

Respiration Rate of Blueberry in Modified Atmosphere at Various Temperatures

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Abstract. Respiration rates of three blueberry (*Vaccinium corymbosum* L.) cultivars (Coville, Blueray, and Jersey) were measured as a function of O₂, CO₂, and temperature using the closed system method. The data conformed well to a recently proposed Michaelis-Menten type respiration (r) model ($r = V_m [O_2] / \{K_m + (1 + [CO_2]/K_i)[O_2]\}$), where V_m was maximum respiration rate, K_m was Michaelis-Menten constant, and K_i was inhibition constant. The model predictions were used to examine the behavior of respiration rates. In general, the respiration rates decreased with increasing CO₂, but were little affected by changes in O₂. 'Coville' blueberries had the highest respiration rate, followed by 'Blueray' and 'Jersey'. The temperature dependence of the respiration rates followed the Arrhenius relationship.

The major causes of spoilage for blueberries are fungal decay and physiological changes (Cappellini et al., 1982). Ceponis et al. (1983) and Smittle et al. (1988) have shown that storing blueberries in a CO₂-enriched atmosphere is a fairly effective way to inhibit postharvest decay without fungicidal treatment. Therefore, controlled atmosphere storage (CA) or modified atmosphere packaging (MAP) possibly can serve as an alternative to fungicides to extend the shelf life of fresh blueberries.

Both CA and MAP use modified atmosphere (usually reduced O₂ and elevated CO₂ levels) to reduce respiration rates and retard softening and physiological changes in the product. MAP has the advantage of not requiring the expensive equipment that is used by CA for regulating the gas composition. To maintain constant gas composition in the package, MAP relies on matching the respiration of the product with the permeation rates of the package. The design of MAP requires knowledge of the product (optimum gas concentrations and respiration rates), the package (surface area, film thickness, and film permeability), and the environmental factors (storage temperature and relative humidity). The optimum ranges of O₂ and CO₂ concentrations can be determined using chemical analysis and/or sensory analysis once the quality attributes of the product are defined. The respiration rates can be measured using the closed system method described by Haggard et al. (1992). After the optimum gas concentrations, respiration rates, and storage conditions are known, the package requirements can be estimated from model predictions (Hayakawa et al. 1975; Lee et al., 1992).

However, the development of MAP of blueberry is impeded by the lack of understanding of the respiration of the fruit. The first objective of this work was to study the respiration rates of three blueberry cultivars as a function of O₂, CO₂, and temperature. Recently, Lee et al. (1992) developed and verified a Mi-

Table 1. Estimated values for V_m, K_m, and K_i at 5, 15, and 25C using multiple linear regression of Eq. [6]. r² is the coefficient of determination.²

Cultivars	Temp (°C)	V _m	K _m	K _i	r ²
<i>Parameters for O₂ consumption curve</i>					
Coville	5	16.602	1.488	7.417	0.996
	15	68.006	0.444	2.914	0.991
	25	127.356	5.200	6.684	0.990
Blueray	5	11.802	1.593	11.725	0.998
	15	34.670	0.130	6.783	0.999
	25	76.247	0.100	11.044	0.999
Jersey	5	9.863	2.106	7.606	0.994
	15	35.868	0.678	3.296	0.996
	25	51.285	0.411	9.361	0.996
<i>Parameters for CO₂ evolution curve</i>					
Coville	5	12.539	0.429	15.486	0.998
	15	51.046	0.177	4.896	0.995
	25	99.032	0.520	13.502	0.996
Blueray	5	8.956	0.705	19.648	0.999
	15	30.203	- 0.104	9.441	0.999
	25	76.049	0.125	19.057	0.997
Jersey	5	7.347	0.797	12.693	0.995
	15	29.942	0.784	4.449	0.998
	25	48.234	0.101	16.701	0.999

¹V_m = maximum respiration rate; K_m = Michaelis-Menten constant; K_i = inhibition constant.

chaelis-Menten enzyme kinetics type respiration model for predicting respiration rate of fresh fruit and vegetables as a function of O₂ and CO₂. The second objective was to test the applicability of this model to the blueberry respiration data obtained from this work.

