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# Modeling Respiration Characteristics of Cucumber to Design a Proper Modified Atmosphere Packaging

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## ABSTRACT

**Having a short postharvest life, cucumber undergoes rapid loss of quality. In this research, the effects of temperature, oxygen, and postharvest storage time on the respiration rate of *Royal* cucumbers were investigated. To design a Modified Atmosphere Packaging (MAP) for cucumber in order to extend its shelf-life, a mathematical model using Michaelis–Menten's equation, with the model constants described by means of an Arrhenius-type relationship, was applied to predict respiration rate at various temperatures (4, 10, and 20ºC) and $O_2$ concentrations. Results revealed that all three factors affected respiration rate of the cucumbers, but the influence of temperature was most pronounced. The model was validated in a commercial passive and active MAP. The model could well predict the $O_2$ change in the package but the modeling of $CO_2$ change, mainly at 20ºC, was not satisfactory, which might be due to occurrence of anaerobic condition. The mathematical model was verified as long as the $O_2$ concentration did not reach anaerobic levels at 20°C. Applying the corresponding respiration, the model will also be applicable for other variety of cucumbers with similar metabolic and respiratory behavior to design the optimal MAP conditions.**

**Keywords :** *Cucumis sativus,* Michaelis–Menten's equation, Respiration rate, Shelf life.

## INTRODUCTION

Cucumber (*Cucumis sativus*) is a widely harvested vegetable in the world and is one of the common ingredients used in salad mixes (Mohammadi *et al.*, 2015). Cucumber is susceptible to postharvest moisture loss, wilting, and chilling injury. Therefore, several techniques have been considered to prolong its postharvest life such as temperature control, coating, controlled and modified atmosphere packaging (Moalemiyan and Ramaswamy, 2012). Packaging of fruits and vegetables under Modified Atmosphere (MA) relies on changing the atmosphere inside the package so that it affects the respiration rate ($O_2$ uptake and $CO_2$ production) of the product and gas transfer ($O_2$ entrance and $CO_2$ moving out) through the polymeric packaging film.

Storing fresh produce in MAP with low $O_2$ and high $CO_2$ levels in package results in longer shelf life due to lower metabolic activity of the product (Montanez *et al.*, 2010). To design a successful MAP, there are several factors that have to be considered: respiration rate of the product, product weight, free volume inside the package, temperature, initial gas composition and film permeability to $O_2$ and $CO_2$ (Hertog and Banks, 2003; Salvador *et al*., 2006; Torrieri *et al*., 2009). Among them, respiration rate is the most considerable factor that is influenced by product age, temperature, gas composition, and humidity inside the package (Özgen *et*

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*al.*, 2002). Different respiration rates have been previously reported for cucumber as 6.90 to 24.39 mL $kg^{-1}$ $h^{-1}$, depending on the cultivar, temperature and gas composition in the package (Mahajan *et al.*, 2006). Therefore, modeling the respiration rate is central to design of MAP for fresh fruits and vegetables (Caleb *et al.*, 2013; Finnegan *et al*; 2013; Sousa-Gallagher and Mahajan, 2013). Several mathematical models for respiration rate have been reviewed by Fonseca *et al.* (2002). Since respiration rate is influenced by temperature, it should be considered in the mathematical modelling of respiration rate (Bal, 2013).

Several studies have been done to design MAP for fruits and vegetables, but there are not enough documents about designing MAP for cucumbers. Aim of this study was to apply a mathematical system to design MAP for cucumber stored at 4, 10, and 20°C, using Michaelis-Menten kinetic and Arrhenius model to describe the effect of $O_2$ and temperature on respiration rate, respectively.

## MATERIALS AND METHODS

### Sample Preparation and Packaging

Cucumbers (*Cucumis sativus L. cv. Royal*) were collected at commercial maturity from an organic farm (Mashhad, Iran). The basic product had the following characteristics: average length 15 cm, firmness of 113±11 N, ºBrix of 1.9±0.2, and chlorophyll content of 0.66±0.03. To do experimental tests and measure the respiration rate of the product, they were washed using tap water, and surface dried in a slow air flow.

