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Full Length Research Paper

Study on effective berry-independent method for grapevine evaluation

***Zhao Yong, Xiamo H. A and Cheng M. O.**

College of Food and Nutritional Engineering, China Agricultural University, Beijing 100083, China.

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The quality and characteristics of grape are fundamentally determined by its biochemical components. Quantitative detection of these components in berries is a classic method to evaluate grapevine resources. However, fruits are not always available for the new generated grape plantlets due to their long juvenile stage (3 to 4 years), as well as for many other potential valuable germplasm resources, such as wild grapes. Therefore, an effective berry-independent method for grapevine evaluation should have great significance. Data were provided from both leaves and berries for 2 groups of grapevine: one group is 12 genotype different varieties or species from environmental similar collections; the other group is one variety of wine grape with 18 different treatments. After quantitative correlation tests, 9 in total 11 detected parameters in genotype different (GD) group and 5 in 9 detected parameters in treatment different (TD) group, respectively, were significantly correlated between leaf and berry, respectively were found. Higher correlation coefficients were found in GD group than in TD group. Parameters of leaf reducing sugar, total flavonoids and superoxide anion scavenging capacity were found significantly correlated to berry, in both groups. These parameters with significant correlation may potentially be used as metabolite markers to estimate the qualities and characters of some new grapevine germplasm, by using the obtained data from leaves. The prospects of this leave-dependent evaluation method have also been discussed in this report.

Key words: Leaf-dependent berry evaluation, leaf/berry quantitative correlation, parameter pair, inter-parameters pair.

INTRODUCTION

Grapevine is one of the most widely planted fruits in the world, and a large proportion is used for wine making. The pursuing for high quality, distinctive features and high adaptabilities of cultivars raises the needs of rapid development of wine grape breeding. At present, thousands of varieties have been developed and many of them broadly utilized in wine industry for their good quality or distinctive adaptation characters all over the world (Alleweldt and Possingham, 1988; This et al., 2004). New cultivars of grape are always generated from crosses using inter or intraspecific grapevine resources or domesticated from wild grapes (*Vitis* species) (Reisch

* Corresponding author. Email: zhao.yong@yahoo.com

et al., 2012). Regardless of the origins of a new variety, the systematic evaluation will be essential before it can be applied in viticulture. In order to select efficiently appropriate cross parents and screen out elite offspring, systematic evaluation of large amount of germplasm resources and cross progeny is indispensable, but a long-term and hard-task process (Alleweldt and Possingham, 1988; Nejatian, 2006). Traditionally, to evaluate potential grapevine germplasm, the candidates should be grown until they produce fruits. Adaptability and other agronomic features can be evaluated during the juvenile stage. The most important procedures are the biochemical evaluation of the berries, which have to wait for 3 to 4 years from planting the cross progeny as a result of the long juvenile stage of grapevine. Nowadays, the quality evaluation procedures is always carried by qualitative and quantitative determinations to berry composition (Guidetti et al., 2010; Shiraishi et al., 2010), and the long delay between juvenile stage to productive stage becomes a bottleneck of rapid selection. Moreover, collection of ripen grape berries from some potentially useful wild species in natural conditions also has difficulties, because of the unpredicted mature time and birds feeding. In contrast, grape leaves of any development stages is easily harvest, especially for wild resources. Therefore, if a leaf-dependent pre-evaluation method can be successfully applied in berry evaluation, time and workload in vine breeding will decrease dramatically. Sine after an earlier leafy compositional and quantitative screening, one could only focus on those most potential candidates. While most of these biochemical characters have not any detectable genetic marks for this purpose.

Moreover, if quantitative responses of certain metabolites in fruits always correlated significantly to their leaves, one may predict the effects of environmental perturbations on berry composition based on results from a leaf or tissue assays. Despite its potential importance, there are no studies assessing the quantitative correlation of biochemical traits between leaf and fruit in plants. In this research, the correlation of several important biochemical traits were tested between leaf and berry from various grape varieties, as well as one cultivar but treated differently. A leaf-dependent prediction method for berry evaluation was then proposed based on the analyzed quantitative correlations of these detected traits. Many of these detected parameters such as sugar, acidity, flavonoids, phenols contents, and anti-oxidative capacities are fundamental in grape quality and characteristic evaluation.

MATERIALS AND METHODS

Plant and experiment design

Commercially ripen fruits and full developed healthy leaves of 11 varietiesofgrapevines(*Vitisvinifera*)andawildspecies(*Vitisheyneana*) were sampled and used in the quantitative analysis for measuring some biochemical and physiological parameters, from vineyards in Qiubei county, Yunnan province, China in 2012 as genotype different (GD) group. Grape cultivars in GD group include Yan73 (v1), Beijixing (v2), Xiahei (v3), Rose honey (v4), Crystal (v5), Cabernet Sauvignon (v6), Red rose (v7), Faguoye (v8), Zhengzhou Dawuhe (v9), America No.1 (v10), Merlot (v11), and a wild species *V. heyeara* (v12). All these field-grown grapevines in a germplasm collection were 5 to 7-year old, spur pruned, with a density of 1.6 m between rows and 1 m between plants. Vineyard management followed the local standards. Another 18 samples as treatment different (TD) group, were harvested from a wine grape cultivar cv. Rose Honey growing in a commercial vineyard (5-year old, also spur pruned, with a density of 1.2 m between rows and 0.9 m between plants) with 18 combinations of fungal regents and pesticides. Vines were separated into 2 parts and one part inoculated with 8 different strains of fungi with a non-fungus inoculation control, and followed the local management for 4 times of pesticides applying. Other parts were also inoculated with the same strains of fungi and a non-fungus inoculation control, but without any pest controlling (pesticides free). Each single treatment con-tains 10 grapevines. The purpose of this treatment was to create the quantitative variation of metabolites in grapevine.

Vines without obvious visible disease symptoms of each variety were sampled randomly from at least 6 plants of GD group. Samples from every 2 vines pooled as one replicate for both leaf and fruit, respectively for each variety, and preserved in an ice box, delivered to lab within 4 h for processing. For berry sampling, 2 ripen clusters for every vine were taken. For leaf samples, almost the same position (4 to 6th from the bottom of the fruit cane), similar size, full developed healthy leaves were sampled. Samples of TD group were harvested with the same method above at berry ripen stage (67 days after treatment). Six grapevines were also sampled and samples (both fruit and leaf) from every 2 grapevines were pooled as one replicate.

Determination of physio-chemical traits

Pre-treatment of leaf and berry samples

All leaf samples were cut into about 1 cm^2 pieces for each sample. Randomly selected ripen berries were picked off from clusters of each replicate sample and well mixed up. About 20 g of leaf pieces and randomly selected berries for each samples were homogenized into fine powder in liquid N_2 with a stainless grinder and transferred to a 50 ml tube, then stored at -80°C for reducing sugar, titratable acidity, total phenols, soluble protein and enzyme activity analysis; the rest of the samples were dried in wind-oven following a program of 110°C for 10 min, 80°C for 48 h (72 h for berries) and then ground into fine powder with a stainless grinder for the measurement of total flavonoids content, 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical, and superoxide anion savaging capacities.