Materials and Methods

Sample preparation. Fruits of 'Coville', 'Blueray', and 'Jersey' highbush blueberry (*Vaccinium corymbosum* L.) were harvested from the New Jersey Blueberry & Cranberry Research Center, Chatsworth, N.J. The blueberries were picked in early July when ≈ 90% of the berry surface was blue with ≈ 10% pink around the scar (Windus et al., 1976) The average weights per berry were 1.61, 1.57, and 1.67 g, and the average widths were 15.3, 15.0, and 15.8 mm for 'Coville', 'Blueray', and 'Jersey', respectively. Sample temperatures were equilibrated for 3 h before the experiment was started.

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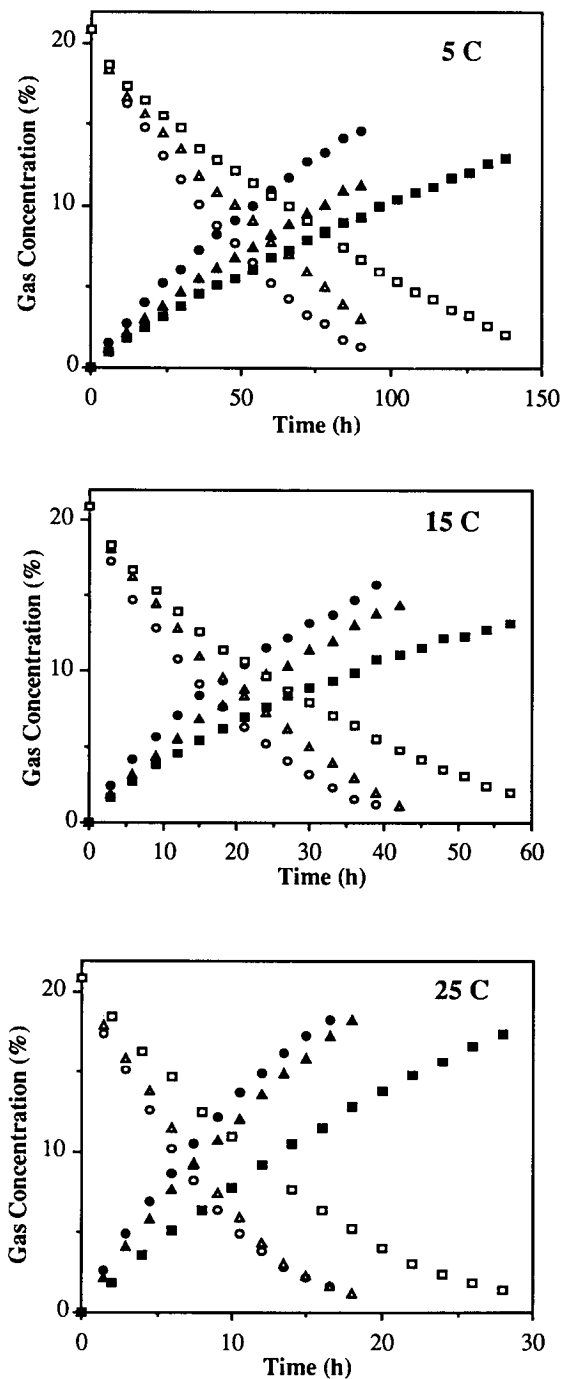
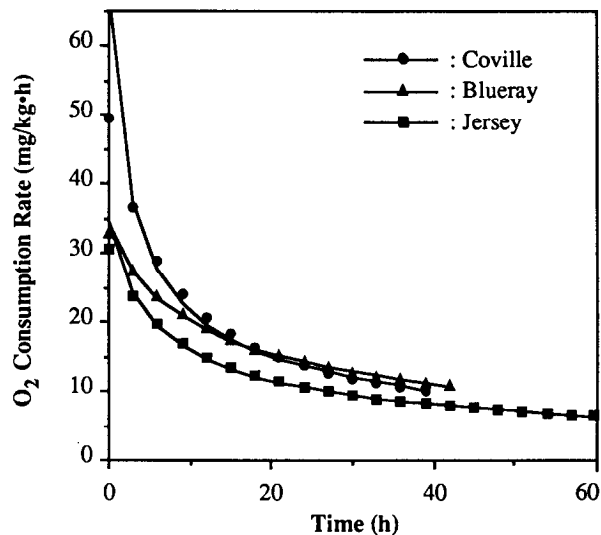


Fig. 1. Plots of O₂ and CO₂ concentration changes inside closed jars containing blueberries at 5, 15, and 25C. Consumption of O₂: 'Coville' (○), 'Blueray' (△), 'Jersey' (□); evolution of CO₂: 'Coville' (●), 'Blueray' (■), 'Jersey' (▲).

