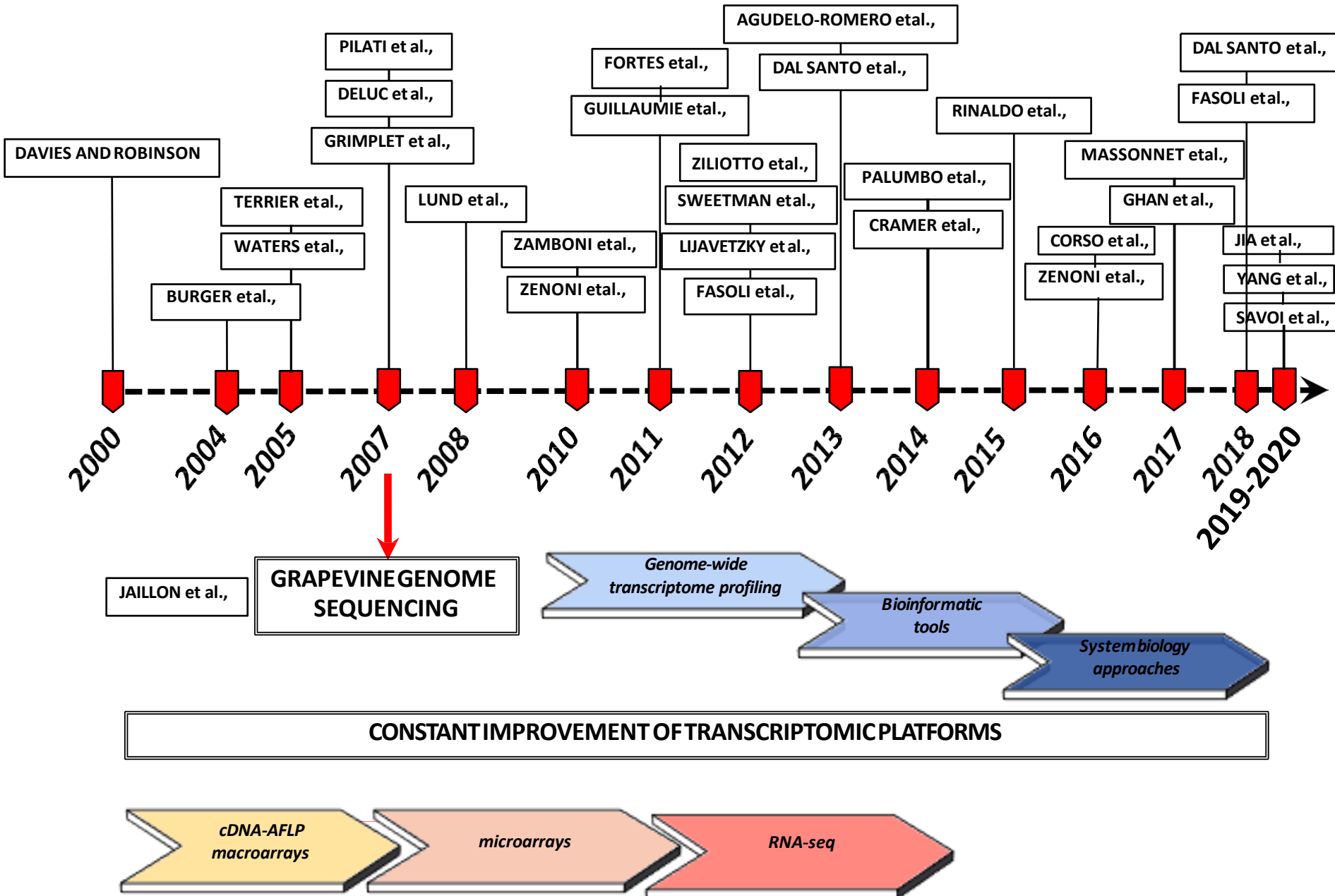
F FANG, F WANG - *食品工业科技*, 2017 - spgykj.com
 ... [36], Liu GT, Wang JF, Grant C, et al. **Transcriptomic analysis** of **grape** (*Vitis vinifera* L.) leaves during and after recovery from heat stress[J]. *BMC Plant Biology*, 2012, 12:174 ... [39], Wu ZM, Yuan XZ, Li H, et al. Heat acclimation reduces postharvest loss of table **grapes** during cold ...
 ☆ 57 Articoli correlati Tutte e 2 le versioni 2

[HTML] Relative quantification of phosphoproteomic changes in grapevine (*Vitis vinifera* L.) leaves in response to abscisic acid

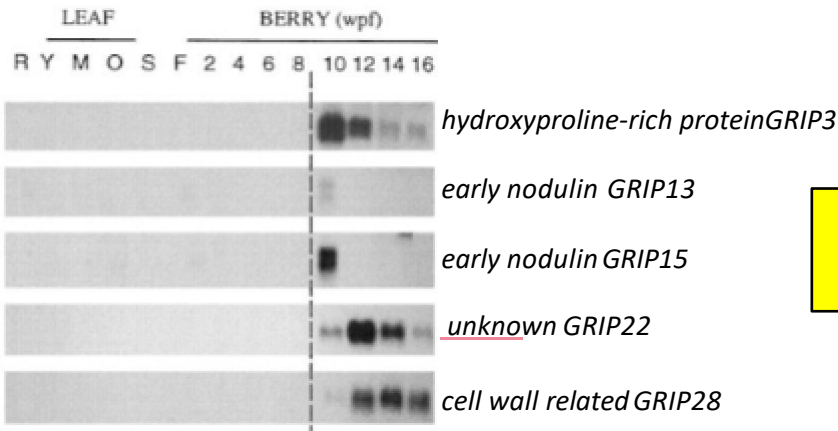
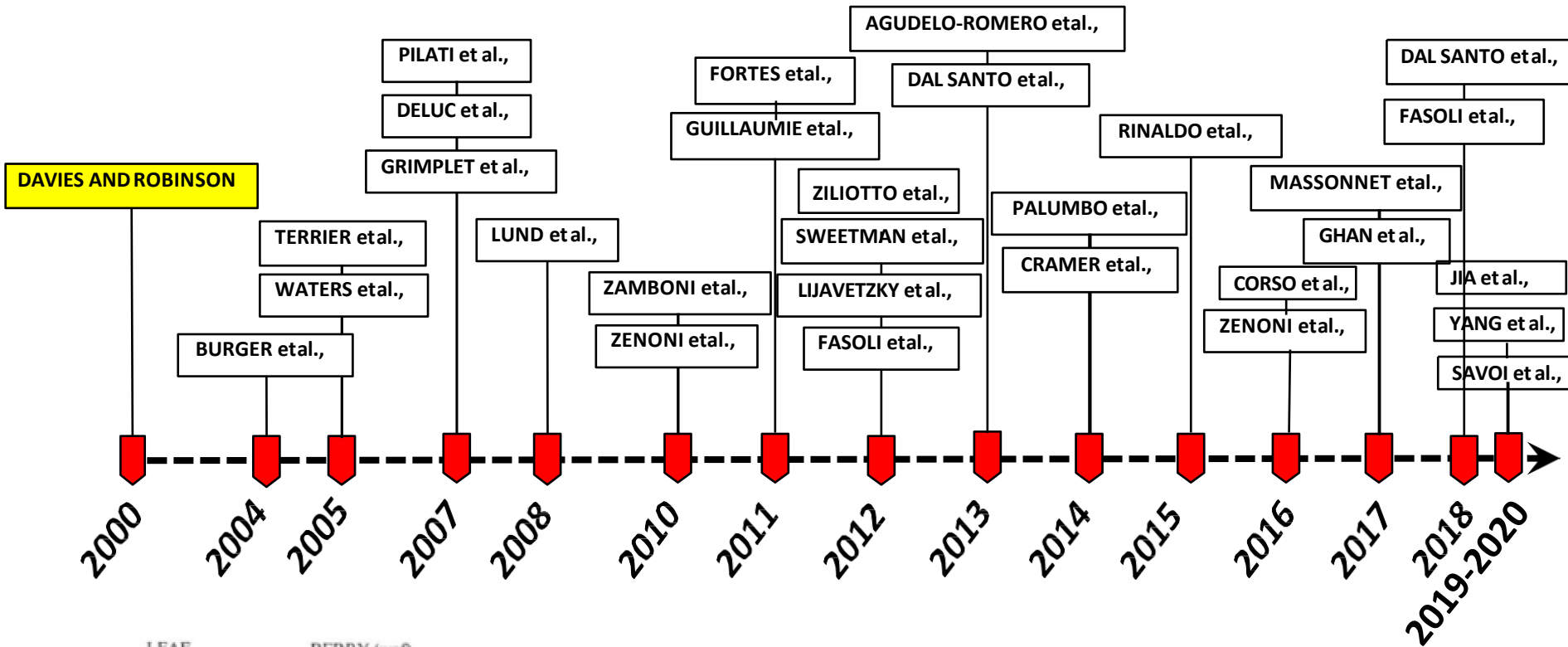
S Rattanakan, I George, PA Haynes, GR Cramer - *Horticulture research*, 2016 - nature.com
 ... In a previous **transcriptomic analysis**, abscisic acid (ABA) was found to affect the abundance of a number of ... They are used for the production of wine, table **grapes**, juice and raisins, and are worth millions of ... Abiotic stresses affect both quality and quantity of **grape** production ...
 ☆ 57 Citato da 13 Articoli correlati Tutte e 15 le versioni 2

[\[HTML\] nature.com Full View](#)

Large-scale transcriptional changes during berry development

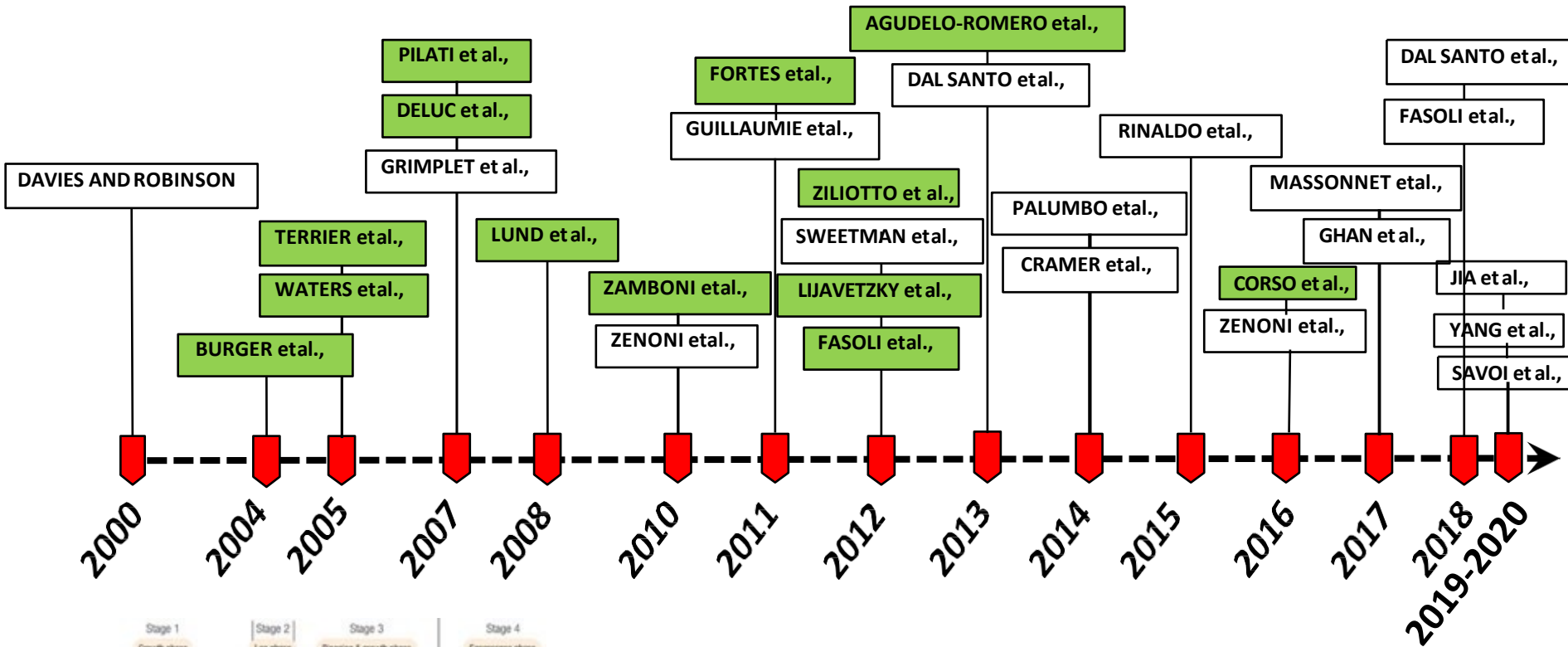


Large-scale transcriptional changes during berry development



*Identification and definition of "GRIP"
grape ripening-induced protein*

Large-scale transcriptional changes during berry development



Expression profile of the principal molecular events during berry development

Transcriptomic rearrangement during the ripening transition

- Photosynthesis
- Cell cycle
- Cellular component organization
- Hormone (auxin) signalling responsive transcripts

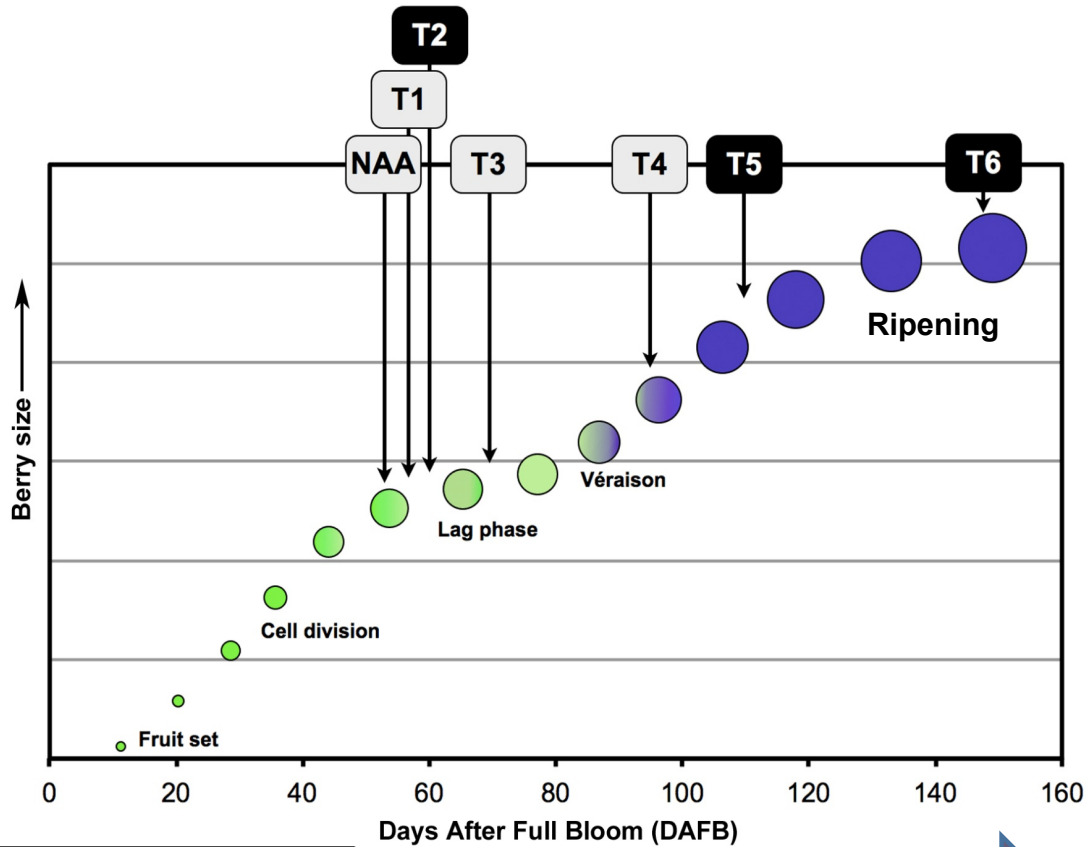


- Secondary metabolism
- Sugar metabolism
- Starch degradation
- Regulation of gene expression
- Biotic stress response
- Cell wall metabolism

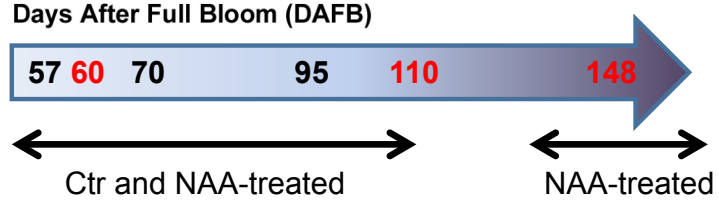
THE SHIFT FROM THE GROWTH TO RIPENING PHASE IN BERRY INVOLVES A PROFOUND TRANSCRIPTOMIC REARRANGEMENT