Determination of reducing sugar (RS) and total sugar (TS)

Fresh sample (1 g) was added with 4 ml 1 mol/L zinc acetate (containing 3% glacial acetic acid) and 4 ml 0.25 mmol/L potassium ferrocyanide, and extracted in 80°C for 10 min with 2 times of vortex. The mixture was centrifuged at 5000 rpm and the supernatants was adjusted to pH=7 by adding calcium carbonate powder. After 30 min in 60°C water bath with several times of vortex, the solution was cooled to room temperature, and metered the volume to 10 ml with distilled water. After 10 min centrifuge at5000 rpm, the supernatant was titrated with alkaline tartrate copper solution A+B (Dygert et al., 1965). The consumption of the supernatant was used to calculate the contents of RS. TS was

obtained by pre-treating the homogenate with 6 mol/L HCL and then follow the same procedure as that of RS.

Titratable acidity (TTA)

Titratable acidity was determined by sodium hydroxide direct titration. About 1 g fresh sample was weighed and extracted in a boiling water bath for 30 min, vortex several times during the bath to get all the organic acids dissolved in the solution. After cooling to room temperature, the solution was centrifuged at 5000 rpm for 10 min, the supernatants was titrated with 0.01 mol/L standard solution of sodium hydroxide. The consumption of sodium hydroxide was used for total acid calculating, and described as the content of tartaric acid (mg/g fresh weight, FW).

Total flavonoids (TF) content

Total flavonoid content of berry and leaf were determined with dried samples by using the aluminum chloride colorimetric method (Willett, 2002), with some modifications. Methanol extracts (0.5 ml), 10% aluminum chloride (0.1 ml), 1 M potassium acetate (0.1 ml) and distilled water (4.3 ml) were mixed after incubation at room temperature for 30 min. The absorbance was measured at 415 nm. Total flavonoid content was calculated by comparing the calibration with rutin trihydrate as standard substance.

Total phenols (TPh) content

Total phenols were determined according to the method of Forint phenol colorimetric. About 1 g fresh frozen sample was used to extract and determined the total phenols (Asami et al., 2003). TPh content was standardized against gallic acid and expressed as milligrams per liter of gallic acid equivalents.

Determination of lipid peroxidation

Lipid peroxidation was estimated by measuring the concentration of thiobarbituric acid reacting substances (TBARS), as described by Dhindsa et al. (1981). Fresh frozen tissue (0.5 g) was extracted with 10 ml trichloroacetic acid (TCA) 0.1% (w/v) for 10 min with 2 times of vortex. The mixture was centrifuged at 6000 rpm at 4°C for 10 min. 2 ml of supernatant were mixed with 2 ml 20% TCA solution (containing 0.5% (w/v) thiobarbituric acid. The mixture was heated at 95°C for 30 min, quickly cooled and centrifuged at 13,000 rpm and 4°C for 10 min. The absorbance of the supernatant was read at 532 nm with the values for non-specific absorption at 600 nm subtracted. TBARS concentration was calculated using the following formula (Heath and Packer, 1965):

TBARS concentration = [(A532 × 1000) - (A600 × 1000)] /155

Determination of total soluble protein and antioxidant enzymes

Fresh tissue (1 g) was added with 10 ml of 0.1 mol/L potassium phosphate buffer (pH 7.0), containing 0.1 mmol/L ethylenediaminetetraacetic acid (EDTA)-Na_2, 0.5 mmol/L ascorbate and 1% polyvinyl polypyrrolidone (PVPP) and stood for 30 min with several times of vortex. The mixture was centrifuged at 13,000 rpm under 4°C for 10 min. The supernatant was used for determinations of protein content and antioxidant enzyme activity. Total soluble protein concentration(SPr) was determined as described by Bradford (1976) using bovine serum albumin as standard. Superoxide dismutase (SOD) was determined by the nitro-blue tetrazolium (NBT) method (Dhindsa et al., 1981), and guaiacol peroxidase (GPX) assay was performed using the method described by Amako et al., 1994).

Activity of phenylalanine ammonia-lyase (PAL)

The extraction and determination of PAL was performed according to the method of Carolyn et al. (1996), with some modifications as described by Xi et al. (2013). Only samples from TD group were analyzed for PAL in this research.

DPPH radical scavenging capacity

Dried sample was ground into fine powder, and about 1 g was accurately weighed into a volumetric flask. DPPH radical scavenging active substances were extracted by adding 50% of ethanol and sonicating for 30 min in an ultrasonic chamber. The mixture was filtered and the filtrate was diluted into gradient concentrations for further detection. DPPH radical scavenging capacity was measured and calculated by using the method of Li et al. (2012); absorbance was read in a spectrophotometer (S22, Biochrom Libra, England) and results were described as percentage of DPPH radical scavenged (Li et al., 2012).

Superoxide anion scavenging capacity (SA)

Preparation of gradient concentrations of sample extract is as the same process as DPPH radical scavenging capacity determination. The measurement of superoxide anion scavenging capacity was following the method of Li et al. (2012), and the SA scavenging capacity was described as percentage of superoxide anion scavenged (Li et al., 2012).

Data analysis

All data were reported as means ± standard variation values of 3 biological replicates, and analyzed by using the software of SPSS version16.0 (SPSS Inc., Chicago, IL, USA) for windows. One-way analysis of variance (ANOVA) and Duncan's multiple range tests were used for the significance determination with a significant level of 0.05. Pearson's correlation test was conducted to determine the correlations between parameters within or between leaf and berry.

RESULTS

Values of the detected parameters presented as means ± standard variation with different significances of all samples, including leaves and berries in different groups were listed and are shown in Tables 1 to 4, respectively. As genotype different resource, values of each parameter in GD group varied significantly ($P<0.05$), both in leaf and in berry (Tables 1 and 2). In TD group, the same cultivar was subjected to different treatments, and the values of every parameter also varied among treatments (Tables 3 and 4). Therefore, quantitative variations of these parameters can not only be caused by genetic factors but also by the given treatments (environment factors).

However, coefficients of variation caused by genotype are obviously higher than that of the environment factors,

Table 1. Leaf results of detected biochemical traits of genetic different (GD) group of grapevine.

