# Crop cultural and chemical methods to control grey mould on grapes

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# Summary

The efficiency of crop cultural (leaf removal) as well as of chemical methods (plant growth regulator, botryticide) to control grey mould caused by Botrytis cinerea was investigated in two seasons (2008 and 2009) on the varieties 'Pinot blanc' and 'Pinot gris' in the Moselle valley (Luxembourg). The application of the plant growth regulator Regalis® (a.i. prohexadione-Ca) led to a considerably more flexible cluster structure and a slight decrease of grey mould disease severity. The reduction of bunch rot infestation was of the same level than obtained by a single application of a botryticide (a.i. fenhexamid) before berries touching. Leaf removal reduced the cluster density slightly and proved to be more efficient against B. cinerea than the chemical treatments (reduction of grey mould disease severity of 57 % on average). Thus, leaf removal in the cluster-zone shortly after bloom can be recommended as an important tool in integrated as well as in organic bunch rot protection strategies. The best loosening effect on the cluster structure as well as the best B. cinerea reduction efficiency (75 % on average) was achieved when combining leaf removal and Regalis® application. The combination of leaf removal and botryticide application showed comparable results. Simulation of the B. cinerea epidemiology demonstrated that all treatments tested might allow for a longer maturation time due to lower infestation. The longest potential harvest delay until reaching an assumed threshold of 5 % disease severity was achieved by combining leaf removal and application of Regalis® or a botryticide (on average 11.6 or 9.9 days, respectively). The here presented strategies can thus be recommended to maximize wine quality in two ways - through a reduction of fungal contamination and/or an improvement of grape maturity.

K e y w o r d s : *Vitis vinifera*, *Botrytis cinerea*, prohexadione-Ca, leaf removal, cluster structure

# Introduction

Grey mould or bunch rot caused by *B. cinerea* can reduce grape yield and quality (RIBÉREAU-GAYON 1983) causing economic damages worldwide. Indeed, beside the direct loss of yield, attack of *B. cinerea* can cause off-flavours, unstable colour, oxidative damages, premature aging and difficulties in clarification (RIBÉREAU-GAYON 1983,

SMART and ROBINSON 1991). Moreover, other fungi and bacteria can readily invade grey mould infected clusters, which further contributes to off-flavours (SMART and ROBINSON 1991). Especially if the fungal pathogens *B. cinerea* and *Penicillium expansum* both occur simultaneously, this can result in earthy-mouldy off-flavours in the wine (LA GUERCHE *et al.* 2005).

The starting points for the bunch rot epidemiology are often infections occurring on the interior parts of compact clusters. Such clusters with a tight structure exhibit a high interior humidity. Furthermore, berries on the interior parts of tight clusters often split (SMART and ROBINSON 1991) under the increasing pressure after veraison offering favourable conditions for the development of fungal pathogens. Consequently, one strategy to prevent bunch rot is to loosen the structure of the cluster. First trials to disaggregate dense clusters by using a plant growth regulator have been conducted by Weaver (Weaver et al. 1962, Weaver 1975) using gibberellic acid – a hormone naturally occurring in plants and regulating various metabolic processes. However, depending on the application time and dose, the developmental stage of the plant as well as on the weather conditions during application, the effects of a gibberellic acid application can be very different. Untimely usage or overdosage might negatively affect the crop in the current year and, moreover, decrease bud burst and the number of grapes per shoot in the following season (WEYAND and SCHULTZ 2005). Trials in the years 2007 to 2009 on the varieties 'Pinot gris', 'Pinot blanc' and 'Pinot noir' (EVERS et al. 2010) indicate that the application of gibberellic acid GA3 in a dose of 0.016 kg·ha<sup>-1</sup> at full flowering is able to loosen the cluster structure and to reduce the disease severity, confirming the results of Korkutal et al. (2008). Results obtained with the plant growth regulator prohexadione-Ca, the active ingredient of the commercial product Regalis®, are presented in this paper. Lo Giudice et al. (2004) showed that the application of prohexadione-Ca reduced single berry sizes as well as the number of berries per cluster, thereby generally affecting the fruit composition in a positive way.