Trattamenti con auxina—cv. Merlot

Ziliotto et al., 2012



**NAA (200 mg/L)
Treatment: 53
DAFB**

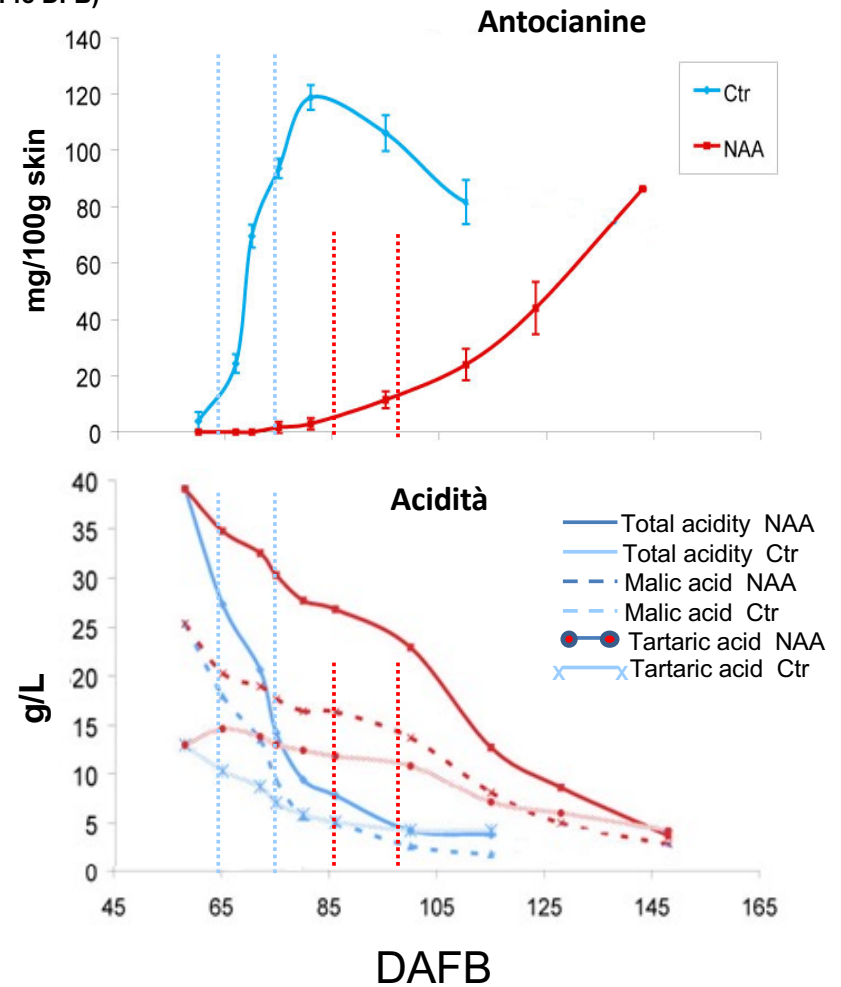
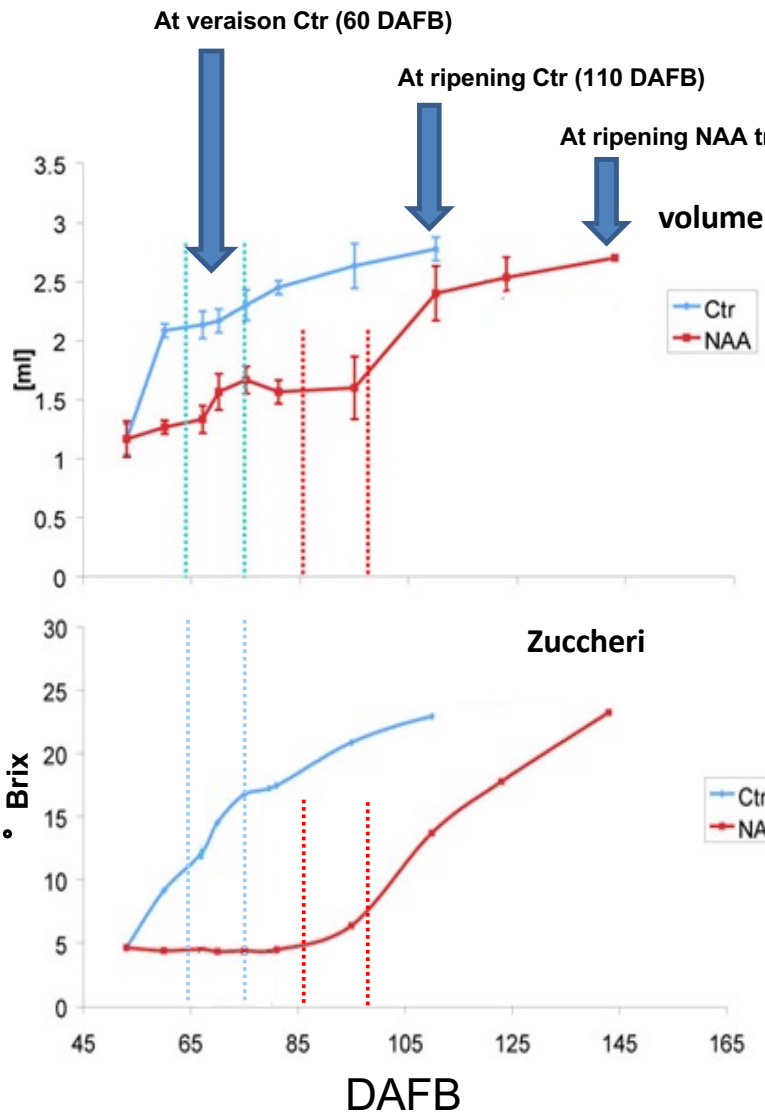


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Sampling (DAFB)

Parametri biochimici

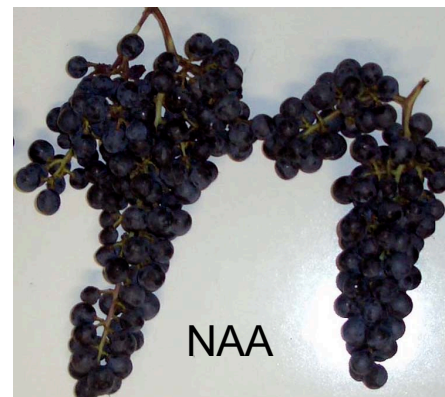
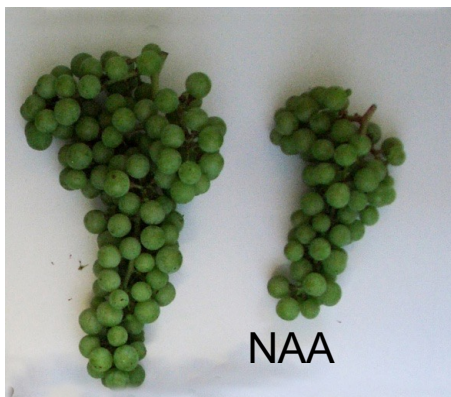
Ziliotto et al., 2012



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Decorso della maturazione in bacche trattate con NAA e non (200 mg/L in preinvaiaatura) Ziliotto et al., 2012



AT VERAISON (60 DAFB)

AT HARVEST OF CTR (110 DAFB)

AT HARVEST OF NAA-TREATED (148 DAFB)



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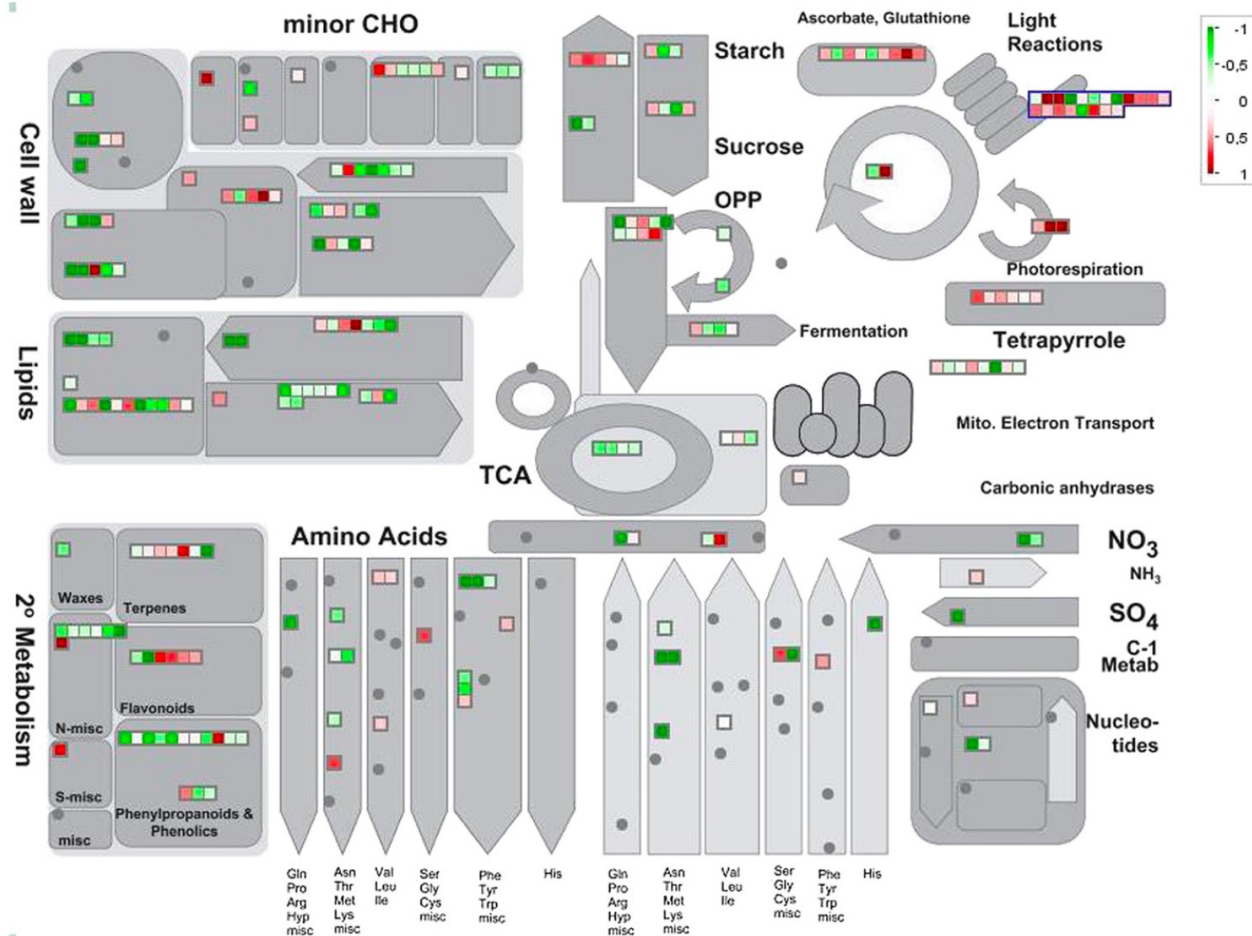


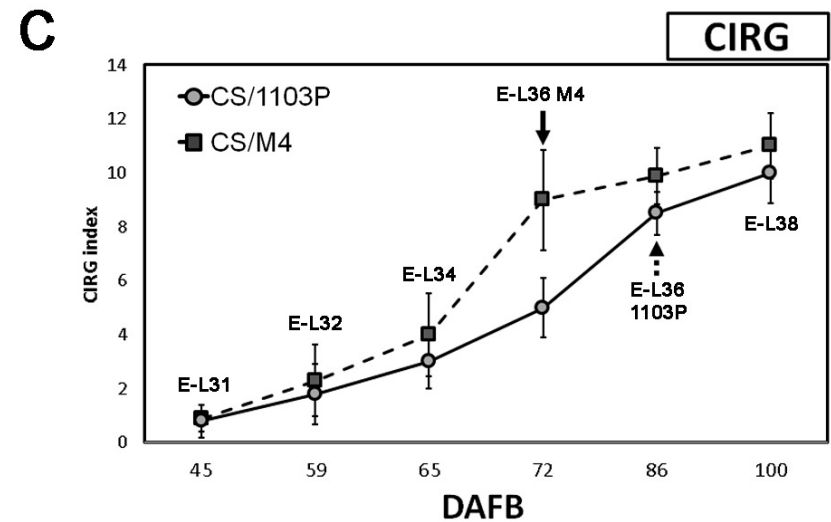
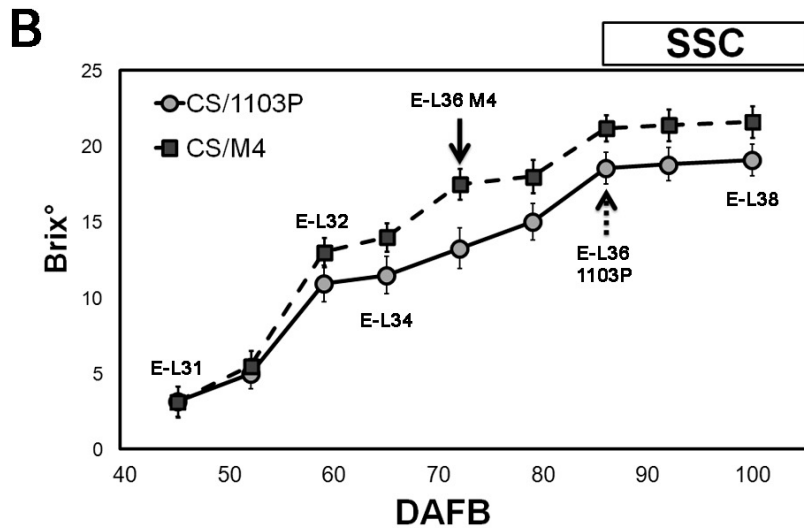
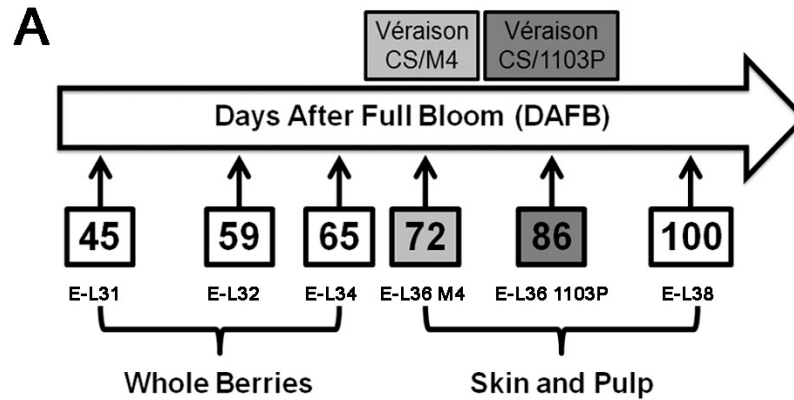
Figure 2 MapMan analysis. MapMan visualization of differences in expression of genes involved in metabolic processes. Classification into bin categories were done by using a mapping file of the grape AROS V1.0 platform (<http://mapman.gabipd.org/web>). Heat maps show genes with statistically significant (P value < 0.084) differential expression identified by comparing NAA-treated and control berries at 60 DAFB (N1/C1). A conventional red-to-green scale was used to indicate up-regulation (red) or down-regulation (green).



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Impatto del portainnesto sulla maturazione delle bacche



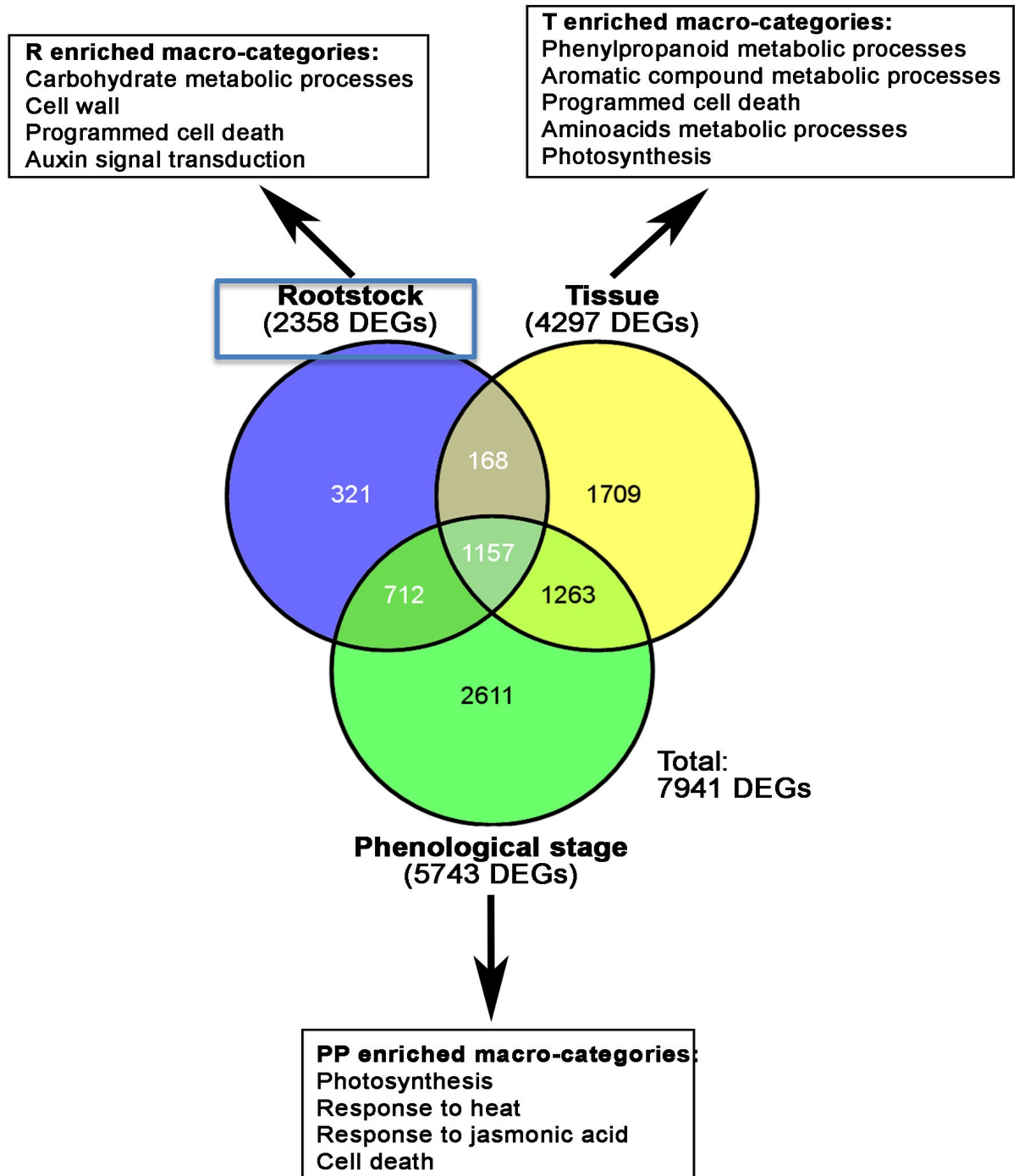
1103P cause a delay in volume and SSC evolution on CS berries



fondazione banfi

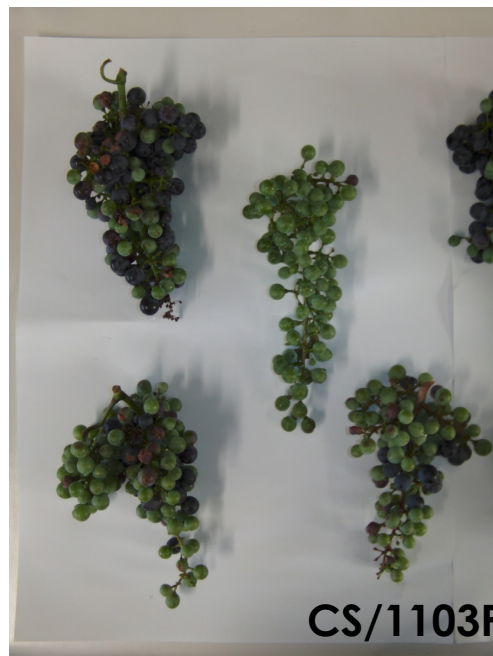
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**Multifactorial analysis:
identification of rootstock role in the ripening modulation**



A large numbers of DEGs related to auxin metabolisms were identified with enrichment and clustering analysis

AUXIN TREATMENT: causes a delay in the onset of ripening



CS/M4 versus CS/1103P at 72 DAFB

NAA treatment at pre-véraison
harvest time (100 DAFB)

