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Sugar accumulation in 'Zweigelt' grapes as affected by "Traubenwelke"

M. Knoll, D. Achleitner and H. Redl

University of Natural Resources and Applied Life Sciences, Vienna, DAPP, Institute of Plant Protection, Vienna, Austria

Summary

"Traubenwelke" (berry shrivel, grape wilting), a physiological disorder during berry ripening, is a severe threat to grape production in numerous wine-growing areas worldwide. Investigations in Austrian viticulture, focusing on the particularly sensitive grape cultivar 'Blauer Zweigelt' (*Vitis vinifera* L.), showed that afflicted clusters exhibited insufficient colouring, loss of turgor, low sugar content and high acidity. The aim of this study was to determine the chronological disease development on the basis of the process of sugar accumulation in both unhealthy and healthy clusters in relation to the emergence of visual symptoms. Sugar accumulation in afflicted clusters stopped shortly after veraison, well (approx. 20 days) before visible symptoms were detectable, and subsequently the degree of ripeness remained unchanged until harvest. The results suggested that development of "Traubenwelke" initiated before veraison. Distal clusters were more frequently affected than proximal clusters, whereas the position of the shoot on the vine did not impact frequency of occurrence. The gas exchange of leaves positioned opposite to healthy clusters was not significantly different than that of leaves opposite to unhealthy clusters.

K e y w o r d s : 'Zweigelt', Traubenwelke, grape wilting, berry shrivel, sugar accumulation, photosynthesis.

Introduction

The grape variety 'Blauer Zweigelt' (*Vitis vinifera* L., 'Blaufränkisch' x 'St. Laurent') is the most important variety for the production of red wine in Austrian viticulture (Statistik Austria 1999, Schmidt 2009). However, over the last 10 years the production of high-quality grapes has been severely endangered by the occurrence of a physiological disorder called "Traubenwelke" (berry shrivel). Berries of afflicted clusters lose turgor soon after veraison (phenological growth stage: beginning of ripening, BBCH 81), and start to shrivel and wilt irreversibly. Contrary to "Stiellaehme" (bunchstem necrosis, grape stalk necrosis), where the rachis becomes nectrotic (Redl 2007), it remains green and visually healthy in the case of "Traubenwelke". Besides the loss of turgor, shrivelled berries exhibit insufficient colouring, higher acidity, lower sugar content and fewer cultivar-specific aromatic compounds than berries of healthy clusters. Consequently, afflicted grapes can be used neither for fresh consumption nor for vinification. Therefore, the disorder has become a serious economic problem for grape growers because infestation rates of up to 80 % have been observed (Redl 2005).

This disorder was first described in 1997 as "Zweigelt disease" (Reisenzein and Berger 1997), but since then it has also been observed to affect a number of other *Vitis vinifera* varieties, both red ('Zweigelt', 'St. Laurent', 'Pinot Noir', 'Blauburger') and white ('Grüner Veltliner', 'Sauvignon Blanc', 'Neuburger', 'Rotgipfler' and 'Zierfandler'), as well as interspecific crossings not only in Austria (Redl 2007), but also in Germany (Stuecklin 2007), Switzerland (Schumacher *et al.* 2007), Italy (Raifer and Roschatt 2001), Northern Africa (Redl 2008) and the USA (Keller 2008). The exact cause of the Traubenwelke disorder has not yet been detected, but as no causal organism has been identified so far, it is generally characterised as a physiological disorder (Reisenzein and Berger 2001, Redl 2005, 2008). The susceptibility of grapes to Traubenwelke is influenced both by environmental factors, like precipitation and temperature (Raifer and Roschatt 2001, Reisenzein 1998, Redl *et al.* 2007), and vineyard management. Frequency of occurrence of Traubenwelke can be reduced through appropriate soil cultivation avoiding soil compaction, adequate potassium supply, early cluster thinning and adequate but not excessive irrigation (Fardossi 2000, Redl *et al.* 2007, 2009).

