# Box–Behnken design optimization of microwave postharvest treatment on peach quality in cold storage

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**Abstract:** The effect of microwave postharvest treatment on peach cv."Mashhad SorkhoSefid" quality in cold storage was optimized using Box–Behnken response surface design coupled with numerical optimization technique. The individual and interactive effect of process variables (microwave power (MP), microwave exposure time (MET) and storage time (ST)) on the quality attributes (decay (%), weight loss (%) and firmness (N)) were studied. The results showed that, MET, MP and ST had significant effect on quality parameters of peaches. The experimental data were analyzed by analysis of variance (ANOVA) and second-order polynomial models were developed using multiple regression analysis. The models developed from the experimental design were predictive and good fit with the experimental data with high coefficient of determination (R2) values. An optimization study using desired function methodology was performed and the optimal conditions were found to be MP; 250.44 w, MET; 49.47 s and ST; 30 days. The corresponding predicted values were 5.18% for decay; 3.35% for weight loss, and 1.25 N for firmness.

**Keywords:** microwave, postharvest, peach quality, cold storage

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## 1 Introduction

Peach (*Prunus persica* (L.) Batsch) is a perishable fruit, and during storage it can undergo postharvest decay which is one of the major factors that limit the extension of storage and market life of fruits (Sasaki et al., 2009). In order to reduce the decay and extend the shelf life of stone fruits, fungicides are available for postharvest treatment. However, consumers' concerns over chemical residues can create health problems, as well as the increasing resistance of many fungi to commonly used fungicides (Zhang et al., 2004). Therfore, devloping new effective postharvest treatments to reduce the impact of agriculture on the environment and on human health is necessary (Elmer and Reglinski, 2006; Mari et al., 2010). Among the alternative treatment methods, microwave heating, also referred to as dielectric heating, has potential to control postharvest decay. Dielectric heating, including radio frequency (RF) and microwave (MW) heating, can be a potential alternative treatment to control postharvest diseases. Dielectric materials, as most agricultural products convert electric energy at RF and MW frequencies into heating (Wang et al,. 2003; Shinya et al., 2014).

Microwave heating is already widely used in the food industry, and its application provides rapid heating (Zhang et al., 2006). A number of researchers have used microwave energy successfully to control postharvest diseases in peaches (Karabulut and Baykal, 2002; Sisquella et al., 2014), however, few studies have been published on the use of microwaves to maintain fruit quality during cold storage. United States Federal Communication Commission has approved two microwave frequencies; 915 and 2,450 MHz for heating application. The 915 MHz microwaves have higher

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energy and penetrate more deeply in fruits and vegetables than 2,450 MHz (Giese, 1992). Karabulut and Baykal, (2002) have successfully used the 2,450 MHz microwave (used in kitchen type microwave ovens) under laboratory conditions to control postharvest diseases of peaches.

Microwave treatment can be used to maintain the quality of fruit during postharvest to reduce the speed of the metabolic processes associated with ripening. From quality point of view, peach ripening is a complex process that can be measured by many parameters, such as; soluble solids concentration (SSC) and titratable acidity (TA) which are related to maturation and to quality of the fruit (Kader, 1999). Zhang et al. (2008) indicated that peach firmness is a significant quality parameter, which is valuable to follow maturity evolution during postharvest storage. Crisosto et al. (2006) identified the importance of the maturity stage in fruit marketing and consumption and proposed that firmness is the most appropriate maturity index.

The main objective of this study was to determine the microwave conditions in a domestic microwave system that could reduce postharvest decay without causing damage to peaches and assess their effect on fruit quality during cold storage.

## 2 Materials and methods

### 2.1 Fruit in the study

Fruit used in this study was peach (*Prunus persica* (L.) Basch) cv. "Mashhad SorkhoSefid". Fruit at commercial maturity was collected from the Research station in Khorasan Razavi Province, Iran. Peaches without visible injuries were selected by hand from fruit bins immediately after harvest. The physico-chemical analysis of fresh peach fruits was carried out as per AOAC (Association of Official Analytical Chemists, 2000). The determination of moisture in peach was effectuated by the drying process in a drying oven (Memmert, Germany) at the temperature of 103℃. PH is a measure of the acidity or basicity of a solution. It is defined as the co logarithm of the activity of dissolved hydrogen ions (H+). The pH of the juices was evaluated using a digital pH meter (Metrohm 691, Swiss) at 27°C. Total soluble solids (TSS) and the refractive index were assayed using the refractometric method, with a Refractometer (ABBE-2WAJ 0-90, China), and corrected to the equivalent reading at 20°C (AOAC, 2000). Acidity was determined by titrating samples with 0.01M NaOH solution up to pH 8.2, and was expressed as malic acid per 100 g juices.