### Respiration Rate

Respiration rate of *Royal* cucumbers was measured using a closed system respirometer connected to $O_2$ and $CO_2$ gas sensors (Vernier, USA). The cucumbers were inserted in the respirometer chamber (1 liter) and stored at 4, 10, and 20°C. $O_2$ and $CO_2$ concentrations in the chamber were recorded at regular time intervals. $O_2$ consumption rate ($R_{O2}$, mL $kg^{-1}$ $h^{-1}$) and $CO_2$ production rate ($R_{CO2}$, mL $kg^{-1}$ $h^{-1}$) of samples were determined as follows (Finnegan *et al*; 2013; Sousa-Gallagher and Mahajan, 2013):

$$R_{O_2} = \frac{(y_{O_{2i}} - y_{O_2})}{100\,(t-t_i)} \times \frac{V_f}{M} \quad (1)$$

$$R_{CO_2} = \frac{(y_{CO_2} - y_{CO_{2i}})}{100\,(t-t_i)} \times \frac{V_f}{M} \quad (2)$$

Where, $y_{O_{2i}}$ and $y_{CO_{2i}}$ are gas concentrations (%) at initial time $t_i$ (h), $y_{O_2}$ and $y_{CO_2}$ are gas concentrations (%) at time $t$ (h), $M$ is the Mass of the sample (0.3 kg) and $V_f$ (450 mL) is the head space of the package obtained by:

$$V_f = V - \frac{M}{\rho} \quad (3)$$

Where, $V$ is the Volume of the package (mL) and $\rho$ the apparent density of the cucumbers (0.987 g $mL^{-1}$).

The Respiratory Quotient (RQ), the ratio of the $CO_2$ released per unit time to the $O_2$ consumed per unit time is a critical point in respiration process of fresh produce:

$$RQ = \frac{R_{CO_2}}{R_{O_2}} \quad (4)$$

Normal RQ values range from 0.7< RQ< 1.3 as reported in literature for apple (Mahajan and Goswami, 2001): 1.01, blueberry (Song *et al.*, 1992): 0.85, chicory (Devlieghere *et al.*, 2000): 0.93, galega kale (Fonseca *et al.*, 2002): 0.93, mango (Charoenchaitawornchit *et al.*, 2003): 0.79, peeled garlic (Lee *et al.*, 1996): 0.74, strawberry (Hertog *et al.*, 1999): 0.92. If RQ is greater than 1.3, anaerobic respiration takes place (Fonseca *et al.*, 2002).

### Modeling the Influence of $O_2$ Concentration and Temperature on Respiration Rate

The respiration rate of *Royal* cucumbers was determined at 4, 10, and 20°C. Modeling the effect of $O_2$ on respiration rate was done based on Michaelis-Menten's equation (Sousa-

Gallagher and Mahajan, 2013; Mahajan and Goswami, 2001; Fonseca *et al.*, 2002; Iqbal *et al.*, 2009):

$$R_{O_2} = \frac{y_{O_2} \times V_m}{y_{O_2} + K_m} \quad (5)$$

Where, $y_{O_2}$ is the $O_2$% (v⁄v) in the initial gas mixture, *Vm* is the maximal respiration rate (mL $kg^{-1}$ $h^{-1}$) and $K_m$ is the $O_2$% corresponding to $\frac{V_m}{2}$.

Arrhenius equation is widely used to model the effect of temperature on respiration rate (Fonseca *et al.*, 2002; Sousa-Gallagher and Mahajan, 2013; Jacxsens *et al.*, 2002):

$$R_{O_2} = R_{ref}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])} \quad (6)$$

Effect of temperature on Michaelis-Menten parameters can be written as:

$$V_m = V_{m_0}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])} \quad (7)$$

$$K_m = K_{m_0}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])} \quad (8)$$

Where, $V_{m_0}$ and $K_{m_0}$ are the values that $V_m$ and $K_m$ assume at a given reference temperature ($T_0$). $E_a$ (kJ $mol^{-1}$) is the activation energy of the process and $R$ is the gas constant (8.314 J $mol^{-1}$ $K^{-1}$).