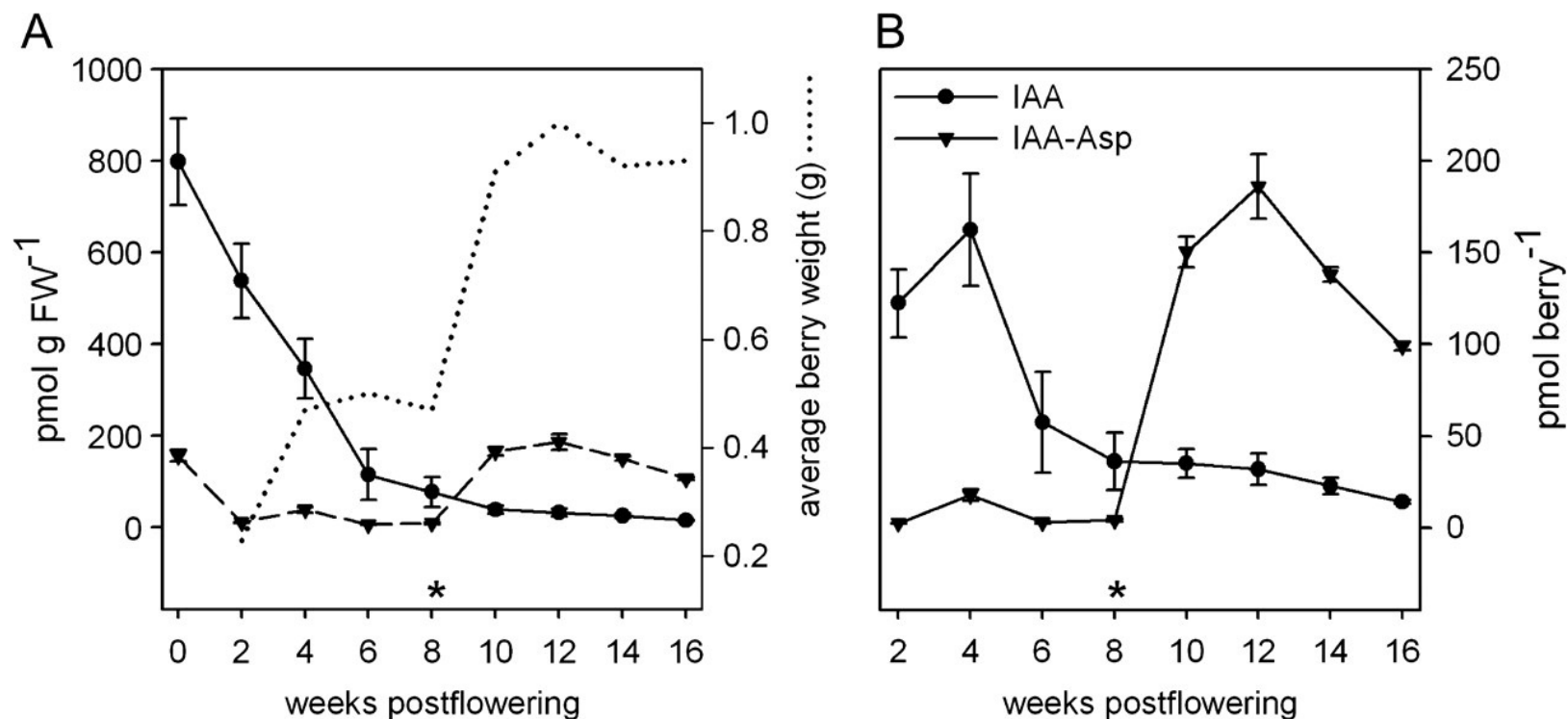


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Corso et al., 2016

Levels of IAA and IAA-Asp in developing grape berries



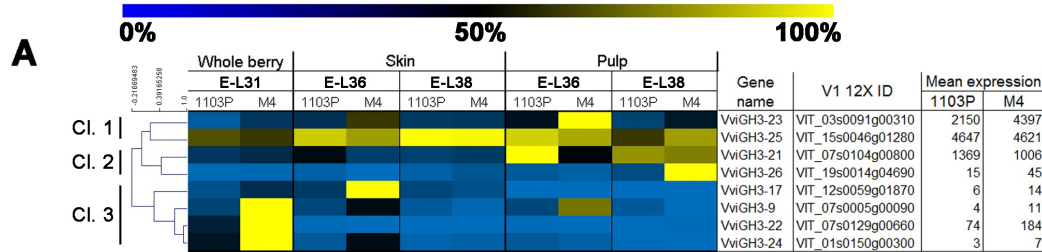
Böttcher C et al. J. Exp. Bot. 2010;61:3615-3625



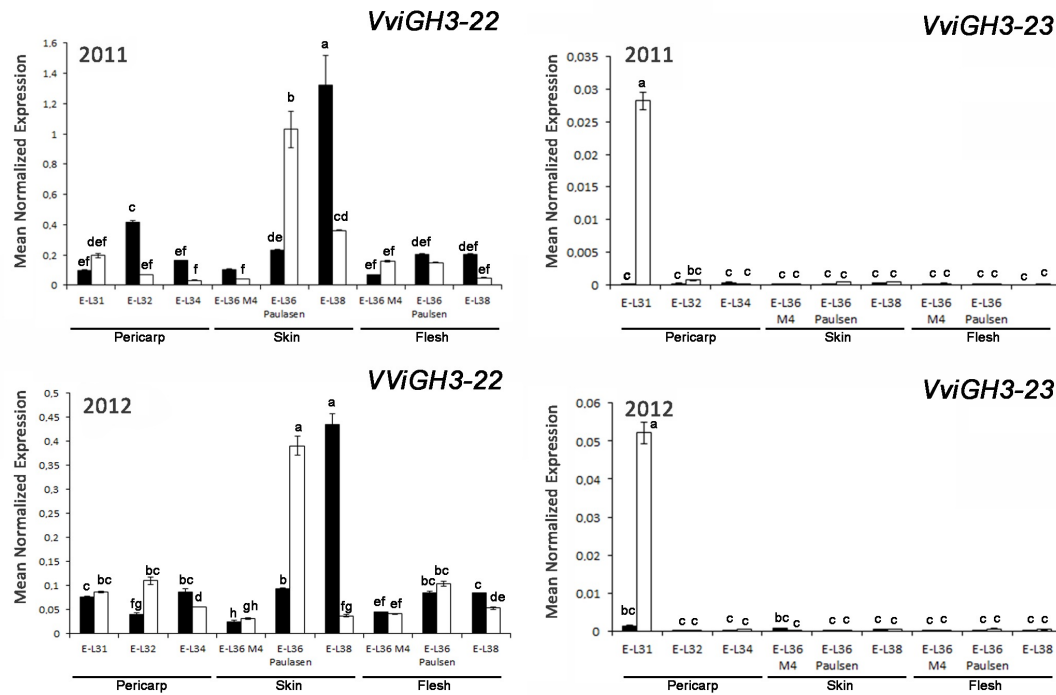
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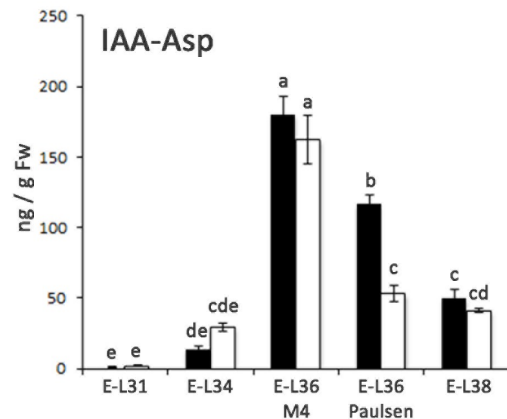
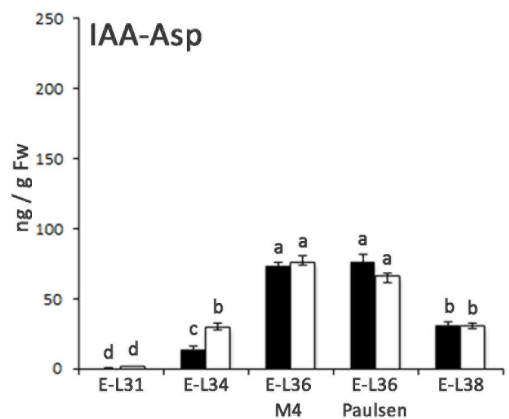
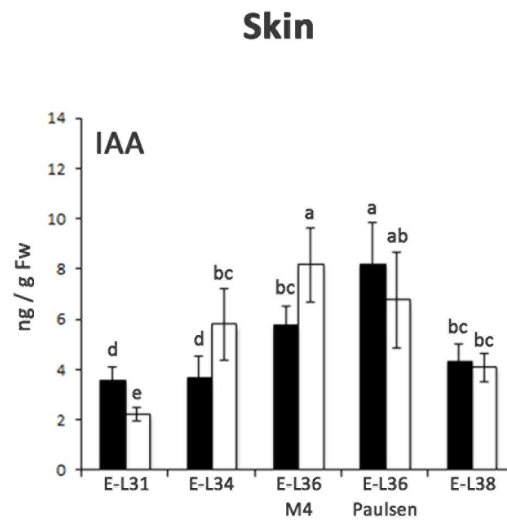
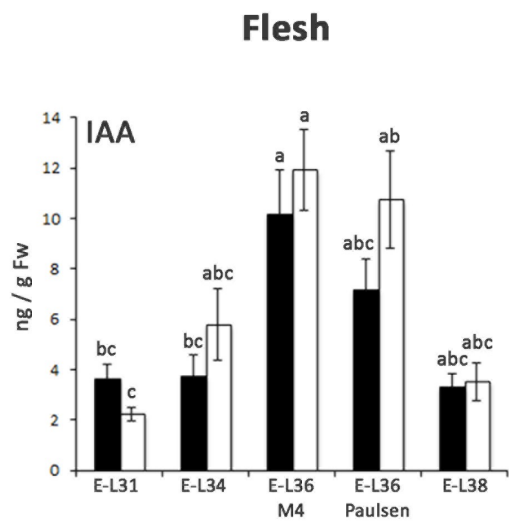
Auxin homeostasis: GH3 genes expression



B



Auxin content in skin and flesh tissues



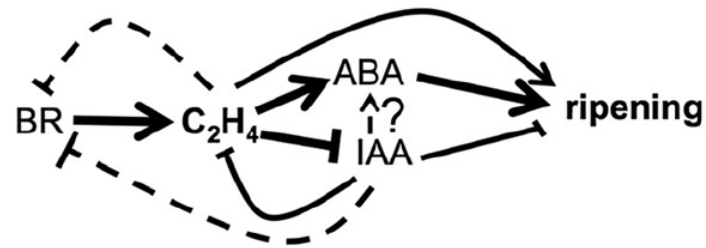
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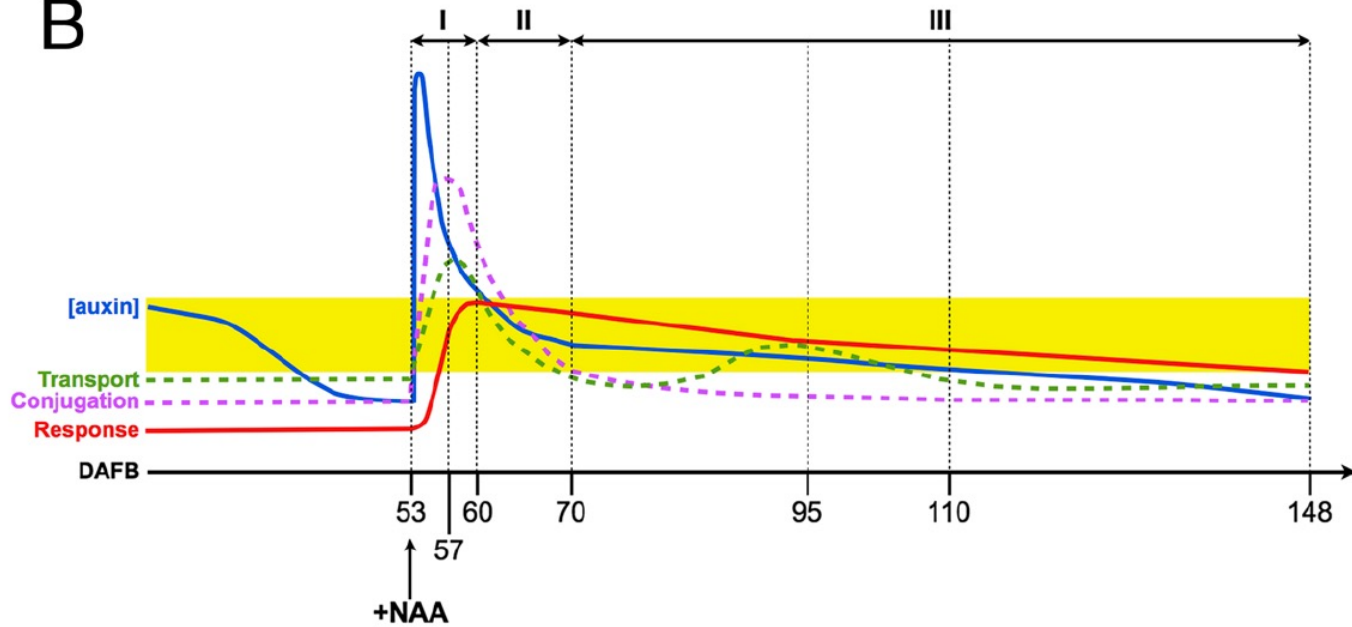
Corso et al., 2016

Interazione ormonale all'invasiatura

A



B

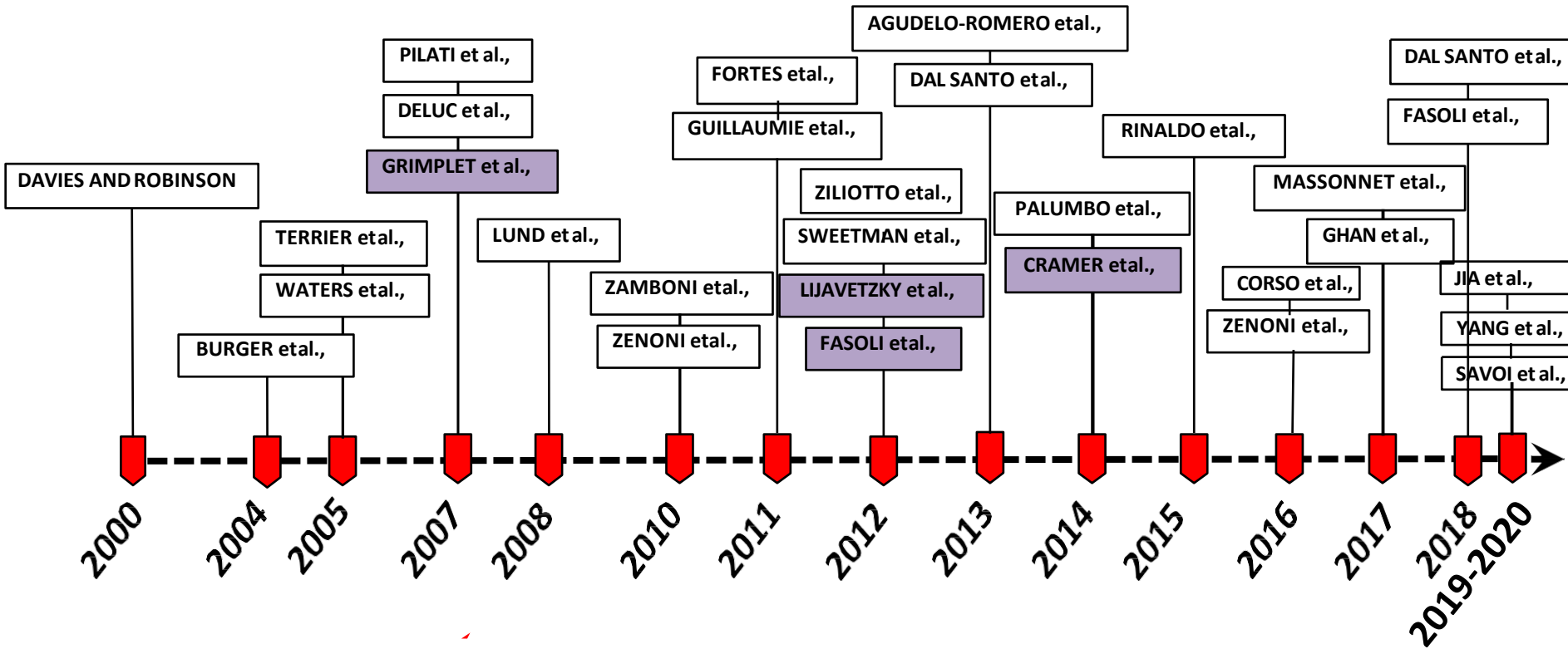


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Ziliotto et al., 2012

Large-scale transcriptional changes during berry development



Transcriptomic changes during berry development in pulp and skin separately

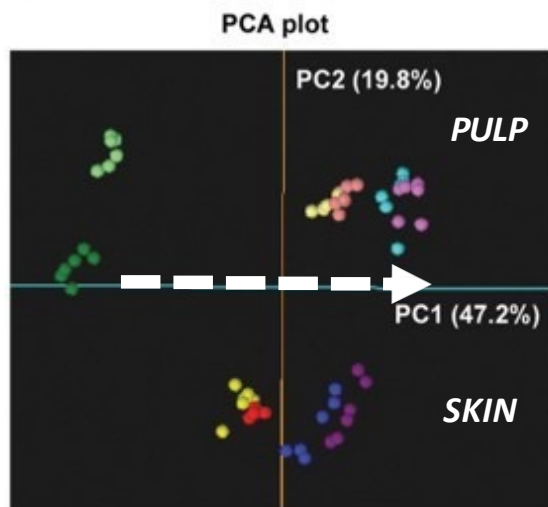
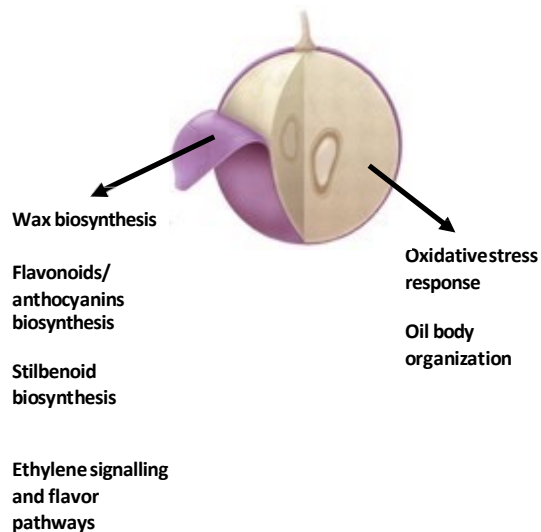


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Modificato da Zenoni et, 2018

