

Modeling of Spore Forming Bacterial Population on the Sliced Chicken as a Function of Period of Storage and Gamma-Irradiation Dose Given

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ABSTRACT: This paper deals with the developing the most suggested model with $R^2 = 0.9092$ for Total Spore forming bacterial count as a function of the irradiation dose and the storage period for a sliced chicken meat.

KEYWORDS: Irradiation, Radiation, Modeling, Cobalt-60, Radiolysis, Design Expert Software 8.0.

1 INTRODUCTION

Poultry carcasses are commonly contaminated with en-teric pathogens such as Salmonella, Campylobacter and *Listeria monocytogenes* (Jacobsreitsma et al., 1994; Murphy et al., 2004); the possibility of cross contamination of poultry carcasses post slaughter is high. Decontamination of poultry carcasses is therefore desirable. Various decontamination technologies have been proposed including the use of various chemical agents such as alkali (Rodriguez et al., 1996), physical methods such as steam treatment (James et al., 2007) and biological control with bacteriophages (Carvalho et al., 2010), but only treat-ment with water supplemented with chlorine or a chlorine-ting agent is used commercially. The effects of such decontaminating treatments are limited (Oyarzabal, 2005; Russell and Axtell, 2005).

The term "radiation chemistry" refers to the chemical reactions that occur as a result of the absorption of ionizing radiation. In the context of food irradiation, the reactants are the chemical constituents of the food and initial radiolysis products that may undergo further chemical reactions. The chemistry involved in the irradiation of foods has been the subject of numerous studies over the years and scientists have compiled a large body of data regarding the effects of ionizing radiation on different foods under various conditions of irradiation. The basic principles are well understood and provide the basis for extrapolation and generalization from data obtained in specific foods irradiated under specific conditions to draw conclusions regarding foods of a similar type irradiated under different, yet related, conditions. The types and amounts of products generated by radiation induced chemical reactions ["radiolysis products"] depend on both the chemical constituents of the food and on the specific conditions of irradiation.

The principles of radiation chemistry also govern the extent of change, if any, in both the nutrient levels and the microbial loads of irradiated foods.

Factors Affecting the Radiation Chemistry of Foods- Apart from the chemical composition of the food itself, the specific conditions of irradiation that are most important in considering the radiation chemistry of a given food include the radiation dose, the physical state of the food (e.g., solid or frozen versus liquid or non-frozen state, dried versus hydrated state), and the ambient atmosphere (e.g., air, reduced oxygen, and vacuum). The temperature at which irradiation is conducted can also be a factor, with more radiation-induced changes occurring with increasing temperature. Temperature is less important, however, than the physical state of the food. The amounts of radiolysis products generated in a particular food are directly proportional to the radiation dose. Therefore, one can extrapolate from data obtained at high radiation doses to draw conclusions regarding the effects at lower doses.

The radiation chemistry of food is strongly influenced by the physical state of the food. If all other conditions, including dose and ambient atmosphere, are the same, the extent of chemical change that occurs in a particular food in the frozen state is less than the change that occurs in the non-frozen state. This is because of the reduced mobility, in the frozen state, of the initial radiolysis products, which will tend to recombine rather than diffuse and react with other food components. Likewise, and for similar reasons, if all other conditions are the same, the extent of chemical change that occurs in the dehydrated state is less than the change that occurs in the fully hydrated state.

The formation of radiolysis products in a given food also is affected by the ambient atmosphere. Irradiation in an atmosphere of high oxygen content generally produces both a greater variety, and greater amounts, of radiolysis products in the food than would be produced in an atmosphere of lower oxygen content. This is because irradiation initiates certain oxidation reactions that occur with greater frequency in foods with high fat content.

With few exceptions, the radiolysis products generated in a particular food are the same or very similar to the products formed in other types of food processing or under common storage conditions. These radiolysis products are also typically formed in very small amounts. Radiation-induced chemical changes, if sufficiently large, however, may cause changes in the organoleptic properties of the food. Because food processors want to avoid undesirable effects on taste, odor, color, or texture, there is an incentive to minimize the extent of these chemical changes in food. Thus, the doses used to achieve a given technical effect (e.g., inhibition of sprouting, reduction in microorganisms) must be selected carefully to both achieve the intended effect and minimize undesirable chemical changes.

Typically, the dose or dose range selected will be the lowest dose practical in achieving the desired effect. Irradiation also is often conducted under reduced oxygen levels or on food held at low temperature or in the frozen state.