Applying Equations (7) and (8) in Equation (5) can describe the simultaneous effect of $O_2$ concentration and temperature on cucumber respiration rate:

$$R_{O_2} = \frac{y_{O_2} \times V_{m_0}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])}}{y_{O_2} + K_{m_0}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])}} \quad (9)$$

Fitting the model to the experimental data by non-linear regression using MATLAB software (Version 7.13), the model constants were estimated. To express error between modeling and experiments, percentage error was determined as follows:

$$\%E = \frac{100}{N} \sum_{1}^{N} \frac{|Rexp - Rpre|}{Rexp} \quad (10)$$

Where, E is percentage Error (%); N is Number of experimental data; $R_{exp}$ is experimental data of Respiration rate ($mO_2$ $kg^{-1}$ $h^{-1}$); $R_{pre}$ is modeled data of Respiration rate ($mLO_2$ $kg^{-1}$ $h^{-1}$).

## Modeling Approach in MAP

Since MAP is a dynamic process, polymeric film regulates the flow of $O_2$ in and $CO_2$ out of the package (Torrieri *et al.*, 2009). Assuming that there is no gas stratification inside the package and that the total pressure is constant, the equations of unsteady material balance are as follows (Finnegan *et al.*, 2013):

$$\frac{dy_{O_2}}{dt} = \frac{A \cdot P_{O_2}}{L \cdot V_f}\,(y_{O_2}^{out} - y_{O_2}^{in}) - R_{O_2}\frac{M}{V_f} \quad (11)$$

$$\frac{dy_{CO_2}}{dt} = \frac{A \cdot P_{CO_2}}{L \cdot V_f}\,(y_{CO_2}^{out} - y_{CO_2}^{in}) + R_{CO_2}\frac{M}{V_f} \quad (12)$$

Where, $P_{O2}$ and $P_{CO2}$ are permeability (mL µm $m^{-2}$ $h^{-1}$ $atm^{-1}$) of the packaging film to $O_2$ and $CO_2$, $R_{O2}$ and $R_{CO2}$ are the respiration rate expressed as oxygen consumption rate and carbon dioxide production rate, *W* is the mass of the product (kg) and *L* is the thickness (µm) of the package film.

To properly design MAP, Equations (11) and (12) must be combined with Equation (9) to consider the effect of $O_2$ and temperature on respiration rate:

$$\frac{dy_{O_2}}{dt} = \frac{P_{O_2}\,(y_{O_2}^{out} - y_{O_2}^{in})}{L \cdot V_f} - \frac{y_{O_2} \times V_{m_0}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])}}{y_{O_2} + K_{m_0}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])}} \times \frac{M}{V_f} \quad (13)$$

$$\frac{dy_{CO_2}}{dt} = \frac{P_{CO_2}\left(y_{CO_2}^{out} - y_{CO_2}^{in}\right)}{L \cdot V_f} + \frac{y_{O_2} \times V_{m_0}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])}}{y_{O_2} + K_{m_0}\, e^{\,(-E_a/R[(1/T)-(1/T_{ref})])}} \times RQ \times \frac{M}{V_f} \quad (14)$$

Where, $V_{m0}$ and $K_{m0}$ are the values that $V_m$ and $K_m$ assume at a given reference Temperature ($T_0$), $E_a$ (kJ $mol^{-1}$) is the activation Energy of the two processes, and *R* is the gas constant (8.314 J $mol^{-1}$ $K^{-1}$).

## Model Validation

To validate the model, cucumbers were stored in Polyethylene (PE) bags (Thickness: 80 µm; $O_2$ permeability: 3,617 mL µm $m^{-2}$ $h^{-1}$

atm$^{-1}$; $CO_2$ permeability: 4241 mL μm m$^{-2}$ h$^{-1}$ atm$^{-1}$; and heat-sealed. A MAP Henkelman (Gustav Muller and Co., Bad Homburg, Germany) was used to perform MAP packaging and the 2 initial internal gas compositions were set. Passive MAP: 21% $O_2$ and 0% $CO_2$, active MAP: 10% $O_2$ and 5% $CO_2$. Samples were stored under 3 temperatures (4, 10, and 20°C) and RH: 85-90%.