In the past, the strategies to control *B. cinerea* were mainly focused on the use of organic-synthetic botryticides. Following the recommendations of Integrated Pest Management, meanwhile, crop cultural methods are of increasing interest in sustainable plant protection strategies. Furthermore, in organic viticulture, these crop cultural practices represent a major option for pathogen control, as organic-synthetic fungicides are not allowed. Especially defoliation of the cluster-zone close to bloom has shown its potential to reduce bunch rot in several trials under dif-

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ferent climatic conditions (Zoecklein *et al.* 1992, Percival *et al.* 1994, Ollat and Gaudillere 1998, Poni and Intrieri 2001, Intrieri *et al.* 2008, Tardaguila *et al.* 2008).

The aim of the present study was the assessment of crop cultural (leaf removal in the cluster-zone after bloom) as well as chemical methods (plant growth regulator, botryticide) and combinations of them in terms of their impact on (i) the cluster structure, (ii) harvest parameters such as yield and sugar level (iii) the grape health status and finally (iv) the possibility to prolong the ripening period.

# **Material and Methods**

Vineyard sites and experimental design: The studies were carried out in 2008 and 2009 along the river Moselle between Remich and Machtum in Luxembourg in three commercial vineyards planted with the *Vitis vinifera* L. varieties 'Pinot gris' (PG) (2 vineyards) and 'Pinot blanc' (PB). The experimental vineyards 'Pinot gris I' (planted in 1980, soil: loamy clay, rootstock: SO4, clone: Colmar 52) and 'Pinot blanc' (planted in 2001, soil: keuper, rootstock: SO4, clone: Dreher 209) were southeast exposed with an inclination of around 30 % and a southwest-northeast orientation of the rows. The vineyard 'Pinot gris II' (planted in 1998, soil: shell limestone, rootstock: SO4, clone: Pépinière, Colmar) was southeast exposed with an inclination of around 5 % and a row orientation from southeast to northwest.

The plantation density was 1.8 (PG I), 2.0 (PG II) and 2.2 (PB) m<sup>2</sup> per plant. All vineyards were vertical shoot positioning trained.

The field experiments were performed using a randomized block design, consisting of 4 replicates (except: 'Pinot gris I' (2008) – which consisted of 8 replicates) of 10-12 vines per treatment, hereafter referred to as plots. The treatments were defined as given in Tab. 1.

Overall, each treatment was tested six times (3 experimental fields, 2 years).

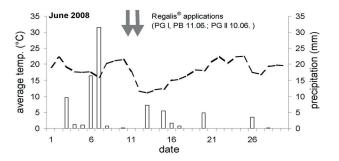
Regalis® treatments were conducted at full flowering (BBCH 65) (LORENZ et al. 1995) early in the morning at high relative humidity levels. To adjust the pH of the spraying mixture, 0.1% of citric acid was added. All experiment-specific applications (botryticides as well as Re-

galis®) were carried out manually with a backpack sprayer. Water amounts used were 400 l·ha<sup>-1</sup> for both Teldor® and Regalis®. Leaf removal consisted in manually removing leaves exclusively in the cluster-zone on the north or east exposed sides of each row just after the end of flowering (BBCH 71). Per shoot, two to four leaves were removed (depending on the number of clusters per shoot).

Pruning as well as canopy and soil management were done by the farmers in a comparable way. Herbicides were used to suppress weed development. Background coverage applications against *Plasmopara viticola* and *Erysiphe necator* were carried out from the air (helicopter) and from the ground (tractor driven fungicide sprayers) by the farmers. No background coverage against *B. cinerea* was carried out

Daily average temperatures and daily precipitation sums for both years were recorded at the weather station Remich (Fig. 1).

Determination of grape bunch structure: To determine the impact of the different treatments



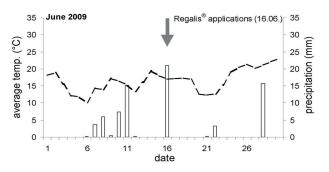


Fig. 1: Daily average temperatures and daily precipitation sums in June 2008 and 2009 at the weather station Remich.

