Modeling the Formulation and Shelf Life of Avocado (*Persea americana*) Fruit Spread

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ABSTRACT

The aim of this study was to use a systematic scientific mixture design to formulate avocado fruit spread in order to retain its aesthetic properties. A two level-factorial with four formulation factors; masses of avocado pulp, sugar, gum and miscellaneous were varied to give a total of eight coded products, which were quantitatively sensory evaluated by eight trained panelists. The data obtained were modeled and used to predict the fruit spread formulation as 50 g of avocado pulp, 5 g of sugar, 9 g of miscellaneous, 0.5 g of xanthan gum on the sensory scores of color (0.52), taste (0.64), aroma (0.71), spreadability (0.72) and finger feel (0.80). The shelf life studies were done by running three tests: Peroxide value, sensory evaluation of spread color intensity on a score card with 1-10 cm scale and total colony forming unit. Though the shelf lives were modeled on three parameters it was rather concluded on microbial safety consideration as 47.50 days at a refrigeration temperature since color and peroxide deterioration could be easily controlled by additives.

Key words: Avocado pear, fruit spread, product development, sensory optimization, spread shelf life, spread deterioration energy, optimum formulation

INTRODUCTION

There have been intensified campaigns by nutritionists against the consumption of high fat diets which is now discussed and backed by media analysts. In the process, many consumers have become aware of the dangers involved in piling up bad fats such as saturated fats in favour of unsaturated fats such as monounsaturated and polyunsaturated fats. Though, the facts still remains that some moderate amount of fat is needed in any healthy diet. One good reason is that there are a number of essential vitamins, such as vitamins A, D, E and K that are fat-soluble and therefore they can only be absorbed with fatty foods. Since the media education and lecture is so intense, the quest for alternative low fat spreads for vulnerable consumers is justifiable and worthy to address through research. A number of confectionery table spreads have been in production and these are the favourites of a large number of consumers. These confectionary table spreads are defined by the EFSA (2004), to be fat-based or made from fruits and vegetables. This definition includes products such as margarine, cheese and butter and those obtained from fruits and vegetables such as jams, preserves and marmalades. Cheeses which are made from curd produced from the coagulation of souring milk by rennin constitute yet another type to spread. Jams, jellies
and marmalades constitute yet another category of spreads that are saddled with numerous consumption problems centering on weight gain. Further consumption problems in the form of trans fats or saturated fats associated with spreads like cheeses and margarines, has always justified the search of alternatives. These novel spread are supposed to deliver the functionalities required in our traditional spreads which abound with consumption problems. Nutritionists recommend that we reduce the amount of fat, especially saturated fat in the human diet and this has resulted in the reduction in the consumption of full-fat products and a justifiable growing interest in low-fat spreads. The idea of producing spreads with a reduced fat content would continue in the laboratories of many foods related establishments, including the R and Ds of many food industries.

Many researches discuss the nutritional value of margarine and other spreads largely around two aspects: the total amount of fat and the types of fat (saturated fat, trans-fat) as components of the formulation. It has always been known that the saturated fatty acids in triglycerides contribute to elevated blood cholesterol levels (Mensink et al., 2003), which in turn has often been linked to cardiovascular diseases. Hayakawa et al. (2000) have indicated a strong link between early death and consumption of high amounts of trans-fats, which had been present in many spread formulations not quite too long ago. Trans-fats, which do not occur naturally in vegetable fats, are a consequence of partial hydrogenation of the fats, a requirement for some spread formulation procedures.

According to some researchers (Flöter and Duijn, 2006) many industries have gradually moved away from using partially hydrogenated oils since the mid-nineties and now produce new spreads that contain less or no trans fats. This is certainly good news but there is several tonnage of precious food raw materials in the form of avocado which rather go waste yearly because of their shelf instability. Avocado pear fruit (Persea americana) comes in handy because it has high levels of oleic acid (51-68%) according to Enig (2000). It is a seasonal fruit and native to the tropics and sub-tropics. The flesh of the fruit of the avocado as generally pale yellow-green and softly succulent with buttery consistency even though inferior varieties may be fibrous in nature. Extensive marketing systems studies has suggested that the global avocado market is about $7.5 billion in retail prices and that production would reach 4.7 metric tones by 2012 (Shumeta, 2010). The problem on hand would be to eliminate the shelf instability of the produce by presenting to consumers a processed product which ultimately eliminates any possibility of waste in the major season of the fruit while preserving the fruit. At the same time action will preserve the fruit and make the fruit and its nutritional value available to many people all year round.