What poses a serious problem for grape growers is that afflicted grapes are often not detected until shortly before harvest, because the loss of turgor in the berries and poor coloration are often not visible to the untrained eye in the early stage (Redl 2008). This has led to the assumption that Traubenwelke occurs late in the ripening process, causing hitherto healthy grapes to shrivel suddenly. But previous investigations by Reisenzein and Berger (2001), as well as our personal experience, have indicated that this disorder begins its course much earlier, even before "Traubenwelke" can be visually detected.

Thus, the aim of this study was to identify the onset of the "Traubenwelke" disorder during berry development and to ascertain the point in time when the sugar content between afflicted and healthy berries begins to differ. Therefore, periodical measurements of sugar concentration in randomly selected, tagged clusters were taken at different sites, and the occurrence of visual "Traubenwelke" symptoms was recorded. Furthermore, the irregularity in the occurrence of afflicted clusters within a vine was investigated and observed if it was thought to be explained with the help of gas exchange measurements on grapevine leaves.

Correspondence to: Dr. M. Knoll, University of Natural Resources and Applied Life Sciences, Vienna, DAPP, Institute of Plant Protection, Peter Jordan-Str. 82, 1190 Vienna, Austria. E-mail: mathilde.knoll@boku.ac.at

Material and Methods

E x p e r i m e n t a l   v i n e y a r d s : The field studies were conducted during the 2004 and 2005 growing seasons in three vineyards (A2, C4 and C6) and during the 2006 growing season in two vineyards (A2, C6) in Austria's Neusiedlersee winegrowing district (Seewinkel, Burgenland). The region is characterised by the typical traits of the Pannonian climate, with a mean annual rainfall of 574 mm and an average temperature of 10.1 °C (ZAMG 2002). All vineyards were planted with *Vitis vinifera* L. 'Zweigelt' ('Blaufränkisch' x 'St. Laurent') grafted on the *Vitis berlandieri* x *Vitis riparia* selections Kober 5BB (vineyards C4 and C6) and 161-49 C (vineyard A2). Plant spacing (distance between rows x distance between vines) was 2.6 x 0.8 m, 2.6. x 1.0 m and 1.8. x 0.8 m in C4, C6 and A2, respectively. All vines were on high trellis (stem height 0.9-1.0 m) and cane pruned (one cane and one small spur), resulting in 7 to 10 shoots per vine. Annual vineyard cultivation was carried out according to viticultural practices adopted by the winegrowers. All vines were cluster-thinned to a maximum of two clusters per shoot.

S e l e c t i o n   o f   c l u s t e r s : At the onset of ripening, when berries started to change colour from green to blue and before any symptoms of "Traubenwelke" were visible, 5 to 9 healthy vines per row in each vineyard were randomly selected for observing occurrence of "Traubenwelke" and sugar accumulation. Of these vines, all clusters were tagged beginning with the first vine until the number of 50 clusters was reached. At the same time the position of each cluster within the vine (position of the shoot on the cane, spur or old wood, and position of the cluster on each shoot) was recorded.

M e a s u r e m e n t s   o f   b e r r y   s u g a r   c o n t e n t   a n d   i d e n t i f i c a t i o n   o f   " T r a u b e n w e l k e "   i n c i d e n c e : From veraison (BBCH 81) until ripening (BBCH 89) the sugar content of each cluster was measured in intervals of 7 to 10 d according to weather conditions and investigation year. From each cluster two berries were collected, one from the shoulder and one from the distal part. Immediately after sampling, must density was measured separately for each berry with a handheld refractometer (T/C American Optical, Buffalo, NY, USA). Must density was expressed as °Oe. At the same time, every cluster was visually inspected for incidence of Traubenwelke. Clusters were considered to be afflicted with Traubenwelke when typical symptoms (turgor loss, insufficient pink-purple colouring) could clearly be detected visually or by touching the cluster.