Table 1

Description of the treatments

	Measures	Active ingredient(s)	Developmental stage (LORENZ et al. (1995))	Application dose (g·ha <sup>-1</sup> or mL·ha <sup>-1</sup> )
T1	untreated control			
T2	Regalis®	prohexadione-Ca (100 g·kg <sup>-1</sup> )	BBCH 65	1500
T3	Teldor®	fenhexamid (510 g·kg <sup>-1</sup> )	BBCH 77	1600
T4	leaf removal		BBCH 71	
T5	Regalis®	prohexadione-Ca (100 g·kg <sup>-1</sup> )	BBCH 65	1500
	+ Teldor®	fenhexamid (510 g·kg <sup>-1</sup> )	BBCH 77	1600
T6	Regalis®	prohexadione-Ca (100 g·kg <sup>-1</sup> )	BBCH 65	1500
	+ leaf removal		BBCH 77	
T7	Teldor®	fenhexamid (510 g·kg <sup>-1</sup> )	BBCH 71	1600
	+ leaf removal		BBCH 77	

on cluster structure, an assessment of the "density index" according to the protocol of IPACH *et al.* (2005) was used. In each plot, 100 bunches (50 on each side of the row) were evaluated at the phenological growth stage BBCH 79 and the cluster structure was classified between 1 and 5 according to Tab. 2. Values are the means of the observations on 100 bunches per plot, averaged for the four replicates.

Determination of B. cinerea infestation level: Disease incidence and severity were assessed at several time-points during the ripening period by examining 100 randomly selected clusters per plot (50 on each side of the row) according to the EPPO (European and Mediterranean Plant Protection Organization) guideline PP1/17.

Determination of sugar level and yield: Close to harvest, 25 berries were randomly collected on both sides of the row in each plot and thereafter pooled for every treatment to achieve a sufficiently large sample. After pressing, the juice was centrifuged. Sugar level was assessed with a digital refractometer.

Experimental harvest was conducted on the same date for all variants of one trial. Each plot was harvested separately and the yield recorded.

Description of disease progress: To describe the epidemiology of *B. cinerea*, the assessed disease severity values were plotted against the date of assessment (expressed as days after bloom (BBCH 68)). Progress curves were fitted to these data according to the equation  $y = e^{a(x-x_0)}$ , where y = disease severity, a = slope of the curve, x = days after bloom (BBCH 68) and  $x_0 =$  days after bloom (BBCH 68) reaching a disease severity of 1 %.

Using this equation allows to estimate the time-point at which a certain disease severity value would be reached. We assumed an acceptable grey mould threshold of 5 % for the production of high quality white wines.

S t a t i s t i c a 1 a n a l y s i s: Data were analyzed by one-way ANOVA. For multiple comparison procedures between means, Tukey tests ( $\alpha = 5$  %) were performed using SPSS 16.0 for Windows.

#### Results

Density of the clusters: The application of Teldor® (T3) showed no effect on the cluster density. A reduction of the density index was observed in each of the six trials as a result of a Regalis® treatment (T2) (relative density index 83.1 %, on average). This reduction was statistically significant in all three trials of the year 2008 and in the 'Pinot blanc' trial of the year 2009. A reduction of the density index was also observed in all six trials as a result of leaf removal (T4) (relative density index 90 %, on average). However, this reduction was not significant in any of the six trials in comparison to the control. Regalis® treatment coupled to leaf removal (T6) showed the lowest density index in each of the six trials (relative density index 75 %, on average) (Tab. 3, Fig. 2).

