Received: 22 August 2020 | Accepted: 21 December 2020 | Published: 3 February 2021
DOI:10.20870/oeno-one.2021.55.1.4476

Metrics for studying berry growth kinetics in seedless grape cultivars (*Vitis vinifera* L.)

George Letchov[1], Venelin Roychev[2], and Neli Keranova[1]

[1]Department of Mathematics and Physics, Agricultural University, Plovdiv, Bulgaria
[2]Department of Viticulture, Agricultural University, Plovdiv, Bulgaria

[*]corresponding author: letchov@au-plovdiv.bg

ABSTRACT

The process related to the changes in dimensions and mass of grape berry passes through two growth phases separated by a lag phase, and can be described by a double sigmoid curve. The onset of the growth phases and their duration are important factors for understanding the growth processes in grape berries. A new method for their quantitative determination was developed in the present study. In this method, the phase transition dates correspond to the times at which the rate of change of the curvature of the logistic (sigmoid) curve reaches an extreme value. The method was tested on three seedless grape varieties, Sultanina, Ruby Seedless and Rusalka 3, and the changes in grape berry dimensions and mass were tracked from anthesis to harvest. For each of the varieties, a double logistic model of change in berry length, width and mass from anthesis to harvest was developed and the metrics of growth - beginning, stabilisation and end of growth - for each of the two phases were determined. It was found that the metrics in mass and berry dimensions do not match and shift relative to each other over time. A comparison of growth metrics with phenological metrics, such as anthesis, veraison and ripening, showed that phenophases cannot be used as a time scale to record the acceleration of growth processes, as they shifted in time with growth metrics. An exception was veraison, which coincided with the beginning of the accelerated growth of grapes during the second growth phase, following the lag phase. The time scale presented in the current research is a new tool for monitoring growth processes and could help clarify the links between visible changes in the grape berries and the ongoing processes within them. The developed method can also be used for the analysis of various growth processes that follow the logistic law.

KEYWORDS

grape berry, growth, phenology, growth metrics, double logistic model, sigmoid curve, seedless grape varieties, Sultanina, Ruby Seedless, Rusalka 3

Supplementary data can be downloaded through: https://oeno-one.eu/article/view/4476

INTRODUCTION

The process of grape berry development has been intensively studied to improve the quantity and quality of production. Grape berry growth is a dynamic process, which includes a complex sequence of molecular genetics and biochemical changes (Conde *et al.,* 2007; Roubelakis-Angelakis, 2009; Serrano *et al.*, 2017). Vast research has been carried out on the relationship between changes in dimensions (or mass) of berries and the processes shaping berry chemical composition. As a result of these studies, it has been found that grape berry development passes through two growth phases following the double logistic growth curve (Coombe, 1960; Harris *et al.,* 1968; Staudt *et al.,* 1986; Coombe and McCarthy, 2000; Dokoozlian and Christensen, 2000; Ollat *et al.,* 2002). The two growth periods are separated by a lag phase. The first phase begins at anthesis and is characterised by a high rate of cell division in the pericarp, the completion of nucellus and endosperm growth and a rapid increase in berry volume. During the first phase, the berries still have high levels of chlorophyll and accumulate organic acids (malic and tartaric acids, hydroxycinnamic). During the lag phase, berry growth rate decreases, the embryos develop rapidly and reach their maximum dimensions, and the level of auxin reaches a maximum. At the end of the lag phase, the berries begin to soften and to lose their chlorophyll. It was found that the lag phase can be displaced in time and that its length is variable (Harris *et al.,* 1968; Lavee, 1986), depending on genotype (Coombe, 1995), environmental factors and possible differences in growing conditions. Veraison launches the second growth phase when fruits continue to grow as a consequence of the enlargement of mesocarp cells. This last phase is characterised by softening and rapid changes in berry colour - from green to red or purple in the blue-black berried varieties and toyellowish in the white varieties - and by a further increase in berry volume, which initially grows very rapidly, but gradually slows down with the ripening of the fruit. Pigments and sugars accumulate while chlorophyll and organic acids break down.

The current study was mostly inspired by Coombe's research (Coombe, 1960), which provided an intuitive rationale behind the use of the double logistic curve for modelling the development of fleshy fruits, including grape, and relating the development and growth of fruit from several varieties to changes in their sugar, auxin, and gibberellin contents between anthesis and maturity. Over the years, various models have been developed to describe the change in grape berry dimensions or mass during the two growth periods. Fanizza and Colonna (1996) have developed a double logistic model to fit berry diameter of table grape varieties. A double logistic function model of berry mass has also been applied by Ollat and Gaudillère (1998) in a study on the effects of limiting leaf area on the development and composition of berries. Price *et al.* (2008) have developed a two-component mixture model based on normal distribution functions. Dai *et al.* (2009) applied a combination of monomolecular and logistic functions to analyse the dependencies between the function parameters and berry quality features. In a study on dry matter growth, García de Cortázar-Atauri *et al.* (2009) proposed a classical double logistic model based on thermal time (Growing Degree Days) and dry mass with two complementary dynamics: exponential and logistic growth. It has recently been shown that changes in berry density (Letchov and Roychev, 2017) and the development of grapevine bunch compactness (Tello and Forneck, 2018) and grape bunch mass (Ellis *et al.,* 2020) also follow the double logistic curve.

The accurate identification of the onset and duration of growth phases is important for producers and processors, as this can affect the quality and management of the grape crop. Tracking the growth of berries is usually done by observing phenological events such as budburst, anthesis, veraison and ripening. In the present study a double logistic model was applied to the timing of events in grape berry growth in terms of berry mass (M) and the berry dimensions length (L) and width (W). To describe growth, two simple sigmoids are considered in a single model, explaining two growth cycles. The sigmoid model is similar to Ollat and Gaudillère (1998) model, as its parameters have good biological relevance and interpretation (Thornley and Johnson, 2000).

A method based on the double logistic model has been developed for timing the main events in berry growth: beginning, stabilisation and end of growth for each of the two growth phases. The growth metrics are defined following the methodology of Zhang *et al.* (2001) and Zhang *et al.* (2003), known as the curvature change rate (CCR) method, and are calculated as day of year (DOY) when CCR of growth curve reaches minimum or maximum values. The CCR method was applied in remote sensing to study the land surface phenology of vegetation cover (Zeng *et al.,* 2020). In the present study, an approximation of the curvature of the logistic curve was developed, which allows basic growth metrics to be explicitly obtained, as simple formulas, providing new information about the growth process. The method was applied to define and determine the berry growth metrics of three table grape cultivars with stenospermocarpic seedlessness: Sultanina, Ruby Seedless and Rusalka 3. The growth metrics were compared to the phenological growth stages of the grapevine (*Vitis vinifera* L. ssp. *vinifera*).