Closed system respiration experiments. Closed system experiments (Hagggar et al., 1992; Lee, 1987) were used to measure the respiration rates as a function of O₂ and CO₂ concentrations at 5, 15, and 25C. One liter glass jars, each containing ≈ 265 g blueberries, were tightly covered with metal caps equipped with silicone sampling ports. The jars were immediately stored in a refrigerator with temperature controlled within ±0.5C. Head space gas was periodically sampled and the O₂ and CO₂ concentrations analyzed using a Hewlett Packard 5890A gas chromatograph equipped with a thermal conductivity detector. An Alltech CTR I column (Alltech Associates,



(a)

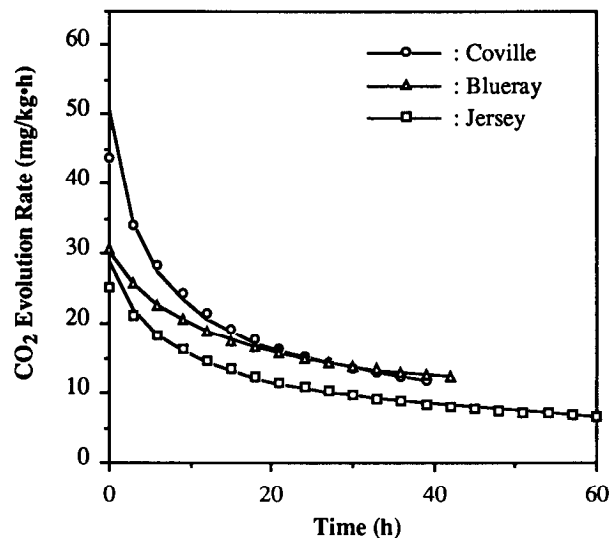


Fig. 2. Actual and predicted respiration rates at 15C; (a) O₂ consumption, (b) CO₂ evolution. The symbols represent actual data entered into Eqs. [3] and [4], and the solid lines are predicted values from Eq. [6].

Deerfield, Ill.) was used with helium as a carrier gas at a flow rate of 65 ml·min⁻¹ and a column temperature of 30C. Sampling of the head space gas was terminated when the CO₂ level inside the jar reached 20% and/or the O₂ level reached 1.5%, because the model is valid only for aerobic respiration (Lee et al., 1992). The measured free volume was 717 to 735 ml, and the sample weight was 264 to 265 g.

Results and Discussion

Respiration rates. The O₂ and CO₂ concentrations inside the closed jars containing blueberry changed with time at 5, 15, and 25C in the expected pattern (Fig. 1). Following the procedure of Hagggar et al. (1992), we fitted the data with the