G a s   e x c h a n g e : Photosynthesis (A, µmol $CO_2 \cdot m^{-2} \cdot s^{-1}$), stomatal conductance (gs, $mol \cdot m^{-2} \cdot s^{-1}$) and transpiration (E, mol $H_2O \cdot m^{-2} \cdot s^{-1}$) were measured with an LCpro photosynthesis system, equipped with a broad leaf chamber (ADC Bioscientific Ltd, Hoddesdon, York). Water use efficiency (WUE) was calculated as follows: WUE $= A \cdot E^{-1}$. Measurements were taken on leaves positioned opposite to each tagged cluster. Measurements were carried out on 2nd Sept. 2004 in the vineyard A2 and on 26th Aug. 2005 in every vineyard, shortly after visual symptoms of "Traubenwelke" had already become clearly detectable. Each leaf was measured once (50 leaves per site), and readings were taken at midday, between 11:00 and 14:00. All measurements were taken under saturated light conditions (Q = 1000 $\mu mol \cdot m^{-2} \cdot s^{-1}$) using an LED-light source mounted on the leaf chamber.

S t a t i s t i c a l   a n a l y s i s : Statistical analysis was performed using SPSS statistics 11.0. Sugar content of clusters and gas exchange were analyzed by ANOVA; influence of cluster and shoot position on the incidence of "Traubenwelke" was analyzed using a chi-square test. Significance levels reported were *, **, *** at 0.05, 0.01, 0.001.

Results

The average frequency of "Traubenwelke" occurrence in 50 tagged clusters in each vineyard over three years varied from 12 to 56 %, depending on the site and year (Tab. 1). It was highest in the year 2004, when 50 % of the investigated clusters showed "Traubenwelke" symptoms on the sites C4 and C6, and lowest in 2006, with only 12 to 18 % frequency of occurrence. The progress of sugar accumulation in healthy and afflicted clusters is shown in Figs 1, 2 and 3 for the years 2004, 2005 and 2006, respectively. The abscissa represents the date expressed as d after full bloom, in order to compare the results of different years, independent of the calendar date.

T a b l e 1

Clusters with Traubenwelke (%) during each year

	Frequency of occurrence (%)		
Site	2004	2005	2006
A2	34	12	18
C4	54	22	
C6	52	56	12

In the year 2004 (Fig. 1) the first measurements were taken 56 days after bloom, shortly after the onset of ripening (BBCH 81). At this time must density in berries of healthy clusters (*i.e.* clusters that remained healthy until harvest) was between 38 °Oe (9.6 °Brix) (C4, C6) and 48 °Oe (11.9 °Brix) (A2), and must density in berries of afflicted clusters (*i.e.* clusters that exhibited the first "Traubenwelke" symptoms 19 d later) did not differ from this in a statistically significant manner, with 35 °Oe to 42 °Oe (8.8-10.5 °Brix). In berries of healthy clusters, sugar content quickly increased to 59-62 °Oe (14.6-15.3 °Brix) 67 d after bloom, whereas there was only a minimal increase in sugar content in berries of afflicted clusters, to 48-52 °Oe (12.0-12.9 °Brix). The first visual symptoms of "Traubenwelke" appeared 75 d after bloom, when berries of healthy clusters had already attained 70 °Oe (17.1 °Brix), but berries of afflicted clusters only 48-51 °Oe (12.0-12.7 °Brix), the same level as at the initial measurement. Thenceforward, must density in berries of healthy clusters continued to increase, up to 80-85 °Oe (19.4-20.5 °Brix) 95 d after bloom, whereas must density in berries of afflicted clusters remained at the same low level of 50-53 °Oe