MATERIALS AND METHODS

The data were collected in 2010 from the vineyards in the experimental fields of the Agricultural University of Plovdiv in Bulgaria (42.05N, 24.77E; 300 m asl), located at the foot of the Rhodope Mountains. Table grape cultivars with stenospermocarpic seedlessness were studied: Sultanina, Ruby Seedless and Rusalka 3 - part of the Ampelographic Collection of the Department of Viticulture at the University. The vines are grown on a Moser training system with a pruning load of 8 to 10 spurs with two winter buds and one cane with 11 winter buds. The vines are grafted onto a *Berlandieri* x *Riparia* SO4 rootstock. The plantation is 20 years old. The average plant density is 2600 vines/ha, the distance between rows is 3.20 m, and 1.20 m between vines, rows oriented north–south. The grape yield was, 7500 kg/ha for Sultanina, 18600 kg/ha for Ruby Seedless, and 12000 kg/ha for Rusalka 3.

In the vineyards, 30 vines of each variety were selected to study grape growth. During the year, every seven days from the emergence of flower buttons to harvest, five bunches of each studied cultivar were selected. Each cluster was conditionally divided into three longitudinal sectors (Harris *et al.,* 1968), with ten berries selected and picked from each sector for the purpose of measuring length (L), width (W) and fresh mass (M). The linear dimensions were measured with an accuracy of 0.01mm using electronic callipers (Black & Decker©), and the fresh mass with an accuracy of 0.001g using analytical scales (Bosch© SAE 200 and Bosch©-waagen seit 1852).

In parallel with the measurements described above, systematic phenological observations were carried out each week to record the following phenological growth stages: budding (BBCH code: 07 and 08), first leaf unfolded and spread away from shoot (code: 11), inflorescence emerge (code: 53), flowering (code: 61, 65, 69), ripening of berries (code: 81, 83, 85), berries ripe for harvest (code: 89)/ technological maturity (brix/acid ratio ≥ 25).

1. Double logistic model

Grape berry growth consists of two successive cycles, each with distinctive characteristics (Combe, 1992); therefore, berry growth was modeled using two growth functions, g_1 and g_2, which describe the first and second growth stages:

(1) $$G(t) = g_1(t; p_1) + g_2(t; p_2);$$

where p_1 and p_2 are the vectors of the parameters

During each phase, the growth parameters (berry fresh mass M, width W and length L) follow a sigmoid pattern, and therefore their change can be described by the logistic function (Thornley and Johnson, 2000; Tsoularis, 2001):

(2) $$g(t) = \frac{a.b}{a+(b-a).exp\,(-\mu.t)} + d;\ p = (a, b, \mu)$$

where μ is the specific growth rate at the beginning of the growth phase,

and $\mu = \frac{1}{g}\left(\frac{dg}{dt}\right)$ when $(g(t)/g_f) << 1$), $a = g(0)$

is the value of the growth function g(t) at the beginning of the growth phase and $b = g_f$ is the value of the growth function at the end of the growth phase; *d* is the accumulated biomass or the dimensions reached at the beginning of the first and second phases. It should be noted that we studied the growth function g(t) in terms of the growth parameters, grape berry fresh mass M and dimensions W and L. The growth rate reaches maximum value at the inflection point and it is exactly half the carrying capacity *b*:

(3) $$g_{inf} = \frac{1}{2}b,\ which\ occurs\ at\ t_{inf} = \frac{1}{\mu}ln\left(\frac{b-a}{a}\right)$$

The growth function parameters (*a, b, μ, d*) were estimated using a least-square method for each growth phase. The fit was assessed using the root mean square error (RMSE) and regression coefficient (R^2) between measured data and model estimates. "Lmfit" high-level interface to non-linear optimisation and curve fitting problems for Python was used to fit the logistics model (Equation 2) to the measured data (https://lmfit.github.io/lmfit-py/index.html).

2. Curvature change rate (CCR) method to identify growth metrics

After fitting the logistic model to the measured M, L and W, the curvature change rate (CCR) method was applied to identify the moments of onset and end of active growth, as well as the point of time when the growth processes became stable. In this method, the moments of acceleration and stabilisation of growth processes are defined as being when the CCR of the growth curve reaches extreme values (Zhang *et al.,* 2003): beginning of growth (t_0), stabilisation of growth (t_{inf}), and end of growth (t_f). The metrics t_0 and t_f are the moments at which the CCR of the logistic growth curve and the acceleration of growth reach maximum values; t_{inf} is the inflection point of the logistic curve at which time the CCR has a minimum value (Figure 1) and the growth proceeds at maximum rate. The metrics t_0, t_f and t_{inf} are defined for every growth phase (Figure 2). Here, we determine the extremes of CCR explicitly by applying an approximation to the curvature of a plane curve (see Supplementary data - Appendix 1). In this approximation, the curvature of the logistics curve can be calculated as the second derivative of the logistics function (Equation 2). Then CCR is defined as the first derivative of the curvature of the curve. The extremes of the CCR of the logistics curve give the growth metrics as:

(4a) $$t_0 = \frac{1}{\mu} ln\left[\left(5 - 2\sqrt{6}\right)\left(\frac{g_f - g_0}{g_0}\right)\right]$$

(4b) $$t_{inf} = \frac{1}{\mu} ln\left(\frac{g_f - g_0}{g_0}\right)$$

(4c) $$t_f = \frac{1}{\mu} ln\left[\left(5 + 2\sqrt{6}\right)\left(\frac{g_f - g_0}{g_0}\right)\right]$$

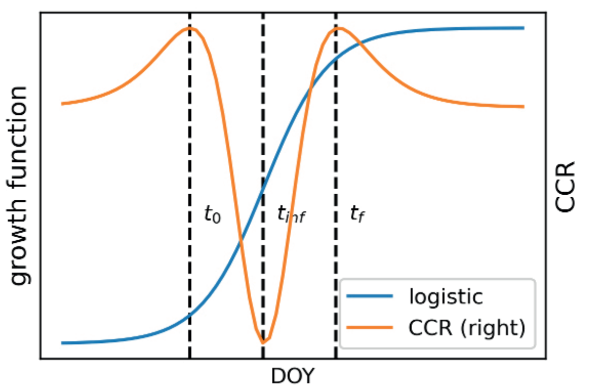

FIGURE 1. A schematic diagram showing how transition dates are calculated using minimum and maximum values in the rate of change in curvature (CCR) of growth function (logistic).

The vertical lines indicate transition dates: beginning of growth (t_0), stabilisation of growth (t_{inf}), and end of growth (t_f). DOY = day of year.