In general, during inactivation of microorganisms on surfaces, the rate of inactivation is inversely proportional to the initial cell concentration (Shintani, 2000). Food irradiation is being considered an important tool, in ensuring safety and extending shelf-life of fresh meat and poultry (Yoon, 2003). Thus irradiation can eliminate food-borne pathogenic microorganisms in meat. Furthermore, the use of gamma irradiation as a safety technological treatment in food preservation has now become legally accepted in many countries of the world (Abdel-Daium, 2007).

Mathematical modeling is an effective way of representing a particular process. It can help us to understand and explore the relationship between the process parameters. Mathematical modeling can help to understand and quantitative behavior of a system. Mathematical models are useful representation of the complete system which is based on visualizations. Mathematical modeling is an important method of translating problems from real life systems to conformable and manageable mathematical expressions whose analytical consideration determines an insight and orientation for solving a problem and provides us with a technique for better development of the system.

The objective of the study is modeling of the total Spore Forming bacterial count as a function of the irradiation dose and the storage period of irradiated sliced chicken meat.

2 MATERIALS AND METHODS

²⁶Sliced chicken were purchased from local market (Benha, Qaliohia governorate, Egypt). All samples were transported to the laboratory food irradiation unit, Nuclear Research Center in ice-box (0°C) and surveyed for microbiological counts spore forming bacteria. Then, sliced chicken samples were packed in tightly sealed polyethylene pouches and divided into seven groups and stored in freezing till irradiation treatments.

2.1 Gamma irradiation treatments²⁶

Four bags from each of sliced chicken were gamma irradiated at 0, 2, 4, and 6 kGy doses using cobalt-60 gamma chamber (1.367 kGy/h) in Cyclotron Project, Nuclear Research Center Atomic Energy Authority, Inshas, Cairo, Egypt. After irradiation, all samples were stored at 4±1°C.

2.2 Microbial analysis²⁶

Spore forming bacteria count according to (FDA, 2002).

2.3 Statistical analysis²⁶

The statistical evaluation of the mean data was compared using one-way analysis of variance (ANOVA) according to Zar (1984). The chosen level of significance was $P \leq 0.05$.

The experimental data²⁶ obtain using the previous procedures were analyzed by the response surface regression procedure using the following higher-order polynomial equations:

like, $y = \beta_0 + \sum \beta_i x_i + \sum \beta_{ii} x_i^2 + \sum \beta_j x_j + \sum \beta_{jj} x_j^2 + \sum \beta_{ij} x_i x_j$, where y is the response, x_i and x_j are the uncoded independent variables (factors), and β_0 , β_i & β_j , β_{ii} & β_{jj} and β_{ij} are intercept, linear, quadratic, and interaction constant coefficients, respectively. Design Expert software package 8.0 was used for regression analysis, analysis of variance (ANOVA) and developing of models of different forms by transformation (linear and of higher order) based on above mentioned principles of forming a functions. Confirmatory experiments were carried out to validate the equations using the combinations of independent variables which were not part of the original experimental design but were within the experimental region. Various models were compared for the best fit summary and there R^2 values were compared to choose the best appropriated model for particular data design and selected runs. In this the Total Spore forming bacterial count was the response and the dependent two factors were the Storage time and the gamma-irradiation Dose given to the sliced chicken.

3 RESULT AND DISCUSSION

The result of statistical Analysis are shown below:

Table 1. shows the fit summary the models

Response	1	Spore formir Transform:	None		
*** WARNING: The Quartic Model and higher are Aliased! ***					
Summary (detailed tables shown below)					
Source	Sequential p-value	Lack of Fit p-value	Adjusted R-Squared	Predicted R-Squared	
Linear	0.0195		0.2966	-0.2306	
2FI	0.2241		0.3206	-0.3175	
Quadratic	0.0056		0.6298	-0.2218	
<u>Cubic</u>	<u>0.0177</u>		<u>0.8276</u>	<u>-0.1223</u>	<u>Suggested</u>
Quartic	0.0514		0.9278	-0.2326	Aliased

Table 2. Shows the model sum of square

Sequential Model Sum of Squares [Type I]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
<u>Mean vs Total</u>	<u>1.712E+007</u>	<u>1</u>	<u>1.712E+007</u>			<u>Suggested</u>
Linear vs Mear	3.233E+007	2	1.617E+007	5.01	0.0195	
2FI vs Linear	4.989E+006	1	4.989E+006	1.60	0.2241	
Quadratic vs 2	2.612E+007	2	1.306E+007	7.68	0.0056	
<u>Cubic vs Quad</u>	<u>1.588E+007</u>	<u>4</u>	<u>3.969E+006</u>	<u>5.01</u>	<u>0.0177</u>	<u>Suggested</u>
Quartic vs Cub	5.929E+006	4	1.482E+006	4.47	0.0514	Aliased
Residual	1.988E+006	6	3.314E+005			
Total	1.044E+008	20	5.218E+006			

