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Berry shrivel of grapes in Austria – Aspects of the physiological disorder with cultivar Zweigelt (*Vitis vinifera* L.)

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ABSTRACT

Berry shrivel (BS) is a physiological disorder of grapevine that affects berry development with decreases in quantity and quality of the crop. The causes of BS are unknown and no treatment exists for prevention or cure. BS seems to occur temporally and spatially at random in vineyards even among and within vines. The detailed description of symptoms and their development is an important step towards the understanding of the molecular background behind BS. Herein symptoms of BS are described with parameters aiming to clearly differentiate between healthy and BS grapes. Analyses were conducted on *Vitis vinifera* (L.) cv. Zweigelt in 2009/2010 at five Austrian vineyards. Berry diameter and berry deformability were severely reduced by BS, and this effect was observed on whole clusters. BS berries had significantly reduced contents of soluble solids and total anthocyanin concentration 67–73 days after anthesis (DAA), both representing key markers for the early detection of BS clusters. A systemic effect of BS on other no symptoms showing clusters of the same plant could not be observed. Additionally the concentrations of several amino acids in grape juice were affected by BS: thirteen were reduced, two were induced and four did not change significantly.

1. Introduction

Berry shrivel (BS) is a complex phenomenon with severe effects on grape quality and yield and therefore has major economic impact for the wine industry. Symptoms are turgor loss, stop of sugar accumulation, high acidity contents and disturbed color development through delayed anthocyanin synthesis (red varieties), while the rachis and pedicels do not show visible necrosis on their surfaces (Bondada and Keller, 2007; Knoll et al., 2010; Krasnow et al., 2009). Shriveling of grape berries during ripening can have manifold causes through physiological disorders, as, e.g. BS, bunch stem necrosis (BSN) or late-season dehydration. BSN is initially characterized by necrotic lesions, leading to full necrosis of the rachis tissue and thereby stopping the transport of assimilates towards ripening berries. The symptoms comprising shriveling berries with reduced sugar content are similar to BS symptoms, depending on the stage of ripening when necrotic lesions start to form on the rachis (Christensen and Boggero, 1985; Capps and Wolf, 2000). Late-season dehydration leads to shrinking berries at the end of the ripening stage resulting in weight loss of berries and higher sugar concentration through dehydration. These symptoms have been described on *Vitis vinifera* cv. Syrah (McCarthy, 1999; Rogiers et al., 2006) and recently cell death in berry mesocarp tissue (Fuentes et al., 2010) and backflow of water through the xylem to the parent vine (Tilbrook and Tyerman, 2009) were proposed as possible causes of late-season dehydration of grape berries.

In Austria grapes of the cultivar Zweigelt (*V. vinifera* L., Blaufränkisch × St. Laurent) are mainly damaged by BS and high yield losses have been reported (Redl, 2005). The incidence of BS in vineyards may be reduced by, e.g. yield reduction or an increase in leaf area (Redl, 2005), however no vineyard management treatment so far has been shown to eliminate BS.

The causes of BS are not known. Cell death pathways in the rachis are postulated to be involved in symptom development of BS in Cabernet Sauvignon (Hall et al., 2011) and disturbed or disrupted assimilate transport towards the fruit may explain the ceased sugar accumulation and the shriveling of berries in addition to water loss (Hall et al., 2011). The blocking of the phloem through the deposition of callose seems not to be a cause of the ceased sugar accumulation, since a differential transcriptional regulation of genes coding for callose synthases could not be confirmed (Griesser et al., 2012). Observation of reduced cell viability in shriveling berries (Krasnow et al., 2009) led to the assumption that the loss of cell membrane integrity is one factor leading to water loss and shrinking of berries as observed with BS.

The key to the identification of the causes of BS is to understand the biochemical and physiological basis of BS symptom development. A prerequisite of such analyses are temporally and spatially

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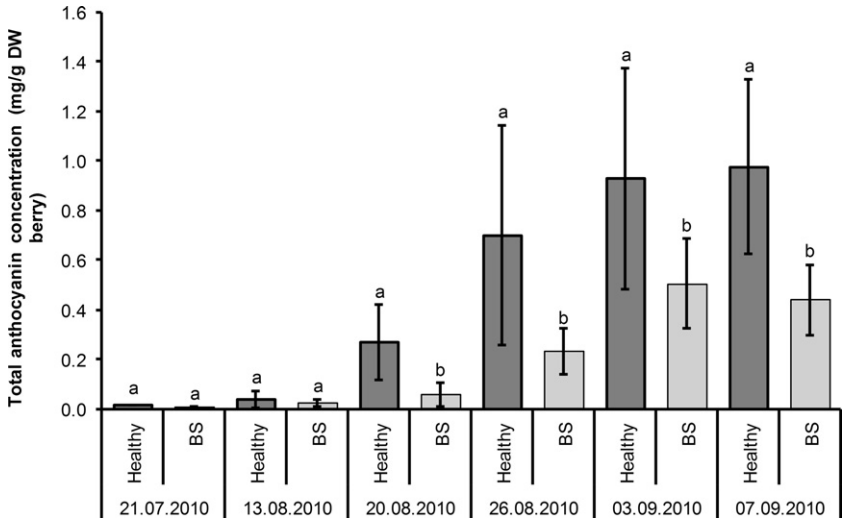