The equations (4a, 4b, 4c) give the main transition points of grape berry growth as functions of the berry dimensions (or mass) g_0 at the beginning of a phase, the maximum of berry dimensions (or mass) g_f, and the specific growth rate μ at the beginning of a growth cycle. As can be seen, equation (4b) coincides exactly with the inflection point of the logistic curve (see Equation 3). Equations (4a) and (4c) allow the length of the active growth period to be determined as follows:

(4d) $$\left(t_f - t_0\right) = \frac{1}{\mu} ln\left(\frac{5+2\sqrt{6}}{5-2\sqrt{6}}\right)$$

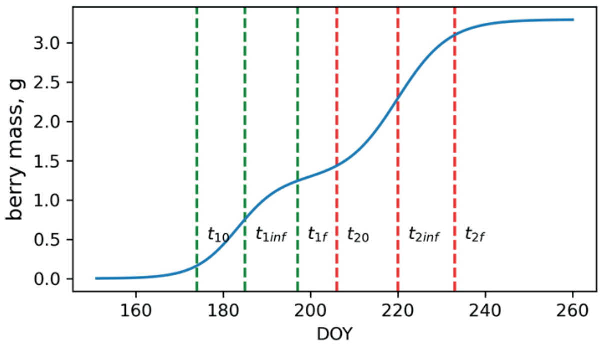

FIGURE 2. Double logistic model of berry mass and growth metrics during the first t_{10}, t_{1inf}, t_{1f} and second t_{20}, t_{2inf}, t_{2f} growth phases. Sultanina variety.

DOY = day of year.

Very often the logistic function is used in another form (Fanizza and Colonna, 1996; Tello and Forneck, 2018; Cao *et al.,* 2019; Ellis *et al.,* 2020). We have derived the growth metrics for this case as well (see Supplementary data - Appendix 2).

RESULTS

1. Phenology

The inflorescence began (BBCH code 53) around 20 April (Table 1). The beginning of anthesis was at the end of May and the beginning of June. It was earliest fo Sultanina (25 May) and it was 10 days late for Rusalka 3 (4 June). Full anthesis for all three varieties was in early June. The longest anthesis was for Ruby Seedless (14 days), while for Sultanina and Rusalka 3 it was about half as long (7 to 9 days).

Veraison began at the end of July (between 20 and 27 July) and continued 12 days for Ruby Seedless, 9 days for Sultanina and 8 days for Rusalka 3. It is noteworthy that for Rusalka 3, anthesis and veraison took place most rapidly, lasting for about a week, while for Ruby Seedless, these processes lasted about two weeks. It should be noted that the anthesis-veraison period was about twice as long as the veraison–ripening period for the Rusalka 3 variety.

TABLE 1. Timing of phenological phases of Sultanina, Ruby Seedless and Rusalka 3 grape vine varieties, Plovdiv region, Bulgaria, 2010.

Varieties	Inflorescence	Anthesis			Veraison			Ripening
	beginning	beginning	full	end	beginning	full	end	
Sultanina	23 April (113)	25 May (145)	30 May (150)	2 June (153)	20 July (201)	24 July (205)	28 July (209)	10 September (253)
Ruby Seedless	April 20 (110)	May 31 (151)	June 5 (156)	June 13 (164)	27 July (208)	3 August (215)	7 August (219)	29 September (272)
Rusalka 3	19 April (109)	4 June (155)	8 June (159)	10 June (161)	25 July (206)	28 July (209)	1st August (213)	26 August (241)

DOY in the brackets

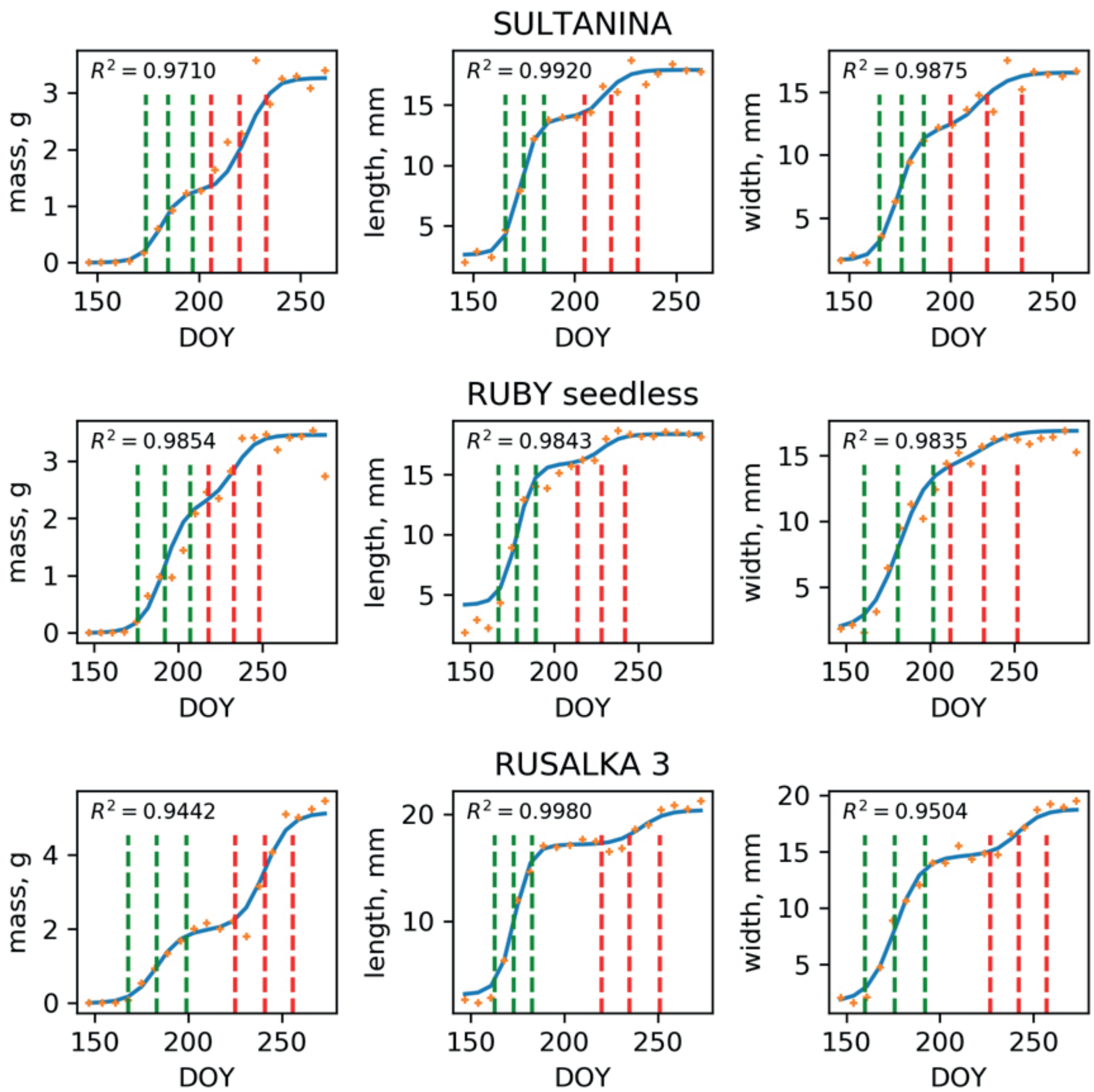

FIGURE 3. Double logistic model (solid lines) fitted to the measured (cross) mass (M), length (L) and width (W) values of grape berries.