Ripening program is anticipated in pulp



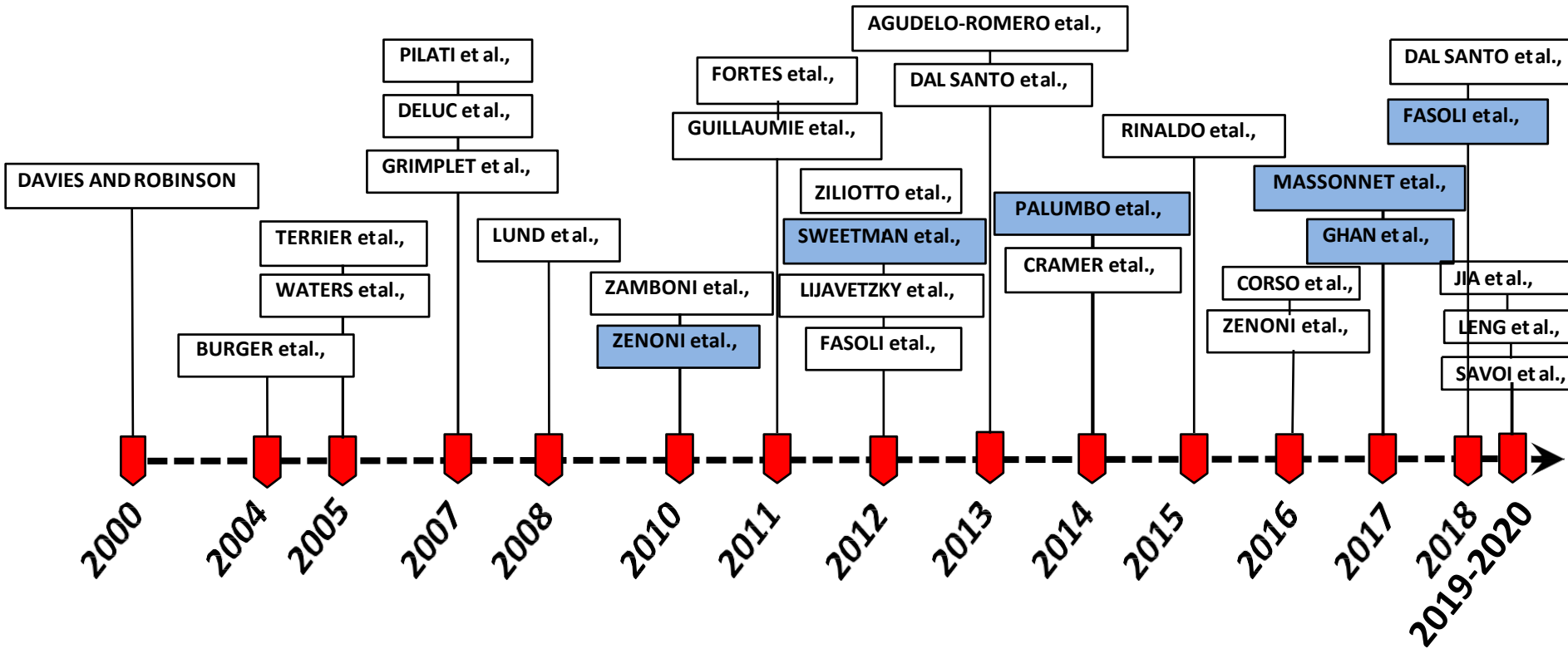
Lijavetzky et al., 2012

Functional terms		P	V1-V2	R1	R2
Auxin signalling, bHLH TF, Homeobox TF, GA biosynthesis	S	█	█	█	█
	F	█	█	█	█
Wax biosynthesis	S	█	█	█	█
	F	█	█	█	█
ABC transporters, Cellulose biosynthesis, Glycerolipid metabolism	S	█	█	█	█
	F	█	█	█	█
Photosynthesis	S	█	█	█	█
	F	█	█	█	█
Microtubule organization and biogenesis	S	█	█	█	█
	F	█	█	█	█
Flavonoid and anthocyanin biosynthesis, ABC transporters	S	█	█	█	█
	F	█	█	█	█
C2C2-DOF TF, Macromolecule transport	S	█	█	█	█
	F	█	█	█	█
Oxidative stress response	S	█	█	█	█
	F	█	█	█	█
Oil body organization and biogenesis, C2H2 TF	S	█	█	█	█
	F	█	█	█	█
Biotic stress response, Xyloglucan modification	S	█	█	█	█
	F	█	█	█	█
Stilbenoid biosynthesis, WRKY TF, Macromolecule transport	S	█	█	█	█
	F	█	█	█	█

TRANSCRIPTOMIC PROGRAM IS ANTICIPATED IN PULP IN COMPARISON TO THE SKIN

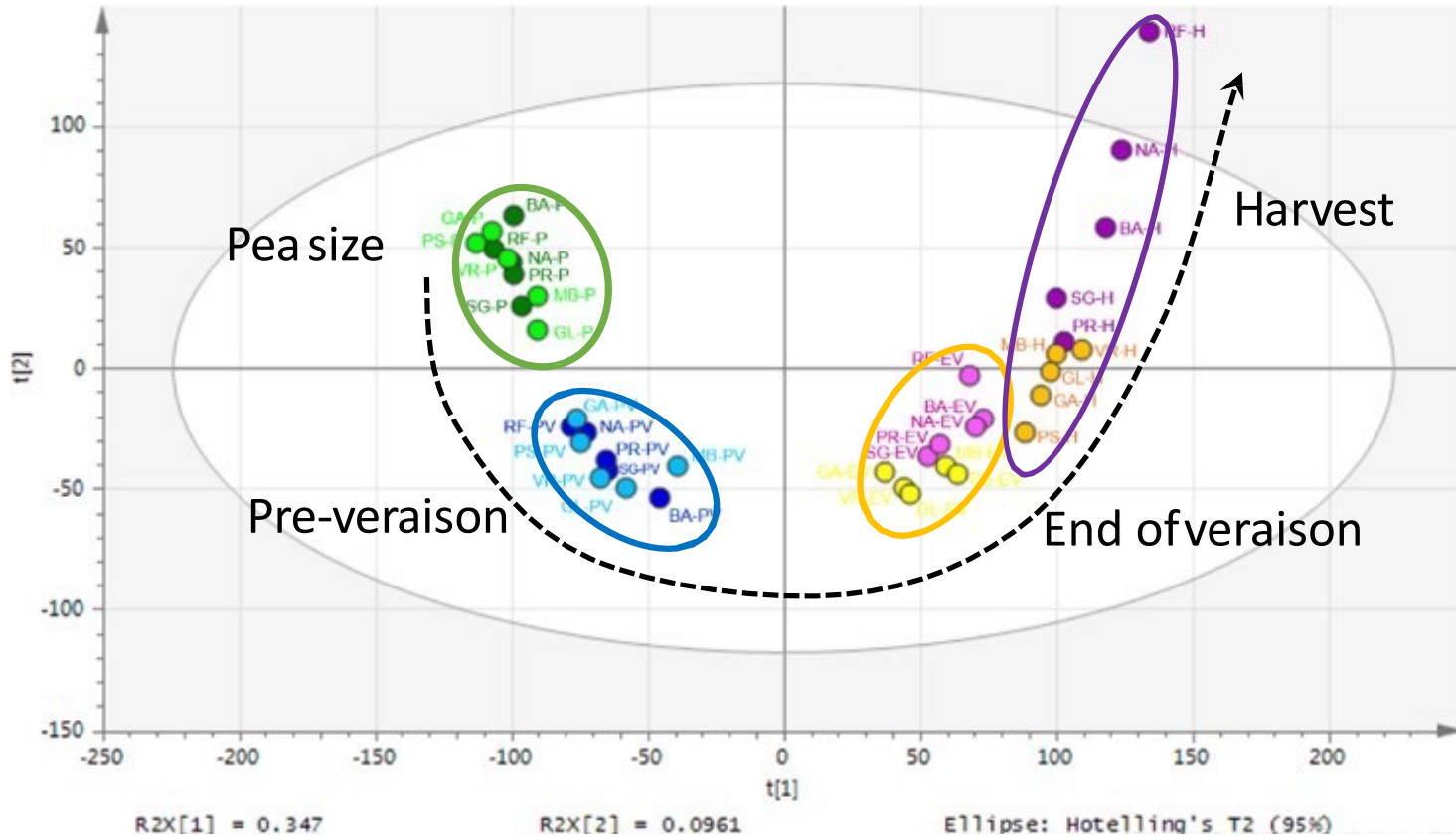
Zenoni et, 2018

Large-scale transcriptional changes during berry development



*RNA-seq approach to dissect the **transcriptional complexity** during berry development*

Core transcriptomic traits during berry development



Red varieties

White varieties

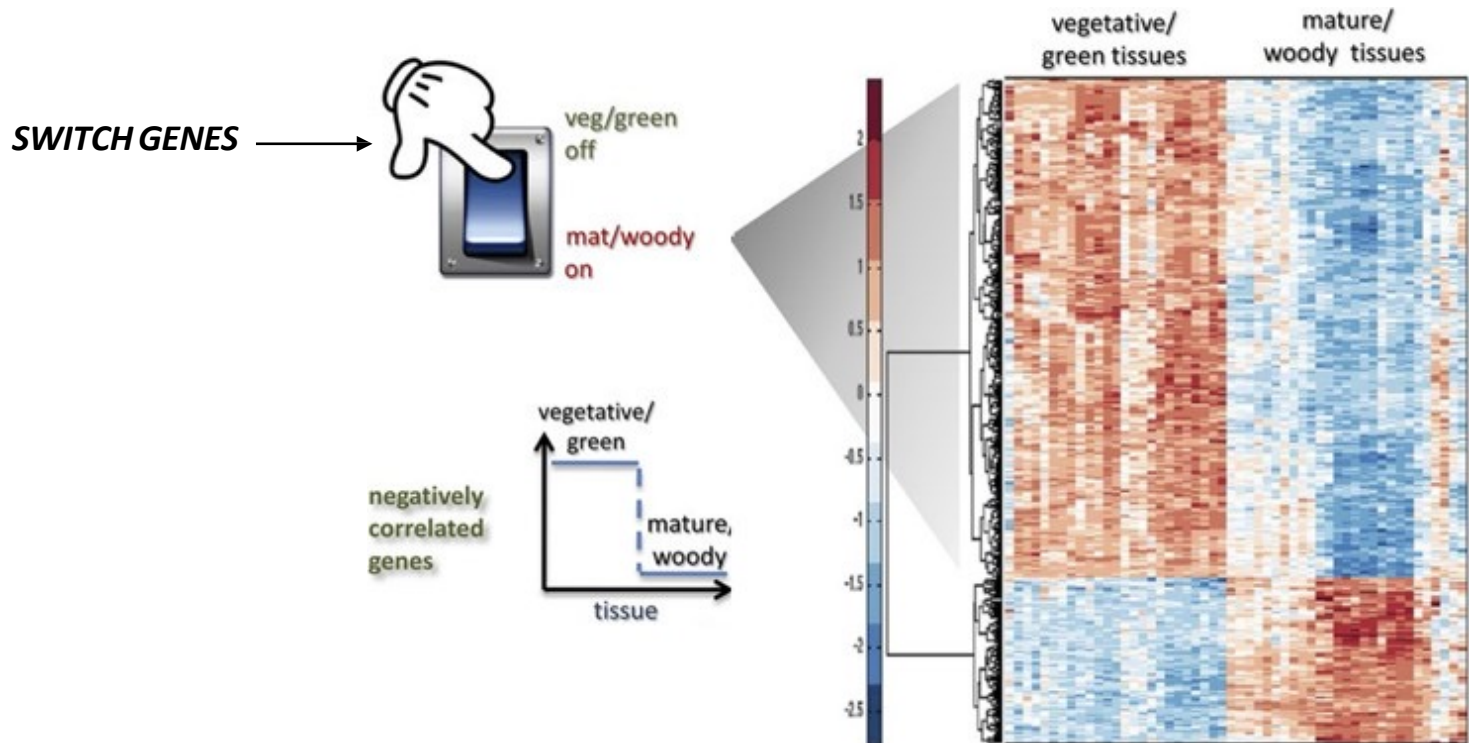
- Pea-sized
- Prévéraison
- End of véraison
- Harvest

-
-
-
-

**CORE TRANSCRIPTOMIC TRAITS
WERE PROFILED**

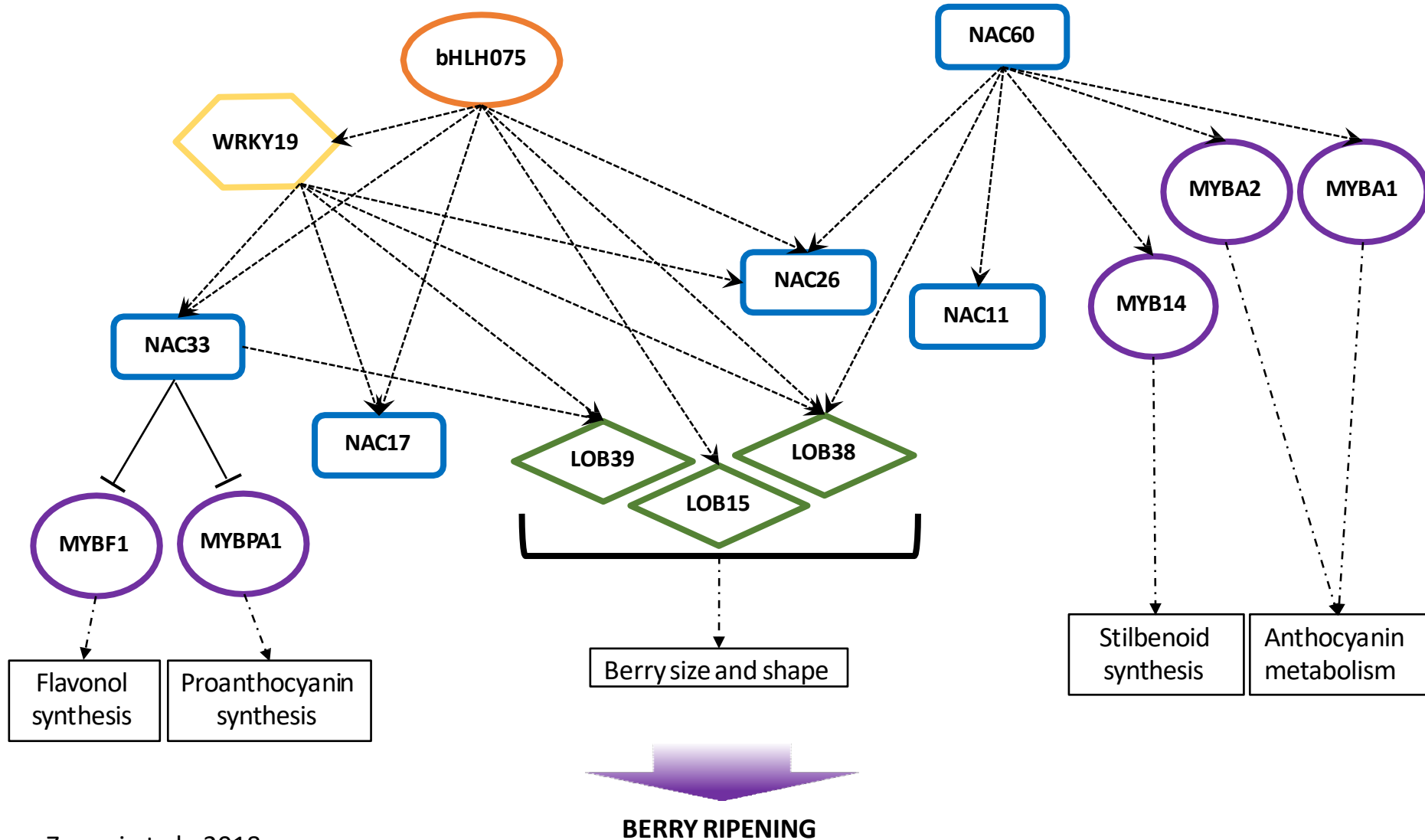
Switch genes model of action

- ✓ All switch genes are down regulated during growth phase and up during mature phase
- ✓ Switch genes could act as an electric switch able to switch-off the expression of vegetative-related genes and to switch-on the expression of mature-related genes