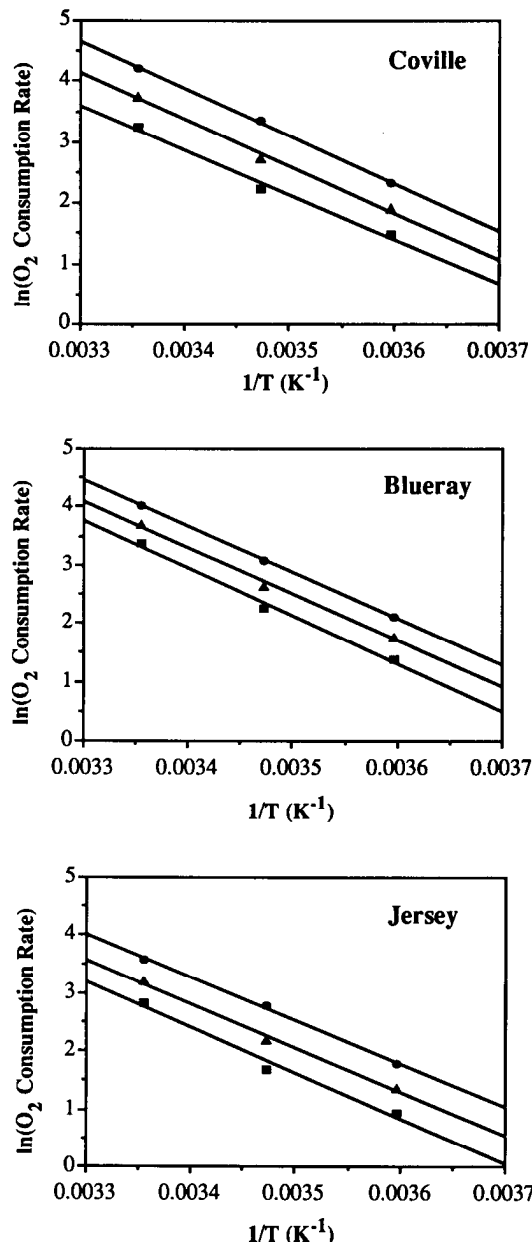


Fig. 3. Effect of temperature on O₂ consumption rates under three gas mixtures. (●) 18% O₂, 4% CO₂; (■) 10% O₂, 10% CO₂; (▲) 4% O₂, 18% CO₂.

following equations using the Software Kaleidagraph (Synergy Software, 1990) to perform nonlinear regression analysis.

$$[O_2] = 21 - \frac{t}{(A_1 + B_2) C_1} \quad [1]$$

$$[CO_2] = \frac{t}{(A_2 t + B_2) C_2} \quad [2]$$

where [O₂] and [CO₂] were O₂ and CO₂ concentrations, respectively; t was time; A₁, A₂, B₁, B₂, C₁, and C₂ were adjustable parameters. The regression equations fitted the data very well with coefficients of determination r² > 0.999. The derivatives

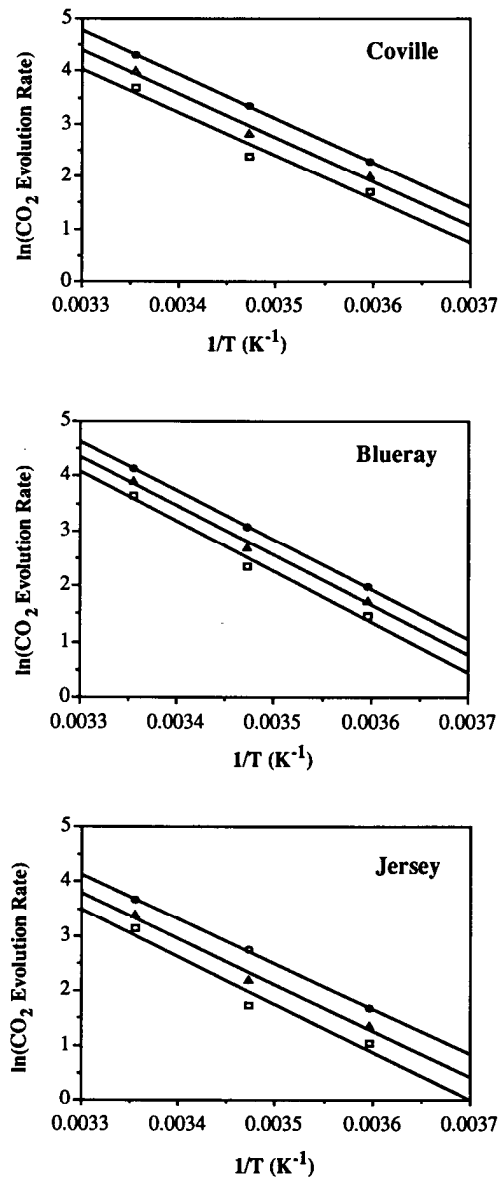


Fig. 4. Effect of temperature on CO₂ evolution rates under three gas mixtures. (○) 18% O₂, 4% CO₂; (□) 10% O₂, 10% CO₂; (△) 4% O₂, 18% CO₂.