### Statistical Analysis

The experimental results were presented as the mean and standard deviation of three measurements. Repeated measures ANOVA (SPSS 12.0, version 2008) was carried out to assess several measurements on the same samples under different conditions. Duncan's Multiple Range Tests (DMRT) at 5% probability were performed to compare the means of different treatments.

## RESULTS AND DISCUSSION

### Respiration Rate

Figure 1 shows the Respiration rate ($R_{O2}$) as a function of time. At 4ºC it decreased from 8.39 to 3 after 44 hours; at 10ºC it decreased from 18.04 to 5.1 after 22 hours; at 20ºC it decreased from 33.64 to 0.34 after 15 hours. $R_{O2}$ decreased over time and at higher temperature, this happens at shorter time with sharper slope, which can be due to higher interaction and respiration rate at accelerated temperatures. $O_2$ was consumed rapidly at early hours at 20ºC and the product faced lack of $O_2$ leading to a fast reduction in $R_{O2}$. Moreover, more $CO_2$ was produced at higher temperatures, which acted as an inhibitor on respiration.

### RQ Parameter

The Respiration Quotient (RQ) shows the transition of aerobic to anaerobic respiration.

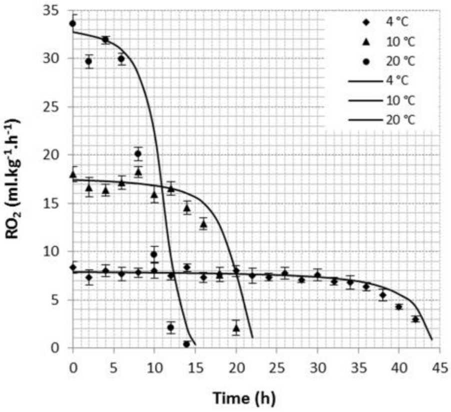


**Figure 1.** $O_2$ consumption rate of Royal cucumber ($R_{O2}$) as function of time at different temperatures. Dots and lines represent experimental and predicted data (Michaelis-Menten's model), respectively.

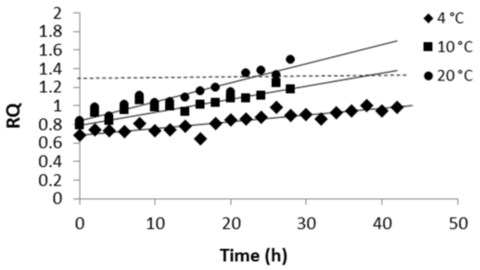


**Figure 2.** Respiration Quotient (RQ) change vs. time.

RQ in range of 0.7-1.3 represents aerobic respiration. According to Figure 2, RQ of samples stored at 4°C stayed at aerobic range by 42 hours. In samples stored at 10 and 20°C, after 32 and 21 hours, respectively, RQ passed the critical limit of 1.3. Del Nobile *et al.* (2006) and Wang *et al.* (2009) reported the same results for lettuce, guajava, and lettuce showing the time and temperature dependence of RQ.

### Modeling the Effect of $O_2$ Concentration and Temperature on Respiration Rate

Figure 3 illustrates $R_{O2}$ of cucumbers stored at 4, 10, and 20ºC as a function of $O_2$ concentration (%). Lower $O_2$ concentration and temperature led to reduction in $R_{O2}$ (dots





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and lines, respectively, represent experimental data and predicted $R_{O2}$ by Michaelis-Menten model).

Michaelis-Menten parameters are presented in Table 1. Temperature has a significant effect on $V_m$ and $K_m$, which increased exponentially with temperature (Figure 4).

Percentage Error (%E) showed a satisfactory agreement between experimental and predicted results (Table 1). Higher $K_m$ value at increased temperature indirectly shows that respiration rate accelerates at higher temperatures. Higher %E about the cucumbers stored at 20°C compared with 4 and 10°C might be because of increased respiration at higher temperature leading to use up almost all $O_2$ in the first short time followed by starting of anaerobic respiration. Therefore, most of $CO_2$ in the package has been produced from anaerobic respiration. Activation Energy ($E_a$) and exponential factor (A) obtained by plotting $K_m$ and $V_m$ against 1/T (Figure 5) are presented in Table 2. The scatter plot (Figure 6) verifies the satisfactory agreement between predicted and experimental $R_{O2}$ ($R^2$= 0.992; r= 0.996) proving that the model describes quite well the effect of $O_2$ concentration and temperature on respiration rate.