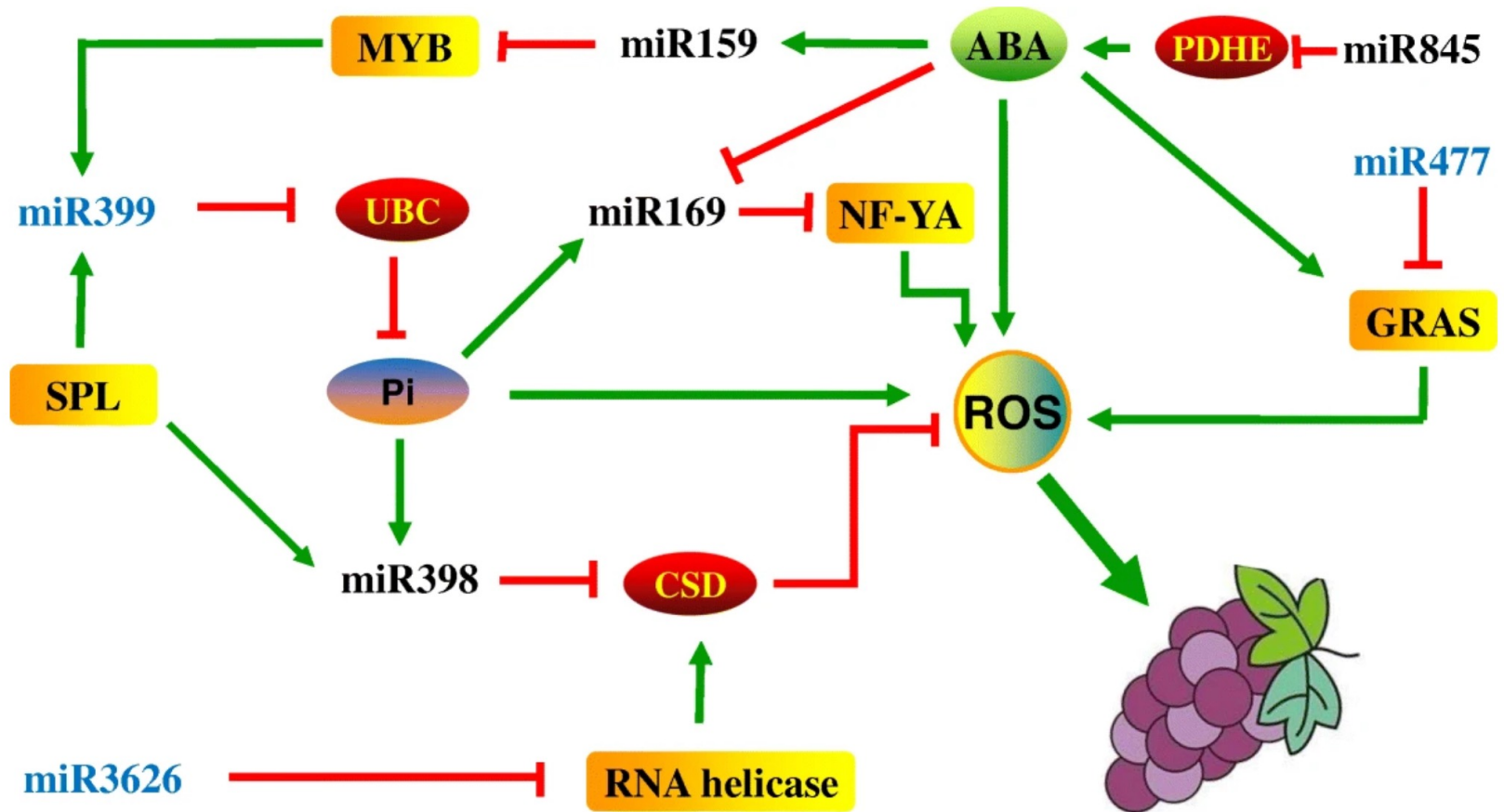


In switch genes are present transcription factors

Putative hierarchy of transcription factors in the onset of berry ripening

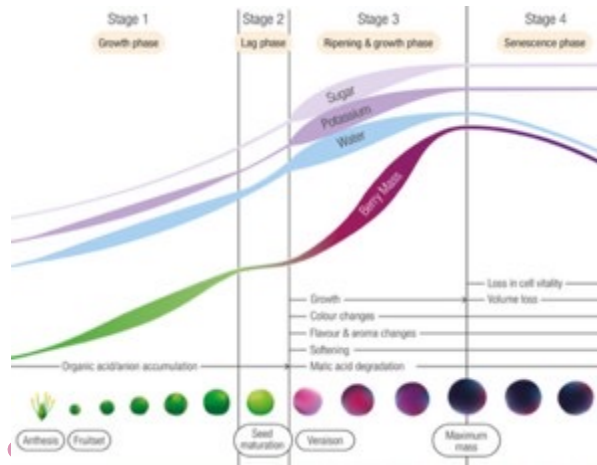
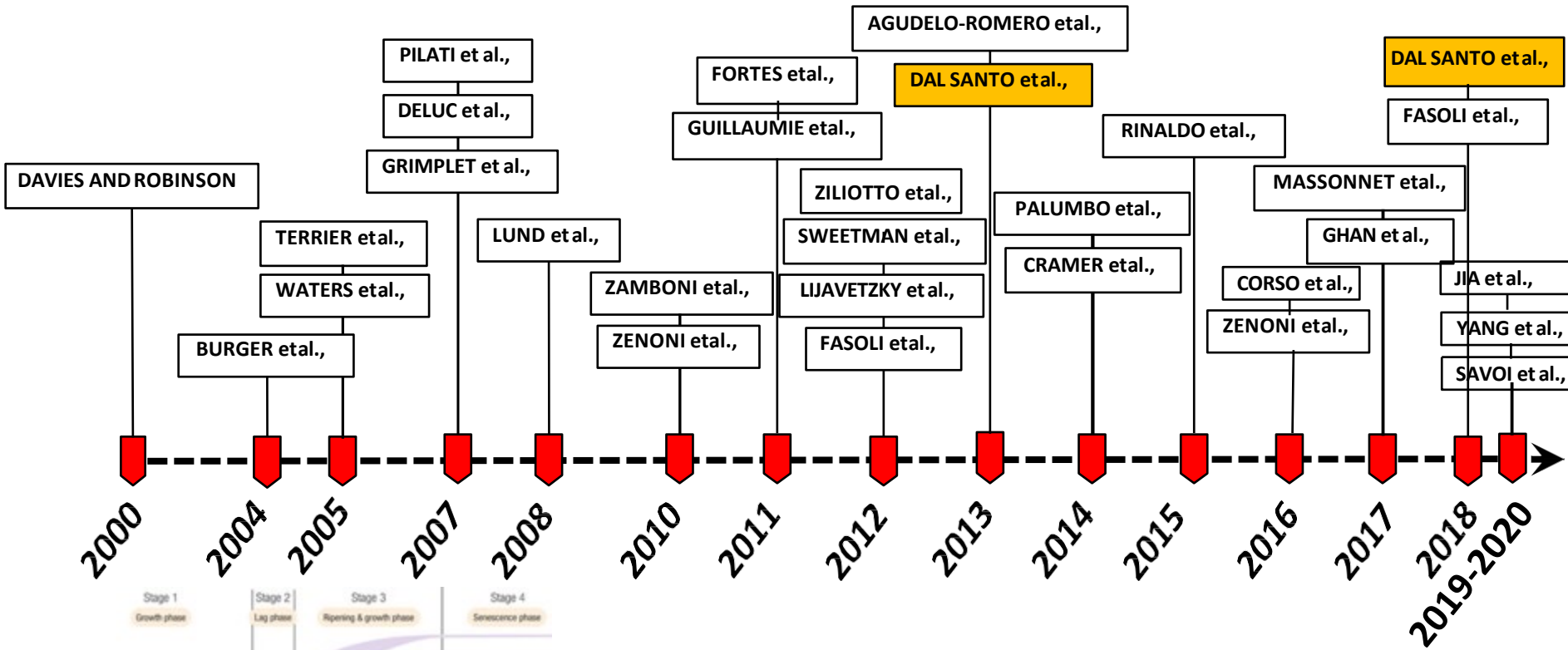


Role of microRNAs in the control of Transcription factors regulating grape berry ripening



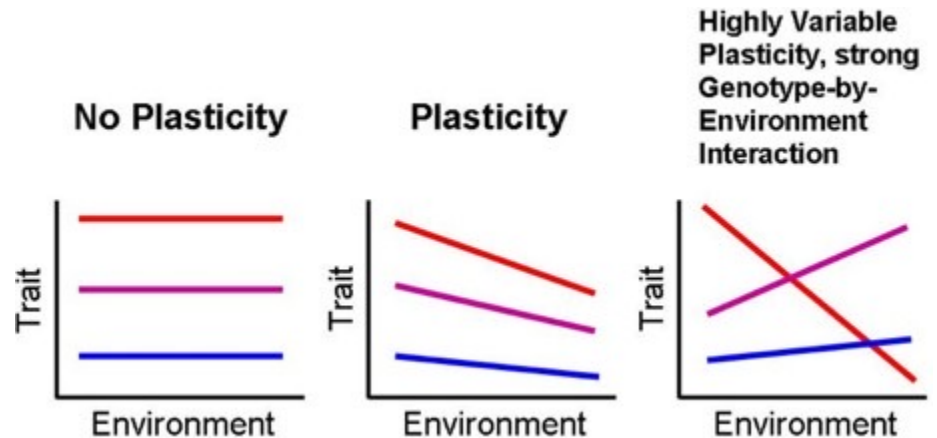
The speculated possible regulatory network of miRNAs-TFs-Genes in early-ripening of 'Fengzao'. PDHE: pyruvate dehydrogenase, CSD: superoxide dismutase, ROS: reactive oxygen species, NFYA: nuclear transcription factor Y subunit A, PHO2: PHOSPHATE2, SPL: squamosa-promoter binding protein-like

Large-scale transcriptional changes during berry development



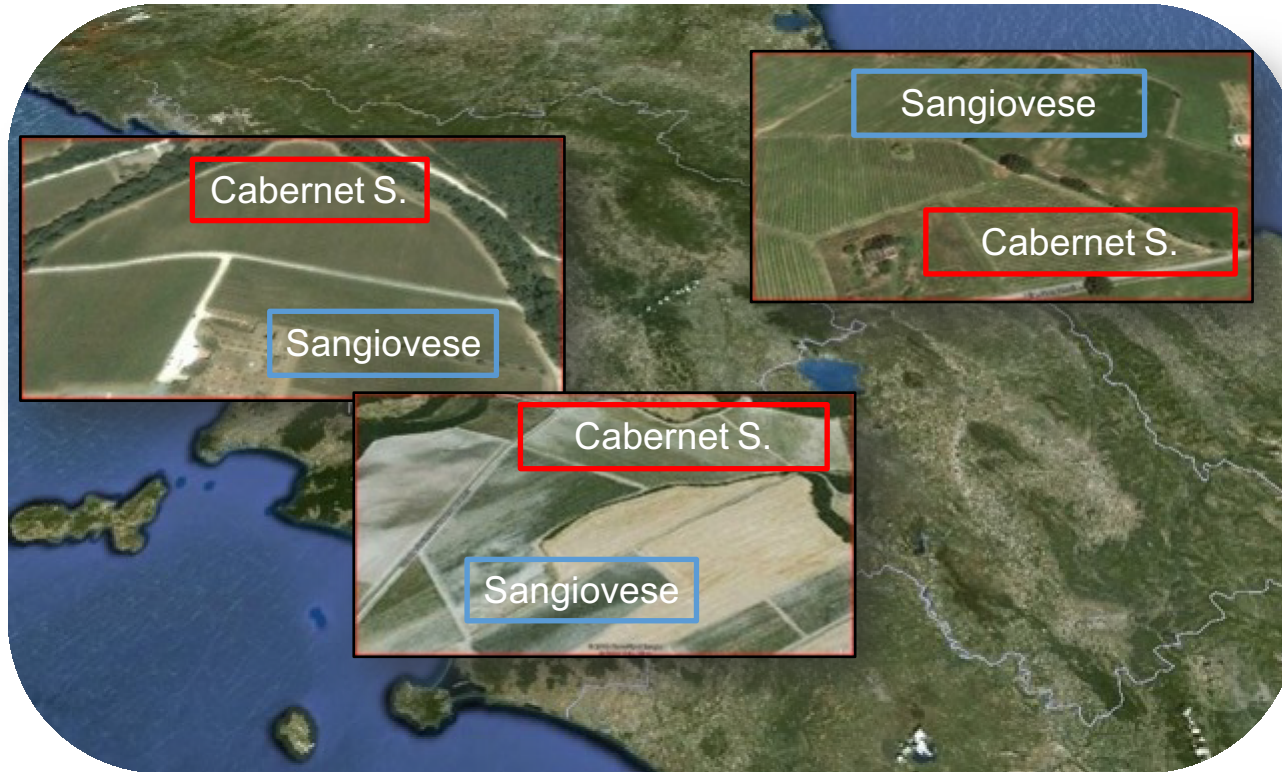
SANGUIS JOVIS

Modificato da Zenoni et al., 2018



Molecular dissection of the grapevine GXE interaction

Changes in performance of genotypes in different environments are defined as genotype X environment (**GXE**) interaction



2 Genotypes
Sangiovese and Cabernet sauvignon

4 Developmental stages

2 Years

3 Areas
Adjacent Vineyards per each Area

Pea Size



Pre-Veraison



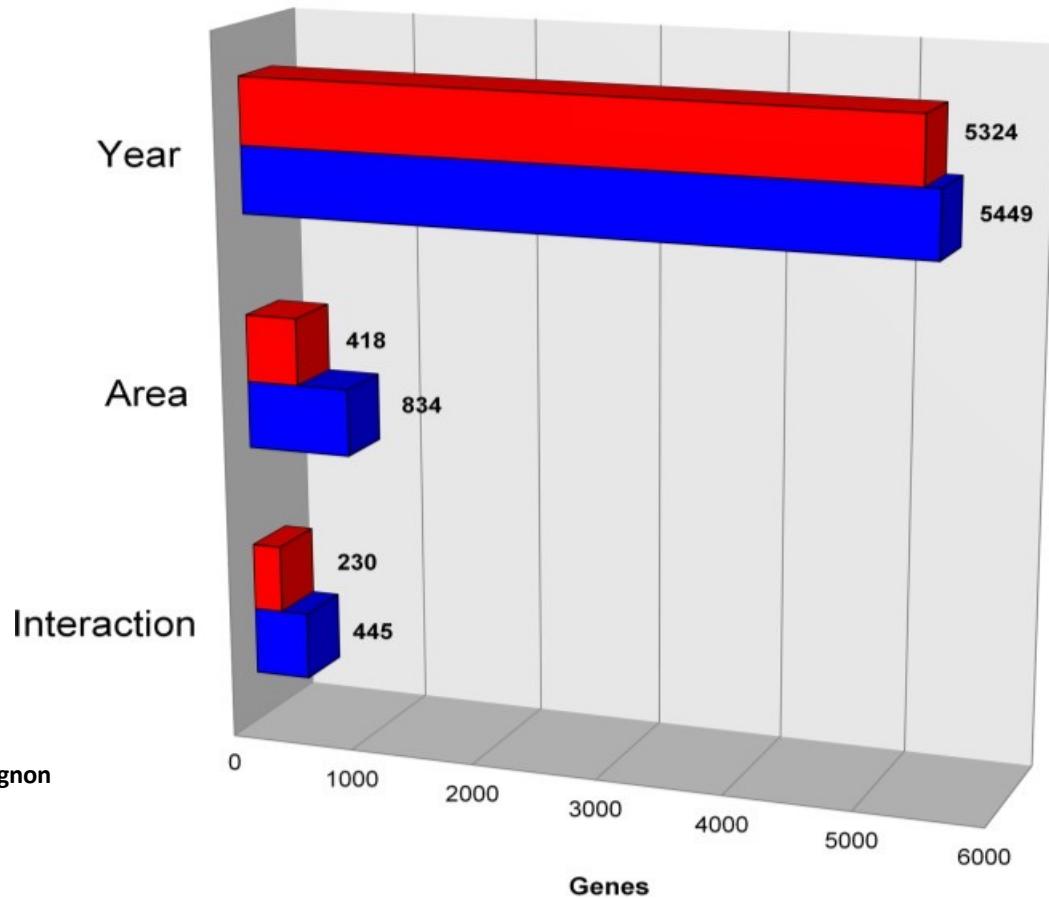
Mid-Ripening



Ripe



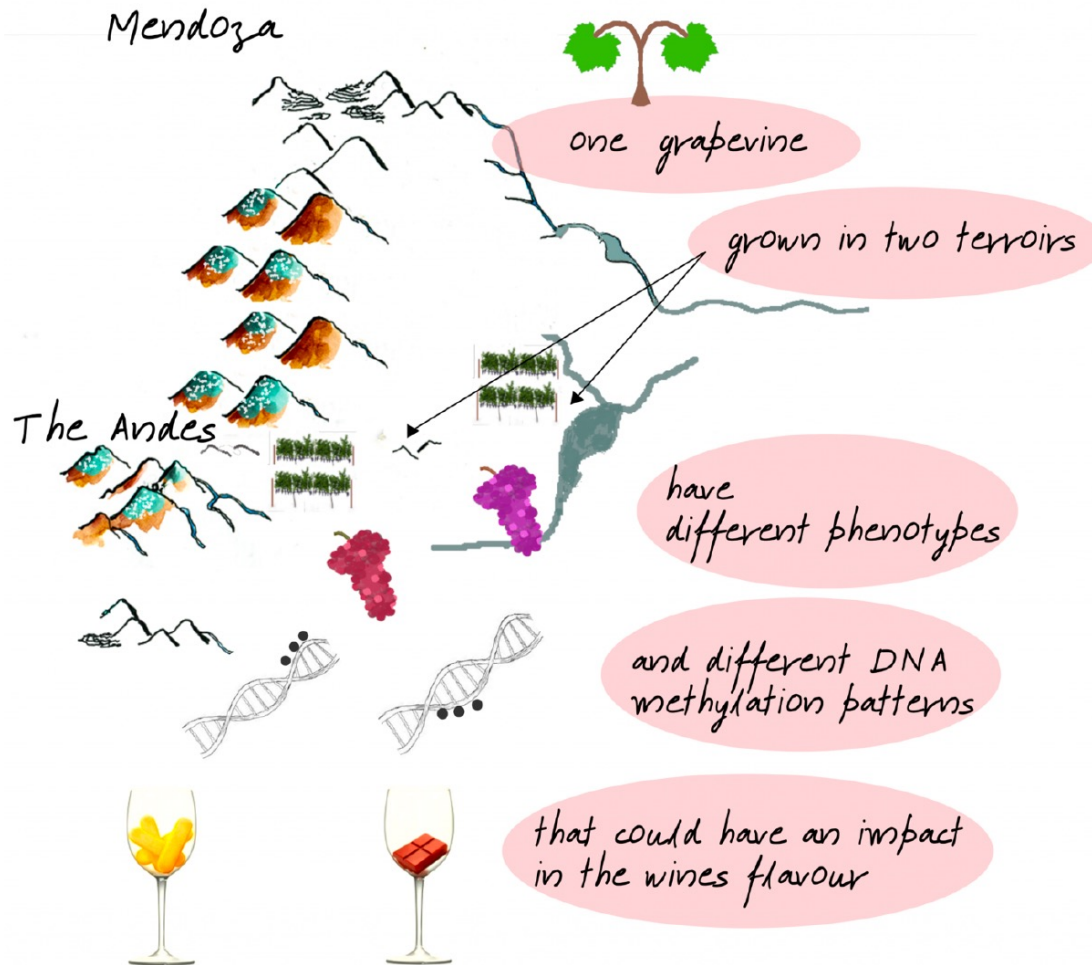
Molecular dissection of the grapevine GXE interaction
ANOVA 2-way (p<0.01)



■ Cabernet Sauvignon
■ Sangiovese

SANGIOVESE RESULTED MORE RESPONSIVE THAN CABERNET SAUVIGNON

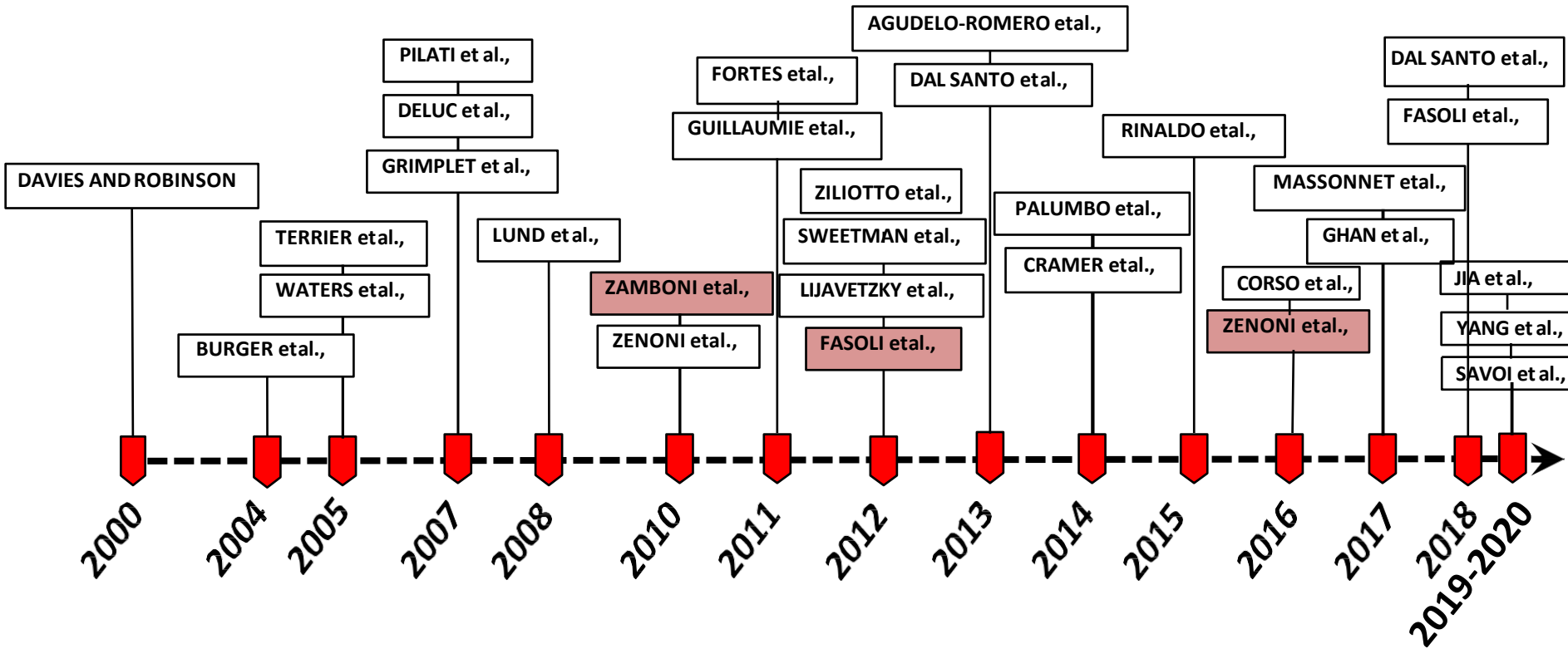
Epigenetic regulation (DNA-methylation) can determine different transcriptome landscape in plant growing in different climate zones