Cultivar	TS mg/g (FW)	RS mg/g (FW)	TF mg/g (DW)	TTA mg/g (FW)	SPr mg/g (FW)	TPh mg/g (FW)	SOD U/g (FW)/min	GPX U/g (FW)/min	TBARS μ mol/g (FW)	DPPH (%)	SA (%)
v1	66.674±3.752$_{e}$	32.841±1.835i	39.485±1.562$_{b}$	15.5166±1.3364$_{d}$	3.470±0.135$_{a}$	2.431±0.054$_{d}$	40.0043±1.2896$_{h}$	62.0238±3.5089j	0.4663±0.0233$_{a}$	93.9332±2.9428$_{a}$	76.9021±5.9730$_{ab}$
v2	108.850±8.365$_{a}$	95.090±0.369$_{a}$	27.773±1.274$_{fg}$	9.6905±0.1922$_{fg}$	1.850±0.070$_{f}$	2.902±0.007$_{b}$	120.129±8.9406$_{e}$	161.8998±2.1628$_{e}$	0.0240±0.0012$_{e}$	68.0597±1.5056$_{d}$	43.7202±3.3958$_{f}$
v3	98.622±0.597$_{bc}$	84.353±0.729$_{b}$	63.279±0.535$_{a}$	10.8333±0.8036$_{e}$	3.273±0.032$_{b}$	2.749±0.017$_{bc}$	102.77±10.8121$_{f}$	85.1755±2.8866i	0.0226±0.0012$_{e}$	63.0239±1.9744$_{e}$	73.2932±5.6927$_{bc}$
v4	105.13±5.340$_{ab}$	66.649±1.389$_{d}$	36.855±1.740$_{c}$	11.4268±0.5326$_{e}$	1.900±0.070$_{ef}$	2.572±0.017$_{cd}$	41.5189±2.1059$_{h}$	97.6904±1.6192$_{h}$	0.0577±0.0029$_{b}$	51.9366±2.5669$_{f}$	83.1652±6.4595$_{a}$
v5	65.924±2.013$_{e}$	46.899±1.438$_{g}$	26.252±0.740$_{g}$	21.2505±0.4992$_{bc}$	1.150±0.101$_{h}$	2.565±0.016$_{cd}$	84.7543±1.4738$_{g}$	111.7384±2.0095$_{f}$	0.0245±0.0012$_{e}$	69.4929±1.8638$_{d}$	73.7941±5.7316$_{abc}$
v6	40.573±1.484$_{f}$	26.455±2.494j	31.800±0.699$_{d}$	8.8768±0.3338$_{g}$	2.207±0.055$_{d}$	1.849±0.451$_{e}$	155.326±1.7262$_{d}$	279.7306±2.5099$_{b}$	0.0316±0.0016$_{de}$	92.9329±2.9114$_{a}$	78.6981±6.1125$_{ab}$
v7	75.178±0.885$_{d}$	36.486±0.912$_{h}$	30.567±0.370$_{de}$	8.7334±0.1714$_{g}$	1.180±0.165$_{h}$	2.370±0.044$_{d}$	188.839±7.0074$_{c}$	203.0260±3.4238$_{d}$	0.0464±0.0023$_{bc}$	85.0873±1.7258$_{b}$	55.2208±4.2890$_{e}$
v8	65.175±2.525$_{e}$	51.929±1.546$_{f}$	29.129±1.233$_{ef}$	21.7514±0.3871$_{b}$	0.2057±0.0104$_{de}$	2.473±0.043$_{cd}$	125.064±7.2309$_{e}$	100.9820±1.5992$_{h}$	0.0225±0.0011$_{e}$	67.1970±1.7919$_{d}$	58.3772±4.5342$_{de}$
v9	97.762±3.788$_{c}$	77.894±2.318$_{c}$	16.307±0.329i	10.6596±0.2115$_{ef}$	2.440±0.085$_{c}$	2.584±0.034$_{cd}$	99.2377±2.1318$_{f}$	106.1515±1.9090$_{g}$	0.0196±0.0010$_{e}$	53.7264±1.3699$_{f}$	33.4142±2.5953$_{g}$
v10	78.210±3.030$_{d}$	62.315±1.854$_{e}$	23.581±0.781$_{h}$	20.3501±0.4037$_{c}$	1.573±0.055$_{g}$	2.392±0.098$_{d}$	215.656±5.6372$_{b}$	245.8244±4.4209$_{c}$	0.0382±0.0019$_{cd}$	79.2567±1.5431$_{c}$	66.7575±5.1851$_{cd}$
v11	45.622±1.768$_{f}$	32.351±1.082i	39.677±2.572$_{b}$	8.7215±0.1730$_{f}$	2.648±0.061$_{c}$	3.685±0.216$_{a}$	342.6612±6.3095$_{a}$	357.5628±6.4304$_{a}$	0.0424±0.0029$_{bc}$	90.0392±2.8208$_{ab}$	75.2043±5.8412$_{ab}$
v12	44.398±0.920$_{f}$	20.472±1.345k	24.207±1.349$_{h}$	24.5099±1.8114$_{a}$	2.841±0.252$_{bc}$	2.341±0.077$_{d}$	101.8998±4.8944$_{f}$	125.0646±5.9322$_{ef}$	0.0183±0.0038$_{e}$	81.8854±2.0066$_{c}$	38.0097±4.3428$_{fg}$

Values in the table are illustrated as mean ± standard variation. The same letters indicating the values are not different significantly. Otherwise, different letters indicating the values are significantly different (P<0.05) Post hoc Duncan test. TS: Total sugar content; RS: reducing sugar content; TF: total flavonoid content; TTA: total titratable acids; SPr: soluble protein content; TPh: total phenolic content. SOD: activity of superoxide dismutase; GPX: activity of Guaiacol peroxidase; TBARS: concentration of thiobarbituric acid reacting substances; DPPH: percentages of DPPH radical scavenged at the concentration of 5 mg/ml; SA: percentages of superoxide anion radical scavenged at the concentration of 10 mg/ml.

Table 2. Berry results of detected biochemical traits of genotype different (GD) group of grapevine