Vertical lines = growth metrics during the first (green) and second (red) development phase. Sultanina, Ruby seedless and Rusalka 3 grape varieties. DOY = day of year. The coefficient of determination R^2 refers to the entire growth period.

TABLE 2: Logistic model parameters a, b, d and μ for the first and the second growth phases of grape berry length (L), width (W) and mass (M) development.

Sultanina						
Parameter	M_1	M_2	L_1	L_2	W_1	W_2
a	3.96E-07	1.92E-08	1.90E-06	1.68E-08	1.94E-05	3.73E-06
b	1.270	2.023	11.29	3.97	10.27	4.58
d	0.002	1.258	2.62	13.86	1.68	11.85
b+d	**1.272**	**3.281**	**13.91**	**17.83**	**11.95**	**16.43**
µ	0.207	0.170	0.243	0.180	0.202	0.132
SSE	0.008	0.656	0.330	4.801	0.669	7.697
R^2	0.999	0.969	1.000	0.948	0.999	0.934
RMSE	0.019	0.173	0.122	0.467	0.174	0.592
Ruby seedless						
Parameter	M_1	M_2	L_1	L_2	W_1	W_2
a	1.34E-05	1.04E-08	1.63E-05	9.42E-09	4.20E-03	2.85E-06
b	2.276	1.179	11.680	2.489	12.570	2.521
d	0.018	2.316	4.151	15.640	1.810	14.380
b+d	**2.294**	**3.495**	**15.831**	**18.129**	**14.380**	**16.901**
µ	0.149	0.152	0.202	0.166	0.114	0.113
SSE	0.160	0.286	4.241	1.426	0.987	0.479
R^2	0.995	0.981	0.996	0.964	0.986	0.992
RMSE	0.085	0.085	0.439	0.255	0.714	0.110
Rusalka 3						
Parameter	M_1	M_2	L_1	L_2	W_1	W_2
a	3.67E-05	9.36E-09	1.02E-05	9.88E-09	1.19E-03	7.77E-09
b	1.983	3.151	14.06	1.33	12.95	4.11
d	0.003	1.986	3.11	17.23	1.70	14.77
b+d	**1.986**	**5.137**	**17.17**	**18.56**	**14.65**	**18.88**
µ	0.151	0.152	0.227	0.151	0.143	0.154
SSE	0.039	1.977	1.623	1.330	4.035	4.860
R^2	0.998	0.959	0.999	0.974	0.995	0.942
RMSE	0.041	0.300	0.272	0.246	0.428	0.470

Grape varieties are Sultanina, Ruby and Rusalka 3. Indices 1 and 2 refer to the first and second growth phases.
SSE = sum of squares due to error, R^2 = coefficient of determination, RMSE = Root mean squared error

TABLE 3. Measured grape berry mass, length and width at the end of the growth period.

Measured	Sultanina	Ruby seedless	Rusalka 3
Mass (M), g	3.3975 ± 0.7332	3.5278 ± 0.3235	5.4493 ± 0.3214
Length (L), mm	17.70 ± 0.75	18.50 ± 0.74	20.49 ± 0.98
Width (W), mm	16.52 ± 1.37	16.91 ± 0.39	19.52 ± 0.57

Sultanina, Ruby seedless and Rusalka 3 grape varieties. Data are mean ± standard deviation (SD).

2. Double logistic model of berry growth

The mass and linear dimensions (length and width) of the berries in all three grape varieties, Sultanina, Ruby Seedless and Rusalka 3, followed the double logistic growth pattern throughout the growing season (Figure 3), with the coefficient of determination R^2 being between 0.944 and 0.998. The fitting parameters of the logistic models g_1 (first phase) and g_2 (second phase) are presented in Table 2. During each of the two phases, grape berry length, width and mass followed the logistic model with very high coefficients of determination values and low SSE and RMSE. The logistic curve had a better fit for measured sizes and mass during the first growth phase; this is due to the larger differences between the measured and model values of grape size and weight at the end of growth when they reached maturity. One of the possible reasons for this is that the berries begin to dry after reaching maturity, which leads to a reduction in their size and mass.

The parameters $p = (a, b, \mu)$ of the growth models g_1 and g_2 provided valuable information about the growth processes during the first and second growth phases of the grape berries. For both processes, parameter *a* was close to zero, since it gives the biomass produced (or dimensions reached) at the beginning of the growth process. It should be noted that this is the new biomass (dimensions) produced at the beginning of a given phase, and differs from the available biomass (dimensions) resulting from previous phases defined by the parameter *d*. For the first phase, $d_1 \approx 0$, as the accumulated biomass (or dimensions) during anthesis, was not taken into account in the present study. For the second phase, $d_2 = b_{1f} + d_1 \approx b_1$, that is, d_2 represents the available biomass (or dimensions reached) at the end of the first growth phase. As can be seen in Tables 2 and 3, the estimates for M, L and W of grape berries given by the double logistic model are very close to the measured values. For both Sultanina and Rusalka 3 varieties, 39 % of biomass accumulated during the first growth phase, while for Ruby seedless variety, 65 % of the total biomass accumulated. For all three varieties, most of the increase in the linear dimensions L and W occurred during the first growth phase: 63 % of the final length L and width W of the berries for Sultanina, 64.4 % of L and 75 % of W for Ruby Seedless, and 76 % of L and 69 % of W for Rusalka 3.

At the beginning of the first phase, for all three grape varieties, the specific rate of biomass accumulation μ_{1M} varied between 0.15 and 0.21 per day, the specific rate of elongation $\mu_{1L} \in [0.20, 0.24]$ per day, and the specific rate of widening of the berries $\mu_{1W} \in [0.11, 0.20]$ per day. At the beginning of the second growth phase, μ_{2M} varied between 0.15 and 0.17 per day, $\mu_{2L} \in [0.15, 0.18]$ per day, and $\mu_{2W} \in [0.11, 0.15]$ per day. For Sultanina, μ_{1M} was 18 % greater than μ_{2M}, μ_{1L} was 26 % greater than μ_{2L}, and μ_{1W} was 18 % greater than μ_{2W}. For the Ruby Seedless and Rusalka 3 varieties, the specific growth rates at the beginning of the two phases were almost the same in mass and width: $\mu_{1M} \approx \mu_{2M}$ and $\mu_{1W} \approx \mu_{2W}$. The specific rate of lengthening of the berries at the beginning of the first phase was higher than that at the beginning of the second phase for all three varieties; *i.e.*, $\mu_{1L} > \mu_{2L}$, with μ_{1L} being greater than μ_{2L} by 18 % for the Ruby Seedless variety, 26 % for Sultanina and 34 % for Rusalka 3.