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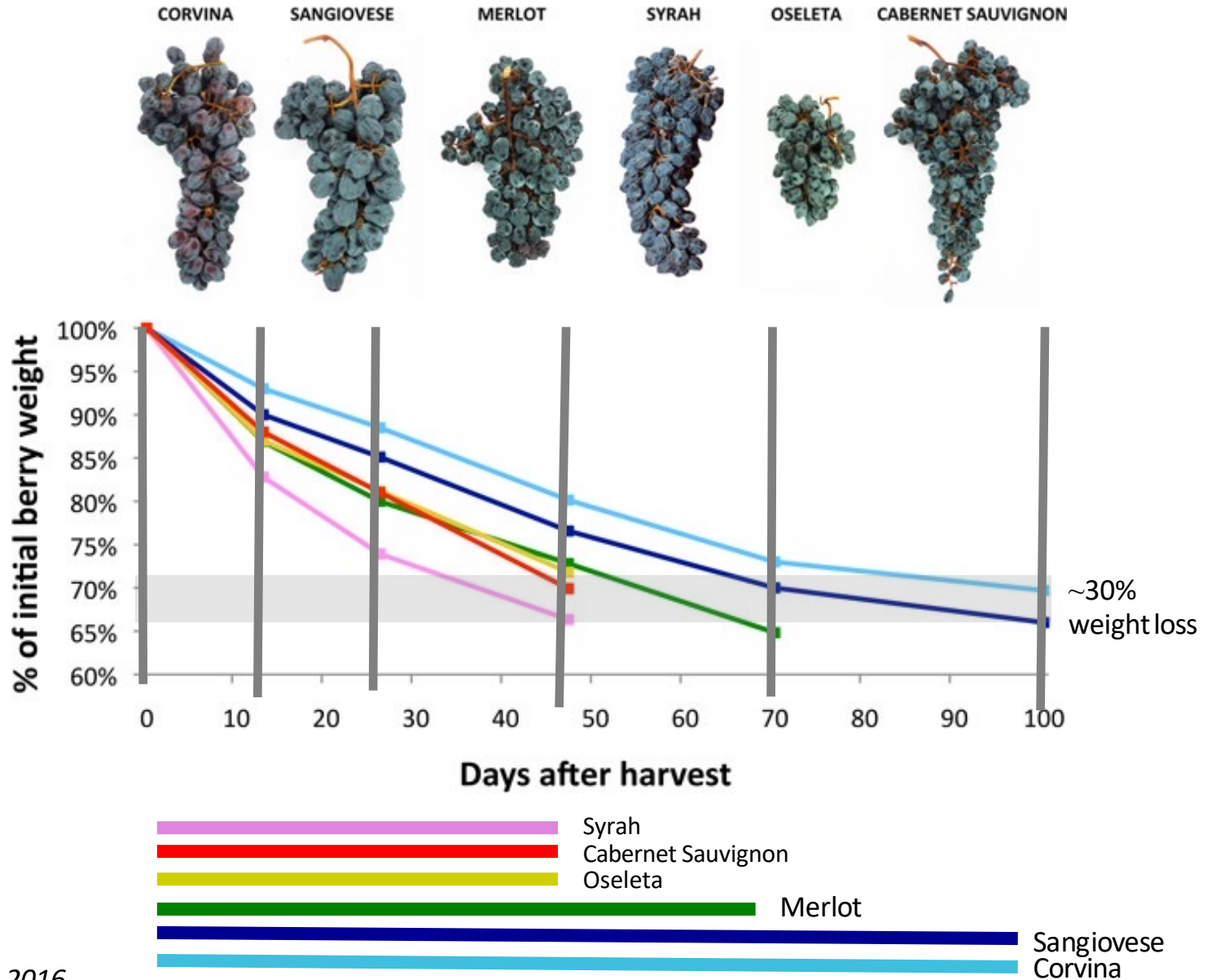
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Large-scale transcriptional changes during berry development



Expression profile of the principal molecular events during postripening phase

Transcriptional changes during berry post-harvest



Transcriptional changes during berry post-harvest

Detachment

Dehydration

Overripening
Senescence



UP
REGULATION

Hormone metabolism

ACD1
Ethylene-responsive protein (2)
Auxin efflux carrier
Nitrilase (4)
VvJAZ9

Secondary metabolism

PAL (11) GENERAL PHENYLPROPANOID
C4H (2)
4CL
STILBENES
STS (19)
ROMT
COMT
CCR
LIGNIN
Dirigent protein (3)
Laccase (8)

Oxidative stress

GST (2)
Peroxidase
Respiratory burst oxidase

Aerobic/Anaerobic respiration

Pyruvate kinase
Lactate dehydrogenase

Defense

Pathogenesis protein (2)
Beta-1,3-glucanase

Osmotic stress

Osmotin (2)

Transcription factors

VvNAC60 VvNAC61
WD40 (2)
WRKY16 WRKY29
Zinc finger C3HC4-type
Myb domain protein

Hormone metabolism

VVERF045
IAA (3)
Auxin-independent growth promoter
Brassinolide-6-oxidase

Secondary metabolism

CCR
LDOX
CYP85
AnthoMATE1
GST4
VvMYBA1 similar
VvMYBA2
ANTHOCYANINS

Oxidative stress

Catalase (2)
Lipoxygenase LOX2

Aerobic/Anaerobic respiration

Alcohol dehydrogenase

Carbohydrate metabolism

Malic enzyme
Sucrose synthase (2)
Ribulose-1,5-bisphosphate carboxylase
Tonoplast monosaccharide transporter

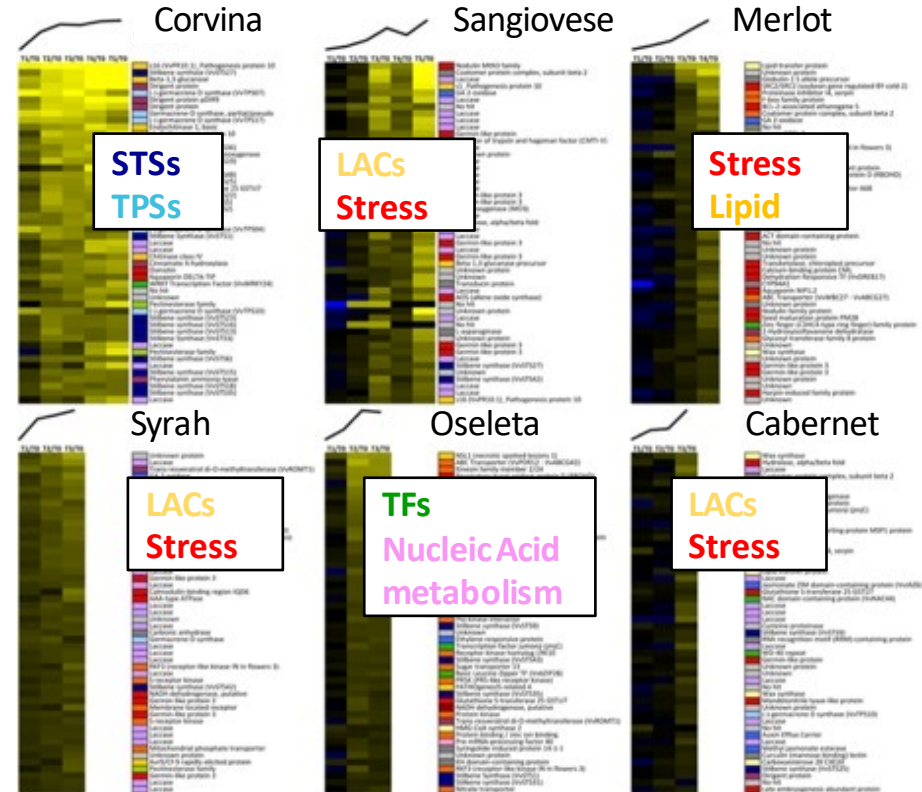
Cell wall metabolism

1,4-beta mannan endohydrolase
Cellulase
Endo-1,4-beta glucanase
Expansin (4)
Esterase-like
Pectate lyase
Pectinacetylesterase (2)
Polygalacturonase
XTH

Transcription factors

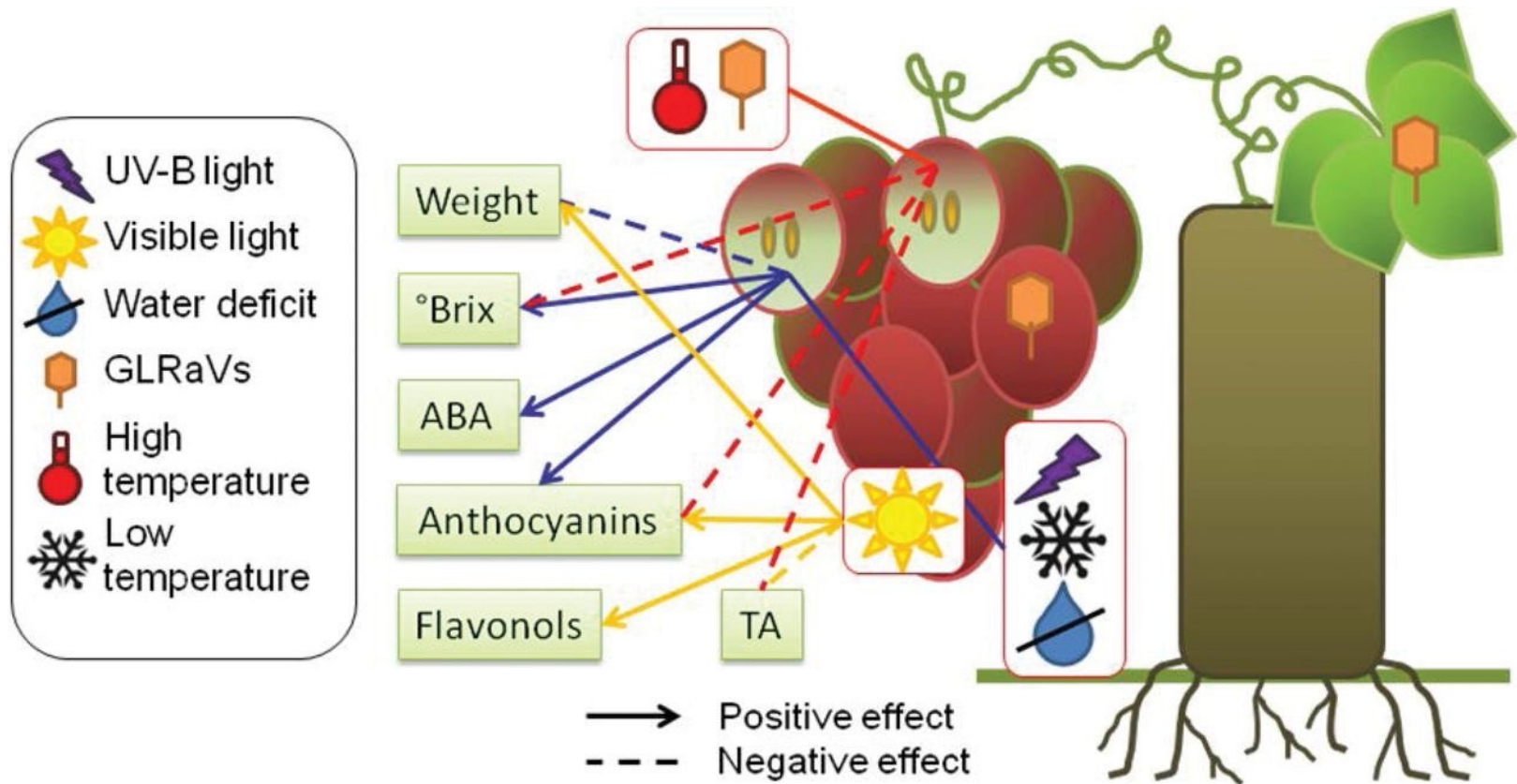
bHLH (2)
VvZIP07
Zinc Finger
MYB Divergata
Zinc finger C3HC4-type

DOWN
REGULATION

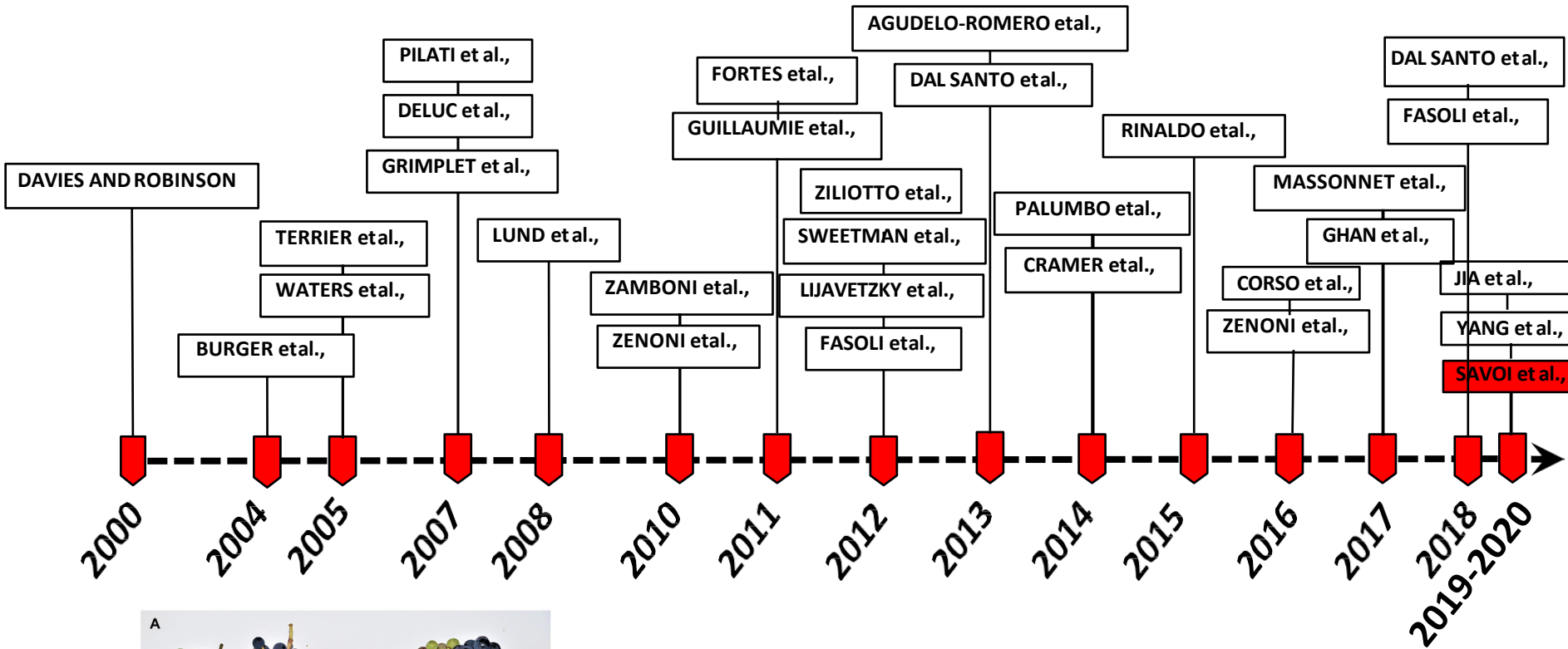


DURING THE POST-HARVEST PHASE THERE IS AN ACTIVE METABOLIC REARRANGEMENT AND NOT ONLY A PASSIVE CONCENTRATION

Fattori ambientali in grado di modificare parametri biochimici associati alla maturazione di bacche pigmentate



Large-scale transcriptional changes during berry development



EST



1.29 g
15.2° Brix

Light and Temperature

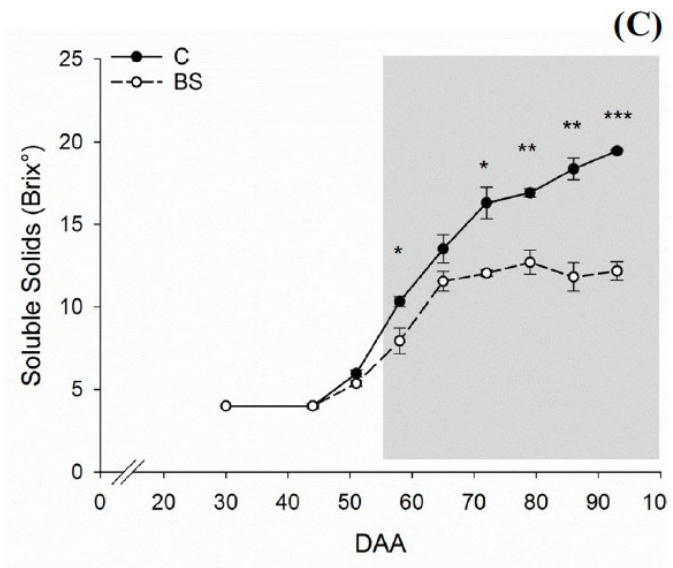
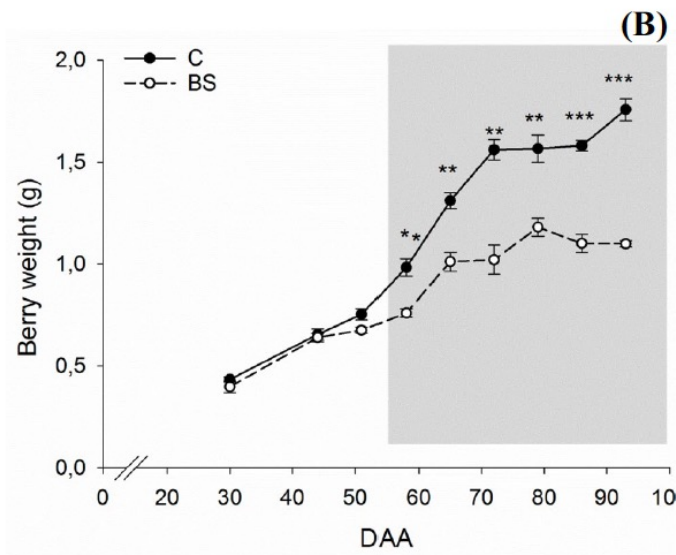
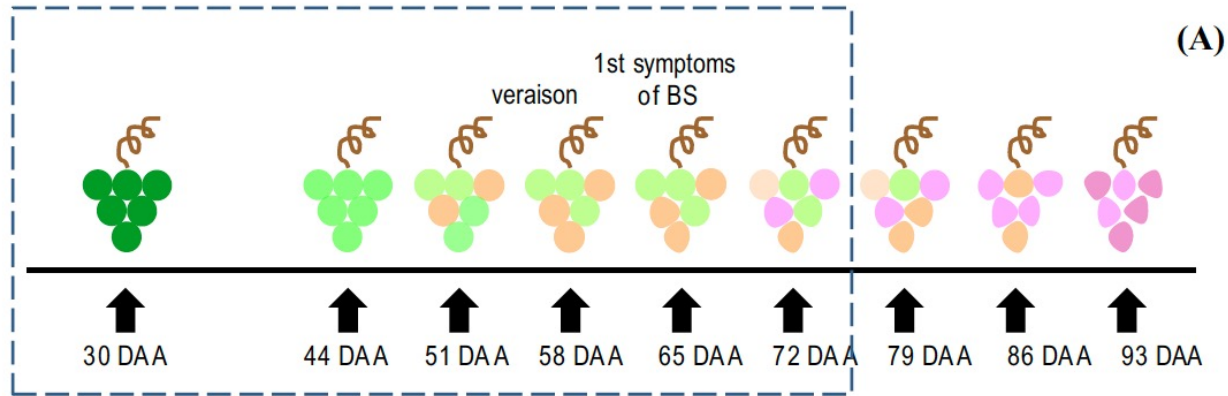
Minimal suburn damages are accompanied by significant differences in berry physical and biochemical parameters

OVEST



1.04 g
8.7° Brix

No metabolic alterations in the berry transcriptome and in the metabolite content was observed in pre-symptomatic and pre-veraison samples.