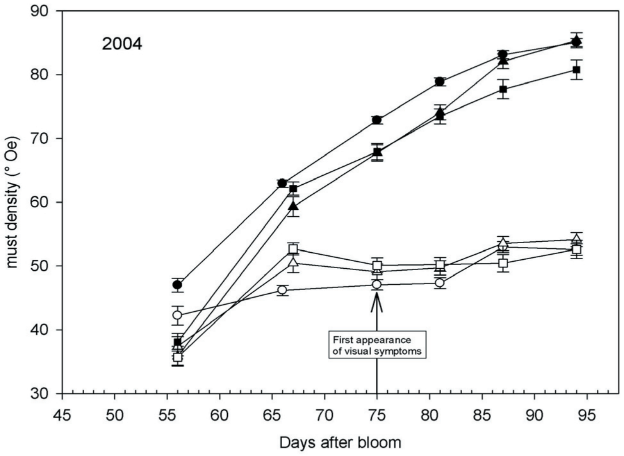

Fig. 1: Must density (°Oe) of healthy (closed symbols) and afflicted (open symbols) clusters during ripening in the year 2004 on the three investigation sites (○ = A 2, □ = C4 and Δ = C6). Error bars show standard error of means; the arrow indicates the date when symptoms of "Traubenwelke" (berry shrivel) were visible for the first time. Differences between healthy and afflicted clusters were not statistically significant (ns) on the first date (56 d after bloom), but highly significant (***) on measuring dates 2 to 6 (67, 75, 81,87 and 94 d after bloom) on every site.

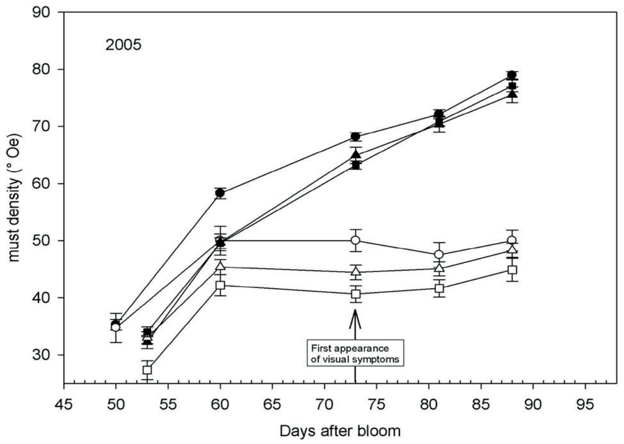

Fig. 2: Must density (°Oe) of healthy (closed symbols) and affected (open symbols) clusters during ripening in the year 2005 on the three investigation sites (○ = A 2, □ = C4 and Δ = C6). Error bars show standard error of means; the arrow indicates the date when symptoms of "Traubenwelke" (berry shrivel) were visible for the first time. Differences between healthy and afflicted clusters were not statistically significant (ns) on the first date (53 d after bloom) on site A2 and C6, but highly significant on C4 (**) and highly significant (***) on measuring dates 2 to 5 (60,73,81 and 87 d after bloom) on every site.

(12.4-13.0 °Brix). The trend of sugar accumulation was similar in the year 2005 (Fig. 2). The first measurement took place 50 d after bloom on the earlier ripening site A2 and 53 d after bloom on C4 and C6. At this time on C4 must density of berries from healthy clusters was significantly higher (34 °Oe; 8.6 °Brix) than must density of berries from afflicted clusters (27 °Oe; 6.9 °Brix), but on A2 and C4 must density of berries from healthy and afflicted clusters was equal and ranged between 32 and 35 °Oe (8.1 and 8.8 °Brix). On all three sites must density increased until the second measurement, 60 d after bloom, in berries from