3. Growth metrics

Table 4 gives the growth metrics during the first and second phases of grape berry development: t_0 (beginning of active growth), t_{inf} (moment at which growth stabilises) and t_f (end of active growth), defined by the CCR method.

3.1. First growth phase

3.1.1. t_0 (beginning of active growth)

The increase in linear dimensions, L and W, of the berries began one to two weeks earlier than the start of active biomass production, so that by the end of anthesis there was a slight decrease in the density of the berries. For Sultanina, the onset of the active increase in linear dimensions began 12 days after anthesis had ended (in early June), and the active accumulation of biomass began about three weeks after anthesis. For Ruby Seedless and Rusalka 3, the increase in linear dimensions accelerated at the end of anthesis, while biomass accumulation was delayed by about a week.

3.1.2. t_{inf} (moment when growth stabilised at a constant rate)

The changes in grape mass, length and width stabilised earlier for the Sultanina and Rusalka 3 varieties, with $t_{inf} \approx 178$ DOY (26 June), while for the Ruby Seedless variety, growth was delayed by about a week. The stabilisation of the biomass accumulation rate was delayed by about a week compared to the stabilisation of berry growth in width and by about 11 days in length. It should be noted that the stabilisation of growth occurred in the middle of the anthesis-veraison period; *i.e.*, late June and early July.

TABLE 4. Growth metrics t_0, t_{inf}, t_f and Δ of grape berries mass M, length L and width W during the first and second phases in days of the year (DOY).

Sultanina						
Metric	M_1	M_2	L_1	L_2	W_1	W_2
t_0	172	206	166	205	165	200
t_{inf}	183	220	175	218	176	218
t_f	194	233	185	231	187	235
Δ	22	27	19	26	22	35
length of lag phase	12		20		13	
Ruby seedless						
Metric	M_1	M_2	L_1	L_2	W_1	W_2
t_0	176	218	167	214	161	212
t_{inf}	192	233	178	228	181	232
t_f	207	248	189	242	202	252
Δ	31	30	22	28	41	40
length of lag phase	11		25		10	
Rusalka 3						
Metric	M_1	M_2	L_1	L_2	W_1	W_2
t_0	168	225	163	220	160	227
t_{inf}	183	241	173	235	176	242
t_f	199	256	183	251	192	257
Δ	31	31	20	31	32	30
length of lag phase	26		37		35	

Sultanina, Ruby Seedless and Rusalka 3 varieties. Indices 1 and 2 refer to the first and second growth phases.

3.1.3. t_f (end of active growth)

At the end of the first phase, the growth processes rapidly slowed down and growth entered the lag phase. The accumulation of biomass in the first phase subsided about two weeks earlier than the elongation of the berries, and one week earlier than the widening; *i.e.*, a slight densification of the berries was observed before veraison. In the Ruby seedless variety, the end of the first phase shifted one to two weeks later compared to the other two varieties. In Sultanina, the increase in berry width and length ended two weeks before veraison in early July, and the significant delay in biomass accumulation began one week before veraison. In Ruby seedless, the attenuation of growth processes began 19 days before veraison with a decrease in the rate of grape berry elongation. Growth in width ended one week before veraison, and the rate of biomass accumulation slowed down noticeably at the beginning of veraison. For the Rusalka 3 variety, the accelerated slowing down of growth processes began first with respect to the length of the berries, 23 days before the onset of veraison. Two weeks before veraison; berry widening and biomass accumulation in the berries slowed down a week before veraison.

3.1.4. $\Delta_1 = (t_f - t_0)$ (duration of first phase)

For all three varieties studied, the anthesis-veraison phase lasted one and a half months (45 days). The period of active growth Δ_1 was shorter than the duration of the anthesis-veraison period; this is because active growth did not begin immediately after the end of anthesis and it ended before veraison. The Sultanina variety had the shortest growth period, which lasted from 19 to 22 days (L, W and M); *i.e.*, for half of the anthesis-veraison period. In the Ruby Seedless and Rusalka 3 varieties, the longest growth was in W (41 days and 32 days respectively) and biomass accumulation M lasted one month. The first phase was the cell division phase throughout mid July. Cell division and cell enlargement both contributed to pericarp growth in the early post-anthesis phase. Cell division in the grape pericarp began 5 to 10 days before anthesis and continued for approximately 25 days (Harris *et al.,* 1968).

3.2. Lag phase

The duration of the lag phase (defined by growth metrics as the period between the end of the first growth phase t_{1f} and the beginning of the second growth phase t_{20}) is presented in Table 3.

For all three varieties, the duration of the lag phase for berry mass Δ_M and width Δ_W was shorter than that for berry length. For Sultanina and Ruby Seedless, the deceleration of berry growth in terms of mass and width lasted for less than two weeks (10 to 13 days), while the deceleration in the lengthening of the berries lasted for 3 weeks (20 to 25 days). For the Rusalka 3 variety, Δ_L and Δ_W lasted for approximately 35 days, while Δ_M lasted for 26 days. The slowing down of the growth processes was greater with respect to L and W compared to that of berry mass M. During the lag phase, berry mass increased almost twice as much as growth in L and W, which led to some compaction of the berries. During the first phase, accumulated mass was 24 % for Rusalka 3, approximately 22 % for Sultanina, and 14 % for Ruby seedless. At the end of the first phase, Sultanina and Ruby seedless had increased in length by 9 % and Rusalka 3 by 11 %. The growth in width W during the lag phase was smallest for the Ruby Seedless variety with an increase in berry width during the first phase of 11 %, while for Rusalka 3 and Sultanina it was 12 and 14 % respectively.

3.3. Second growth phase

3.3.1. t_0 (beginning of active growth)

The onset of active development of Sultanina and Ruby Seedless berries during the second phase began during veraison, with active growth beginning earliest in terms of W, followed by L and then M. For Sultanina, accelerated growth in M and L began during full veraison, while W development accelerated at the beginning of veraison. For Ruby seedless, W and L growth began during full veraison, while accelerated biomass production began at the end of veraison. For the Rusalka 3 variety, the growth processes accelerated after veraison. Berry elongation started one week after veraison, while widening and biomass accumulation began two weeks after veraison. In the second phase, the accelerated growth of Sultanina berries began the earliest at the end of July, followed by Ruby Seedless in early August and Rusalka 3 in late August. For Ruby Seedless, accelerated biomass accumulation began about a week later than the accelerated development of the linear dimensions of the berries. Acceleration was delayed by almost a week for Rusalka 3 (5 days) and for Sultanina (6 days) in terms of length and width respectively.