To make inroads into the business of formulating and storage of this product for the lean season, it must be borne in mind that every food product faces constant thermodynamic instability (Van Boekel, 2005). Therefore careful considerations can be made along its microbial, biochemical and physical deteriorating pathways in other to control spoilage while in storage. The specific objectives of this research attempts to tackle just issues raised: to formulate avocado fruit spread using modeling of the sensory parameters data and formulation variables and to study the shelf life using three deteriorating parameters of the formulated avocado fruit spread packaged in glass bottles.

**MATERIALS AND METHODS**

**Materials:** Avocado (Persea americana) fruits used for this work were obtained from an out-grower at Ahinsan Estate in Kumasi, Ghana. Food grade citric acids and ascorbic acid were obtained from Sigma-Aldrich Inc., USA. Xanthan gum was obtained from BDH Chemicals Limited, UK. Sarsons
malt vinegar, sugar, Annapurna iodized salt, and Raysse vanilla essence was obtained from Opoku Trading Store, Kumasi, Ghana. Sodium benzoate and potassium sorbate were obtained from Scharlau Chemie S.A., Spain.

**Avocado pulp preparation:** The avocado fruits were hand-picked in November 2007 from the Akimesan Estate at commercial maturity and kept at room temperature for three days to complete the ripening process. They were then washed in the Department’s laboratory the same day under running potable water and subsequently soaked for 15 min in Milton’s chlorinated tablets. The fruits were peeled, deseeded and cut into four wedges using stainless steel knives and eventually into small pieces. They were finally pulverized using a food blender (Binatone 5080 MP, UK) at speed 4 and stored in plastic bags pending further investigation.

**Methods**

**Formulation of avocado spread:** A two-level factorial of four factors; masses of pear pulp, sugar, gum and miscellaneous were varied statistically (Design Expert, 2007) to give eight formulations. The miscellaneous comprised of the preservatives (sodium benzoate and potassium sorbate), citric acid, ascorbic acid, iodized salt, vinegar and vanilla essence (Table 1).

According to the particular experimental run, the specified amounts of the ingredients were weighed separately within three days after pulverizing in the laboratory. The pulp was placed in a bowl to which was added the ingredients, gum, sugar, salt and preservatives in that order. This was then homogenized in a Hand mixer (Binatone CA 3090, UK) starting at speed 1 and accelerating to speed 5. Vanilla essence and vinegar were thoroughly mixed and added to the puree. Mixing then continued until a smooth paste was achieved. Approximately 20 g of the product was then scooped into previously sterilized (chlorine tablets washings followed by heating between 75-85°C) transparent glass bottles, pasteurized in a water bath (Scientz, SC-15) at a temperature of 65°C for 5 minutes, sealed and labeled as M1 (654), M2 (732), M3 (754), M4 (296), M5 (254), M6 (336), M7 (963) and M8 (557).

**Sensory evaluation of formulated spread**

**Training of sensory panel:** For the quantitative descriptive test, eight subjects who have had previous training at the Council for Scientific and Industrial Research (CSIR), Food Research Institute, Accra were used. These assessors were further trained for 2 h per week for a two-week period, using model products like margarine, groundnut paste and hard cheese to classify the selected parameters for this study: Color, taste, aroma, spreadability and finger feel as defined in Table 2.

**Table 1: Quantities of ingredients used for the eight formulations**

<table>
<thead>
<tr>
<th>Runs</th>
<th>A: Avocado (g)</th>
<th>B: Sugar (g)</th>
<th>C: Gum (g)</th>
<th>D: Mise (g)</th>
<th>E: Grace (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>40</td>
<td>5</td>
<td>5</td>
<td>1.0</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>50</td>
<td>5</td>
<td>9</td>
<td>0.5</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>40</td>
<td>1</td>
<td>9</td>
<td>0.5</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>50</td>
<td>5</td>
<td>9</td>
<td>1.0</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>50</td>
<td>1</td>
<td>5</td>
<td>0.5</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>50</td>
<td>1</td>
<td>5</td>
<td>1.0</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>40</td>
<td>1</td>
<td>9</td>
<td>1.0</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>40</td>
<td>5</td>
<td>5</td>
<td>0.5</td>
<td></td>
</tr>
</tbody>
</table>
Table 2: Definition of sensory parameters as used in the text