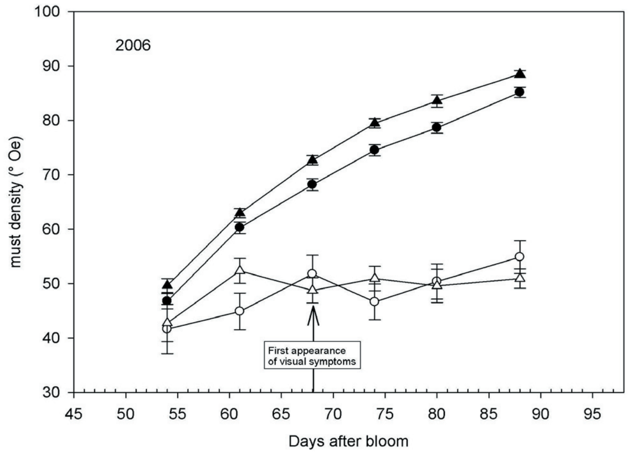

Fig. 3: Must density (°Oe) of healthy (closed symbols) and affected (open symbols) clusters during ripening in the year 2005 on two investigation sites (○ = A 2, □ = C4 and Δ = C6). Error bars show standard error of means; the arrow indicates the date when symptoms of "Traubenwelke" (berry shrivel) were visible for the first time. Differences between healthy and affected clusters were not statistically significant (ns) on the first date (53 d after bloom) but highly significant (***) on measuring dates 2 to 5 (60, 73,81 and 87 d after bloom) on both sites.

both healthy and afflicted clusters. Nevertheless, on all three sites the increase in sugar content was significantly higher in berries from healthy clusters than in those from afflicted clusters. Similar to the year 2004 the first visual symptoms appeared 73 d after bloom, when must density of healthy clusters was 63-68 °Oe (15.5-16.6 °Brix) and only 40-50 °Oe (10.0-12.4 °Brix) in afflicted clusters. The divergence between must density in healthy and afflicted clusters further increased as sugar accumulation continued in healthy clusters to 75-79 °Oe (18.2-19.1 °Brix) 88 d after bloom, whereas must density in afflicted clusters remained at the same level, between 44 and 52 °Oe (11.0 and 12.9 °Brix), from the second sampling date until the end of the sampling period.

In the year 2006 measurements were performed only on the two sites A2 and C6. Already 54 d after bloom, berries from healthy clusters had a slightly higher must density (47-50 °Oe; 11.7-12.4 °Brix) than berries from afflicted clusters (42-43 °Oe; 10.5-10.8 °Brix); the difference was statistically significant for the mean of both sites. There was only little increase of sugar content in berries of afflicted clusters, to 45-52 °Oe (11.3-12.9 °Brix) 61 d after bloom, but a strong increase of sugar content in berries of healthy clusters (60-63 °Oe; 14.8-15.5 ° Brix). When visually detectable symptoms appeared on the third date, 68 d after bloom, must density in healthy clusters was between 68 and 73 °Oe (16.6 and 17.8 °Brix), significantly higher than in afflicted clusters (49-52 °Oe; 12.2-12.9 °Brix). With afflicted clusters there was neither an increase nor a decrease of must density from 68 d after bloom to the last sampling date, 88 d after bloom, whereas must density in healthy clusters steadily increased to 85-88 °Oe (20.5-21.1 °Brix).

In summary, sugar content in afflicted clusters tended to be lower already shortly after veraison, 50 to 56 d after bloom, increased only slightly until 60 to 61 d after bloom and remained at the same level thereafter. Unlike sugar content, in all three years the typical visually detectable symptoms of "Traubenwelke" (loss of turgor and insufficient change to a pink-purple colour) did not occur until 68-75 d after full bloom. Thus, the time span from the first difference in sugar content between healthy and afflicted clusters to the appearance of visual Traubenwelke symptoms was approximately 20 d.

Since afflicted and healthy clusters were often found on the same shoot (Fig. 4), it was tested whether the position of the cluster on the shoot (proximal cluster or distal cluster) had an influence on frequency of occurrence. Distal clusters tended to be more frequently affected by Traubenwelke than proximal clusters at all three locations

Fig. 4: 'Zweigelt' cluster afflicted by "Traubenwelke" (bottom) adjacent to a healthy cluster (top).