Cultivar	TS mg/g(FW)	RS mg/g(FW)	TF mg/g(DW)	TTA mg/g(FW)	SPr mg/g(FW)	TPh mg/g(FW)	SOD U/g(FW).min	GPX U/g(FW)·min	TBARS μ mol/g(FW)	DPPH %	SA %
v1	76.723±2.016$_{g}$	61.976±1.755$_{e}$	12.937±1.216$_{a}$	19.3149±0.7255$_{e}$	4.159±0.459$_{b}$	0.902±0.036$_{cd}$	35.2284±3.6363$_{g}$	92.6452±3.1893j	0.6407±0.0641$_{a}$	94.0354±1.5413$_{a}$	88.2055±4.2270$_{b}$
v2	177.849±5.282$_{a}$	162.70±6.753$_{a}$	3.859±0.129$_{fg}$	15.0122±0.5639$_{f}$	0.918±0.053$_{f}$	0.919±0.0177$_{bc}$	87.1901±9.5897$_{a}$	253.8754±2.4133$_{e}$	0.1509±0.0151$_{d}$	35.3508±0.5794$_{h}$	22.7818±1.7908$_{e}$
v3	120.32±6.605$_{bc}$	104.067±5.713$_{bc}$	10.882±0.297$_{b}$	25.1913±2.0731.	0.778±0.084$_{f}$	0.8.84±0.013$_{cde}$	53.088±1.3616$_{de}$	101.3510±2.2645$_{i}$	0.1404±0.0141$_{de}$	82.2641±1.3484$_{c}$	82.5355±3.9420$_{bc}$
v4	123.31±2.907$_{bc}$	98.528±5.408$_{bc}$	7.362±0.576$_{c}$	28.6425±1.0759$_{c}$	4.626±0.143$_{a}$	0.977±0.013$_{ab}$	23.4415±2.5245$_{h}$	104.1430±2.2610$_{i}$	0.3161±0.0316$_{b}$	88.3319±1.4478$_{b}$	94.5748±4.5522$_{a}$
v5	97.133±5.332$_{e}$	74.321±4.080$_{d}$	4.773±0.405$_{e}$	41.6238±1.5634$_{a}$	4.863±0.084$_{a}$	0.999±0.005$_{a}$	57.991±2.3602$_{cd}$	170.8425±2.2672$_{f}$	0.0152±0.0016$_{f}$	66.9622±1.0976$_{d}$	85.0449±4.0676$_{b}$
v6	48.718±2.674$_{i}$	34.643±1.902$_{f}$	8.087±0.218$_{c}$	21.4225±1.9315$_{e}$	3.090±0.113$_{c}$	0.679±0.097$_{h}$	49.028±4.5257$_{ef}$	336.9288±4.8217$_{c}$	0.2214±0.0222$_{c}$	90.2109±1.4786$_{b}$	88.0319±4.2183$_{b}$
v7	114.985±6.312$_{c}$	97.330±5.343$_{c}$	4.279±0.251$_{ef}$	11.8331±1.5713$_{g}$	2.980±0.128$_{c}$	0.772±0.013$_{fg}$	80.426±0.3265$_{ab}$	296.3325±2.5611$_{d}$	0.0281±0.0028$_{f}$	64.9301±1.0643$_{e}$	64.1567±3.0615$_{d}$
v8	128.341±7.045$_{b}$	106.424±5.842$_{b}$	11.225±0.126$_{b}$	31.7831±1.1938$_{b}$	1.301±0.210$_{e}$	0.825±0.022$_{ef}$	61.8468±1.7291$_{c}$	137.2536±3.1980$_{h}$	0.1031±0.0104$_{e}$	44.4341±0.7283$_{g}$	69.3667±3.3025$_{d}$
v9	106.709±3.169$_{d}$	97.621±4.051$_{c}$	3.101±0.535$_{g}$	16.5134±0.6203$_{f}$	2.101±0.102$_{d}$	0.727±0.016$_{gh}$	43.6158±2.5919$_{f}$	162.3003±2.1539$_{g}$	0.0028±0.0003$_{f}$	30.0247±0.4921$_{i}$	11.9519±1.8325$_{f}$
v10	83.589±2.482$_{f}$	76.470±3.174$_{d}$	5.951±0.165$_{d}$	31.5256±1.1842$_{b}$	4.995±0.380$_{a}$	0.845±0.033$_{de}$	77.6177±3.7469$_{b}$	375.8534±4.9878$_{b}$	0.1250±0.0125$_{de}$	52.3952±0.8588$_{f}$	77.8890±3.7123$_{c}$
v11	45.622±1.768$_{i}$	47.556±1.891$_{ef}$	11.123±1.224$_{b}$	13.5110±0.5074$_{g}$	3.270±0.184$_{c}$	1.064±0.061$_{a}$	93.6076±1.6535$_{a}$	546.6959±7.2550$_{a}$	0.2535±0.0293$_{c}$	84.7174±1.3886$_{c}$	86.4790±4.1398$_{b}$
v12	44.398±0.920$_{i}$	30.998±2.311$_{f}$	3.940±0.654$_{g}$	33.8944±1.0094$_{b}$	3.500±0.234$_{bc}$	0.570±0.45$_{i}$	42.0238±5.1122$_{f}$	188.8393±8.9200$_{f}$	0.1111±0.0087$_{e}$	63.4611±1.9452$_{e}$	36.9856±3.9873$_{e}$

Values in the table are illustrated as mean ± standard variation. The same letters indicating the values are not different significantly. Otherwise, different letters indicating the values are significantly different (P<0.05) Post hoc Duncan test. TS: Total sugar content; RS: reducing sugar content; TF: total flavonoid content; TTA: total titratable acids; SPr: soluble protein content; TPh: total phenolic content. SOD: activity of superoxide dismutase; GPX: activity of Guaiacol peroxidase; TBARS: concentration of thiobarbituric acid reacting substances; DPPH: percentages of DPPH radical scavenged at the concentration of 5 mg/ml; SA: percentages of superoxide anion radical scavenged at the concentration of 10 mg/ml.

Table 3. Leaf results of detected biochemical traits of treatment different (TD) group of grapevine

T	GPX U/g (FW)/min	SOD (U/g (FW)/min)	RS (mg/g (FW))	SPr (mg/g (FW))	TF (mg/g (DW))	TPh (mg/g (FW))	PAL (U/(g (FW)/min))	DPPH (%)	SA (%)
1	42.67±11.39^{g}	367.44±44.85^{de}	66.3±01.8^{b}	1.03±0.02^{cd}	28.4±1.13^{a}	2.17±0.06^{i}	243.06±2.86^{c}	89.13±0.24^{ab}	77.83±4.98^{a}
2	93.33±15.72^{e}	271.32±82.72^{f}	54.7±0.9^{de}	1.17±0.02^{bc}	26.4±1.31^{ab}	2.08±0.12^{i}	219.22±21.33^{de}	69.81±4.68^{ef}	70.20±1.53^{abc}
3	213.33±41.63^{cd}	494.57±29.90^{bc}	78.2±3.7^{a}	1.02±0.02^{cd}	26.9±0.64^{ab}	4.13±0.42^{b}	231.89±10.34^{de}	87.48±00.31^{ab}	76.91±0.82^{ab}
4	180±20^{cd}	514.73±11.71^{bc}	60.9±3.1^{c}	1.02±0.02^{cd}	26.3±0.39^{ab}	3.65±0.06^{de}	212.44±14.94^{e}	59.35±2.53^{h}	68.83±0.76^{b}
5	262±49.52^{bc}	368.99±25.62^{de}	61.5±3.0^{c}	1.38±0.09^{a}	26.0±0.48^{ab}	4.49±0.23^{a}	240±13.86^{c}	73.47±9.03^{de}	67.70±3.97^{bc}
6	52±3.46^{fg}	331.78±60.52^{e}	50.3±1.9^{g}	1.15±0.09^{bc}	20.6±0.38^{abc}	3.89±0.34^{d}	184.56±21.45^{hi}	29.62±1.16^{j}	63.38±1.26^{cd}
7	410.67±227.04^{a}	368.99±75.62^{de}	62.7±1.5^{c}	0.74±0.11^{e}	26.7±0.41^{ab}	4.43±0.23^{ab}	168.44±7.90^{ij}	42.45±2.02^{i}	62.10±3.23^{cd}
8	76.67±25.17^{ef}	243.41±32.67^{g}	51.0±0.3^{f}	1.06±0.10^{c}	24.6±0.44^{ab}	3.58±0.13^{e}	228.22±5.87^{d}	43.95±5.34^{i}	60.11±2.16^{d}
9	222±37.04^{cd}	527.13±20.97^{b}	57.1±3.3^{d}	1.23±0.07^{b}	20.9±0.17^{abc}	4.59±0.32^{a}	218.89±16.98^{de}	81.71±3.52^{bc}	56.30±2.25^{de}
10	68±8^{f}	578.29±18.80^{ab}	55.6±3.3^{de}	0.84±0.07^{de}	22.7±0.68^{abc}	3.79±0.06^{d}	197±14.38^{ef}	76.04±1.85^{d}	54.22±5.58^{e}
11	282.67±8.33^{bc}	590.70±20.27^{ab}	54.0±2.0^{de}	1.09±0.02^{c}	19.4±1.34^{b}	4.70±0.21^{a}	242±5.81^{bc}	64.50±0.50^{g}	50.29±1.46^{f}
12	304±141.32^{b}	601.55±11.71^{a}	46.5±1.2^{g}	0.84±0.04^{de}	27.7±0.82^{a}	4.08±0.01^{bc}	144.56±2.22^{g}	73.06±1.93^{e}	51.60±2.08^{ef}
13	303.33±49.89^{b}	488.37±58.28^{bc}	52.0±0.6^{f}	1.05±0.05^{c}	28.3±0.85^{a}	4.53±0.01^{a}	260.11±7.03^{b}	80.01±3.10^{c}	66.82±2.75^{bc}
14	240.67±133.17^{cd}	443.41±33.65^{c}	46.6±1.1^{g}	0.49±0.04^{g}	14.4±2.88^{c}	2.53±0.14^{h}	187.11±4.53^{f}	62.29±3.89^{gh}	51.82±3.11^{ef}
15	194.67±46.54^{cd}	393.80±42.20^{d}	50.4±1.9^{fg}	0.94±0.03^{d}	27.2±0.68^{ab}	2.57±0.04^{h}	300.22±15.41^{a}	82.43±0.99^{b}	65.53±1.62^{c}
16	276.33±21.01^{bc}	435.66±28.04^{cd}	45.0±1.0^{g}	0.62±0.08^{f}	24.0±1.83^{abc}	3.56±0.11^{e}	187.33±5.04^{f}	79.34±5.15^{cd}	60.07±2.37^{d}
17	154±22^{d}	404.65±9.30^{d}	41.3±0.9^{h}	1.09±0.08^{c}	14.2±0.96^{c}	2.95±0.04^{g}	210.78±8.70^{e}	79.9±2.13^{cd}	46.75±2.05^{f}
18	253.33±53.97^{c}	355.04±39.01^{de}	36.6±0.3^{i}	0.85±0.10^{de}	15.0±1.00^{c}	3.29±0.07^{e}	187.33±4.18^{f}	90.2±2.48^{a}	49.88±1.87^{f}