D i s e a s e s e v e r i t y: The treatment including only the application of the botryticide Teldor® (T3) resulted on average in a reduction of disease severity of 25.9 % compared to the untreated control. However, the reduction was not significant in any of the six trials in comparison to the untreated control. The efficiency of the Regalis® treatment (T2) ranged between 9 ('Pinot gris II' (2009)) and 61 % ('Pinot gris II' (2008)) (28 % on average). Only in the trial 'Pinot gris II' (2008), a significant reduction of the disease severity in comparison with the untreated control was observed. The combination of both chemical measures (T5) provided a more stable efficiency level concerning the disease severity (between 44 and 62 % and an aver-

T a b l e 2

Description of the bunch density index according to IPACH et al. (2005)

Index	Description
1	Very loose; berries do not touch; bending of the stem to 90° possible
2	Loose, berries touch; bending of the stem to 45°-90° possible
3	Dense bunch structure; berries still flexible; bending of the stem to 10-45° possible
4	Compact bunch structure; berries not flexible; bending of the stem up to 10°
5	Very compact bunch structure; berries not flexible; bending of stem not possible

T~a~b~l~e~~3 Average bunch density index. Treatments not marked with the same letter differ significantly according to Tukey test (\$\alpha\$ = 5 %)

	Pinot gris I		Pinot gris II		Pinot blanc	
	2008	2009	2008	2009	2008	2009
T1	3.48 a	3.80 a	3.85 a	3.60 a	3.66 a	3.82 a
T2	2.93 b	3.45 ab	2.97 c	3.08 abc	2.98 bc	3.02 bc
T3	3.28 ab	3.77 a	3.80 a	3.44 ab	3.70 a	3.70 a
T4	3.11 ab	3.30 ab	3.49 ab	3.13 abc	3.49 ab	3.47 ab
T5	2.90 b	3.24 ab	3.15 bc	3.04 bcd	2.97 bc	2.99 bc
T6	2.90 b	2.94 b	2.86 c	2.55 d	2.70 c	2.85 c
T7	3.19 ab	3.29 ab	3.52 ab	2.96 bcd	3.68 a	3.50 ab

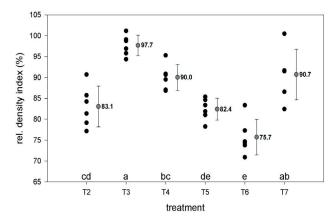


Fig. 2: Relative bunch density index. Each spot represents the relative density index compared to the untreated control (100 %) in one single trial. Treatments not marked with the same letter differ significantly according to Tukey test ( $\alpha = 5$  %). The bars indicate the standard deviation and the grey spots in the middle of the bars the relative density index on average of all six trials.

age of 48 %). The reduction was statistically significant in three of six trials in comparison to the untreated control. The crop cultural measure leaf removal (T4) turned out to be more efficient than the chemical treatments Regalis® or Teldor® when applied alone. The efficiency level compared to the untreated control was never below 38 % and reached 68 % in both 'Pinot gris' trials (2009) as well as in 'Pinot blanc' (2008) (average efficiency level of 57 %). In those three trials and in addition in 'Pinot gris I' (2008), the reduction was statistically significant in comparison to the untreated control. The combination of leaf removal and Teldor® (T7) provided an efficiency level between 58 and 80 % (70 % on average) and in five of six trials a significant reduction in comparison to the untreated control was observed. The highest average efficiency of all treatments was achieved by the combination of leaf removal and Regalis® (T6) (75 %). The efficiency never ranged below 64 % and even reached 84 % in the trials 'Pinot blanc' (2008) and 'Pinot gris II' (2009) with a significant reduction of the disease severity in comparison to the untreated control in all six trials (Tab. 4, Fig. 3).

Disease progress curves: The equation to describe the disease progress ( $y=e^{a(x-x_0)}$ ) proved to be well adapted to the epidemics of *B. cinerea* under the present experimental and climatic conditions. The coefficient of determination ( $R^2$ ) ranged between 0.963 and 0.999. As an example, the disease progress curves of the trial 'Pinot gris I' (2008) are shown in Fig. 4.

According to these calculations, all treatments allowed for a potential delay of the harvest date (time-point reaching grey mould threshold of 5 %) compared to the untreated control ranging between 2.5 (T2 and T3) and 11.6 d

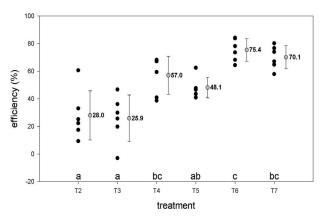


Fig. 3: Efficiency of the treatments concerning the disease severity of *Botrytis cinerea*. Each spot represents the efficiency compared to the untreated control in one single trial. Treatments not marked with the same letter differ significantly according to Tukey test ( $\alpha = 5$ %). The bars indicate the standard deviation and the grey spots in the middle of the bars the efficiency on average of all six trials.