<table>
<thead>
<tr>
<th>Sensory parameter</th>
<th>Definition of sensory parameter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Color</td>
<td>Green to yellow</td>
</tr>
<tr>
<td>Taste</td>
<td>Uncharacteristic pear taste to characteristic pear taste</td>
</tr>
<tr>
<td>Aroma</td>
<td>Unpleasant to pleasant smell</td>
</tr>
<tr>
<td>Spreadability</td>
<td>Aggregate to easy-to-spread</td>
</tr>
<tr>
<td>Finger feel</td>
<td>Rough to smooth-to-touch</td>
</tr>
</tbody>
</table>

**Sensory evaluation of samples:** The sensory evaluation was conducted using eight of the trained panelists, employing the graphic scale methodology (Larmond, 1977), which run from 0 to 1. A general factorial design with two factors: products (A) (coded from M1 to M8) and panelists (B) (coded from P1 to P8) were used to plan the serving order for the sampling of the eight products. The panelists were provided with unsalted cream crackers and water to rinse their mouths between tests.

**Statistical methods**

**Fitting the data collected:** The sensory evaluation data collected were loaded and fitted to models that could explain the behaviour of the sensory parameters of the avocado spread. This involved studying such coefficients as the regression $(R^2)$, adjusted regression $(\text{adj } R^2)$, prediction regression $(\text{pred } R^2)$ and adequate precision $(\text{adeq precision})$ of the models selected. After fitting and ANOVA studies, diagnostic plots were carried out where the normal probability plot of the studentized residuals were analyzed to check for normality of residuals. To check for constant error, the studentized residuals against predicted values were studied. Subsequently, externally studentized residuals were also studied to look for outliers or the influential factors after. The Box-Cox plot for power transformations were investigated to see the need for any transformation operations into pre-treated data that could give a better interpretation of the model. When all the model statistics and diagnostic plots were evaluated to be good, the model graphs were obtained.

**Optimizing the parameters of the sensory data:** The color of the fruit spread was set at midpoint of yellowish-green, because avocado fruit has shades of both green and yellow. The rest of the sensory parameters were set at maximum. Since it was prudent for the statistical tool to select the optima based on the constraints imposed, the ingredients and their respective values were set in range. For the interactions between the products and the panelists, the lower and upper limits were M1 (654) and P2 and M8 (657) and P8 respectively. As shown in Table 3, the scores for the lower limits of the sensory parameters ranged between 0.05 in spreadability to 0.24 in finger feel. The upper limits had scores ranging from 0.75 in aroma to 0.88 in finger feel. At the end of the sensory evaluation optimisation process, sample M2 (732) was rated the best with panelist P8 as a the best assessor. Sample M2 (732) was thus formulated and used for the rest of the study.

**Analytical methods**

**Determination of peroxide value:** Peroxide value determination proposed by Kirk and Sawyer (1991) was determined by first extracting the oil from the avocado fruit spread according to the method described by Ilkuoria and Maliki (2007). In this method, a volume of 100 mL of chloroform was added to 50 g of the sample, stoppered and vortexed thoroughly at 150 rpm with a Griffin flask shaker (M008B, Mumbai-India) for 30 min and repeated five times. The extracts were pooled in a conical flask and left opened for most of the chloroform to evaporate and the oil siphoned.
Table 3: Constraints showing the importance of factors and responses goals set for optimization

<table>
<thead>
<tr>
<th>Name</th>
<th>Goal</th>
<th>Lower limit</th>
<th>Upper limit</th>
<th>Importance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Products</td>
<td>Is in range</td>
<td>M1(654)</td>
<td>M8(657)</td>
<td>3</td>
</tr>
<tr>
<td>Panelist</td>
<td>Is in range</td>
<td>F1</td>
<td>F8</td>
<td>3</td>
</tr>
<tr>
<td>Color</td>
<td>Is target = 0.52</td>
<td>0.21</td>
<td>0.82</td>
<td>5</td>
</tr>
<tr>
<td>Taste</td>
<td>Maximize</td>
<td>0.23</td>
<td>0.80</td>
<td>5</td>
</tr>
<tr>
<td>Aroma</td>
<td>Maximize</td>
<td>0.18</td>
<td>0.75</td>
<td>5</td>
</tr>
<tr>
<td>Spreadability</td>
<td>Maximize</td>
<td>0.05</td>
<td>0.83</td>
<td>5</td>
</tr>
<tr>
<td>Finger feel</td>
<td>Maximize</td>
<td>0.24</td>
<td>0.88</td>
<td>5</td>
</tr>
</tbody>
</table>