Values in the table are illustrated as mean ± standard variation. The same letters indicating the values are not different significantly. Otherwise, different letters indicating the values are significantly different (P<0.05) Post hoc Duncan test. T: Numbers of treatments; GPX: activity of Guaiacol peroxidase; SOD: activity of superoxide dismutase; RS: reducing sugar content; SPr: soluble protein content; TF: total flavonoid content; TPh: total phenolic content. PAL: activity of phenylalanine ammonia lyase; DPPH: percentages of DPPH radical scavenged, at the concentration of 15 ug/ml; SA: percentages of superoxide anion radical scavenged at the concentration of 10 mg/ml.

Table 4. Berry results of detected biochemical traits of treatment different (TD) group of grapevine

T	GPX U/g (FW)/min	SOD (U/g (FW)/min)	RS mg/g (FW)	SPr (mg/g (FW))	TF (mg/g (DW))	TPh (mg/g (FW))	PAL (U/(g (FW)/min))	DPPH (%)	SA (%)
1	21.33±2.31^{h}	88.37±25.90^{e}	116.6±0.9^{b}	0.38±0.13^{c}	3.17±0.6^{b}	0.44±0.01^{cd}	53.44±7.94^{ab}	25.66 ±1.11^{bc}	44.52±0.43^{ab}
2	40±20.25^{fgh}	165.89±51.86^{cd}	85.3±0.2^{f}	0.43±0.03^{ab}	3.08±0.04^{b}	0.22±0.02^{f}	41.72±2.70^{cd}	23.34±1.63^{c}	41.13±1.54^{b}
3	134±6.61^{b}	106.20±90.68^{cde}	134.5±0.4^{a}	0.53±0.05^{ab}	3.68±0.07^{a}	0.41±0.02^{cd}	42.83±4.37^{c}	36.01±2.49^{a}	47.90±0.82^{a}
4	97.47±4.40^{cd}	130.23±66.23^{cde}	97.2±0.5^{d}	0.45±0.09^{ab}	3.05±0.04^{bc}	0.50±0.06^{b}	41.67±0.93^{cd}	27.92±6.05^{b}	45.11±0.59^{ab}
5	175±12.95^{a}	227.13±34.91^{bc}	103.5±1.4^{c}	0.54±0.06^{a}	3.62±0.06^{ab}	0.42±0.05^{c}	44.17±4.65^{bcd}	15.97 ±4.41^{f}	27.18±3.95^{e}
6	20.97±1.89^{h}	133.33±12.8^{d}	100.0±1.6^{cd}	0.43±0.07^{b}	3.02±0.05^{bc}	0.31±0.01^{e}	30.61±1.29^{fg}	21.79±2.3^{cd}	33.83±1.26^{d}
7	25.67±5.13^{h}	89.92±20.05^{de}	134.0±2.2^{a}	0.46±0.04^{ab}	1.76±0.04^{e}	0.42±0.03^{c}	45±1.67^{bc}	14.94±2.38^{fg}	31.13±3.23^{de}

Table 4. Contd.

8	48±18.52^{ef}	208.53±11.47^{c}	99.4±1.2^{d}	0.35±0.07^{cd}	1.36±0.06^{ef}	0.65±0.08^{a}	48.89±2.55^{b}	20.45±2.48^{d}	30.10±2.16^{de}
9	42±4.36^{f}	334.52±13.87^{a}	121.6±3.2^{b}	0.40±0.08^{b}	1.79±0.04^{e}	0.37±0.05^{cd}	45.22±5.32^{bcd}	15.77±2.15^{f}	33.09±2.25^{d}
10	60±3^{e}	302.17±13.67^{ab}	114.2±1.2^{bc}	0.51±0.02^{ab}	2.83±0.06^{c}	0.24±0.04^{f}	40.89±2.18^{cd}	18.65±0.88$^{e\ efgh}$	21.14±5.58^{f}
11	105.33±14.74^{c}	275.35±14.27^{ab}	94.4±1.8^{e}	0.38±0.06^{bc}	2.98±0.02^{c}	0.38±0.06^{cd}	49.06±1.58^{bc}	25.04±1.57^{bc}	38.30±1.46^{c}
12	20.32±3.39^{h}	188.56±50.56^{cd}	97.9±0.5^{d}	0.32±0.07^{d}	2.04±0.06^{e}	0.64±0.02^{a}	36.5±2.85^{de}	21.17±0.31^{cd}	37.11±2.08^{c}
13	99.33±0.95^{cd}	259.57±23.71^{bcd}	91.6±0.6^{f}	0.37±0.03^{c}	2.50±0.04^{d}	0.64±0.03^{a}	40.67±2.25^{cd}	17.10±1.39^{ef}	28.80±2.75^{de}
14	89.57±1.30^{d}	314.01±17.76^{ab}	96.1±0.6^{e}	0.29±0.11^{e}	1.65±0.04^{e}	0.39±0.00^{cd}	31.39±13.14^{f}	5.51±2.72^{i}	20.72±3.11^{f}
15	29.07±2.10^{g}	318.74±34.33^{ab}	111..5±0.4^{bc}	0.46±0.05^{ab}	1.43±0.05^{ef}	0.34±0.00^{d}	57.39±1.77^{a}	10.46±4.51^{j}	27.51±1.62^{e}
16	18.88±11.34^{h}	245.36±20.68^{bc}	110.5±0.7^{bc}	0.30±0.04^{e}	1.46±0.02^{ef}	0.41±0.02^{cd}	37.83±1.09^{d}	12.88±0.64^{h}	27.92±2.37^{e}
17	51.73±2.69^{ef}	271.40±7.61 ^{b}	98.7±1.2^{d}	0.53±0.05^{ab}	1.05±0.06^{f}	0.50±0.02^{b}	34.11±1.18^{e}	8.96±0.27k	29.41±2.03^{de}
18	42.9±1.97^{f}	265.88±14.27^{b}	84.2±0.5^{f}	0.38±0.02^{bc}	1.12±0.03^{f}	0.40±0.01^{cd}	25.89±1.35^{g}	18.19±1.31^{e}	27.93±1.86^{e}