Table 4

Disease incidence and disease severity of *Botrytis cinerea* at the final assessments in the years 2008 and 2009. Treatments of one year not marked with the same letter differ significantly according to Tukey test ( $\alpha = 5 \%$ )

		Pinot	gris I	Pinot gris II		Pinot blanc	
year		disease	disease	disease	disease	disease	disease
		incidence (%)	severity (%)	incidence (%)	severity (%)	incidence (%)	severity (%)
2008	T1	65.1 a	17.7 a	36.8 a	4.4 a	52.3 a	5.3 a
	T2	56.8 ab	13.7 ab	20.8 b	1.7 b	41.3 ab	3.6 abc
	T3	55.6 abc	11.3 abc	25.3 ab	2.3 ab	46.3 a	4.3 ab
	T4	43.1 bcd	7.2 bc	26.8 ab	2.7 ab	25.8 ab	1.7 bc
	T5	51.0 abcd	10.0 bc	17.0 b	1.7 b	35.0 ab	2.8 abc
	T6	38.3 d	5.6 c	16.3 b	1.6 b	18.5 c	0.8 c
	T7	38.8 bcd	5.8 c	14.5 b	1.0 b	31.0 ab	2.2 bc
2009	T1	89.0 a	33.5 a	71.8 a	14.6 a	86.9 a	19.4 ab
	T2	80.3 a	25.1 ab	65.4 ab	13.3 ab	73.8 abc	16.0 abc
	T3	83.5 a	24.9 ab	57.6 abc	10.3 abcd	83.0 ab	20.0 a
	T4	58.0 bc	10.9 cd	40.6 cde	4.7 bcd	69.5 bcd	11.5 abcd
	T5	76.6 ab	19.8 bc	50.0 abcd	7.7 abcd	65.3 cd	10.1 bcd
	T6	48.5 c	7.3 d	28.2 de	2.4 d	44.3 e	5.1 d
	T7	50.1 c	8.7 cd	25.5 abc	2.9 abcd	52.9 de	6.8 cd

(T6), on average. This latter treatment (T6) allowed for a potential prolongation of the ripening period of at least 7.8 ('Pinot gris' (2008)) and up to 16.8 d ('Pinot blanc' (2009)) (Fig. 5).

Harvest parameter: On average of all six trials the use of Teldor® led to an insignificant increase of the yield (+2% compared to the control). All other treatments decreased the total yield level on average between 11 and 25%. The lowest total yield was observed in the treatment T6 (Regalis® + leaf removal) with on average 75% of the yield harvested in the control. For this treatment, the variation between the trials was high. The relative total yield in this treatment ranged between 55 ('Pinot gris II' (2009)) and 105% ('Pinot gris I' (2008)) compared to the untreated control (data not shown).

All treatments increased, on average of the six trials, the sugar level at harvest marginally. The highest average sugar level of all treatments was reached in the treatment T6 (Regalis® + leaf removal). This treatment showed a higher sugar level than the control in each of the six trials (103.6 % of the control, on average) (data not shown).

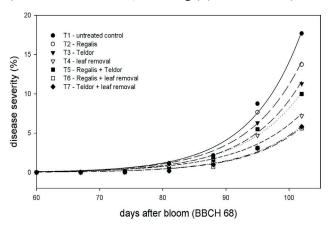


Fig. 4: Disease progress curves of the trial 'Pinot gris I' (2008) plotted against the days after bloom (BBCH 68). The spots represent the observed disease severities, the lines the calculated progress according to the equation  $y = e^{a(x \cdot x_o)}$ .