Fig. 1. Total anthocyanin concentration (mg/g DW berry) of healthy berries and berries collected from BS clusters was determined a six sampling dates during berry development. Ripening phase (veraison) started within the week of 13.08.2010 (55–60 DAA) and first BS symptoms were observed on 20.08.2010 (67 DAA). Anthocyanin concentrations are presented as mean values with standard deviations ($n = 5$ replicates per sampling date and cluster symptoms) Significant differences between healthy and BS clusters of each sampling date are indicated with different letters ($p < 0.05$).

defined samples of different target tissues during early BS development. The intention of this study is to collect phenological and phenotypical data for symptom description and differentiation of healthy and BS grapes with the focus on an early detection of BS grapes in vineyards.

2. Materials and methods

2.1. Vineyard locations and experimental setup

Experiments 2009 were conducted in three (V1–V3) vineyards (Göttlesbrunn, AT, LAT: 48.0594917, LON: 16.7382982; row direction: V1 and V2 NE-SW; V3: N-S) and experiments 2010 in two vineyards (Krems, AT, LAT: 48.401571, LON: 15.692725; row direction N-S). All vineyards in Göttlesbrunn were *V. vinifera* cv. Zweigelt grafted on Kober 5BB (*V. berlandieri* × *V. riparia*) between 7 and 10 years old and trained as a vertical shoot positioned (VSP) trellising system with a uni-lateral cane (6–8 nodes per vine; "Flachbogen"). In Krems Zweigelt scions were grafted on Teleki 5C (*V. berlandieri* × *V. riparia*) and SO_4 (*V. berlandieri* × *V. riparia*) rootstocks trained as VSP with a uni-lateral cane (7–9 nodes per vine; "Flachbogen").

Berry parameters and nutrient contents in grape juice (K^+, Mg^{2+}, and Ca^{2+}) were determined in 2009 and 2010. Berry diameter and deformability were measured 2009 in all vineyards. Pedicel diameter and amino acids content in grape juice was determined 2010 as additional parameters. BS clusters were identified visually through dehydrated, flaccid berries and delayed coloring (anthocyanin synthesis). First BS symptoms appeared 67 days after anthesis (DAA) and 62 DAA in 2009 and 2010, respectively. Assessment of BS in vineyards was conducted 2009 by visual observation of all clusters of 50 vines located in four different rows of V1–V3 ($n = 200$ per vineyard). BS assessment in 2010 was conducted with 138 randomly selected vines throughout the vineyards.

Climatic conditions in Göttlesbrunn, AT are mean annual precipitation of 550 mm, mean annual temperature of 9.7 °C (mean daily temperature per month (mean 7:00 + mean 19:00 + mean max. temperature + mean min. temperature)/4) and growing degree days from 1700 to 1800 (Huglin-Index). Climatic conditions in Krems, AT are: mean annual precipitation of 515 mm, mean annual temperature of 9.4 °C and growing degree days from 1750 to 1800 (ZAMG, 2002; Soja, 2010).

2.2. Berry development: berry diameter, berry deformability and pedicel diameter

Berry diameter and berry deformability (measured as percentage of berry deformation) was determined non-destructively with a modified digital caliper rule. A force sensor (Honeywell FSG15N1A, Illinois, USA) was attached to one extended arm of the caliper rule and applied pressure was observed with a handheld visual indicator (Matthews et al., 2009). Batteries were changed at the beginning of each measurement date and accuracy of the instrument was evaluated with a spring balance after 100 measurements each. Berry diameter (R0) was measured without any pressure applied. To determine berry deformability the berry diameter was measured after applying a pressure of 150 g (R150) and was calculated as percentage (%) berry deformability (100 − (R0/R150) * 100). In total 50 healthy and 100 BS clusters were analyzed 85–90 DAA ($n = 150$ clusters) in vineyard V1–V3 in 2009.

Diameter of pedicels was determined with a digital USB microscope (DinoLite, Sotac Computer, Stuttgart, Germany). Measurements were conducted at 13th September 2010 (91 DAA) in Krems. The diameter of three pedicels per cluster was determined (23 healthy and 23 BS clusters; $n = 69$) at three different positions (Fig. 2D). The distance to the samples and the magnification of the microscope was fixed to obtain sharp pictures at the beginning of the measurement. Calibration measurement was conducted using a standardized calibrated ruler and a final resolution of 1 μm was obtained.

2.3. Berry ripening parameters, nutrients, amino acids and total anthocyanin content

Samples collected 85–90 DAA (2009: healthy, healthy on BS vines, BS clusters; $n = 18$ each; 2010: healthy, BS clusters; $n = 14$ each) were used to determine fruit parameters and nutrient contents in grape juice. The ripening parameters soluble solids, titratable acids and pH value, were determined with FT-IR scanner Typ WineScan FT 120 (FOSS GmbH, Rellingen, Germany). The method was calibrated with standard solutions and reference measurements according to recommendation of the manufacturer.