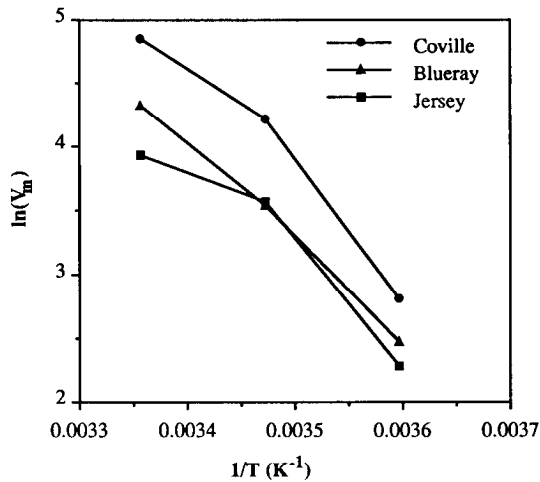
of the equations were used to obtain the experimental respiration rates (O₂ consumption and CO₂ evolution):

$$r_{O_2} = - \frac{d[O_2]}{dt} \left(\frac{M_{O_2} P V}{100 R W T} \right) \quad [3]$$

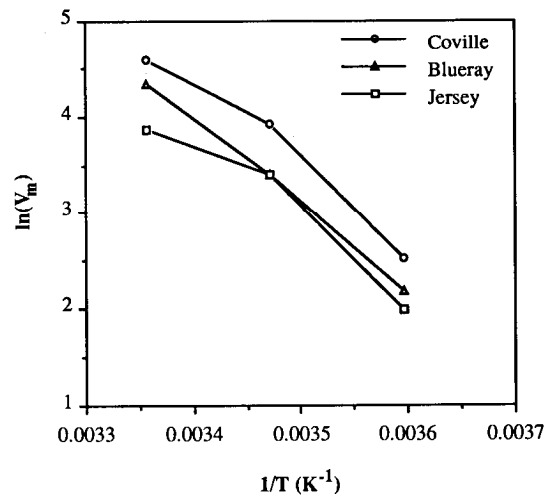
$$r_{CO_2} = \frac{d[CO_2]}{dt} \left(\frac{M_{CO_2} P V}{100 R W T} \right) \quad [4]$$

where r_{O₂} and r_{CO₂} were O₂ consumption and CO₂ evolution, respectively; M_{O₂} and M_{CO₂} were molecular weights of O₂ and CO₂, respectively; W was product weight; P, V, and T was pressure, free volume, and temperature in the respiration chamber, respectively. As an example, Fig. 2 shows the experimental respiration rates as a function of time at 15C.

Enzyme kinetics respiration model. The respiration rates determined were used to test the recently proposed enzyme kinet-



(a)



(b)

Fig. 5. Arrhenius plots for the model parameter V_m ; (a) O_2 consumption and (b) CO_2 evolution. (V_m = maximum respiration rate; T = °C; K = Kelvin).

ics respiration model of Lee et al. (1992), which can be represented by the equation:

$$r = \frac{V_m [O_2]}{K_m + (1 + [CO_2]/K_i) [O_2]} \quad [5]$$

or its linearized form

$$\frac{1}{r} = \frac{1}{V_m} + \frac{K_m}{V_m} \frac{1}{[O_2]} + \frac{1}{K_i V_m} [CO_2]$$

We used the software JMP (SAS Institute, 1991) to perform multiple linear regression using Eq. [6] to estimate V_m , K_m , and K_i . The estimated parameter values and the r^2 are listed in Table 1. With the exception of K_m value (-0.104) for blueberry at 15C, all the parameter values are positive, as expected. However, the negative value is quite close to zero. To confirm the applicability of the model, predicted respiration rates were obtained from the estimated parameter values and compared to the actual values. The predicted and the actual values at 15C com-

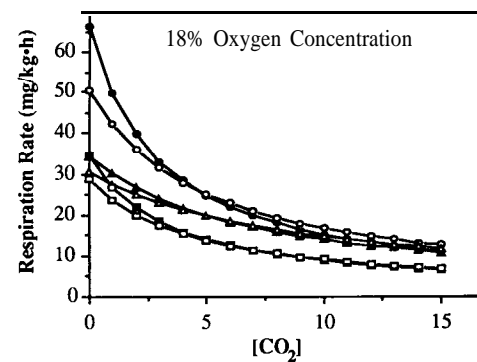
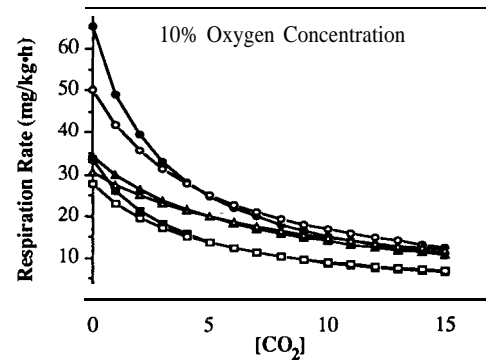
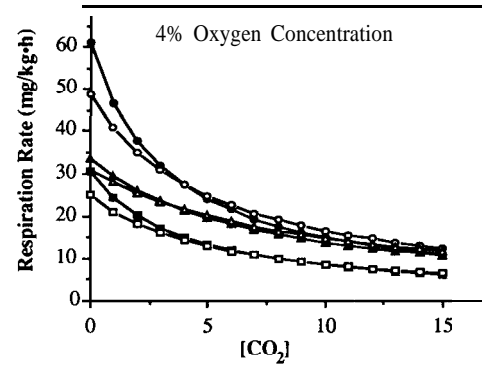