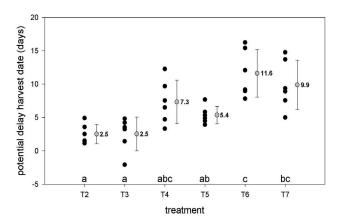


Fig. 5: Potential delay of the harvest date. Each spot represents the potential delay reaching the calculated threshold of 5 % disease severity of *Botrytis cinerea* compared to the untreated control in one single trial. The bars indicate the standard deviation and the grey spots in the middle of the bars the potential delay of the harvest on average of all six trials.

# Discussion

Loose clusters lead to an increased airflow and a better sun and wind exposure of the interior parts of the grape. This results in a reduced interior humidity and, in consequence, in less favourable conditions for the development of fungal pathogens (Zoecklein *et al.* 1992). Moreover, a loose structure diminishes the risk of berry burst due to high pressure inside the cluster induced by berries touching each other (SMART and ROBINSON 1991). Burst berries are easy to colonize by fungal pathogens.

In the present study, the use of Regalis® as well as the defoliation of the cluster-zone led to less compact cluster structures most likely as results of increased abortion of inflorescences or young berries (lower number of berries per grape) as well as smaller berry sizes.

In the case of the plant growth regulator Regalis® (active ingredient: prohexadione-Ca), the loosening effect is most likely due to the perturbation of pollination and cell division processes. Prohexadione-Ca is a structural mimic of a co-substrate in the biosynthesis of gibberellins (RADEMACHER 2000). The promotion of flower or berry abortion could thus be due to a disturbance in the proportion of biologically active and inactive gibberellins.

Leaf removal after flowering led in all six trials to a reduction of cluster density. However, this effect was not as clear as in the case of the use of Regalis® and mostly not statistically significant compared to the untreated control. The observed disaggregation might be caused by the reduction of assimilating leaves. This presumably led to a reduced supply of assimilates required for the post-bloom cell division processes and, in consequence, to a higher berry abortion and smaller berry sizes. Indeed, the degree of abscission is related to the sugar concentration in the inflorescence shortly after anthesis (VASCONCELOS *et al.* 2009). Observations by Intrieri *et al.* (2008) as well as OLLAT and GAUDILLERE (1998) confirm the reduced berry number and size on clusters of defoliated vines.

In the present study, the most important loosening effect was observed as a result of the combination of a Regalis® application and leaf removal. The combination of the disturbance of the plant hormone status and the additional reduction of the assimilate supply probably led to a considerable berry abortion and reduction of berry size.

Reduction of the grey mould infestation level of *B. cinerea*. The application of the botryticide Teldor® before berries touching – a standard method in Luxembourg's wine growing region – showed an efficiency level of around 25 % on average of all six trials. Especially in trials with a high disease pressure, the effects of this treatment were low.

The exclusive use of the plant growth regulator Regalis® at full bloom reached the same efficiency level (around 28 % on average) as the application of the botryticide Teldor®. Vail & Marois (1991) found a significant correlation between cluster density and the disease severity of *B. cinerea*. However, the significant loosening effects on the

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cluster structure as a result of the Regalis® application observed at BBCH 79 (majority of berries touching) were not that obvious at the end of the ripening period. This might be due to a compensation reaction and may explain why grey mould infestation level at harvest was higher than expected at earlier developmental stages.

Good results were achieved through leaf removal just after flowering (T4) with an efficiency against *B. cinerea* of more than 50 % without any input of chemical substances. The reason for this is the combination of the described reduced cluster compactness and the better sun and wind exposure. The efficiency of leaf removal closely after bloom has been confirmed in several studies (ZOECKLEIN *et al.* 1992, PERCIVAL *et al.* 1994, OLLAT and GAUDILLERE 1998, INTRIERI *et al.* 2008, TARDAGUILA *et al.* 2008) and can hence be recommended as a standard procedure in quality-orientated grape protection strategies. Furthermore, its efficiency makes leaf removal the most powerful tool in organic viticulture where organic-synthetic botryticides as well as plant growth regulators are not allowed.