Three grams of the extracted oil was weighed into a previously cleaned and dried 250 mL stoppered conical flask and 10 mL of chloroform added to solubilize the fat. After vortexing with 15 mL of glacial acetic acid and 1mL fresh saturated aqueous potassium iodide the flask was immediately stoppered and shaken for 1 min and placed in the dark for 5 min. It was later tipped with 75 mL of distilled water, mixed and titrated with 0.01M sodium thiosulphate solution using 1% soluble starch solution as indicator. The peroxide value was calculated as:

\[
\frac{(V-V_0)T}{M} \times 10^6 \text{mEq kg}^{-1}
\]

where, \(V-V_0\) is the titre value, \(T\) is the exact molarity of the sodium thiosulphate solution and \(M\) is the mass in grams of the sample.

**Color determination of sample formulation:** Fifty grams of the sample spread were placed in petri dishes, photographed using a digital camera (564 MCD) and printed on a printer (HP 3900, UK). Eight trained panelists were then made to measure the color intensity on a sensory evaluation card with a scale running from 1 to 10 at the end of the final day of sampling.

**Microbial load:** A sub-sample mass of 1.00 g of the sample fruit spread was prepared by mixing with 1mL of distilled water. Six test tubes labeled \(10^{-1}\) to \(10^{-6}\) were then filled with 9 mL of Ringers solution (used as diluents) after which 1 mL of the mixture was pipetted into the first tube labeled \(10^{-1}\). The \(10^{-2}\) dilution was subsequently obtained by taking 1 mL of the \(10^{-1}\) dilution solution into the \(10^{-2}\) labeled tube and the rest of the serial dilutions were similarly done. Using a fresh sterile pipette tip for each dilution, 1 mL each of the six dilutions was aseptically added to six test tubes containing molten plate count agar at 45°C. This was mixed by rotating the tubes between the palms taking care to avoid the formation of bubbles. The agar was then aseptically poured into fresh petri dishes, which were labeled \(10^{-1}\) to \(10^{-5}\) allowed to solidify and incubated in inverted positions at 37°C for 24 h and Electronic colony counters (Fisher Scientific 7-910, Canada) used to count the colonies formed.

**Analysis of shelf life:** The results obtained were loaded and analyzed by plotting the polynomial regression in a statistical tool (Stat Point, 2008) and the data fitted to models. The order was deemed adequate where the largest R-squared statistics, interpreted as the percentage of the variability in the response variable which has been explained by the model was significant; i.e., \(p<0.05\). Such models were subsequently chosen to calculate the shelf-lives.
Calculating shelf life

**Peroxide value:** Kirk and Sawyer (1991) reported that a rancid taste is noticeable when the peroxide value is between 20 and 40 mEq kg\(^{-1}\) in oily products. For this study, the limit for rancidity was pegged at 10 mEq kg\(^{-1}\). Furthermore, based on Polhemus (2005) deduction that in predicting shelf life, the largest number of weeks for which the degrading parameter has reached 90% must be used translated into 9 mEq kg\(^{-1}\).

**Fruit spread color:** The scale used during the sensory descriptive test for color determination ranged from 1 to 10 indicating color shade from yellow to green. For this study, the midpoint of the highest color intensity was used in calculating for the shelf life reflecting the yellowish green color of the avocado flesh. The highest value from the study was 9.5, half of which was 4.75. With reference to Polhemus (2005) deduction for shelf life determination, 90% of 4.75, which is 4.275, were used for the shelf life determination.

**Calculating** \(Q_{t0}\): The activation energy or the deterioration energy, \(E_a\) of the fruit spread which represented the inertia that ought to be overcome in order for spoilage reactions to occur, were then calculated by first determining the \(Q_{t0}\) according to the Labuza (1984) procedure as:

\[
Q_{t0} = \frac{\text{shelf life}(t)}{\text{shelf life}(t+10)}
\]

\[E_a = \log (Q_{t0}) \times t [1 + 10] 4.57 \times 10^4\]

**RESULTS AND DISCUSSION**

**Fitting the model for the sensory parameters:** To explain the behaviour of the responses against the variable factors, a summary of the statistics of model fitted, indicated the standard deviation, the \(R^2\), adjusted \(R^2\) and predicted \(R^2\). These were indicators of how well the selected sensory models fitted the data. For goodness-of-fit, low standard deviations, \(R^2\) near 1 were desired (Design Expert, 2007).