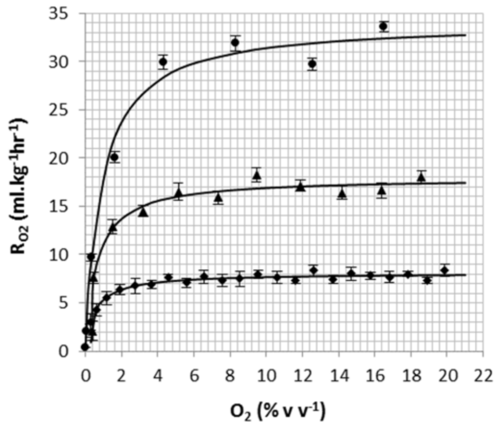


**Figure 3.** $O_2$ consumption rate of Royal cucumber vs. $O_2$ concentration at tested temperatures: ◆ 4; ▲ 10, ● 20°C. Lines show predicted data using the Michaelis-Menten's model.

## MAP Design

Calculating $R_{O2}$ parameters at each temperature and gas concentration from Equations (13) and (14), differential equations were obtained.

Solving the obtained equations using Runge-Kutta method by MATLAB software (version 7.13), gas changing profile inside the packages was estimated. Figure 7 shows the gas changing profile inside permeable

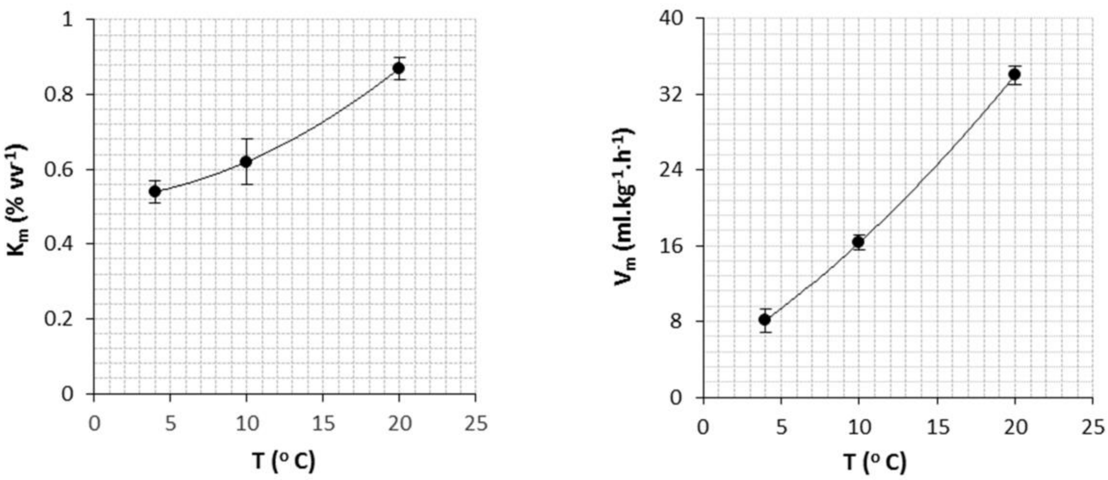


**Figure 4**. Michaelis-Menten model's parameters vs. temperature (lines represent the parameters predicted by the Arrhenius model). The bars represent the standard deviation.