The success of the combination Regalis® with leaf removal (average efficiency level 75 %) is most likely caused by the double interference of the grape physiology at and just after flowering which leads to an enhancement of the effects described for each single treatment. Especially the density of the clusters is considerably reduced and it seems as if the compensation reaction was less intensive than in the exclusive Regalis® treatment.

Regalis® is expected to be metabolized in grapes between two to three weeks after application without leaving any residues neither in the vineyard nor in the wine (Rademacher, personal communication). Thus, the combination Regalis® and leaf removal could represent a strategy for sustainable viticulture to reduce pesticide input into the environment.

Potential prolongation of the maturity period: In cool climate winegrowing regions the timing of the harvest date is more often determined by the decreasing health status (due to fungal attack) of the grapes than by their maturity. With increasing disease severity of *B. cinerea* and secondary mould pathogens, like *Penicillium expansum*, the risk of loss of fruitiness and of the appearance of off-flavours increases. Thus, for the production of high quality wines, the fungal rot has to be minimized in order to optimize grape maturity. Especially under cool climate viticultural conditions, a delay of harvest date leads to an increase in wine quality (Spring 2004) and, furthermore, a decreased risk of atypical aging flavours.

As shown, the disease severity of *B. cinerea* progresses approximately exponentially. Assuming a tolerable threshold of grey mould disease severity of 5 % for high quality white wines, the values for the potential delay of the harvest in the present studies reached between 2.5 (Regalis® or Teldor®) and 11.6 d (Regalis® + leaf removal).

In fluence on harvest parameters: The total yield per plant was slightly reduced by all treatments, apart from the application of the botryticide (T3). The marginally higher yield in this treatment can be explained by the on average lower disease severity compared to the untreated control. With increasing bunch rot infestation level the yield decreases due to drying out of berries and shedding of grapes due to rotten stems. The reduced yield in the other treatments is most likely the result of the impact on pollination and cell-division caused by leaf removal and/or the application of Regalis®. Yield reduction in these treatments ranged between 12 and 18 % but the reduction was in many cases not significant compared to the untreated control. The degree of the yield reductions due to a Regalis® application seems to be dependent on timing and dosage.

Intrieri *et al.* (2008) also found a clear reduction of the yield due to a defoliation of the cluster-zone, whereas no significant yield reductions were observed in the trials of Tardaguila *et al.* (2008), Zoecklein *et al.* (1992) and Bledsoe *et al.* (1988). Probably, the impact of leaf removal on the yield is depending on the weather conditions and the general yield level of the vineyard.

In the present trials, the highest yield reduction (25 % on average) was observed if Regalis® and leaf removal were applied in combination – probably due to the double interference at the grape physiological level. In the year 2009, reductions of up to 45 % were observed. We suppose that this was caused by the cold and rainy flowering period (Fig. 1). Cool rainy weather in general leads to poor pollination (VASCONCELOS *et al.* 2009) and this natural effect is most likely enhanced by both measures applied. In consequence, it might be safer to decrease the dose of Regalis® under such conditions or not to use it at all, if considerable yield reductions are not acceptable.

The differences in sugar level observed in the trials were low and most likely mainly caused by the differences in the yield following quantity-quality-correlation (Howell 2001).

# **Conclusions**

Manual leaf removal in the cluster-zone just after bloom provided a significant reduction of bunch rot infestation without any input of chemical substances. Subsequently, this treatment can be recommended as an important tool in any bunch rot protection strategy for integrated as well as organic viticulture.

Application of the commercial plant growth regulator Regalis® (a.i. prohexadione-Ca) at full flowering (BBCH 65) as well as the application of the botryticide Teldor® (a.i. fenhexamid) before bunch closure (BBCH 77) also slightly decreased disease severity, but the efficiency of these treatments was lower than of the leaf removal.

Excellent efficiency levels against *B. cinerea* were obtained when combining leaf removal with the application of Regalis® or a botryticide (efficiency levels of more than 70 %, on average). Both strategies provide options to maximize wine quality in two ways – in terms of a reduction of fungal contamination of the crop and/or an improvement of grape maturity through a prolongation of the ripening period. However, in case of the combination of Regalis® and leaf removal, potential yield reductions have to be taken into consideration.

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