The Physico-chemical analysis results of fresh peach fruits are presented in Table 1.

**Table 1 Physico-chemical analysis results of peach**

| Physic chemical parameter | Mean values ±SD |
|---|---|
| Moisture,%wb | 87.5±0.10 |
| TSS, °Bx | 11.3 ±0.20 |
| pH | 4.14 ±0.02 |
| TA ,% | 0.33 ±0.01 |
| Firmness,N | 1.55 ±0.04 |

### 2.2 Fruit quality attributes

2.2.1 Microwave heating system and suitable treatment conditions

A domestic microwave oven (Samsung, USA), with 1.2 kW nominal maximum powers and a frequency of 2450 MHz was used to carry out the experiments. A turntable enhanced uniformity of the microwaves in the cavity. For all experiments, the desired output power levels at different exposure times were applied for 3 fruits, and the experiment was repeated three times. A set of fruit was not treated and used as a control. After treatment, fruits were kept in the laboratory for 20 min to allow the heat to redistribute and to equilibrate with room temperature. Fruits were then placed in a corrugated carton box(9 peaches), and stored at 0±0.5℃ with relative humidity of 90± 5% for 30 days followed by 2 days at 24℃.

2.2.2 Quality parameters

To evaluate the effect of microwave powers and exposure times on fruit quality, the parameters (decay (D), firmness (F), and weight loss (WL)) were measured after harvest and storage days. Each treatment comprised 18 fruit (3 in microwave turn table; 9 in courgated box , and 18 (2boxes) for each treatment)and the experiments were done in 3 replicates. The percentage of decayed fruit including; the presence of exudate (juice leak), macroscopic fungal growth, on the fruit surface were visually evaluated. Fruits presenting any of those characteristics were considered as damaged fruits (Civello et al., 1997). For weight loss measurement (Wang et al., 2010), the weight of 18 fruit from each replicate was recorded by an electronic analytical balance (±0.001 g) (Mettler Toledo, PB303, Switzerland) before (A) and after (B) storage, respectively, and the weight loss was calculated as:

$$[(A - B)/A] \times 100\% \qquad (1)$$

Fruit firmness (N) was measured by using a penetrometer (Wagner Fruit Firmness Tester model FT-327, USA). An 8 mm plunger tip was used after removing the epidermis from the opposite equatorial sides of each fruit with the help of a peeler (Tareen et al., 2012).

### 2.3 Experimental design and statistical analysis

The Box-Behnken designs (BBD) (Box and Behnken, 1960) of response surface methodology with three levels were used for the study. The process was optimized based on three input variables whose interactions were studied as three major responses. Based on preliminary single factor experiments the levels of input variables were determined. The levels of various input variables selected were as follows: microwave power: 200-800 w, microwave exposure time: 0-120 s, and storage time 0 – 30 days. All the responses were analyzed in triplicates and the average value was reported. The responses selected were decay, weight loss and firmness. Low and high levels of each factor were coded as –1 and +1 keeping 0 as mid-point. The symbols and levels are shown in Table 2. Five replicates at the center of the design were used to allow for estimation of a pure error sum of squares. Since the various responses were the result of various interactions of independent variables, therefore the following second order polynomial regression equation was fitted to the experimental data of all responses, Equation (2).

$$Y = \beta_0 + \sum_{j=1}^{k} \beta_j X_j + \sum_{j=1}^{k} \beta_{jj} X_j^2 + \sum_{i=1}^{j-1} \sum_{j=2}^{k} \beta_{ij} X_i X_j + \varepsilon \qquad (2)$$

where, $Y$= predicted response, $\beta_0$= a constant, $\beta i$ = linear coefficient, $\beta jj$= squared coefficient, and $\beta ij$ = interaction coefficient, $Xi$ and $Xj$ are the independent variables and $\varepsilon$ is noise or error (Yousefi et al., 2015).

The Design Expert software (version 8.0.5.2, 2010, Minneapolis MN, USA) was employed for regression and graphical analysis of the obtained data. The quality of the fitted polynomial models was expressed by the coefficient of determination ($R^2$), adjusted $R^2$, coefficient of variation (CV), the prediction error sum of squares (PRESS) and adequate precision.