Values in the table are illustrated as mean ± standard variation. The same letters indicating the values are not different significantly. Otherwise, different letters indicating the values are significantly different (P<0.05) Post hoc Duncan test. T: Numbers of treatments; GPX: activity of Guaiacol peroxidase; SOD: activity of superoxide dismutase; RS: reducing sugar content; SPr: soluble protein content; TF: total flavonoid content; TPh: total phenolic content. PAL: activity of phenylalanine ammonia lyase; DPPH: percentages of DPPH radical scavenged, at the concentration of 15 ug/ml; SA: percentages of superoxide anion radical scavenged at the concentration of 10 mg/ml.

especially on some primary metabolites, such as sugar and protein contents (Figure 1). Except few parameters, variations of different parameters between leaves and berries in GD group are very similar, however, greater variations of the detected parameters were found in berries than in leaves of the TD group (Figure 1). These variations promted us to assess the correlations of these traits between leaf and berry.

Inter-parameters' correlation within leaf or berry for both GD and TD groups were detected and the result of correlation coefficients were listed and are shown in Tables 5 and 6, respectively. Only smaller proportion (<11%) of inter-parameter pairs tested were with significant correlation within leaf or berry samples in both groups. In GD group, parameter pairs TS/RS, GPX/SOD and TF/SA showed significant correlations both in leaf and in berry. Significant correlation of inter-parameter pairs in TD group were also found, but only one inter-parameter pairs TF/SA showed significant correlation both in leaf and in berry in this group (Table 6). Correlation of different parameters within leaf (berry) implies the possible correlations of these parameters in metabolisms or functions. Interestingly enough, in coefficients of variation, there was also significant correlations between leaves and berries in both GD group (r=0.918; P<0.001) and TD group (r=0.870; P<0.05). Lower proportion of correlation as well as lower correlation coefficients of inter-parameter pairs within leaf (berry) samples ensured the effectiveness of following quantitative correlation analysis between leaf and berry.

Correlations between leaf and berry for every coordinate traits were tested for both GD and TD groups and the correlation coefficients are shown in Tables 7 and 8, respectively. Out of the 11 detected biochemical parameters, 9 showed significant correlations between leaf and berry in GD group. Amongst, parameters of TS, RS, TTA, TBARS, GPX, SOD, and SA were correlation significant at P<0.01 level, with the correlation coefficients as high as 0.84, 0.87, 0.83, 0.80, 0.98, 0.87, and 0.97, respectively. In total flavonoids (TF) and total phenols (TPh), there was also significant correlation between leaf and berry in GD group at P<0.05 level, with the correlation coefficients of about 0.68 (Table 7). Therefore, data of these correlation significant traits from leaf of GD group may be used to estimate the values of coordinating berry traits. In TD group, values of parameters RS, SPr, SA, and PAL correlate significantly between leaves and berries at P<0.01 level, with the correlation coefficients as 0.67, 0.52, 0.58, and 0.69, respectively. TF showed significant correlation at P < 0.05 level between leaf and berry with a correlation coefficient of 0.51. Parameters of TPh, SOD, and GPX showed significant correlations in GD group, while in these parameters, there was no significant correlation in the TD group (Table 8). Amongst the parameters that detected simultaneously in both GD and TD groups, RS, TF, and SA showed significant cor-relation between leaves and berries. Therefore, significant correlations in values of some biochemical

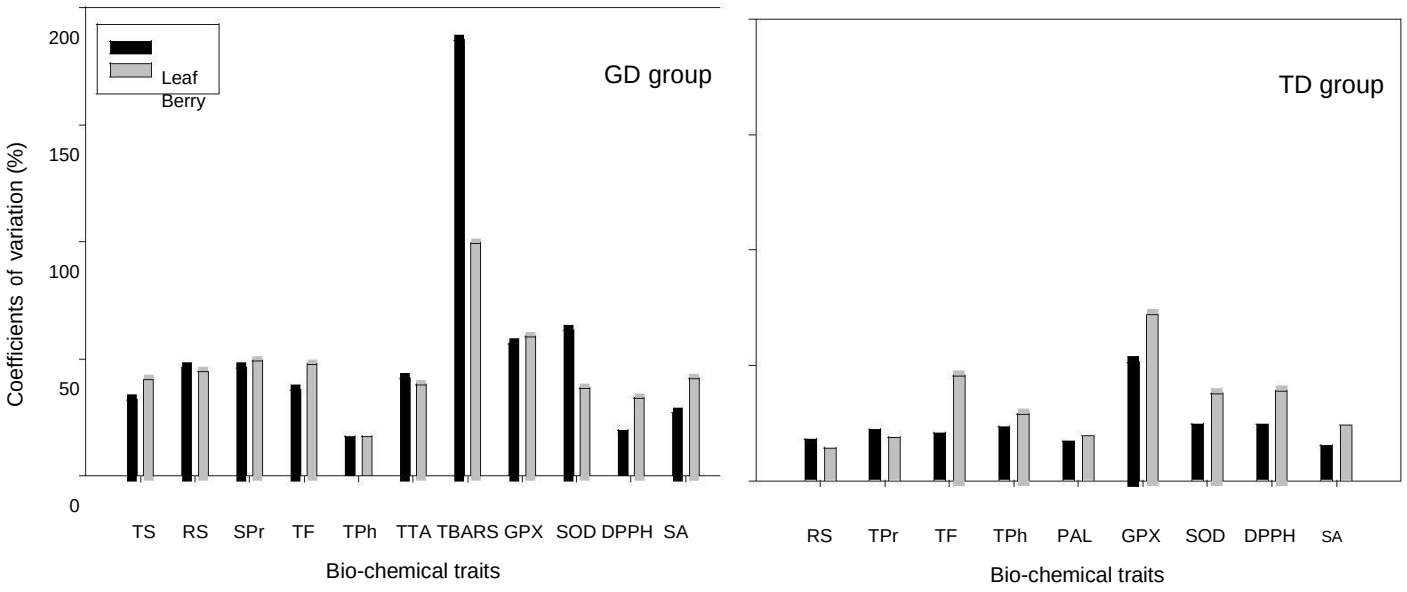

Figure 1. Comparison of coefficients of variation of detected biochemical traits within leaves and berries in both genotype different (GD) and treatment different (TD) groups of grapevine.

traits between leaves and berries of grapevines broadly existed. No doubt that the ranges of these correlated traits in berries could be primarily estimated by the detected leaf values.

Beside leaf/berry same-parameter pairs afore-mentioned, significant correlations of inter-parameter pairs between leaf and berry were also detected, such as TS.L&B/RS.L&B ("L" represents leaf and "B" represents berry), TF.B/TPr.L, TF.B/SA.L, GPX.L&B/SOD.L&B, TBARS.B/TPr.L, TF.L/DPPH.B, and SA.L/DPPH.B. Parameters RS.B and TS.B were foundto be both significantly negative correlated with DPPH.L in GD group (Table 7). Compared to GD group, more of such inter-parameter pairs between leaf and berry were found with significant correlation in the TD group. For some examples, leaf RS correlated significantly to berry TF (r=0.72), PAL (r=0.52), DPPH (r=0.65), SA (r=0.60); berry TF significantly correlated to leaf RS and SA (r=0.54); and berry PAL also significantly correlated to leaf RS, TF (r=0.62) and SA (r=0.48) at the same time (Table 8).