Table 3. Model summary Statistics

Model Summary Statistics						
Source	Std. Dev.	R-Squared	Adjusted R-Squared	Predicted R-Squared	PRESS	
Linear	1797.09	0.3707	0.2966	-0.2306	1.074E+08	
2FI	1766.23	0.4278	0.3206	-0.3175	1.149E+08	
Quadratic	1303.66	0.7273	0.6298	-0.2218	1.066E+08	
<u>Cubic</u>	<u>889.81</u>	<u>0.9092</u>	<u>0.8276</u>	<u>-0.1223</u>	<u>9.790E+07</u>	<u>Suggested</u>
Quartic	575.64	0.9772	0.9278	-0.2326	1.075E+08	Aliased

"Model Summary Statistics": Focus on the model maximizing the "Adjusted R-Squared" and the "Predicted R-Squared".

Table 4. Shows the ANNOVA tables

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	7.932E+007	9	8.813E+006	11.13	0.0004	significant
A-storage pe.	2.716E+006	1	2.716E+006	3.43	0.0937	
B-GID	1.200E+006	1	1.200E+006	1.52	0.2464	
AB	1.592E+006	1	1.592E+006	2.01	0.1865	
A ²	7.339E+006	1	7.339E+006	9.27	0.0124	
B ²	83041.84	1	83041.84	0.10	0.7527	
A ² B	1.600E+006	1	1.600E+006	2.02	0.1855	
AB ²	17300.05	1	17300.05	0.022	0.8854	
A ³	1.395E+007	1	1.395E+007	17.62	0.0018	
B ³	69003.04	1	69003.04	0.087	0.7739	
Residual	7.918E+006	10	7.918E+005			
Cor Total	8.724E+007	19				

The Model F-value of 11.13 implies the model is significant. There is only a 0.04% chance that an F-value this large could occur due to noise.

Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case A², A³ are significant model terms.

Values greater than 0.1000 indicate the model terms are not significant.

Table 5. ANNOVA Summary

Std. Dev.	889.81	R-Squared	0.9092
Mean	925.20	Adj R-Squared	0.8276
C.V. %	96.18	Pred R-Square	-0.1223
PRESS	9.790E+007	Adeq Precisor	15.492

A negative "Pred R-Squared" implies that the overall mean is a better predictor of your response than the current model.

"Adeq Precision" measures the signal to noise ratio. A ratio greater than 4 is desirable. Your ratio of 15.492 indicates an adequate signal. This model can be used to navigate the design space.

The developed equation in terms of coded factor:

Final Equation in Terms of Coded Factors:

$$\begin{aligned} \text{Spore forming bacterial count} = & \\ & +1501.81 \\ & +6844.32 * A \\ & -2455.13 * B \\ & -8088.21 * AB \\ & +13155.88 * A^2 \\ & +525.45 * B^2 \\ & -5928.82 * A^2B \\ & +322.92 * AB^2 \\ & +7819.60 * A^3 \\ & -329.56 * B^3 \end{aligned}$$

The equation in terms of coded factors can be used to make predictions about the response for given levels of each factor. By default, the high levels of the factors are coded as +1 and the low levels of the factors are coded as -1. The coded equation is useful for identifying the relative impact of the factors by comparing the factor coefficients.

The developed Fit Suggested required Model:

Final Equation in Terms of Actual Factors:

$$\begin{aligned} \text{Spore forming bacterial count} = & \\ & +821.60580 \\ & +36.32223 * \text{storage period} \\ & -563.16208 * \text{GID} \\ & +69.41304 * \text{storage period} * \text{GID} \\ & -19.44036 * \text{storage period}^2 \\ & +132.35734 * \text{GID}^2 \\ & -8.78344 * \text{storage period}^2 * \text{GID} \\ & +2.39202 * \text{storage period} * \text{GID}^2 \\ & +2.31692 * \text{storage period}^3 \\ & -12.20606 * \text{GID}^3 \end{aligned}$$