**Table 2 Experimental levels in the experimental design and responses**

| Run | Real values | | | Responses | | |
|---|---|---|---|---|---|---|
| | X1<br>MP(W)[a] | X2<br>MET(s)[a] | X3<br>ST (day)[a] | Decay<br>,% | Weight Loss<br>,% | Firmness<br>,N |
| 1 | 500 | 0 | 0 | 0.00 | 0.00 | 1.30 |
| 2 | 500 | 60 | 15 | 3.52 | 3.91 | 1.23 |
| 3 | 200 | 60 | 30 | 5.23 | 3.08 | 1.26 |
| 4 | 200 | 120 | 15 | 2.33 | 5.69 | 1.20 |
| 5 | 500 | 120 | 30 | 6.16 | 5.83 | 1.01 |
| 7 | 800 | 60 | 0 | 0.00 | 0.00 | 1.07 |
| 8 | 200 | 60 | 0 | 0.00 | 0.00 | 1.28 |
| 9 | 500 | 0 | 30 | 6.58 | 2.47 | 1.03 |
| 10 | 500 | 120 | 0 | 0.00 | 0.00 | 0.97 |
| 11 | 500 | 60 | 15 | 3.25 | 4.09 | 1.38 |
| 12 | 800 | 120 | 15 | 6.87 | 6.28 | 0.85 |
| 13 | 800 | 60 | 30 | 8.56 | 3.50 | 1.05 |
| 14 | 500 | 60 | 15 | 3.34 | 4.23 | 1.27 |
| 15 | 200 | 0 | 15 | 4.32 | 2.71 | 1.12 |
| 16 | 500 | 60 | 15 | 2.89 | 3.67 | 1.25 |
| 17 | 800 | 0 | 15 | 4.55 | 2.71 | 1.12 |

Note: [a]: MP,MET and ST are Microwave Power, Microwave Exposure Time, and Storage Time.

## 3 Results and discussion

### 3.1 The results of various combinations on independent variables

The various combinations of independent variables generated by BBD of RSM are depicted in Table 3. The results of analysis of variance (ANOVA) indicate that the contribution of the quadratic models was very significant ($p < 0.0001$) for the responses. The high $R^2$ (0.8864 – 0.9925) and adjusted $R^2$ (0.7729 – 0.9829) values indicate the precision of the constructed models. At the same time, the CV values were between 6.43% and 15.96%, indicating high degree of precision and reliability of the experimental values. The suitable PRESS values also suggest the adequacy of the fitted quadratic models for predictive applications (Table 3). The adequate precision measures the signal-to-noise ratio with a ratio greater than 4 being desirable. The high adequate precision values (9.03 – 33.76) indicated that the fitted models could be used to navigate the design space.

Figure 1 (a-c) shows the comparison between the observed and the model predicted values. The results demonstrate that the polynomial regression models were in good agreement with the experimental data. The perturbation plot was used to study the influence of three factors simultaneously on the fruit quality attributes (Figure 2(a-c)). The relationships between the experimental variables and the responses are illustrated in three-dimensional representations of the response surfaces. These plots are presented in Figure 3(a-f).