**Optimizing the sensory evaluation of the process:** For the fruit spread color (Table 4), the predictive \(R\)-squared of 0.36 obtained is in reasonable agreement with the adjusted \(R\)-squared of 0.51. The negative predictive \(R\)-squared obtained for taste (-0.22), aroma (-0.01), spreadability (-0.13) and finger feel (-0.38) implied the overall mean was a better predictor of the response than those models obtained for the sensory parameters under investigation. According to Design Expert (2007) adequate precision measures the signal to noise ratio and a ratio greater than 4 is desirable.

<table>
<thead>
<tr>
<th>Sensory parameters</th>
<th>Mean</th>
<th>CV (%)</th>
<th>(R^2)</th>
<th>Adj (R^2)</th>
<th>Pred (R^2)</th>
<th>Adep precision</th>
</tr>
</thead>
<tbody>
<tr>
<td>Color</td>
<td>0.50</td>
<td>21.79</td>
<td>0.62</td>
<td>0.51</td>
<td>0.36</td>
<td>9.31</td>
</tr>
<tr>
<td>Taste</td>
<td>0.56</td>
<td>27.31</td>
<td>0.38</td>
<td>0.08</td>
<td>-0.22</td>
<td>4.15</td>
</tr>
<tr>
<td>Aroma</td>
<td>0.53</td>
<td>21.59</td>
<td>0.41</td>
<td>0.24</td>
<td>-0.01</td>
<td>5.45</td>
</tr>
<tr>
<td>Spreadability</td>
<td>0.60</td>
<td>26.56</td>
<td>0.33</td>
<td>0.15</td>
<td>-0.13</td>
<td>4.74</td>
</tr>
<tr>
<td>Fingerfeel</td>
<td>0.70</td>
<td>16.38</td>
<td>0.19</td>
<td>-0.04</td>
<td>-0.38</td>
<td>4.21</td>
</tr>
</tbody>
</table>

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Therefore the ratios of 9.31, 4.15, 5.45, 4.74 and 4.21 obtained for color, taste, aroma, spreadability and finger feel respectively indicated adequate signal or precision suggesting that their models could be used to navigate the design space and subsequent optimization.

**Color:** For Fig. 1, the averages for color of spreads ranged between 0.3625 and 0.6675 on a scale of 1.0 and they were evenly distributed. The average scores for sample M1 (654) and sample M8 (657) were statistically different from each other whereas the rest of the products were not significant (p<0.05). In terms of the yellowish-green color of the avocado fruit spread, the panelists rated sample M8 (657) as the best and sample M1 (654) as the least acceptable. The implication is that sample 8 retained most of the yellowish-green color of the avocado fruit after the processing.

**Taste:** The average scores for taste (Fig. 2) ranged between 0.515 and 0.6575. The values for the first three samples M1 (654), M2 (732) and M3 (754) appeared to have similar values. There was however, a gradual decrease in the scores from sample M6 (336) to M8 (657). Samples M5 (254) and sample M4 (296) were respectively rated as the worst and best in terms of taste by the panelists but were statistically insignificant (p<0.05).

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![Color](image1)

*Fig. 1: Results of yellowish-green intensities of avocado spread color as examined by the panellists*

![Taste](image2)

*Fig. 2: Results of taste sensory intensities of avocado spread examined by the panellist*
Aroma: The average scores for aroma had a wide range from 0.465 to a little over 0.6075. There was a general decrease in the average scores from sample M2 (732) to sample M8 (657). Sample M2 (732) was rated as the best in terms of aroma. Sample M6 (638), on the other hand, received the lowest rating by the panelists in terms of its aroma as Fig. 3 shows. Sample M2 (732) was statistically significant compared to all the products from M5 (254) to M8 (657).

Spreadability: As shown in Fig. 4, the average scores for spreadability ranged between 0.44 and a little above 0.635. Sample M7 (963) was adjudged most spreadable by the panelists as against sample M8 (657) being the best in terms of spreadability though they were not statistically significant.