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**Table 1.** Michaelis-Menten parameters at different temperatures for Royal cucumber.

| Temp (°C) | $K_m$ | $V_m$ | %E |
|---|---|---|---|
| 4 | 0.54±0.03 | 8.078±1.2 | 6.10 |
| 10 | 0.62±0.05 | 16.35±0.8 | 7.54 |
| 20 | 0.87±0.03 | 34.01±1 | 10.05 |

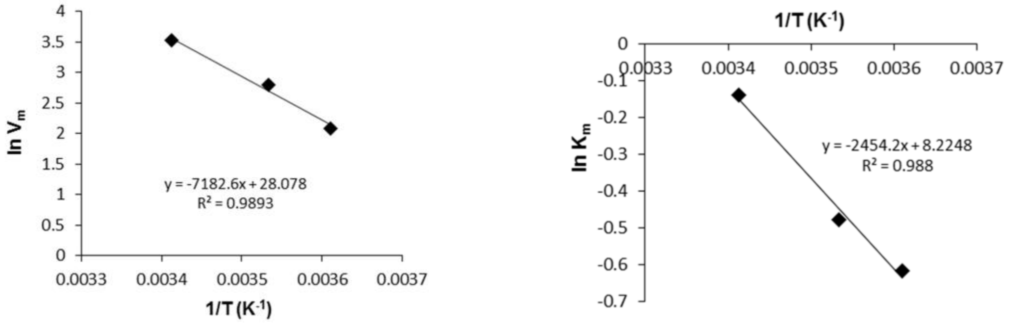


**Figure 5**. Arrhenius equation for Michaelis-Menten parameters for Royal cucumber.

**Table 2.** Exponential factor (A) and activation Energy ($E_a$) of Michaelis-Menten parameters for Royal cucumber.

| Model parameters | A | $E_a$ ($KJ.mol^{-1}$) | $R^2$ |
|---|---|---|---|
| $V_m$ | 28.078±2.6 | 59.7161±3.8 | 0.989 |
| $K_m$ | 8.2248±1.2 | 20.4042±1.9 | 0.988 |

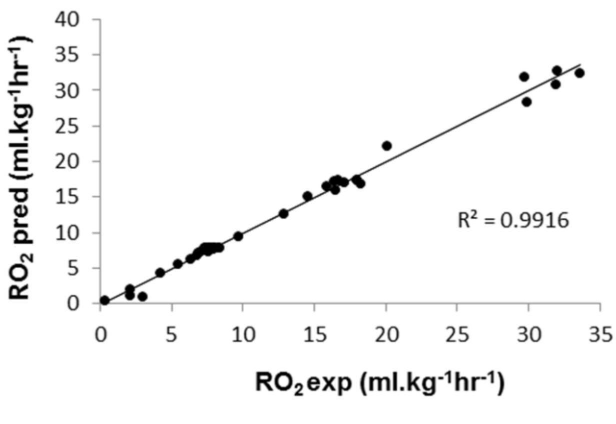


**Figure 6.** Correlation between experimental and predicted $RO_2$ obtained by Equation (9).

packages of cucumbers (active and passive MAP) during storage period at 4, 10, and 20ºC. These results revealed that gas composition inside a package changed as predicted by Equations (11) and (12). Except for 20ºC, predicted data had satisfactory agreement with experimental ones (Table 3).

As expected, $O_2$ decreased and $CO_2$ increased in all packages and reached equilibrium after a specific time. The state of equilibrium is established in the system when the $O_2$ consumption rate by the product is equal to the amount of $O_2$ entering the package and the $CO_2$ production rate of the product is also the same as the $CO_2$ output from the package. Equilibrium Time (ET) is considered as an effective parameter on shelf-life of a product. The shorter the time to reach the equilibrium, the longer the shelf-life of the product is achieved, because the product is sooner exposed to its favorite atmosphere at equilibrium condition.

**Table 3.** Percentage Error (%E) of modeling for gas composition of the packages containing Royal cucumbers according to Figure 6.

| Gas composition | Temp ºC | %E | |
|---|---|---|---|
| | | $O_2$ | $CO_2$ |
| 21% $O_2$+0% $CO_2$ | 4 | 8.57 | 20.57 |
| | 10 | 12.75 | 15.53 |
| | 20 | 44.71 | 24.31 |
| 10% $O_2$+5% $CO_2$ | 4 | 7.73 | 3.75 |
| | 10 | 11.51 | 2.57 |
| | 20 | 12.17 | 10.24 |