**Table 3 Evaluation of mathematical models for the response variables**

| Source | DF | Decay ($Y_1$, %) | | | Weight Loss ($Y_2$, %) | | | Firmness( $Y_3$,N) | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | Cof. | SS | *p-Value* | Cof. | SS | *p-Value* | Cof. | SS | p-Value |
| Model | 6 | 3.33 | 108.10 | < 0.0001 | 4.22 | 66.31 | < 0.0001 | 1.32 | 0.35 | 0.0044 |
| **Linear** | | | | | | | | | | |
| $b_1$(MP, w)[a] | 1 | 1.01 | 8.20 | < 0.0001 | | | | -0.1 | 0.07 | 0.0067 |
| $b_2$ (MET, s) | 1 | -0.01 | 0.00 | 0.9292 | 1.24 | 12.28 | < 0.0001 | -0.07 | 0.04 | 0.0356 |
| $b_3$ (ST, day) | 1 | 3.32 | 87.92 | < 0.0001 | 1.86 | 27.66 | < 0.0001 | -0.03 | 0.01 | 0.2308 |
| **Quadratic** | | | | | | | | | | |
| $b_{11}$ | 1 | 1.08 | 4.64 | 0.0004 | | | | -0.08 | 0.03 | 0.0660 |
| $b_{22}$ | 1 | 0.83 | 2.77 | 0.0018 | | | | -0.16 | 0.11 | 0.0020 |
| $b_{33}$ | 1 | -0.10 | 0.04 | 0.5587 | -2.36 | 23.55 | < 0.0001 | -0.07 | 0.02 | 0.0837 |
| **Interaction** | | | | | | | | | | |
| $b_{12}$ | 1 | 0.73 | 2.21 | 0.0033 | | | | -0.09 | 0.03 | 0.0481 |
| $b_{13}$ | 1 | 0.46 | 0.89 | 0.0277 | | | | | | |
| $b_{23}$ | 1 | -0.61 | 1.56 | 0.0080 | 0.84 | 2.82 | 0.0054 | 0.08 | 0.02 | 0.0690 |
| Residual | 10 | | 0.81 | | | 2.95 | | | 0.05 | |
| Lack- of-fit | 6 | | 0.47 | **0.2844**[b] | | 2.39 | **0.2432** [b] | | 0.01 | **0.9082** [b] |
| Pure error | 4 | | 0.35 | | | 0.56 | | | 0.04 | |
| Total | 16 | | 108.91 | | | 69.26 | | | 0.4 | |
| $R^2$ | | | 0.9925 | | | 0.9574 | | | 0.8864 | |
| Adj- $R^2$ | | | 0.9829 | | | 0.9431 | | | 0.7729 | |
| PRESS | | | 8.05 | | | 10.23 | | | 0.16 | |
| *CV(%)* | | | 9.47 | | | 15.96 | | | 6.43 | |
| Ad- precision | | | 33.76 | | | 23.03 | | | 9.03 | |

Note: [a] MP,MET and ST are Microwave Power, Microwave Exposure Time, and Storage Time.
[b] Not significant ($p > 0.05$).