DISCUSSION

Correlative growth of different parts of plant has been well known, because of the continuously exchange of nutrients, metabolites, and signal molecules (Srivastava, 2002; Teale et al., 2006). Compositional correlations between different organs of plants had also evidences. Some special substances detected in certain species of plant can always be detected more or less from other parts or organs in this species of plants (Neto et al., 1992), and some of these compounds and existent patterns have been used as chemo-taxonomical parameters (Herl et al., 2008; Loreto, 2002; Figueiredo-González et al., 2012). However, the quantitative correlation of biochemical components between different parts of a plant has not been systematically studied. In some earlier studies, correlations of some nutrients, such as N, Ca, K, P, Mg, etc., between or within plant leaves and fruits has been reported (Dris et al., 1999). Correlations between metabolites in grape berries also had been studied which were focused more on correlations of inter-parameter within or between leaves and fruits, other than purposely designed to investigate the correlations of same-parameter pairs between different organs or parts of plant (Shiraishi et al., 2010). The later work has just tested the significance of correlations of the biochemical traits within grape berries. Plant cells from different parts or organs differentiated as cells with different phenotypes and functions, and will have different patterns of gene expression and the resultant metabolites. However, the high similarities of genetic background of these cells from different parts of one plant or same variety will share higher proportions of metabolic similarities compared to genetically varied cells as has proved again in this study (Figure 1). Furthermore, cells in leaves or berries of a single plant always under similar environmental conditions and may respond to these factors coordinately to produce similar defense metabolites, due to the continuously exchange of all kinds of transportable metabolites, including some defense compounds among different parts of tissues (Jørgensen et al., 2015).

Therefore, the existence of some metabolites with co-ordinating concentrations in leaves and fruits is expected. The obtained data have provided evidence and proved the existence of such kind of values' co-vibaration of some

Table 5. Correlation coefficients among parameters (inter-parameter pair)within berry (right-above) and leaf (left-below) of genotype different (GD) group of grapevine.

Parameter	TS	RS	TF	TTA	TPr	TPh	SOD	GPX	TBARS	DPPH	SA
TS	-	0.985**	-0.209	-0.118	-0.509	0.279	0.141	-0.418	-0.226	-0.509	-0.327
RS	0.916**	-	-0.211	-0.221	-0.551	0.306	0.253	-0.302	-0.234	-0.560	-0.387
TF	0.127	0.129	-	-0.062	-0.087	0.384	-0.100	-0.042	0.673*	0.600*	0.662*
TTA	-0.314	-0.269	-0.336	-	0.397	-0.036	-0.361	-0.372	-0.200	0.005	0.260
TPr	-0.125	-0.087	0.518	-0.062	-	0.159	-0.241	0.152	0.268	0.400	0.476
TPh	0.144	0.242	0.274	-0.273	0.163	-	0.284	0.144	0.282	0.249	0.472
SOD	-0.410	-0.243	-0.013	-0.251	-0.159	0.509	-	0.746 **	-0.349	-0.329	-0.129
GPX	-0.478	-0.333	-0.094	-0.350	-0.228	0.285	0.909**	-	-0.131	0.044	0.096
TBARS	-0.091	-0.270	0.201	0.022	0.493	-0.092	-0.331	-0.307	-	0.649*	0.465
DPPH	-0.773**	-0.764**	0.043	0.003	0.172	-0.103	0.455	0.548	0.422	-	0.818**
SA	-0.170	-0.215	0.578*	-0.163	0.048	0.030	0.056	0.180	0.309	0.236	-

*Correlation is significant at the 0.05 level (2-tailed); **Correlation is significant at the 0.01 level (2-tailed). ; GPX: activity of Guaiacol peroxidase; SOD: activity of superoxide dismutase; RS: reducing sugar content; TS: total sugar; TPr: total protein content; TTA: titratable acidity TF: total flavonoid content; TPh: total phenolic content. DPPH: percentages of DPPH radical scavenged, at the concentration of 15 ug/ml; SA: percentages of superoxide anion radical scavenged at the concentration of 10 mg/ml.

Table 6. Correlation coefficients among traits in berry (right-above) and leaf (left-below) of treatment different (TD) group of grapevine.

Parameter	GPX	SOD	RS	SPr	TF	TPh	PAL	DPPH	SA
GPX	-	0.064	-0.034	0.398	0.639**	0.079	0.048	0.233	0.065
SOD	0.35	-	-0.29	-0.168	-0.27	-0.113	-0.087	-0.645**	-0.704**
RS	-0.061	0.111	-	0.35	0.178	-0.145	0.422	0.157	0.163
SPr	-0.32	-0.172	0.313	-	0.329	-0.325	0.17	0.21	0.13
TF	0.036	0.002	0.601**	0.213	-	-0.078	0.422	0.570*	0.479*
TPh	0.530*	0.484*	0.219	0.24	0.121	-	0.013	0.017	0.075
PAL	-0.202	-0.13	0.28	0.462	0.286	-0.134	-	0.196	0.278
DPPH	0.079	0.303	0.019	0.006	0.027	-0.2	0.335	-	0.844**
SA	-0.248	-0.294	0.759**	0.322	0.751**	-0.153	0.445	0.066	-

*Correlation is significant at the 0.05 level; **Correlation is significant at the 0.01 level (2-tailed). GPX: activity of Guaiacol peroxidase; SOD: activity of superoxide dismutase; RS: reducing sugar content; SPr: soluble protein content; TF: total flavonoid content; TPh: total phenolic content. PAL: activity of phenylalanine ammonia lyase; DPPH: percentages of DPPH radical scavenged, at the concentration of 15 ug/ml; SA: percentages of superoxide anion radical scavenged at the concentration of 10 mg/ml.

parameters between leaf and berry of grapevine as indicated in Tables 7 and 8, implies the possibility to predict some berry quality-related parameter values by using values from leaves or other parts of vines. In addition, the increase of sample numbers will Increase the significance of correlation coordinately, since randomly take off of any sample datum will decrease the significance and coefficient of correlations.

Table 7. Correlation coefficients of detected physio-chemical traits between berry and leaf of the genotype different (GD) group of grapevine.

Parameter	TS.B	RS.B	TF.B	TTA.B	TPr.B	TPh.B	SOD.B	GPX.B	TBARS.B	DPPH.B	SA.B
TS.L	0.84**	0.85**	-0.25	-0.16	-0.31	0.29	-0.09	-0.45	-0.11	-0.37	-0.31
RS.L	0.82**	0.86**	-0.21	-0.10	-0.49	0.29	0.09	-0.30	-0.25	-0.50	-0.37
TF.L	0.04	0.04	0.68*	-0.12	-0.28	0.37	-0.07	-0.15	0.38	0.64*	0.55
TTA.L	-0.20	-0.27	-0.06	0.83**	0.33	-0.23	-0.20	-0.29	-0.13	-0.21	-0.00
TPr.L	-0.32	-0.29	0.58*	-0.19	-0.28	-0.12	-0.37	-0.24	0.58*	0.37	0.02
TPh.L	0.10	0.21	0.22	-0.32	-0.19	0.69*	0.51	0.40	0.04	-0.03	-0.03
SOD.L	-0.36	-0.24	0.09	-0.37	-0.00	0.18	0.80**	0.95**	-0.21	-0.00	0.11
GPX.L	-0.45	-0.35	0.00	-0.37	0.15	0.09	0.66*	0.98**	-0.10	0.15	0.18
TBARS.L	-0.17	-0.18	0.52	-0.19	0.28	0.16	-0.33	-0.29	0.87**	0.44	0.29
DPPH.L	-0.65*	-0.62*	0.31	-0.25	0.27	-0.15	0.31	0.54	0.40	0.45	0.32
SA.L	-0.24	-0.29	0.65*	0.169	0.43	0.57	-0.15	0.08	0.53	0.82**	0.97**

*Correlation is significant at the 0.05 level; **Correlation is significant at the 0.01 level (2-tailed). Parameters with ".B" indicate the berry parameters, and ".L" indicate the leafy parameters.