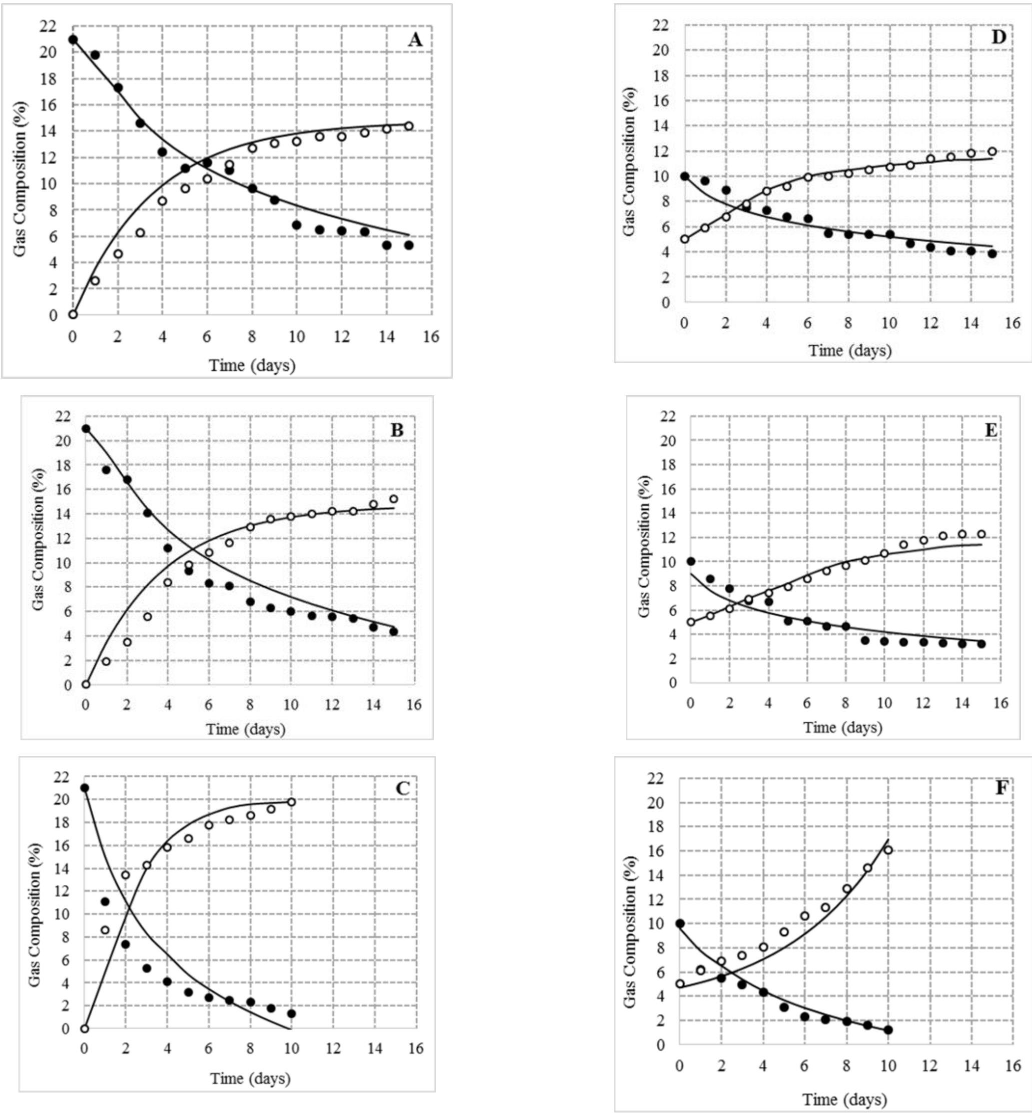


**Figure 7.** Gas composition change in the packages containing Royal cucumber (Passive MAP: A, B, C and Active MAP: D, E, F) vs. time at different temperatures: (A and D) 4ºC; (B and E) 10ºC, (C and F) 20ºC.