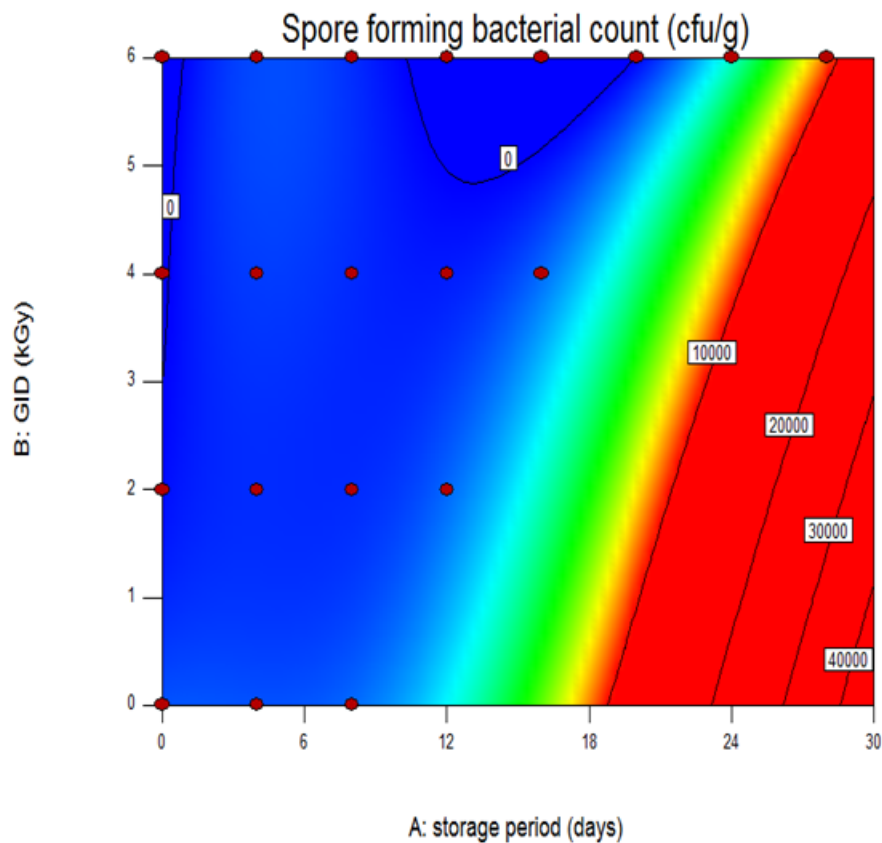


Fig 1. Shows the Contour Graph for the developed Model.

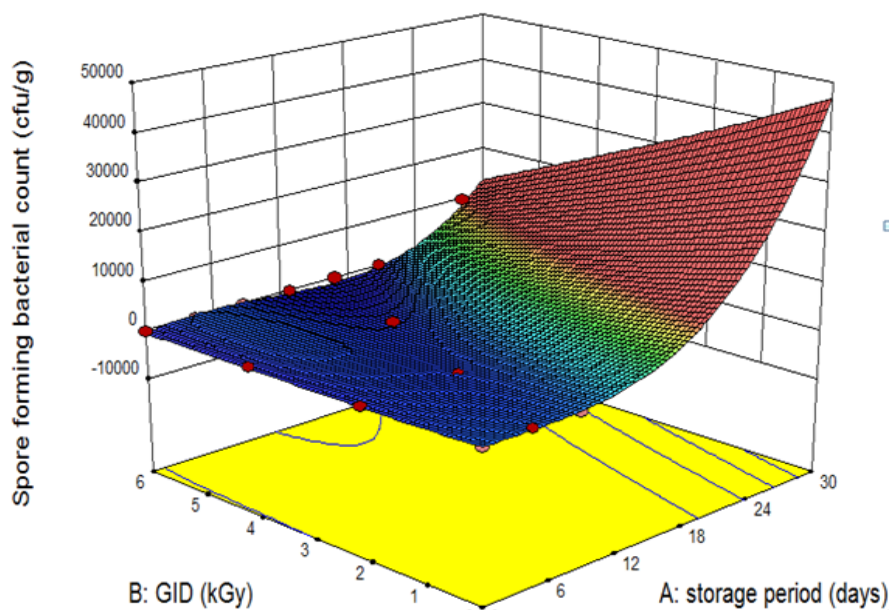


Fig 2. Shows the 3-D Graph Plotted between TSFB, GID and SP. For the developed model.

4 CONCLUSION

Thus we get a most fitted model for the function of total spore forming bacterial (TSFB) with gamma Irradiation dose (GID) and storage period (SP) as the two variants, with $R^2 = 0.9092$, F value 11.13 and P value <0.004 , the suggested model is more significant for the given design data set.

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