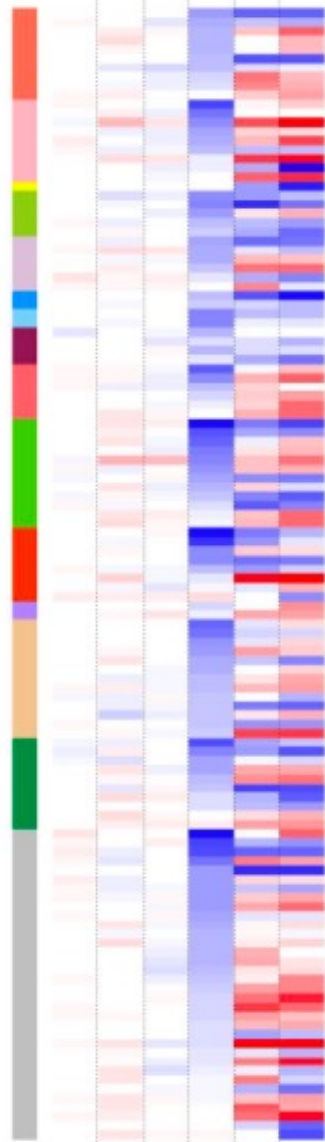




(A)

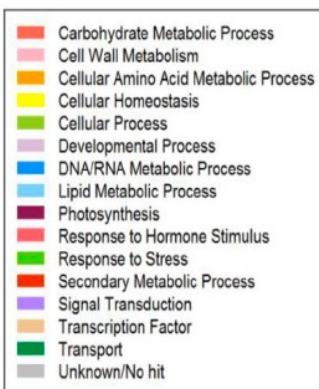
DAA 30 44 51 58 65 72 C

UP in SB
DOWN in SB



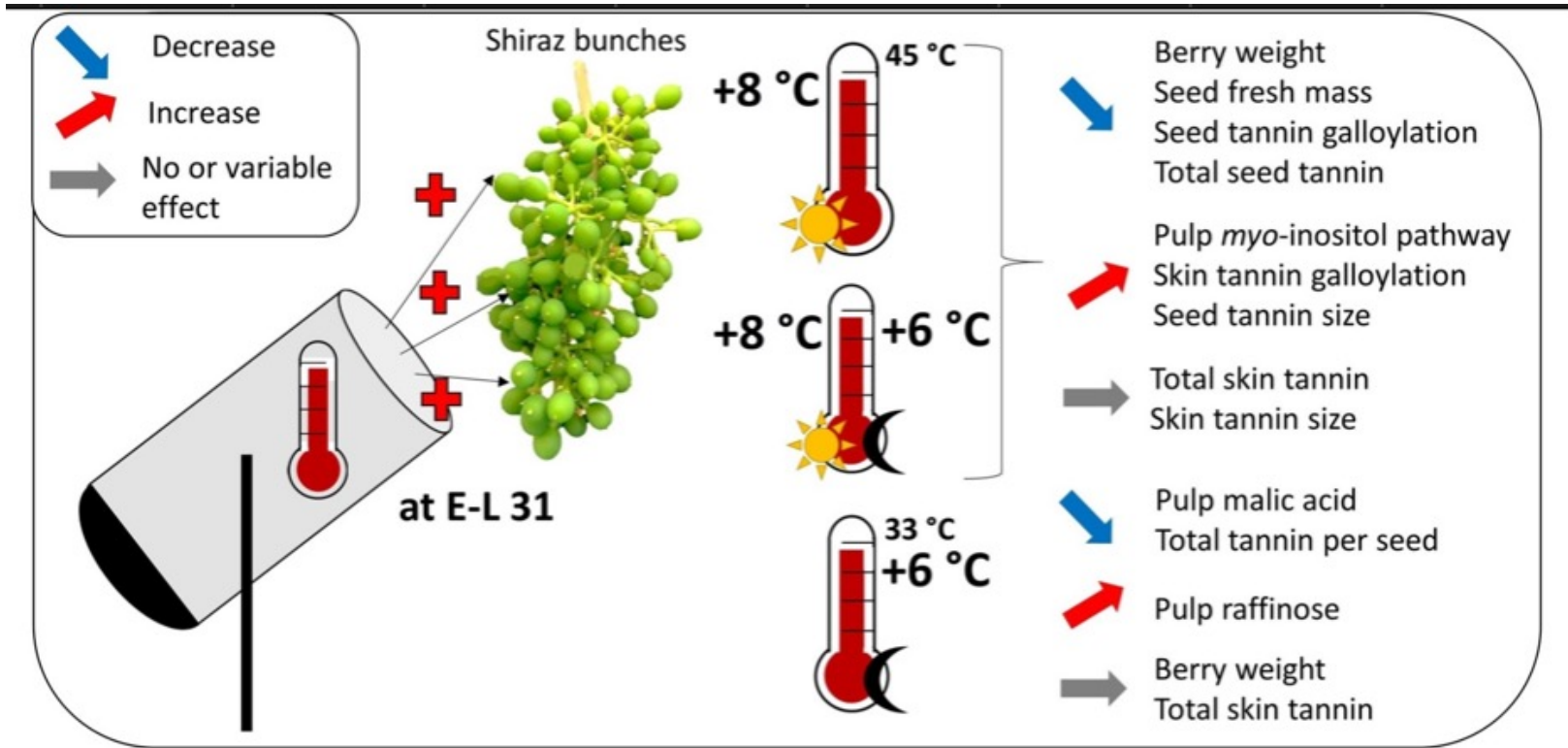
Sunburn berries Transcriptome

Interestingly, at veraison, with still not visible symptoms appearing on the berry, a subset of genes, called **switch genes** previously suggested as master regulators of the ripening onset in grape berries, were strongly lower expressed in BS.



Savoi et al., 2019

Effetto di ondate di calore (20gg dopo fioritura)



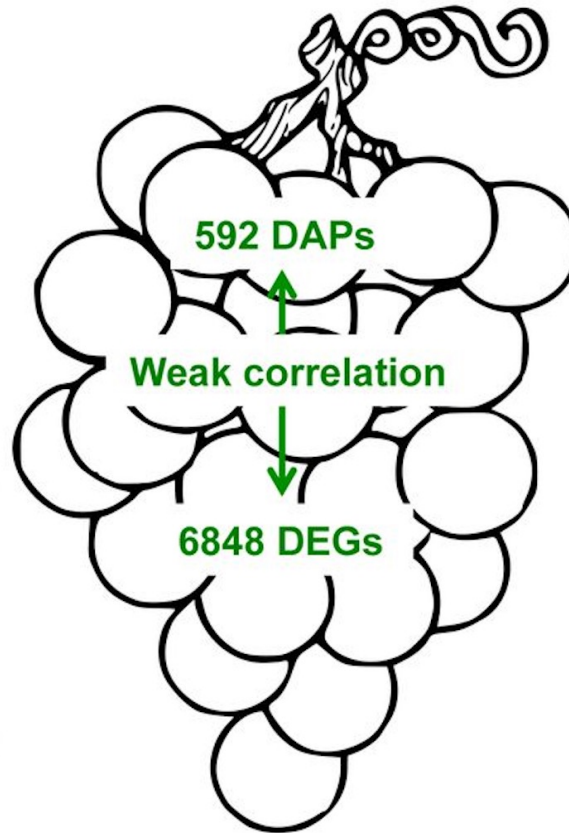
Guot et al., 2019

Proteoma in bacche soggette a colpo di caldo

DAMAGING EFFECTS

- . Protein degradation & unfolding ↗
- . Protein synthesis ↘
- . Transport ↘
 - Sugars
 - K⁺
 - Anthocyanins
- . Carbohydrate & energy metabolism ↘
 - Malic acid
 - Shikimate
 - Sugar phosphate
- . Secondary metabolism ↘
 - Anthocyanins
 - Flavonols
 - Aromas?
- . Hormones & Cell wall ↘

HEAT



TOLERANCE RELATED EVENTS

- . Perception/ Signaling ↗
 - VvCNGC2
 - VvHsf
 - VvAP2/ERF, VvbHLH, VvMYB
 - VvMBF1c
- . Chaperones ↗
 - VvHSP90, VvHSP70, VvHSP101...
 - VvBAG, VvROF1
 - UPR process
- . Protective metabolites ↗
 - GABA
 - Galactinol
 - Vitamines
- . Antioxidants ↗
 - VvGST, VvGR, VvPDI, VvGSNOR,...
- . Protective barrier ↗
 - Lipid metabolism
 - Lignin
 - Cuticule
 - Wax

Differential accumulation of metabolites is induced by light exposure

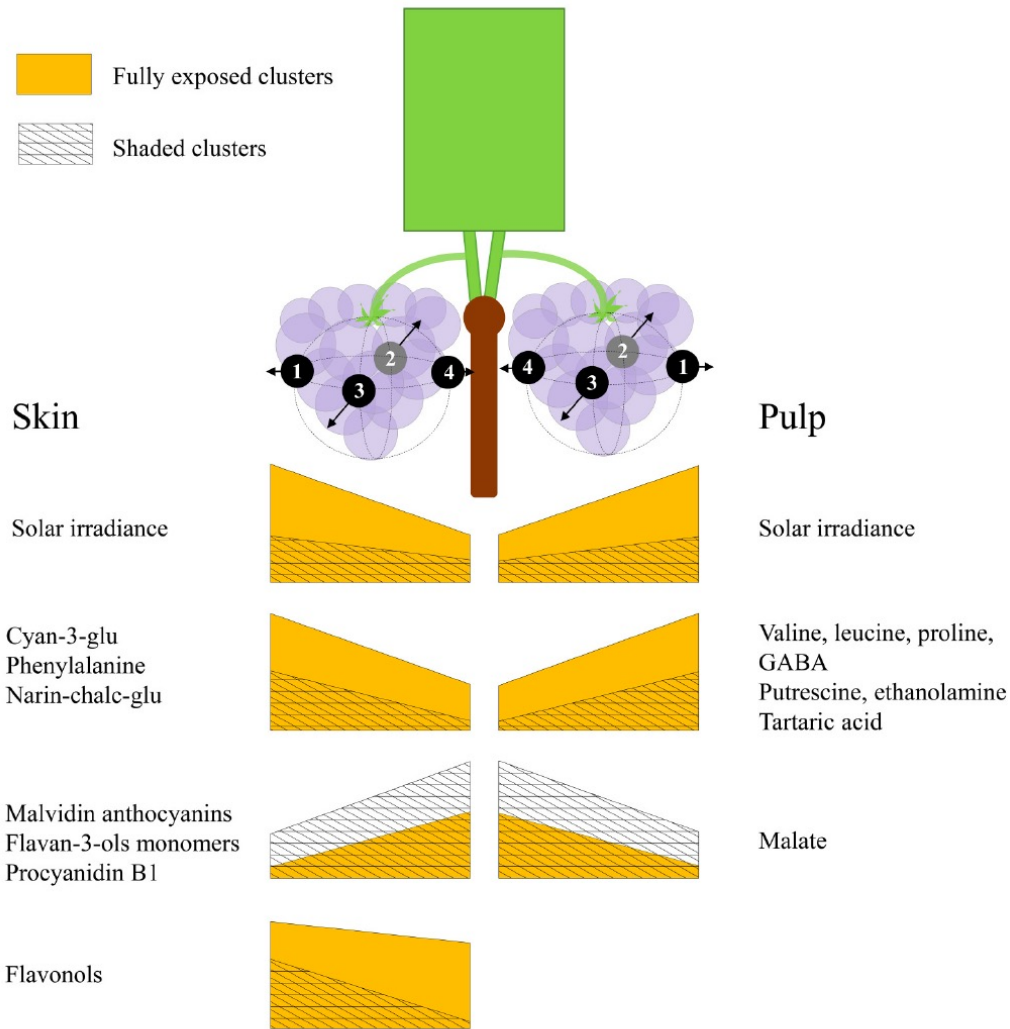
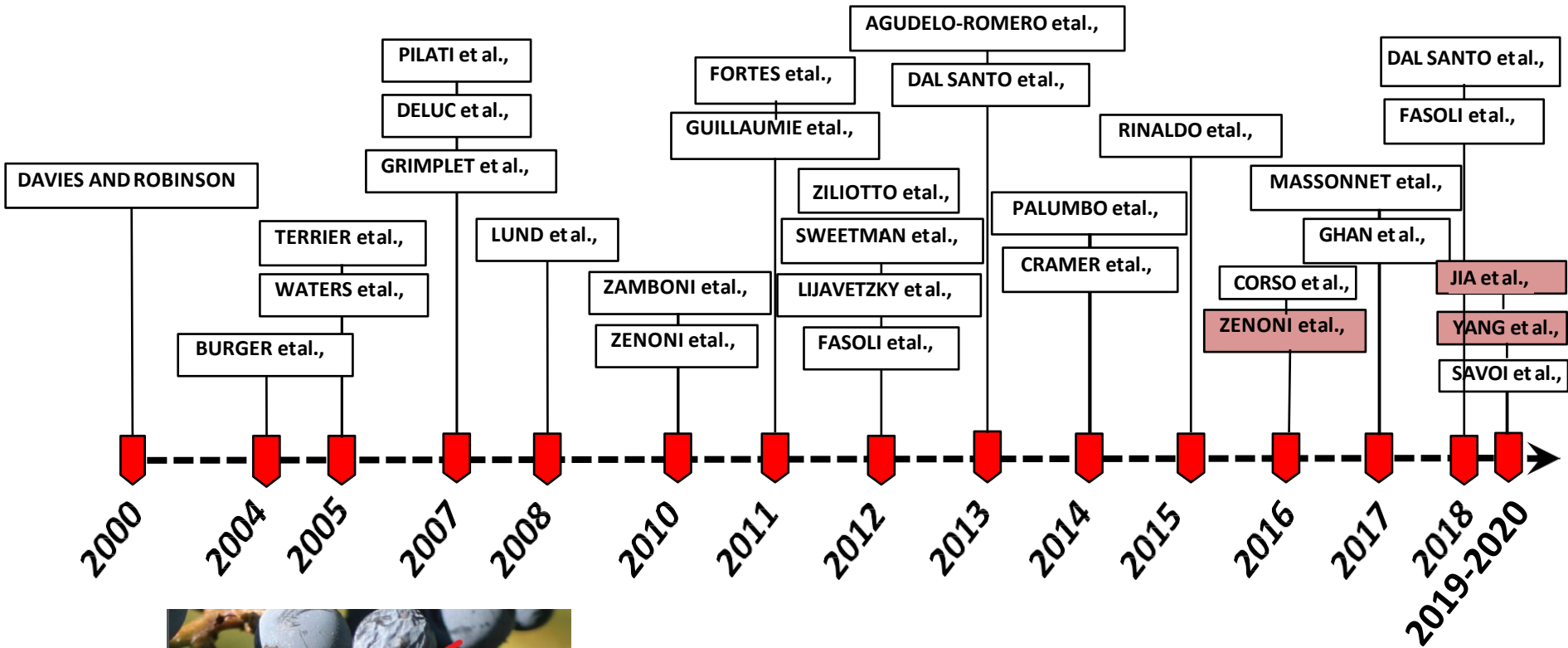


FIGURE 8 | An illustration summarizing the main findings of the study. Both sides of the canopy refer to the same cluster orientations (mirror image), the left side displays the results obtained for skin phenylpropanoids, and the right side for pulp primary metabolites. The trapezoid heights represent metabolite abundance and its pattern of change across the cluster, from internal to external orientations, corresponding with daily incoming SI. Yellow trapezoids represent fully exposed clusters, while mesh trapezoids represent 60% shaded clusters.

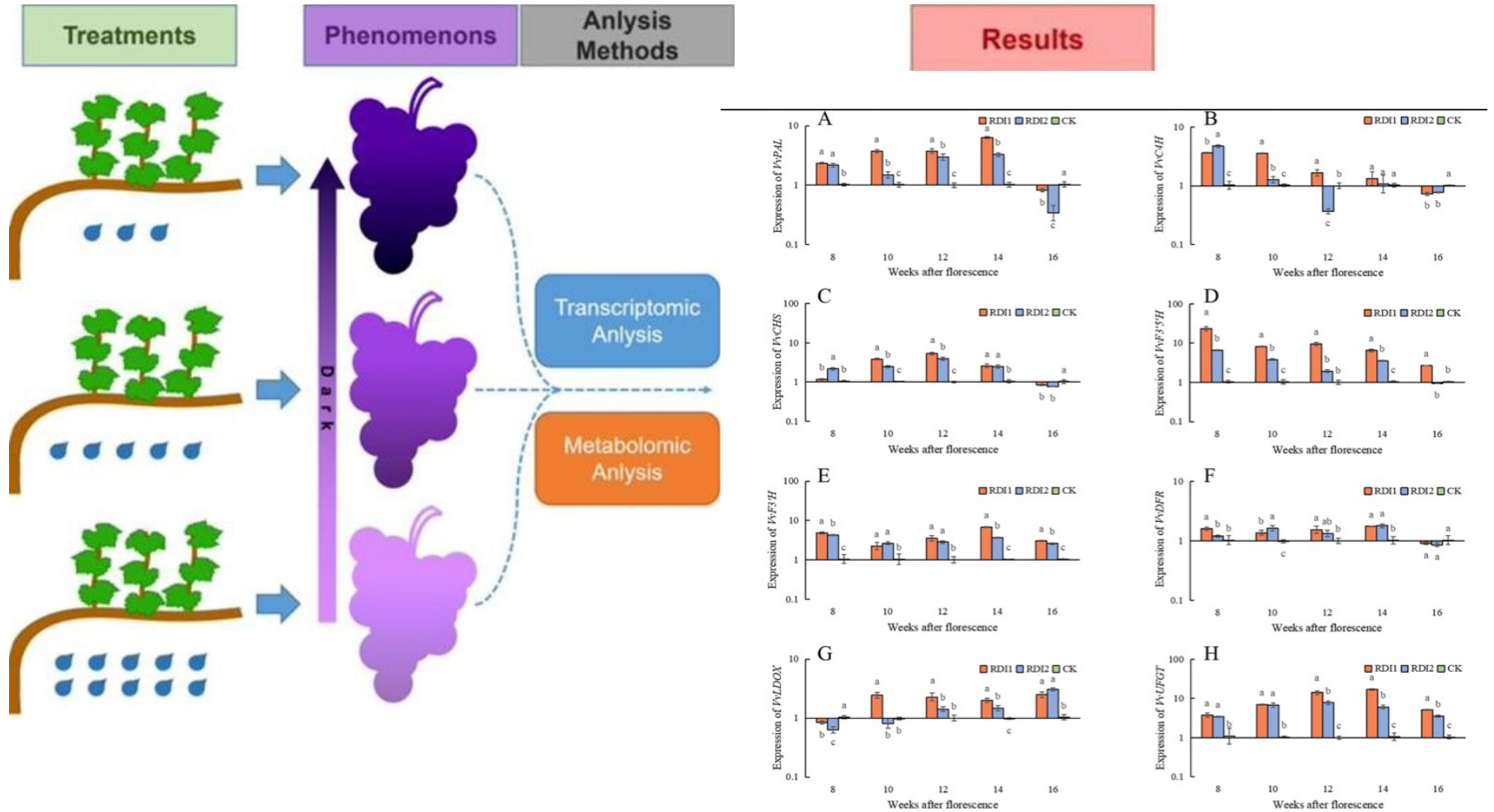
Large-scale transcriptional changes during berry development



Water stress (water-saving irrigation technology)