Table 8. Correlation coefficients of detected physio-chemical traits between berry and leaf of the treatment different (TD) group of grapevine.

Parameter	GPX.B	SOD.B	RS.B	SPr.B	TF.B	TPh.B	PAL.B	DPPH.B	SA.B
GPX.L	0.21	0.114	0.141	-0.173	0.026	0.291	-0.097	-0.244	-0.159
SOD.L	0.228	0.286	0.158	-0.041	0.311	0.085	-0.011	0.145	0.067
RS.L	0.421	0.573	0.673**	0.455	0.723**	-0.12	.535*	0.647**	0.598**
SPr.L	0.343	-0.117	-0.096	0.517*	0.305	-0.019	0.301	0.372	0.373
TF.L	0.026	-0.515*	0.345	0.105	0.510*	0.209	0.615**	0.452	0.443
TPh.L	0.388	0.034	0.253	0.132	0.123	0.274	-0.015	0.198	-0.019
PAL.L	0.339	0.218	0.023	0.279	0.319	-0.043	0.688**	0.07	0.103
DPPH.L	0.136	0.329	0.056	0.086	0.36	-0.034	0.101	0.009	0.059
SA.L	0.2	-0.642**	0.355	0.263	0.536*	-0.073	0.479*	0.549*	0.579*

*Correlation is significant at the 0.05 level; **Correlation is significant at the 0.01 level (2-tailed). Parameters with ".B" indicate the berry parameters, and ".L" indicate the leafy parameters.

According to the aforementioned analysis, it is impossible and unreasonable to expect all metabolites having quantitatively coordinated concentrations in different parts of plants, because of the positional and functional differences of cells.

However, it will be of great significance if some parameters or components of interest such as sugar, organic acids, flavonoids, phenols, etc., to show this kind of correlations. On the base of these correlations, it will be allowed to develop a berry-independent method for berry pre-evaluation. The values of several parameters for 2 groups in both leaf and berry samples of grapevine were measured. GD group were dif-ferent varieties but shared the similar environmental

conditions, and TD group were same variety, but treated with different factors. Nine of 11 and 5 of 9 of the detected parameters in GD and TD group, respectively, showed significant correlation in values' variation between leaf and berry. Parameters of TS, TF and SA in leaves are significantly correlated to berries, both in GD and TD groups. Therefore, values of parameters TS, RS, TF, TTA TPh, GPX, SOD, and SA of leaf can be used to estimate the values in berry for the genotype differed materials but grow in similar environmental condition. Amongst sugar, acidity, and antioxidants such as total flavonoids and phenols are always important parameters for berry quality evaluation. Leaf values of parameters RS, TF, SPr, SA, and PAL can be used to estimate the corresponding values of berries for the same genotype materials but treated differently. Although, all these mentioned parameters have significant correlation in one or both groups, but correlation coefficients in GD group were obviously higher than in the TD group as indicated in Tables 7 and 8; which implies this leaf-dependent berry quality evaluation will be more reliable for those genotype varied candidates.

Theoretically, parameters which have significant correlation in values between leaf and berry, leafy values can potentially be used to estimate the ranges of the corresponding traits of berry. But how to make the estimation more accurate should have some strategies, both in experiment designing and choices of indicator leaves. Genetic differed grapevines growing at similar environment can be evaluated by just comparing the leaf values of certain parameter amongst the candidate materials, because of the higher variation coefficients among different genotypes and higher correlation coefficients of certain traits between leaf and berry (Figure 1, Tables 1 to 4, 7 and 8). More traits including some special groups of metabolites, such as organic acids, free amino acids, flavonoids, tannins, stilbenes or even anthocyanins, etc., that are not only closely related to the quality of grape but also vibrate coordinately in leaf and berry need to be developed. As for the choice of indicator leaves, almost the same physiological conditioned leaves should be chosen as indicator leaf as has been described in materials and experimental design. The fact that the existence of significant correlation of some inter-parameter pairs between leaf and berry, implies that values of some leafy parameters could also be used as indicators of some other parameters in berry, as already suggested in some similar studies (Dris et al., 1999). For examples, values of SA in leaf can be used to estimate the values of TF and DPPH in berries, and the leafy TF values can also be used to indicate the values of certain berry DPPH. As for TD group, beside the different treatments, grapevines of the same cultivar also grew under environ-mental conditions, the effects of different treatments on some leafy parameters could also be used to estimate the effects on corresponding traits of berries. While more parameter pairs including same- and inter-parameter pairs were found to be significantly correlated in TD group than in GD group, the relatively lower coefficients of correlation in TD group may limit the accuracy of estimation. However, as a purpose of primary estimation, it is enough for making a decision.

One might notice that this pre-evaluation method cannot evaluate the resources integrally for multiple agronomic characters. It could only be applied as one of an assistant method in early stages of screening from numerous candidates, especially for certain biochemical trait screening, for some examples, the selection of higher sugar content, special sugar/acid ratio, or higher flavonoids content materials, etc., whereas the final evaluation to resources should still be dependent upon the formal ways of evaluation, but at this time focus only on the mostly potential candidates.

Except for providing a leaf-dependent berry pre-evaluation method, the obtained results have also provided a basic relationship between results from *in vitro* and *in vivo* experiments. Nowadays, many elicitors have proved to be able to induce or modify certain kinds of secondary metabolite in grape suspension cells (Tassoni et al., 2012; Cetin et al., 2014; Cai et al., 2011; Chao et al., 2015), but if all these elicitors or factors can also cause similar responses in grapevine at plant level or fruits, is still lack the theoretic basis. In fact, some elicitors or factors did cause similar secondary metabolic responses both at plant and cell level of grapevine. For some instances, ABA can promote the synthesis of anthocyanins in certain line of grape cells (Gagné et al., 2011), in vitro cultured grape berries (Hiratsuka et al., 2001), and can also promote the coloration of fruits at plant level (Jiang and Joyce, 2003; Pirie and Mullins, 1976). The UV-B can induce the accumulation of resveratrol and other secondary metabolites responses where ever in suspension cells, *in vitro* tissues and *in vivo* of vine (Li et al., 2008; Zamboni et al., 2006). The present study has given a good explanation for these similar responses to same factor, but in different type of experimental materials. However, whether exist significant correlations in value vibration of these responding effects between *in vitro* and *in vivo* experiments needing further evidences.

Conflict of Interests

The authors have not declared any conflict of interest.

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