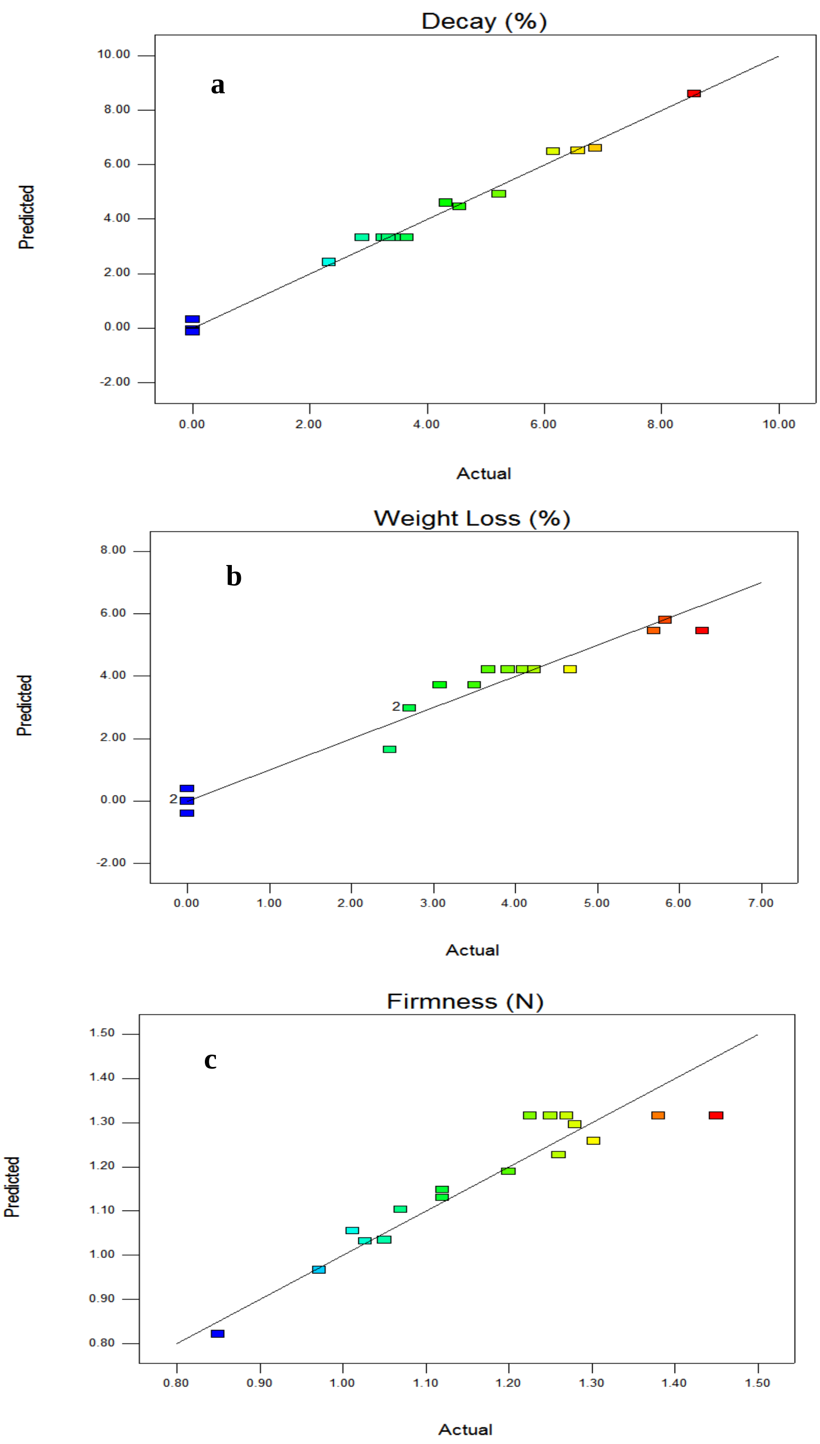


Figure 1 Predicted vs Acual plots on the decay (a), weight loss (b), firmness (c), as a function of Microwave Power (MP), Microwave Exposure Time (MET), and Storage Time (ST)