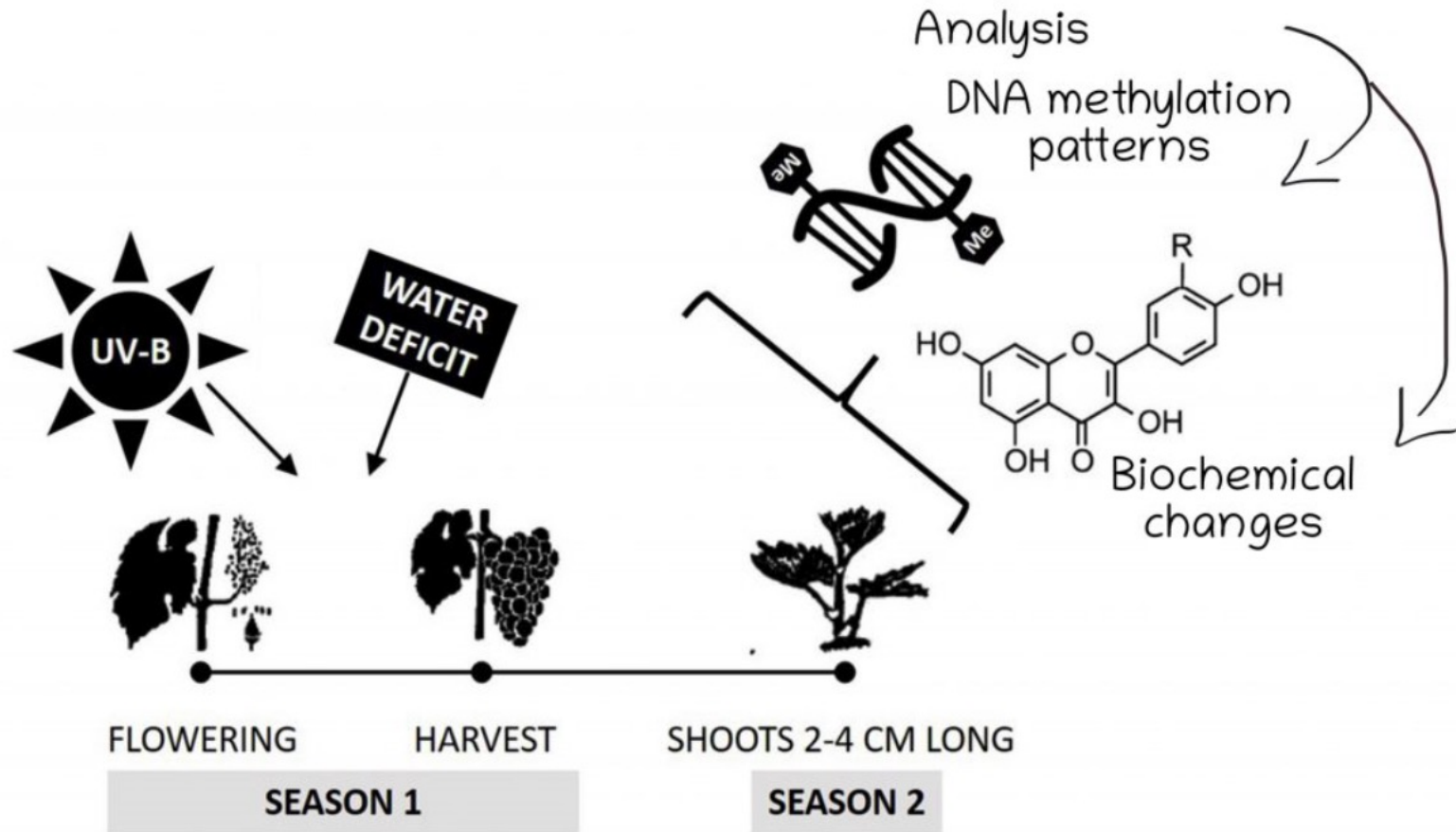
water deficit leads to changes in plant endogenous hormone levels that affect the grape berry's metabolism.

Deficit idrici controllati



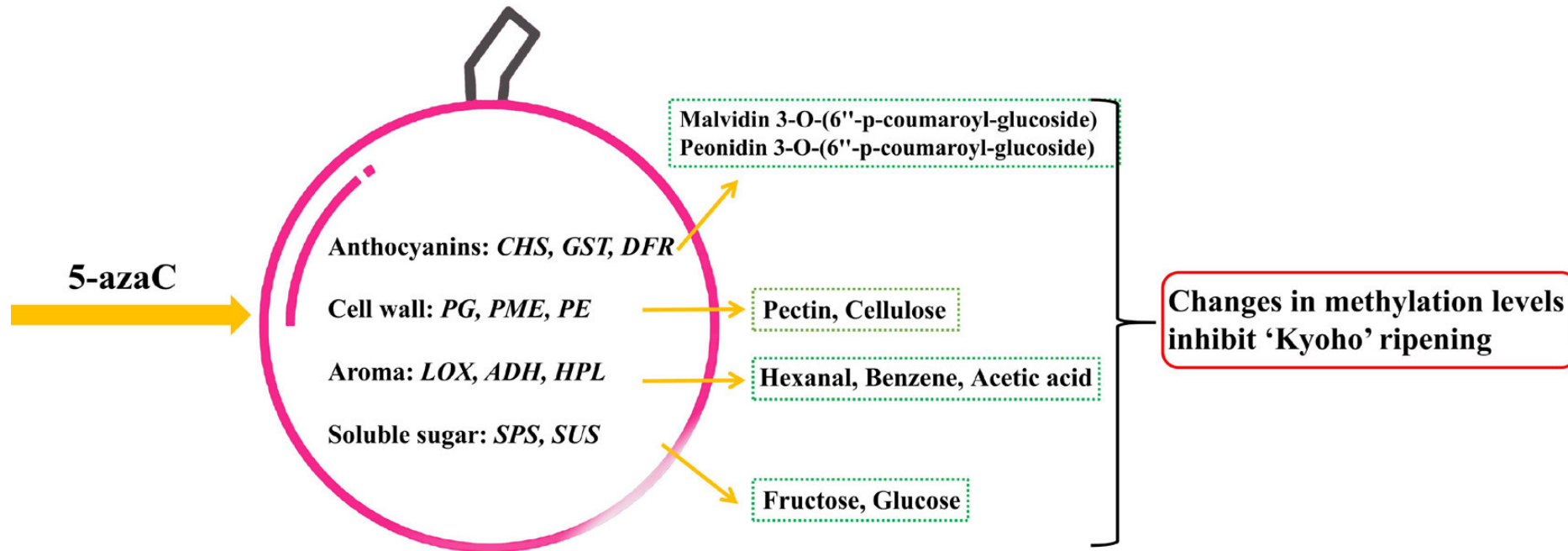
Deficit idrici controllati inducono la trascrizione, quindi, la sintesi di antociani

DNA Methylation

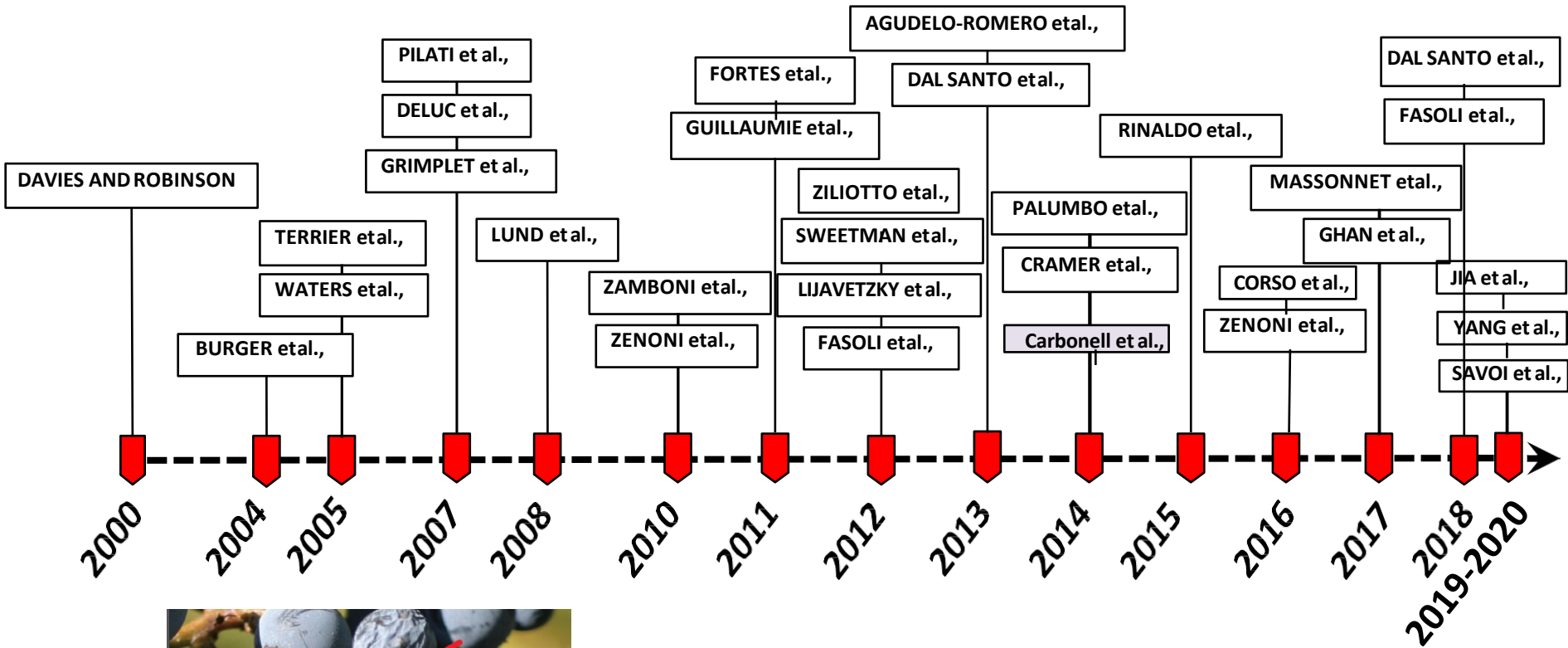


High UV-B and water deficit were the treatments that induced greater number of DNA methylation changes respect to Control (minus UV-B and well-watered). In addition, high UV-B was associated with flavonols accumulation, suggesting that DNA methylation could regulate sensory quality compounds accumulation and participate in acclimation mechanisms.

Modifiche dello stato di metilazione non programmate possono influire sulla progressione della maturazione



Large-scale transcriptional changes during berry development



Circadian clock

Transcriptional changes are activated to prevent biotic and abiotic stresses when they are more probable in the day

Genes differentially expressed in a 24 h cycle

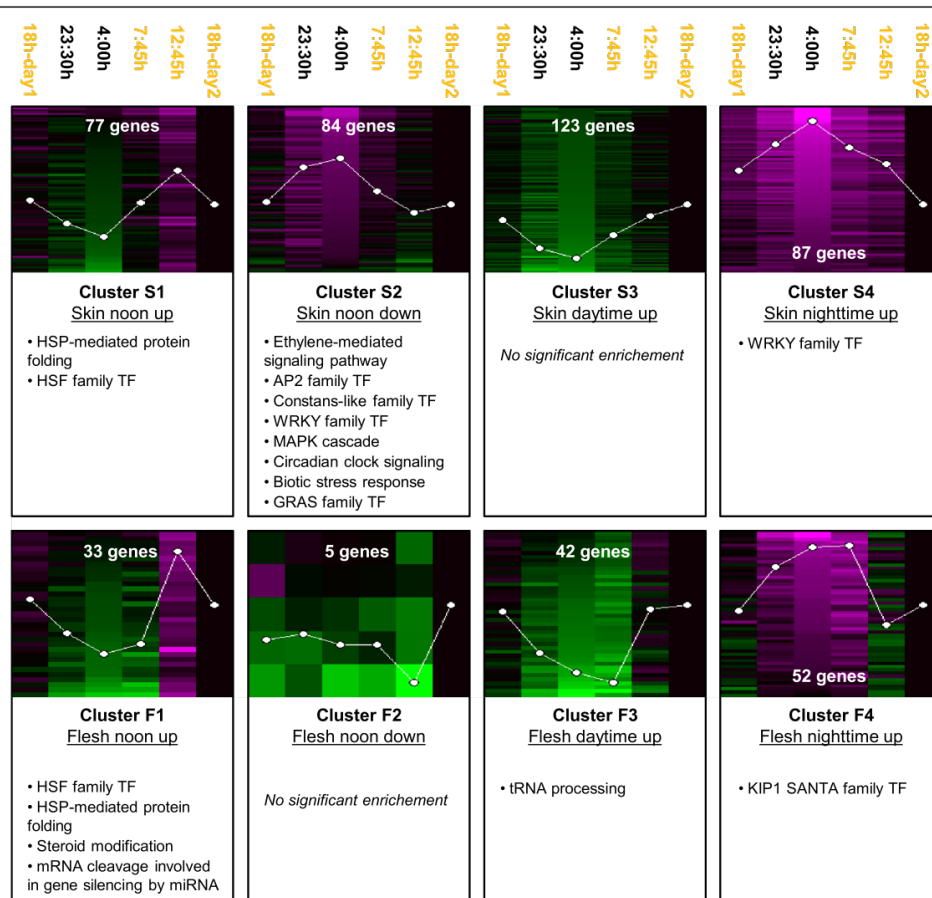


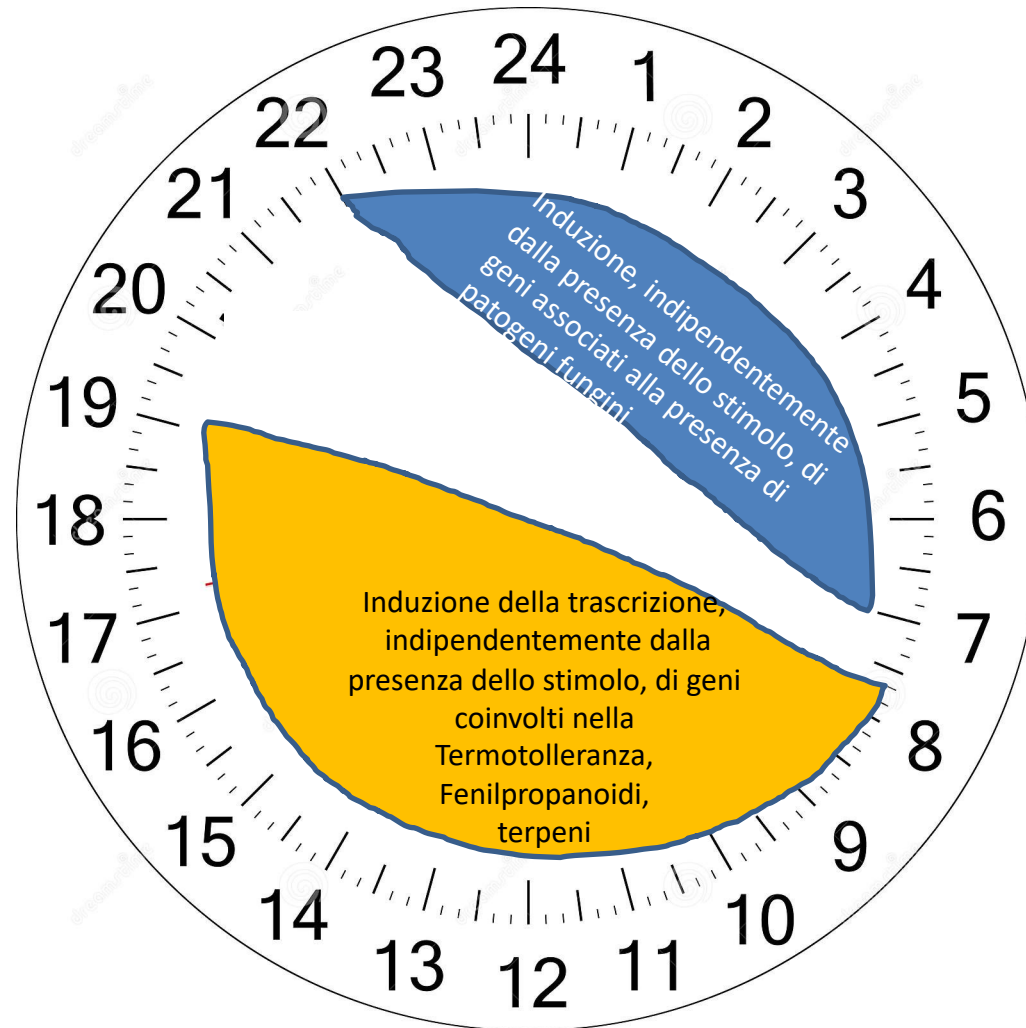
Figure 2 Clustering and functional analysis of Tempranillo skin and flesh DEG throughout a 24 h daily cycle. Significant transcripts identified for each tissue (5% FDR in Limma and ≥ 2 -fold change) were clustered by SOMs. Four main expression profiles were identified in the skin (clusters S1 to S4) as well as in the flesh (clusters F1 to F4) of Tempranillo berries. \log_2 expression normalized to the last time point is represented within each cluster; no difference of expression is represented in black, higher expression in magenta and lower expression in green. Number of genes within each cluster is written in white. Time points in the light period are indicated in yellow. A summary of over-represented functional categories (5% FDR) ordered by their significance level is indicated for each cluster profile.



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Regolazione circadiana della trascrizione di geni associati alla maturazione e alla resistenza a stress biotici ed abiotici



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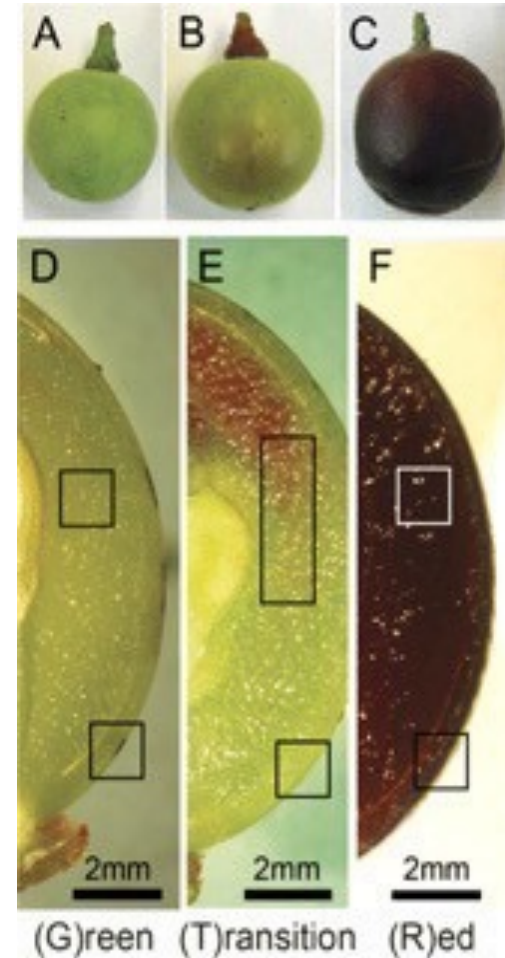
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Conclusioni

- ❖ Le nuove tecnologie hanno prodotto una massa di dati imponenti. Il collo di bottiglia nel loro utilizzo è la capacità di interpretarli
- ❖ L'intricata rete regolativa che opera durante la maturazione si sta dipanando e il pool di geni (switch genes) la cui espressione segna l'inizio della maturazione sono stati individuati sulla base della loro espressione
- ❖ Sono iniziati studi funzionali per identificare la funzione di questi geni

Passi successivi

- Identificazione dei geni target degli «switch genes»
- Investigare sul ruolo delle modifiche epigenetiche sul controllo dell'espressione genica durante la maturazione
- Caratterizzare il ruolo di microRNA sulla regolazione degli «switch genes»



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ALTA SCUOLA DEL SANGIOVESE