Fig. 6. The model predictions of respiration rates vs. CO_2 at 15C for 4%, 10%, and 18% O_2 . Consumption of O_2 : 'Coville' (●), 'Blueray' (▲), 'Jersey' (■); evolution of CO_2 : 'Coville' (○), 'Blueray' (△), 'Jersey' (□).

pared very well with each other (Fig. 2). Good agreements also was found for the other two temperatures. These results demonstrate that the enzyme kinetics model, based on noncompetitive inhibition of CO_2 , applies very well to blueberries.

Effect of temperature on respiration and V_m . The respiration rates at 5, 15, and 25C were calculated for three selected O_2 and CO_2 concentrations using Eq. [6] and the model parameters from Table 1. The Arrhenius equation was used to fit the data, and excellent agreement resulted. The activation energies (E_a) were almost independent of the cultivars; the average E_a for O_2 consumption (Fig. 3) was 35.2 ± 1.2 Kcal/mole, and the average E_a for CO_2 evolution (Fig. 4) was 39.0 ± 1.6 Kcal/mole. This result supports the suggestion of Haggart et al. (1992) that it may be possible to use only one activation energy for various cultivars of a produce.

The model parameter V_{max} was found to increase with temperature. This result is not surprising because V_{max} has the physical meaning of being the maximum respiration rate. The

Arrhenius plots for V_{\max} are nonlinear and seem to display two regions (Fig. 5). A similar phenomenon was observed in the data of Hagggar et al. (1992). This observation may be explained by the following reasoning. There is evidence that high CO_2 levels suppress respiration by inhibiting certain enzymes such as succinic dehydrogenase (EC 1.3.99.1), cytochrome oxidase (EC 1.9.3.1), and phosphofructokinase (PFK; EC 2.7.1.56) (Kubo et al., 1990). For example, a reduction of PFK activity by elevated CO_2 could result from an inhibition of PFK synthesis or by inactivation of preexisting PFK (Kerbel et al., 1990). Thus, V_{\max} in the enzyme catalyzed reactions depends on several rate constants, each of which may be affected differently by changing temperature (Segel, 1976). Possibly, a different temperature becomes rate limiting at a given reaction step, and consequently the Arrhenius plot may show a change in slope. Although the exact transition temperature is difficult to determine, some changes in reaction rate are apparent at $\approx 15\text{C}$. Therefore, the estimated E_a should be considered an apparent or average value.

Model predictions. The model and the parameter values were used to predict the respiration of blueberries at 15C for 4%, 10%, and 18% O_2 . The respiration rates decrease with increasing CO_2 but are very little affected by changes in O_2 . In general, 'Coville' had the highest respiration rate, followed by 'Blueray' and 'Jersey'. As CO_2 increased, however, the differences in respiration rates among the cultivars decreased. The respiratory quotients (RQ) for all cultivars were >1.0 for CO_2 concentrations up to 4% and ≈ 1.0 at higher CO_2 concentrations.

The enzyme kinetics respiration model, based on noncompetitive inhibition of CO_2 , describes the respiration of the three blueberry cultivars very well. The model predictions indicate that 'Coville' has the highest respiration rate, followed by 'Blueray' and 'Jersey'. Increases in CO_2 can greatly decrease respiration, but change in O_2 has little effect. The temperature dependence of respiration follows the Arrhenius relationship. The Arrhenius plots for V_{\max} are nonlinear, indicating the possibility that this parameter may be controlled by several enzyme reactions, and that the rate constants of these reactions are affected differently by changing temperature. From a practical

point of view, the model along the estimated parameter values can be used to design modified atmosphere packages for blueberries.

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