Finger feel: As indicated in Fig. 5, a little below 0.72 and 0.88 were the range for the average scores of the finger feel. There was a gradual score decrease from sample M1 (654) to sample M7 (963) after which it increased sharply in sample M8 (657). Sample M1 (654) was the best, according

![Fig. 3: Results of aroma intensities of the avocado spread examined by the panellists](image)

![Fig. 4: Results of spreadability intensities of the avocado spread examined by the panellists](image)
Table 5: Results of optimization showing the overall acceptability when the constraints were imposed

<table>
<thead>
<tr>
<th>Products</th>
<th>Panelist</th>
<th>Color</th>
<th>Taste</th>
<th>Aroma</th>
<th>Spreadability</th>
<th>Fingerfeel</th>
</tr>
</thead>
<tbody>
<tr>
<td>M2 (732)</td>
<td>P8</td>
<td>0.52</td>
<td>0.64</td>
<td>0.71</td>
<td>0.72</td>
<td>0.80</td>
</tr>
</tbody>
</table>

Table 6: Shelf lives (days since produced) of the selected responses: peroxide value, spread color and total colony forming units at ambient and refrigerated conditions after fitting a first or a second order polynomials

<table>
<thead>
<tr>
<th>Responses</th>
<th>Days since produced</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total cfu (Ambient)</td>
<td>37.49</td>
</tr>
<tr>
<td>Total cfu (Refrigerated)</td>
<td>47.50</td>
</tr>
<tr>
<td>Peroxide value (Ambient)</td>
<td>40.26</td>
</tr>
<tr>
<td>Peroxide value (Refrigerated)</td>
<td>70.15</td>
</tr>
<tr>
<td>Spread color (Ambient)</td>
<td>25.91</td>
</tr>
<tr>
<td>Spread color (Refrigerated)</td>
<td>54.42</td>
</tr>
</tbody>
</table>

Fig. 5: Results of finger feel intensities of avocado spread examined by the panellists
to the panelists, whilst sample M7(963) was adjudged the least. Sample M1 (654) and sample M7 (963) were not statistically significant.

**Optimizing the process:** Producing an avocado spread that optimizes the sensory parameters (Table 5) was the principal aim of this study. In order to achieve this, the sensory parameters were constrained according to our goals as presented in the Table 3.

**Shelf life:** Shelf life represents the period of time through which food products remain safe to eat and retains its essential sensory properties and complies with the label’s nutritional declarations (Doughari et al., 2007). Many products have a limited shelf life because as soon as they are produced, changes in their wholesomeness begin to occur and after some period, the product loses its effectiveness and therefore must be pulled from the shelf. The output of the analysis (Table 6) showed the results of fitting a second order polynomial model to describe the relationship between the responses and days since produced apart from the spread color at ambient condition, which presented a first order. The R-squared of all the models fitted could explain between 94.57 to 98.99% of the variability in the responses studied and the models generated confirm the adequacy of the orders of polynomials (p<0.05).
Fig. 6: Polynomial regression of total cfu at ambient and refrigerated conditions and days since produced

**Microbial:** There was a steady rise in the microbial numbers as the days increased until it tapered off and subsequently declined (Fig. 6) for the refrigerated and ambient curves studied. As was expected, the refrigerated samples slowly deteriorated compared to the samples stored at ambient conditions. From Fig. 6, the shelf life was deduced from the point at which the curve tapered off since the growth was no longer following a linear progression. Following from the Polhemus (2005) deduction, the shelf life was determined at a cfu value of 90% of the tapering off value of 38 cfu which was calculated to be 34.2 cfu. The locate button in the statistical tool (Stat Point, 2008) was then used to locate 34.2 cfu on the curve which gave a shelf life of 37.49 days. For the refrigerated samples, the curve tapered off at 22.8 cfu and 90% of this value gave a total cfu of 20.52 and a shelf life of 47.50 days.

**Peroxoide value:** Peroxide Value (PV) measured the formation of intermediate hydroperoxides in milliequivalents of active oxygen per kilogram of sample. Fresh oils have values less than 10 mEq kg\(^{-1}\) and values below 10 characterize the majority of conventional oils (Kirk and Sawyer 1991). The data obtained for this work were to be expected since the rate of increase of peroxide concentration for the refrigerated samples were lower than the rate of increase for the ambient samples (Fig. 7). Kirk and Sawyer (1991) reported that a rancid taste was noticeable when the peroxide value was between 20 and 40 mEq kg\(^{-1}\) in oily products. However, from the data obtained from this work, the limit for rancidity was pegged at 10 mEq kg\(^{-1}\) since that was a reasonable point where the two curves could be well compared. Deductions at 90% (Polhemus, 2005) of this peroxide value 10 gave 9 mEq kg\(^{-1}\) which was set as the limit. Using the locate button as before, a PV value of 9 mEq kg\(^{-1}\) gave shelf lives of 70.15 and 40.26 days, respectively for the refrigerated and ambient samples. These shelf life values were however, lower than those obtained by Maskan et al. (2007), who recently evaluated the shelf life of margarine samples coded as A and B to be 145 and 134 days, respectively. These shelf lives were
Fig. 7: Polynomial regression of peroxide value at ambient and refrigerated conditions and days since produced