3.3.2. t_{inf} (moment of the most rapid growth (stabilisation of growth))

During the second phase, the growth processes of the Sultanina variety stabilised the earliest at the beginning of August (218 to 220 DOY), while those of the Ruby Seedless variety stabilised in the middle of August. For the varieties Sultanina and Ruby Seedless, growth stabilisation occurred before the middle of the veraison-maturity period, while for Rusalka 3 it was delayed by 10 to 20 days and was reached at the end of August, shortly before this variety reached technological maturity.

3.3.3. t_f (end of active growth)

The end of the second phase (t_{2f}) - marked by accelerated attenuation of growth processes due mainly to the mechanisms of cell size enlargement - occurred the earliest for the Sultanina variety at the end of August, for Ruby Seedless at the beginning of September, and for Rusalka 3 in the first half of September.

3.3.4. $\Delta_2 = (t_f - t_0)$ (duration of the second phase)

The duration of biomass accumulation in the second phase was about one month (30 days), while the growth period in length was the shortest for the Sultanina variety (25 days); for Ruby Seedless it lasted 28 days and for Rusalka 3 it lasted 30 days. In the second phase, the widening period varied, being the shortest for the Rusalka 3 variety (30 days), while Sultanina lasted 35 days and Ruby Seedless 41 days. For the Sultanina variety, the period of active development of the berries began during veraison and ended three weeks before the onset of technological maturity. Because the veraison-ripening period lasted 44 days, it was about 1.5 to 1.7 times longer than the active growth duration Δ_2. In Ruby seedless, the growth processes began immediately after the end of veraison and ended three weeks before technological maturity. The veraison-maturity period lasted 53 days, or about twice as long as the M and L and 1.3 times longer than the W growth period. For the Rusalka 3 variety, the growth processes in the second phase began one week for L and two weeks for M and W after the end of veraison, and ended two weeks after reaching maturity. The duration of the second phase was 30 days and was longer than the period veraison-technological maturity (23 days).

This anomaly is most probably due to the fact that harvest was carried out before reaching technological maturity.

3.4. Comparison of the two growth phases. Average growth rate.

For all three varieties, the duration of the first phase Δ_1 was less than or equal to the duration of the second phase Δ_2. For Sultanina, $\Delta_1 = 0.74\Delta_2$, and for the other two varieties $\Delta_1 \approx 0.9\Delta_2$. The duration of the two phases was almost the same in terms of biomass accumulation and widening; however, the berry lengthening process was longer in the second phase and $\Delta_{1L} \approx 0.74\Delta_{2L}$.

Defining growth metrics allows us to determine the average growth rate during a given phase. During the first phase, the process of biomass accumulation in the berries proceeded at an average rate of 66 mg/day for the three varieties, the process being the fastest in Rusalka 3 at 74 mg/day and slowest in Sultanina at 54 mg/day; meanwhile, the rate was 65 mg/day in Ruby Seedless (Figure 4). Biomass accumulation rates during the second phase of Sultanina and Ruby Seedless were higher: 75 and 104 mg/day respectively. For the Rusalka 3 variety, the rate of biomass growth during the second phase was about half that of the first phase (only 39 mg/day).

The average rates of increase in linear dimensions in the first phase were significantly higher (3 to 6 times) than the growth rates during the second phase. The average lengthening rates were 0.60 mm/day for the first phase and 0.12 mm/day for the second growth phase. The average growth rates in terms of width were 0.11 mm/day in the first phase and 0.39 mm/day in the second phase. Growth processes during the lag phase slowed down, but did not stop, as the rate of biomass accumulation in the berries varied from 0.017 g/day for Rusalka 3 to 0.024 g/day for Sultanina and Ruby Seedless. The slowing down in growth in terms of length and width during the lag phase was significant compared to the first phase. For the Sultanina and Ruby Seedless varieties, the lengthening was about 0.06 mm/day and the width increased by 0.09 mm/day. For the Rusalka 3 variety, the growth rates of L and W during the lag phase were the same (0.04 mm/day). The rates of accumulation of biomass during the first phase was 2.4 (Sultanina), 2.8 (Ruby Seedless) and 4.5 (Rusalka 3) times higher than those during the lag phase. The processes of lengthening and widening of the berries during the first phase were about 10 times and 5 to 7 times faster respectively. After the lag phase, there was a 2-fold (Rusalka 3), 3-fold (Sultanina) and 4.5-fold (Ruby Seedless) increase in biomass accumulation rate, while the lengthening was almost twice as fast, and the growth rate in terms of W was 1.5 times faster.

DISCUSSION

Over the years, several logistic growth models have been proposed to describe the growth of individual plants, such as the logistic and Gompertz models (Thornley and Johnson, 2000; Tjørve and Tjørve, 2010; Paine *et al.,* 2012). As we have seen, the double logistic model describes grape berry growth processes in terms of M, L and W well. The development of a double logistic model is associated with some difficulties when fitting the sigmoid curve to the measured berry dimensions and mass; therefore, in the present study a detailed method is given for developing and applying a double logistic model to describe growth processes. When applying growth functions in plant and crop modelling,

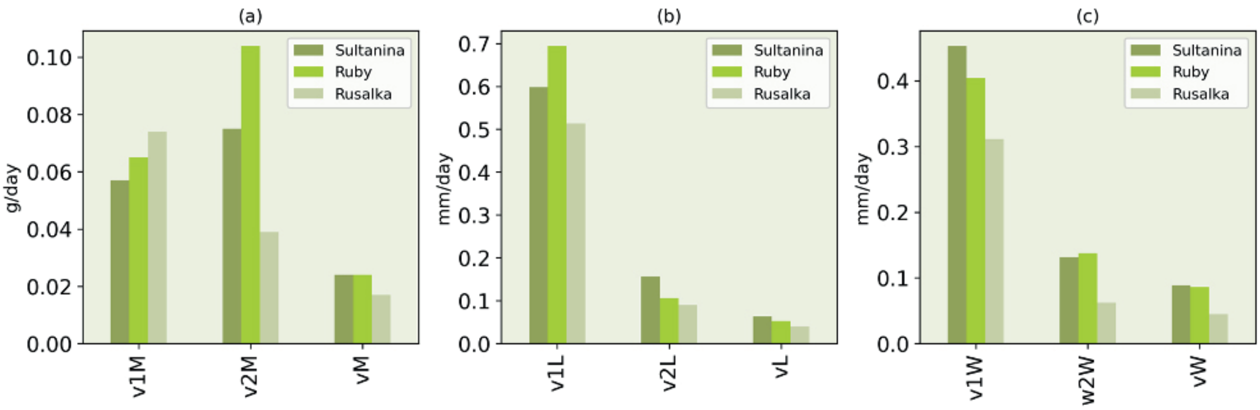

FIGURE 4. Average growth rates of the berries in the first phase (v1), second phase (v2) and lag phase (v).
(a) vM : g/day = berry mass rate, (b) vL : mm/day = berry length rate; (c) vW : mm/day = berry width rate.