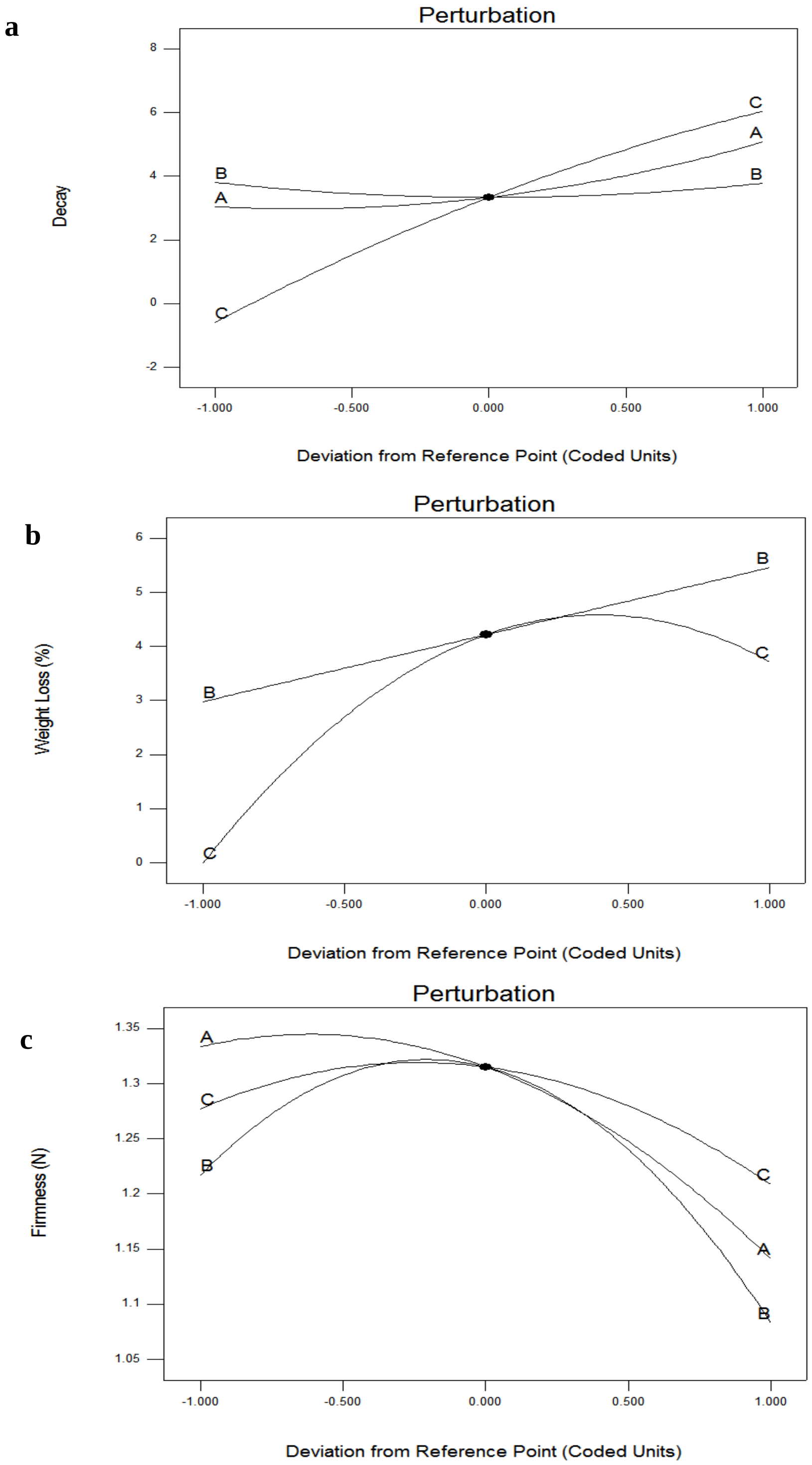


Figure 2 Overlay plot of perturbation of the independent variables on the decay (a), weight loss (b) and firmness (c)

Note: A: Microwave power=500 w ; B: Microwave Exposure Time =60 s ;C: Storage Time=15 days.