Table 2

Influence of cluster insertion (proximal or distal cluster) on the shoot on frequency of Traubenwelke occurrence

site	year	Frequency of occurrence (%) proximal cluster	distal cluster	significance (Chi-Square Pearson)
A2	2004	27,3	47,1	0,162 ns
	2005	12,5	10,0	0,828 ns
	2006	19,6	0,0	0,329 ns
	Total	19,3	29,0	0,240 ns
C4	2004	54,5	52,9	0,914 ns
	2005	20,6	25,0	0,725 ns
	Total	37,3	39,4	0,840 ns
C6	2004	44,8	65,0	0,219 ns
	2005	56,3	50,0	0,670 ns
	2006	6,1	23,5	0,072 ns
	Total	35,1	46,4	0,170 ns
Total		28,9	40,0	0,030*

(Tab. 2). On average 29 % of proximal clusters but 40 % of distal clusters were affected. However, there were great variations between the years, especially on site A2. In contrast to cluster position, shoot position on the vine did not significantly influence frequency of occurrence (Tab. 3). Symptoms of "Traubenwelke" appeared in 31 % of the clusters positioned on the cane, 34 % on the spur and 40 %

Table 3

Influence of shoot position within the vine on frequency of Traubenwelke occurrence in shoots within a vine

Site	year	Frequency of occurrence (%) cane	spur	old wood	significance (Chi-Square Pearson)
A2	2004	33,3	37,5	33,3	0,974 ns
	2005	12,5	0,00	-	0,594 ns
	2006	17,1	25,0	20,0	0,918 ns
	total	20,0	28,6	27,3	0,670 ns
C4	2004	55,6	50,0	50,0	0,939 ns
	2005	12,1	38,5	50,0	0,056 ns
	total	34,8	42,9	50,0	0,570 ns
C6	2004	59,5	25,0	40,0	0,178 ns
	2005	56,8	45,5	50,0	0,799 ns
	2006	7,9	20,0	50,0	0,139 ns
	Total	41,1	31,0	44,4	0,584 ns
Total		31,0	34,4	40,0	0,560 ns

on old wood (fruiting water sprout), respectively. Measurements of gas exchange on leaves positioned opposite to each of the 50 tagged clusters were performed a day after the first observation of "Traubenwelke" (74 and 76 d after bloom, respectively, in the years 2004 and 2005). There were no statistically significant differences in photosynthesis, stomatal conductance or water use efficiency (Tab. 4) of leaves that were positioned on the same node as an afflicted or healthy cluster. Nevertheless, photosynthesis and stomatal conductance tended to slightly higher values on leaves opposite to afflicted clusters, while water use efficiency was lower.

Discussion

Periodical measurements of must density of tagged 'Zweigelt' clusters from veraison until ripening over three years at three different locations in Austria's Seewinkel winegrowing region (Burgenland) clearly showed that from the onset of ripening, sugar accumulation in clusters afflicted by "Traubenwelke" differed from sugar accumulation in healthy clusters. This was an important finding, because it opposed the belief that Traubenwelke affected healthy clusters suddenly before harvest. On the contrary, it provided evidence that sugar accumulation in afflicted clusters stopped shortly after veraison, almost three weeks before any symptoms of "Traubenwelke" were visible, and sugar content remained at the same level thereafter. Thus, our findings suggested that the onset of the development of "Traubenwelke" occurred no later than veraison, because beginning with veraison, differences in sugar content were already detectable.

Reisenzein and Berger (2001) also found that must densities of afflicted and healthy clusters differed from each other from the beginning of the ripening period; however, unlike the trend observed in our study, they noted a sharp increase in the must density of afflicted clusters after veraison from approximately 6,5 °KMW (31 °Oe; 7.8 °Brix) to

Table 4

Gas exchange (photosynthesis, stomatal conductance and water use efficiency) of grapevine leaves positioned opposite to healthy and afflicted clusters