two basic criteria must be satisfied (Thornley and Johnson, 2000): first, the model should derive from a differential equation for the growth process and, second, the parameters in this equation should be biologically meaningful. We hence preferred to work with the logistics function in the form of the equation (2), with parameters satisfying these conditions. Thus, the values of the model parameters obtained in the fitting process were biologically meaningful and allowed us to set realistic initial values to accelerate the convergence of the fitting procedure.

In addition to describing growth well and in detail, the double logistic model allows new metrics to be introduced for tracking the growth of grapes. The developed method - based on the rate of curvature change of the logistic curve - allows the onset of important events in grape berry growth to be explicitly defined. The events in traditional phenology register the visible changes in the berry, while the growth metrics are related to the kinematics of the ongoing processes in the berry, rate and acceleration of the growth processes. Over the years, berry growth has been monitored by examining changes to either one of the dimensions of the berries or berry mass. Naturally, the question arises as to which extensive property (*i.e.*, dimensions or mass) should be used to describe growth. The results of the present study show that the metrics t_0, t_{inf}, t_f shift depending on which physical property of the berries is used (M, L, or W), and the differences between the growth metrics - depending on the property against which they are determined - are greater in the first phase than in the second phase. The onset of the first L and W growth phase for the Sultanina variety began two weeks after the end of anthesis, and the acceleration of M growth began three weeks after anthesis. For Ruby Seedless, the acceleration of L and W growth began at the end of anthesis, while the acceleration of M was delayed by two weeks. In Rusalka 3, the beginning of L and W growth began at the end of anthesis, and M was delayed by about a week. The beginning of the second phase for the Sultanina and Ruby Seedless began during full veraison. For the Rusalka 3 variety, the second phase of L growth began one week after the end of veraison, while active M and W growth was delayed by about two weeks. For Sultanina and Ruby Seedless varieties, berry growth ended 3 to 4 weeks before technological maturity. For the Rusalka 3 variety, growth continued for 2 to 3 weeks after reaching technological maturity. All these differences in the timing of growth metrics were most likely due to the fact that the reasons for these changes differed depending on the physical property that was changed. The shift in time of the growth metrics in relation to phenological events raises the question of interdependence between the visible changes in the berries and the kinematics of the growth processes taking place in them.

When comparing the two types of metrics, phenological and growth metrics, it should be borne in mind that phenological observations were made weekly, which affected the accuracy of these estimates. Therefore, in future studies on the relationship between the two types of metrics, phenological observations and measurements of grape berry mass and dimensions should be performed more frequently, and a more detailed phenological scale should be used (*e.g.*, the BBCH scale; Lorenz *et al.,* 1995).

The value of the specific growth rate μ is an important parameter related to the length Δ of a given phase. The explicit determination of growth metrics (see Equations 4a,b,c) shows that the length of the growth period ($t_f - t_0$) depends on the growth rate μ (Equation 4d), provided there is no external influence during growth: *i.e.*, the higher the rate, the shorter the length of the given phase. The specific growth rates of the mass μ_M, the length μ_L and the width μ_W of the berries at the beginning of the first phase μ_1 were greater than or almost equal to those of the second phase μ_2 for all three varieties. Therefore, the length of growth in the first phase was less than or equal to the length of growth in the second phase.

In general, the duration of growth in a given phase also depends on external factors, such as precipitation and temperature. Therefore, taking into account the relationship between the length of phase Δ and μ, we can assume that μ depends on external factors; nevertheless, this assumption should be further explored.

CONCLUSION

Grapevine phenology observations and measurements of grape berry dimensions and mass of three seedless varieties of *Vitis vinifera,* Sultanina, Ruby seedless and Rusalka 3 were made from anthesis to maturity. The fruit of all three varieties showed a double-logistic growth curve. Here, we used the logistic model in its classical form for the model parameters to have a clear biological meaning and interpretation. The double logistic model allows a new scale for timing growth stages to be introduced, which is different from the traditional phenological scale. The new scale is strictly quantitative and is related to the kinematics of growth processes, their rate and acceleration. The onset and length of the two phases were determined by applying an approximation to the curvature of the logistic curve. We found that growth metrics shifted over time relative to traditional phenophases and could be defined as the moments at which the rate of change of the curvature of the logistic growth curve reached extreme values (maximum and minimum). This raises current questions about, for example, the factors that lead to a change in growth rate, and what happens to the structure and composition of the berries during the moments of maximum acceleration of growth. The time scale introduced in the current research for recording important events during the growth of grapes is a new tool for monitoring growth processes and could help clarify the links between visible changes to grape berries and the ongoing processes within them. The developed method can also be used for the analysis of various growth processes that follow the logistic law.

REFERENCES

Cao, L., Shi, P.-J., Li, L., & Chen Guifen, G. (2019). A New Flexible Sigmoidal Growth Model. *Symmetry, 11*, 204. https://doi.org/10.3390/sym11020204

Conde, C ., Silva, P., Fontes, N., Dias, A. C. P., Tavares, R. M., Sousa, M. J., Agasse, A., Delrot, S. & Gerós, H. (2007). Biochemical changes throughout grape berry development and fruit and wine quality. *Food, 1*(1), 1–22. http://hdl.handle.net/1822/6820

Coombe, B. (1960). Relationship of Growth and Development to Changes in Sugars, Auxins, and Gibberellins in Fruit of Seeded and Seedless Varieties of Vitis vinifera. *Plant Physiology, 35*(2), 241–250. https://doi.org/10.1104/pp.35.2.241

Coombe , B. G. (1992). Research on development and ripening of the grape berry. *American Journal of Enology and Viticulture, 43*(1), 101-110. https://www.ajevonline.org/content/43/1/101

Coombe, B. (1995). Growth Stages of the Grapevine: Adoption of a system for identifying grapevine growth stages. *Australian Journal of Grape and Wine Research, 1*(2), 104–110. https://doi.org/10.1111/j.1755-0238.1995.tb00086.x

Coombe, B., & McCarthy, M. (2000). The dynamics of grape berry growth and physiology of ripening. *Australian Journal of Grape and Wine Research, 6*, 131–135. https://doi.org/10.1111/j.1755-0238.2000.tb00171.x