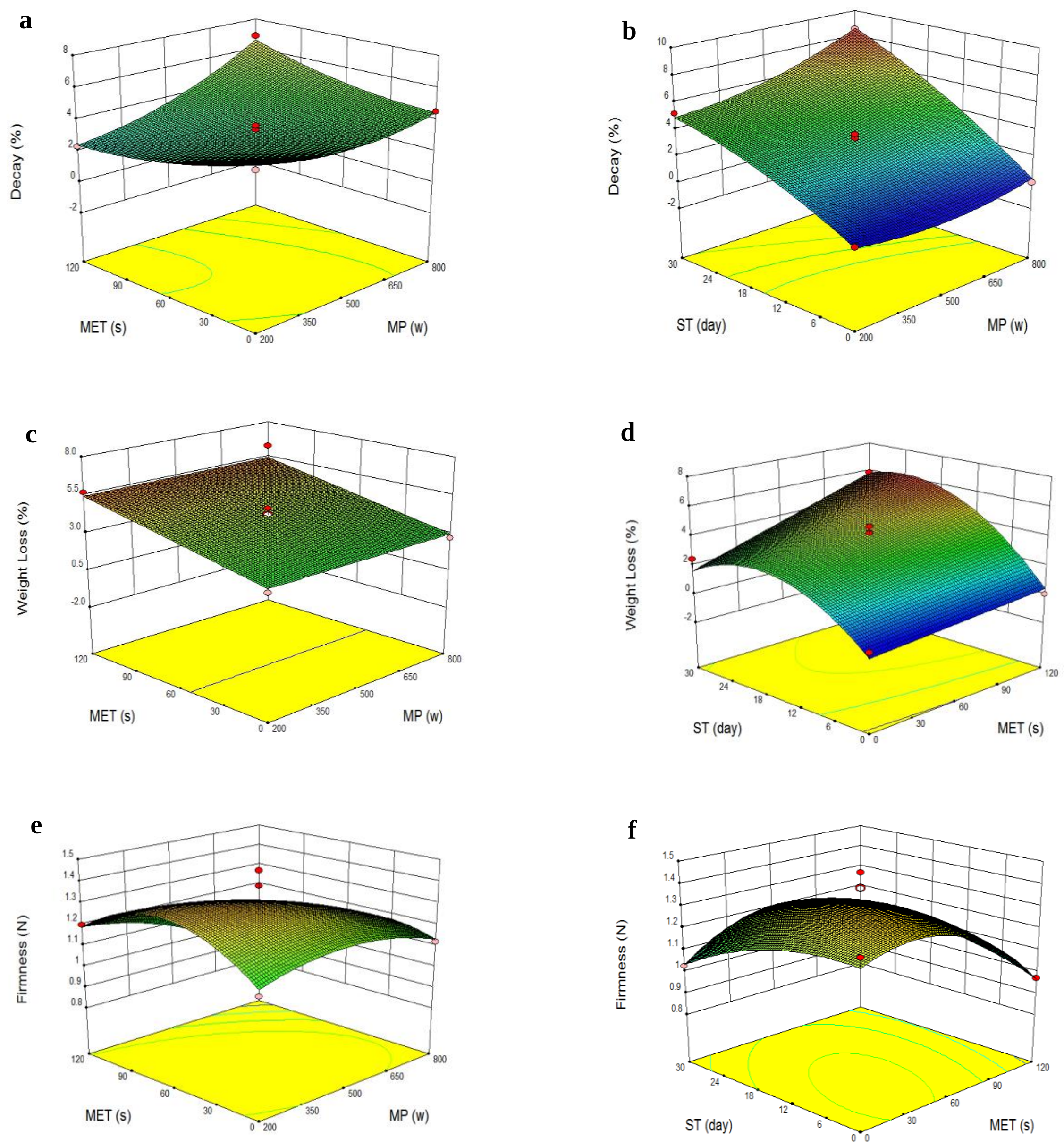


Figure 3 Response surface plots for the significant interaction effects on the decay (a, b), weight loss (c,d), firmness (e,f), as a function of Microwave Power(MP), Microwave Exposure Time (MET), and Storage Time (ST)

### 3.2 Analysis of Various Process Responses

#### 3.2.1 Decay

Table 3 shows that the linear effects of MP and ST were significant ($p < 0.0001$) on the decay ($Y_1$), whereas MET was insignificant. Quadratic effect of MP and MET was significant at $p < 0.05$ in experimental domain studied. The mutual interaction all the independent variables were found to be significant ($p <0.05$).

The effects of independent variables on the decay value represented in perturbation graphs (Figure 2a). This plot showed that MP enhancement caused increasing decay; while increasing in MET up to 60s, decreased decay, and afterwards this factor increased gradually, besides this attribute increased by increasing storage time. Figure 3a shows the three-dimensional response surface plot for MET and MP of decay. As shown in Figure 3 a, b the variation in the decay could be explained as a non-linear function of the contents of all independent variables. The most significant ($p < 0.0001$) effect on decay was shown to be the linear effect of ST and MP followed by the quadratic effects of MP and MET (Table 3). In addition, the interaction terms of MP and MET was significant.

From Figure 3a, it can be seen that the increasing MP increases the decay whereas with increasing MET till 60s, the decay decreased but there after the decay showed an increasing trend. Sisquella et al. (2013a) reported fruit microwave treatment at 17.5 kW for 50 s significantly reduced brown rot incidence to 14% in 'Roigd'Albesa' peaches compared with untreated fruit (100%). In Figure 3b, the effect of MET and ST on the decay was studied when the MET was kept constant. The result showed that increasing MET gradually increased the decay whereas this factor increased sharply by increasing ST. Karabulut and Baykal (2002) evaluated the possible use of microwave power to control postharvest pathogens of peach fruit and reported that the natural decay of 'J.H. Hale' peaches were very low after 45 days of storage and 5 days shelf life by application of 0.4 kW microwave power treatment for 2 min. Other authors (Zhang et al., 2006; Zhang et al. 2004) reported that fruit natural decay reduced after microwave treatments. Their results showed that the use of microwave power was very effective in controlling natural decay during storage.

3.2.2 Weight loss

The linear effects of MET and ST variables were significant on the weight loss ($Y_2$) values ($p < 0.0001$). In quadratic terms, ST had significant effects on this factor (Table 3). Also, the mutual interaction between MET and ST was found to be significant ($p< 0.001$). It can be seen that the variable with the largest effect on the weight loss was the linear effects of ST and MET; respectively. In addition, Figure 3c shows that when MET increased, the weight loss increased slightly, while an increase in ST led to a sharp increase in the weight loss until 24 days and then it reduced gradually. The most significant ($p < 0.0001$) effect on weight loss was shown to be the linear effect of ST and MET followed by the quadratic effects of ST, and interaction effects of MET with ST and (Table 3). Figure 3d also demonstrated that ST had a considerable role in the weight loss of peach fruit. Figure 2b also clearly depicts that the investigated factor increased by increasing MET and ST. Gonzalez-Aguilar *et al*. (2004) found the same results by application UV-C irradiation pre-storage of peaches. Ikediala et al. (1999) treated Sweet cherries (*Prunus avium* L.) by 0.2 to 5 kW microwaves for 2 min holding and 5 min hydrocooling protocol and reported significant differences ($P > 0.05$) in weight of cherries due to the treatments, although the changes were slight.