Year	Site	Healthy	Afflicted	Significance
Photosynthesis (µmol $CO_2 \cdot m^{-2} \cdot s^{-1}$)				
2004	A2	10,52	11,02	0,537 ns
2005	A2	6,70	7,58	0,407 ns
	C4	6,43	6,52	0,908 ns
	C6	6,73	7,02	0,657 ns
	mean	6,61	6,97	0,394 ns
Stomatal conductance (mol·$m^{-2} \cdot s^{-1}$)				
2004	A2	0,157	0,171	0,327 ns
2005	A2	0,264	0,250	0,838 ns
	C4	0,127	0,132	0,801 ns
	C6	0,194	0,210	0,558 ns
	mean	0,198	0,196	0,914 ns
Water use efficiency				
2004	A2	4,26	4,20	0,740 ns
2005	A2	3,25	3,23	0,942 ns
	C4	4,55	3,94	0,495 ns
	C6	3,70	3,61	0,681 ns
	mean	3,83	3,64	0,475 ns

13 °KMW (63 °Oe; 15.5 °Brix). These contradicting results may be explained by the small sample size in their study, because the "Traubenwelke" disorder usually shows a high variability between years and locations, and even within the same vineyard (Knoll *et al*. 2006, Redl 2008). The results of the present study, demonstrating that must density of afflicted 'Zweigelt' berries remained at the same low level of 45-55 °Oe (11.3-13.6 °Brix) throughout berry ripening, also differed considerably from recent findings of Krasnow *et al.* (2009) in Napa Valley, California, who reported an increase in total soluble solids in shrivelled 'Cabernet Sauvignon' berries after veraison of up to 19-25 °Brix (76 to 106 °Oe). Moreover, they observed initial berry shrivel symptoms not before 106 to 112 d after anthesis, almost 40 d later than symptoms of "Traubenwelke" were visually detectable with 'Zweigelt' clusters in the present study. These differences in sugar accumulation and date of appearance of first symptoms between "Traubenwelke" on 'Zweigelt' and shrivel disorder on 'Cabernet Sauvignon', as described by Krasnow *et al.* (2009), lead to the conclusion that there are different forms of shrivel disorder which are not identical. Furthermore, Krasnow *et al.* (2009) concluded from their results that berry shrivel was a phenomenon affecting the whole vine, which means that all clusters of an afflicted vine have some symptoms, whereas our results clearly indicated that "Traubenwelke" affected single clusters, while the other clusters on the same vine remained healthy. Furthermore, we also observed high variability between the years, which means that a single vine could bear numerous afflicted clusters in one year and none in the following year (Redl *et al.* 2007), corresponding with findings by Keller (2008). Our study provided evidence that distal clusters on the shoot were slightly more frequently afflicted by "Traubenwelke" than were proximal clusters. But the results were not statistically significant for every year and site. Contrary to this, Reisenzein and Berger (2001) reported that proximal 'Zweigelt' clusters had higher rates of occurrence than distal clusters. Schumacher *et al.* (2007) studied the occurrence of "Traubenwelke" on different grape varieties in Switzerland and found that proximal 'Blauer Burgunder' clusters tended to higher frequency of occurrence, while with 'Gamaret', distal clusters were more frequently afflicted, but they also pointed out the variability within a single vine. However, higher rates of occurrence of "Traubenwelke" in distal clusters may be associated with a delay in ripening of distal clusters compared to proximal clusters. In our study, berries of healthy 'Zweigelt' clusters in the distal position tended to lower must densities than berries of healthy, proximal clusters. This was most pronounced shortly after veraison, when sugar accumulation of healthy and afflicted clusters began to differ (data not shown).

The gas exchange measurements on leaves positioned opposite to the tagged clusters did not show any significant connections between occurrence of afflicted clusters and gas exchange of leaves. The values for photosynthesis were low, most probably because the measurements were taken on leaves in the fruiting zone, which were already old leaves at this time and showed a decline in photosynthetic activity compared to adult leaves in the middle of the shoot (Knoll 2008). Nevertheless, photosynthetic activity, stomatal conductance and water use efficiency were in a normal range and therefore indicated that the xylem of shoots and leaves where afflicted clusters were attached was not defective.

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