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**Table 4.** Equilibrium concentrations and time in the packages containing Royal cucumber.

| Gas composition | Temp (°C) | Equilibrium $O_2$ (%) | Equilibrium $CO_2$ (%) | Equilibrium Time (h) |
|---|---|---|---|---|
| 21% $O_2$ + 0% $CO_2$ | 4 | 7 | 14 | 262 |
| | 10 | 5.5 | 13 | 264 |
| | 20 | 3 | 19 | 240 |
| 10% $O_2$ + 5% $CO_2$ | 4 | 4 | 12 | 252 |
| | 10 | 3.6 | 12 | 248 |
| | 20 | 1 | - | 240 |

Equilibrium Time (ET) and concentration of $O_2$ and $CO_2$ are presented in Table 4. ET decreased with increase in temperature. However, this fact is not enough and remaining in equilibrium is significant (Mahajan *et al.*, 2006). In spite of the shorter ET in the passive MAP stored at ambient condition, consumption of $O_2$ very soon and turning the atmosphere to anaerobic led to a rapid exit of equilibrium. Moreover, $CO_2$ in the active MAP at 20ºC increased rapidly and never reached equilibrium. Chen *et al.* (2000) reported that ET is longer at lower temperatures to store capsicums using LDPE film. $O_2$ and $CO_2$ concentration reached equilibrium after 11, 14, 27, and 32 days at 30, 20, 12, and 0ºC, respectively. Study on apples packed under modified atmosphere and stored at 5, 8, 14, and 20°C revealed that ET at 5 and 8ºC was 14 days, at 14ºC was 11 days, and at 20ºC all $O_2$ content was consumed due to high respiration rate (Torrieri *et al.*, 2009). They reported that the most influential factor to control ET is the ratio of product weight to package volume or use of protective atmosphere technology.

Change in $O_2$ concentration was well predicted by the model (Table 3). However, no satisfactory agreement was observed between the model prediction and $CO_2$ concentration change. This might be due to anaerobic respiration in the package, which was not considered in the modeling.

## CONCLUSIONS

The respiration rate of the Royal cucumbers was well described by Michaelis–Menten model combined with an Arrhenius-type equation. The model was successfully validated in a commercial type of package. Therefore, it can be used to design best package for different chain scenarios. The mathematical model was verified as long as the $O_2$ concentration did not reach anaerobic levels at 20°C. Applying the corresponding respiration, the model will also be applicable to design the MAP for other varieties of cucumber with similar metabolic and respiratory behavior.

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## مدل سازی خصوصیات تنفسی خیار جهت طراحی بسته بندی اتمسفر اصلاح شده بهینه

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## چکیده

به دلیل کوتاه بودن عمر پس از برداشت خیار، این میوه به زودی در معرض افت کیفیت قرار می گیرد. در این تحقیق اثر دما، اکسیژن و زمان پس از برداشت روی نرخ تنفس خیار رقم رویال مورد بررسی قرار گرفته و مدل ریاضی با استفاده از معادله میکائیلز- منتن اعمال شد. نرخ تنفس در دماهای مختلف (۴، ۱۰ و ۲۰ درجه سانتیگراد) و غلظت های متفاوت اکسیژن پیش بینی شده و در طراحی بسته بندی با اتمسفر اصلاح شده برای نگهداری و افزایش ماندگاری خیار مورد استفاده قرار گرفت. نتایج نشان داد که هر سه فاکتور نرخ تنفس خیار را تحت تاثیر قرار دادند ولی تاثیر دما چشمگیرتر بود. مدل حاصله در بسته های با تمسفر اصلاح شده فعال و غیر فعال اعتبارسنجی شد. مدل تغییرات اکسیژن درون بسته را به خوبی پیش بینی کرد ولی پیشگویی تغییرات دی اکسیدکربن در دمای ۲۰ درجه سانتیگراد رضایت بخش نبود که ممکن است به دلیل ایجاد شرایز بی هوازی باشد. مدل ریاضی تا زمانی که غلظت اکسیژن در دمای ۲۰ درجه سانتیگراد به سطح بی هوازی نرسیده بود اعتبار داشت. با اعمال کردن نرخ تنفس متناظر، مدل برای دیگر واریته های خیار با رفتار متابولیکی و تنفسی مشابه در طراحی بسته بندی اتمسفر اصلاح شده بهینه قابل استفاده می باشد.