3.2.3 Firmness

Table 3 illustrates that among three independent variables, the linear term of MP was the highest effect on the firmness value ($p < 0.001$) followed by linear effect of MET ($p < 0.05$), and quadratic effect of MET, and interaction effect of MP with MET content ($p < 0.05$). Figure 2c clearly depicts that the investigated factor enhanced by increasing MP, MET and ST, while it decreased with an increase in MP, MET and ST at the middle point. Figure 3e shows the significant interactions of independent variables MP and MET on the firmness. Firmness of microwave-treated fruit was significantly higher than that of control fruit after long-term storage and shelf life. The results indicated that MW treatment on peach fruits did not negatively affect fruit firmness. Firmness of fruit after microwave treatment apparently was either higher or similar to that of the Control (Ikediala et al., 1999). Similar trends have been reported by (Sisquella et al., 2013b) who observed higher firmness in treated fruit with microwave in comparison with untreated fruits. The greater firmness observed in peaches might be due to the inactivation of cell wall hydrolytic enzymes, mainly polygalacturonase (Malakou and Nanos, 2005) or could be associated to lower ethylene production from heated fruit in comparison to untreated fruit (Budde et al., 2006). Figure 3f showed that ST had a considerable role on firmness of peach fruit, which deceased during cold storage. Shinya et al. (2014) reported the same results.

### 3.3 Optimization and validation of process

After the development of models for various responses (decay, weight loss, and firmness), the optimization of the process parameters depending upon the results was done. From the various data obtained and by their statistical analysis, the appropriate range or values for various process parameters were selected like MP and MET in range and maximum value for ST. The criteria used for optimization along with predicted value of responses have been presented in Table 4. The optimum conditions for best fruit storage quality in terms of decay, weight loss, and firmness can also be determined by intersection zone of minimum decay and weight loss, and maximum firmness as presented in the overlay plot of MP and MET (Figure 4). Validation step showed a good agreement between the predicted and experimental data. Thus, response surface optimization suitably predicted the optimum conditions.

**Table 4 Criteria used for optimization along with predicted and actual value of responses**

| Constraints | Goal | Lower Limit | Upper Limit | Importance |
|---|---|---|---|---|
| MP,W | In range | 200 | 800 | 3 |
| MET,s | In range | 0 | 120 | 3 |
| ST,day | Maximize | 0 | 30 | 3 |
| Decay,% | Minimize | 0.00 | 8.56 | 3 |
| WL,% | Minimize | 0.00 | 6.28 | 3 |
| F,N | Maximize | 0.85 | 1.45 | 3 |

Note: MP= Microwave Power; MET= Microwave Exposure Time; ST=Storage Time; D=decay; WL= weight loss; F= firmness.

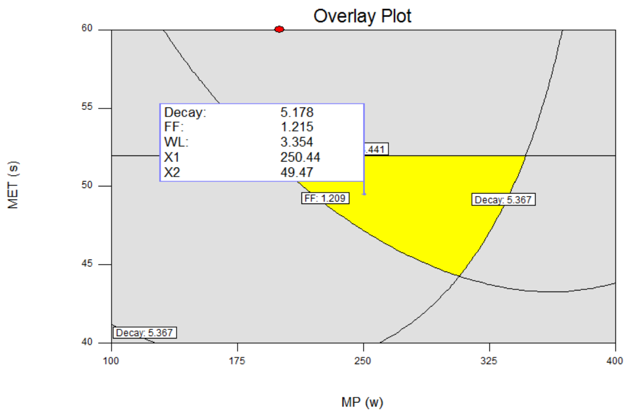


Figure 4 Overlay plot showing the level of input variables and predicted values of responses. X1=microwave power, X2= microwave Exposure Time, (Storage time; 30 days)

## 4 Conclusions

RSM was used for optimization of microwave postharvest treatment of peach cv."Mashhad SorkhoSefid" in cold storage. Decay, weight loss, and firmness of peaches were studied with 17 combinations of microwave power, exposure time, and cold storage. An optimization study using desired function methodology was performed and the optimal conditions were found to be MP; 250.44 w, MET; 49.47 s and ST; 30 days. The corresponding predicted values were 5.18% for decay; 3.35% for weight loss, and 1.25N for firmness.

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