Fig. 8: Regression of spread color at ambient and refrigerated condition and days since produced determined using a similar parameter as peroxide but the values determined in this work were much lower probably due to the presence of more rancid oils in the spread.

**Fruit spread color:** The study indicated that as the number of days increased, the yellowish-green color of the avocado fruit spread generally decreased (Fig. 8). As might be expected, the rate of decrease was faster at the ambient rather than for the refrigerated condition. The scale used during the sensory descriptive test for color determination ranged from 1 to 10 and presented color shades from yellow to green. For this study, the midpoint of the highest color intensity reflecting the yellowish green color of the avocado flesh was used in calculating for the shelf life. The highest value for the two curves from the study was 9.5, half of which gave 4.25, of which 90% target (Polhemus, 2005) gave 4.275. The locate button subsequently projected the shelf lives of 69 and 30.1 days for refrigerated and ambient samples respectively. The retention of the inherent color of
fresh vegetables is often used as a quality indicator and has a substantial impact on consumer acceptance (Roura et al., 2000) and therefore steps should be taken to control the degradation of color of foods.

In this research the degradation of color of the avocado fruit spread could most likely be related to the degradation of chlorophyll, (Lazcano et al., 2001). Since chlorophyll is known to have higher degradation rate at higher temperatures and availability of oxygen, compared to lower temperatures (Taoukis et al., 2001), it is intended that anti browning agents such as cysteine could be used to mitigate these occurrences.

**Q\textsubscript{10} and activation energy:** Chemical deterioration reactions require a certain amount of energy called activation energy (Ea), to get started. The Ea is in fact the energy barrier that molecules need to overcome to be able to react (Petrou et al., 2002; Van Boekel, 2005). The higher the Ea of a reaction, the greater its acceleration with increases in temperature. A simple way to express this acceleration is the use the Q\textsubscript{10} concept (Labuza, 1984). The parameter Q\textsubscript{10} is the increase in the rate of the reaction when the temperature is increased by 10°C (18°F). Van Boekel (2005) argued that the Q\textsubscript{10} parameter shows the temperature dependence of a reaction as the factor by which the reaction rate is doubled when the temperature is increased by 10°C. He further argued that this value could be calculated from the data of most storage tests where the product has been stored at two or more temperatures regardless of whether or not they are 10°C apart. Using the formula proposed by Labuza (1979), the Q\textsubscript{10} parameter was calculated for the peroxide and color values, since these two parameters constitute the chemical reactions. The values obtained were then used to determine the activation energies for degradation of the spread. The Q\textsubscript{10} and Ea for peroxide and spread color responses are as presented in Table 7.

Now, apart from microbial issues, the color and rancidity of the spread could be controlled physically, therefore, it is submitted that the shelf life should be based on safety issue which is the microbial load, whilst the quality issues should be based on the peroxide values. The spread color could guide the aesthetic or the final commercial presentation of the product since color enhancers or coloring agents can be incorporated. The shelf stability of the avocado fruit spread was therefore pegged at 47.50 days under refrigeration conditions. Further studies would be mounted to observe the effect of opening and closing at refrigerating temperatures in a subsequent research.

**CONCLUSION**

The avocado fruit spread sample that recorded the best overall acceptability was M2 (732) which was formulated as 50 g of avocado pulp, 5 g sugar, 9 g miscellaneous, 0.5 g xanthan gum. These recordings were based on the sensory values of color, taste, aroma, spreadability and finger feel of respectively 0.52, 0.64, 0.71, 0.72 and 0.80 scaling from 0 to 1. The shelf stability of the avocado fruit spread was estimated to be 47.50 days under refrigeration conditions. However, the Q\textsubscript{10} and activation energy (Ea) of degradation values were highest for spread color as 2.1 and 13.17 kcal mol\textsuperscript{-1}, respectively.
REFERENCES
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