Dai, Z., Génard, M., Li, S., & Vivin, P. (2009). Analyzing the functional association among seed traits, berry growth and chemical composition in Cabernet-Sauvignon berry (*Vitis vinifera* L.) using a mathematical growth function. *Journal International Des Sciences de La Vigne et Du Vin, 43*, 35–44. https://doi.org/10.20870/oeno-one.2009.43.1.807

Dokoozlian, N., & Christensen, L. P. (2000). *Grape Berry Growth and Development. In Raisin Production Manual* (pp. 30–37). University of California. Agricultural and Natural Resources, Communication Services, Publication 3393: Oakland, CA, USA. http://iv.ucdavis.edu/files/24467.pdf

Ellis, R., Moltchanova, E., Gerhard, D., Trought, M., & Yang, L. (2020). Using Bayesian growth models to predict grape yield. *OENO One, 54*(3), 443–453. https://doi.org/10.20870/oeno-one.2020.54.3.2972

Fanizza, G., & Colonna, G. (1996). A mathematical function to study berry growth variability in grape (*Vitis vinifera*). *Journal of Wine Research, 7*(2), 117–123. https://doi.org/10.1080/09571269608718072

García de Cortázar-Atauri, I., Brisson, N., Ollat, N., Jacquet, O., & Payan, J.-C. (2009). Asynchronous dynamics of grapevine ("*Vitis vinifera*") maturation: experimental study for a modelling approach. *OENO One, 43*(2), 83–97. https://doi.org/10.20870/oeno-one.2009.43.2.801

Harris, J., Kriedemann, P., & Possingham, J. (1968). Anatomical aspects of grape berry development. *Vitis, 7*, 106–109. https://doi.org/10.5073/vitis.1968.7.106-119

Lavee, S. (1986). Grape. In Monselise, S. P. *CRC Handbook of Fruit Set and Development* (1st ed.). CRC Press. https://doi.org/10.1201/9781351073042

Letchov, G., & Roychev, V. (2017). Growth kinetics of grape berry density (*Vitis vinifera* L. 'Black Corinth'). *Vitis, 56*, 155–159. https://doi.org/10.5073/vitis.2017.56.155-159

Lorenz, D., Eichhorn, K., Bleiholder, H., Klose, R., Meier, U., & Weber, E. (1995). Growth Stages of the Grapevine: Phenological growth stages of the grapevine (*Vitis vinifera* L. ssp. vinifera)—Codes and descriptions according to the extended BBCH scale. *Australian Journal of Grape and Wine Research, 1*(2), 100–103. https://doi.org/10.1111/j.1755-0238.1995.tb00085.x

Ollat, N., Diakou-Verdin, P., Carde, J., Barrieu, F., Gaudillère, J., & Moing, A. (2002). Grape berry development: a review. *Journal International Des Sciences de La Vigne et Du Vin, 36*, 109–131. https://doi.org/10.20870/oeno-one.2002.36.3.970

Ollat, N., & Gaudillère, J. (1998). The effect of limiting leaf area during stage I of berry growth on development and composition of berries of *Vitis vinifera* L., cv. Cabernet Sauvignon. *American Journal of Enology and Viticulture, 49*(3), 251–258. https://www.ajevonline.org/content/49/3/251

Paine, C. E. T., Marthews, T. R., Vogt, D. R., Purves, D., Rees, M., Hector, A., & Turnbull, L. A. (2012). How to fit nonlinear plant growth models and calculate growth rates: an update for ecologists. *Methods in Ecology and Evolution, 3*(2), 245–256. https://doi.org/10.1111/j.2041-210X.2011.00155.

Price, W., Shafii, B., Blom, P., Tarara, J., Dokoozlian, N., & Sanchez, L. (2008). *Modeling seasonal wine grape development using a mixture technique* (G. L. Gadbury, Ed.). New Prairie Press, Kansas State University. https://doi.org/10.4148/2475-7772.1098

Roubelakis-Angelakis, K. (2009). *Grapevine Molecular Physiology and Biotechnology*. Dordrecht, The Netherlands: Springer Science+Business Media B.V. https://doi.org/10.1007/978-90-481-2305-6

Serrano, A., Espinoza, C., Armijo, G., Inostroza-Blancheteau, C., Poblete, E., Meyer-Regueiro, C., Arce, A., Parada, F., Santibáñez, C., & Arce-Johnson, P. (2017). Regulatory Networks Associated with Endogenous Processes and Environmental Responses. *Frontiers in Plant Science, 8*, 1486. https://doi.org/10.3389/fpls.2017.01486

Staudt, G., Schneider, W., & Leidel, J. (1986). Phases of berry growth in *Vitis vinifera*. *Annals of Botany, 58*, 789–800. https://doi.org/10.1093/oxfordjournals.aob.a087261

Tello, J., & Forneck, A. (2018). A double-sigmoid model for grapevine bunch compactness development. *OENO One, 52*(4), 307–316. https://doi.org/10.20870/oeno-one.2018.52.4.2132

Thornley, J., & Johnson, I. (2000). *Plant and Crop Modelling: A Mathematical Approach to Plant and Crop Physiology*. Caldwell, New Jersey: Blackburn Press.

Tjørve, E., & Tjørve, K. M. C. (2010). A unified approach to the Richards-model family for use in growth analyses: Why we need only two model forms. *Journal of Theoretical Biology, 267*, 417–425. https://doi.org/10.1016/j.jtbi.2010.09.008.

Tsoularis, A. (2001). Analysis of Logistic Growth Models. *Research Letters in the Information and Mathematical Sciences, 2*, 23–46. https://mro.massey.ac.nz/handle/10179/4341?show=full

Zeng, L., Wardlow, B., Xiang, D., Hu, S., & Li, D. (2020). A review of vegetation phenological metrics extraction using time-series, multispectral satellite data. *Remote Sensing of Environment, 237*, 111511. https://doi.org/10.1016/j.rse.2019.111511

Zhang, X., Friedl, M., Schaaf, C., Strahler, A., Hodges, J., Gao, F., Reed, B., & Huete, A. (2003). Monitoring vegetation phenology using MODIS. *Remote Sensing of Environment, 84*, 471–475. https://doi.org/10.1016/s0034-4257(02)00135-9

Zhang, X., Hodges, J., Schaaf, C., Friedl, M., Strahler, A., & Gao, F. (2001). *Global Vegetation Phenology from AVHRR and MODIS Data. IGARSS 2001. Scanning the Present and Resolving the Future. Proceedings. IEEE 2001 International Geoscience and Remote Sensing Symposium* (Cat. No.01CH37217), 2262–2264. https://doi.org/10.1109/IGARSS.2001.977969.