Predictive modelling for shelf life determination of nutriceereal based fermented baby food

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Abstract A shelf life model based on storage temperatures was developed for a nutriceereal based fermented baby food formulation. The formulated baby food samples were packaged and stored at 10, 25, 37 and 45 °C for a test storage period of 180 days. A shelf life study was conducted using consumer and semi-trained panels, along with chemical analysis (moisture and acidity). The chemical parameters (moisture and titratable acidity) were found inadequate in determining the shelf life of the formulated product. Weibull hazard analysis was used to determine the shelf life of the product based on sensory evaluation. Considering 25 and 50 % rejection probability, the shelf life of the baby food formulation was predicted to be 98 and 322 days, 84 and 271 days, 71 and 221 days and 58 and 171 days for the samples stored at 10, 25, 37 and 45 °C, respectively. A shelf life equation was proposed using the rejection times obtained from the consumer study. Finally, the formulated baby food samples were subjected to microbial analysis for the predicted shelf life period and were found microbiologically safe for consumption during the storage period of 360 days.

Keywords Baby food · Consumer study · Predictive modelling · Shelf life · Weibull hazard analysis

Introduction

Shelf life may be defined as the time period within which the food is considered safe for consumption. In general, a

manufacturer tries to develop a product with the longest shelf life possible, consistent with cost, handling ease and use by distributors, retailers and consumers. Consumer acceptability of a food product is a critical parameter to determine its shelf life. Insufficient shelf life leads to consumer dissatisfaction or complaints. Such dissatisfaction not only affects the acceptance and sales of the product as well as the brand, but also can lead to malnutrition and severe health problems (Freitas and Costa 2006). This is reason enough for food processors to pay enormous attention to adequate storage stability or shelf life of the product.

Powdered baby food products have low moisture content and are generally microbiologically stable products. The shelf life of such products can either be defined by the chemical changes occurring in them and/or by the sensory characteristics of the product. Chemical changes occurring in the product are not easily recognized by the common consumers. On the other hand, the sensory properties of a product are easily identified and analysed by them. Thus, in such products, consumer decides if they would consume a food product after a particular storage period (Gambaro et al. 2006). Consequently, consumers study is the most suitable means of determining the shelf life of a baby food (Hough et al. 2003), by definition, the consumers are babies, however in case of baby food, mothers often purchase and are the first one to try and decide whether or not to give it to their children.

Various methods could be used to determine the sensory shelf life of a food product. One such methodology determines the sensory shelf life as the time required for the overall acceptability scores of the product to fall below a certain predetermined value. However, this will not mean that at that time, the consumers would refuse to consume the food product altogether (Gambaro et al. 2006). Survival analysis is a branch of statistics widely used in clinical studies, epidemiology, biology, sociology and reliability studies (Kleinbaum 1996; Meeker and Escobar 1998; Klein and Moeschberger

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1997; Gomez et al. 2001; Gambaro et al. 2004; Araneda et al. 2008; Cruz et al. 2010). Hough et al. (2003) introduced this method for shelf life estimation of food material. The key concept of survival analysis was to focus the shelf life hazard on the consumer rejecting the product, rather than on the deterioration of the product.

Consumer panel appear to be the most appropriate tool to determine the shelf life and quality of a food product. However, assembling consumer panels for repetitive multiple measurements is impractical and expensive. On the other hand a sensory panel is more suitable for repeated measurements; however its results would be more analytical and will not reflect the consumer's perception. Therefore by correlating the consumer data with that of the data obtained from the trained panel a fair estimation of the shelf life of the food product could be obtained (Gambaro et al. 2006).

The nutriceal based fermented baby food was developed using germinated and fermented blends of cereals viz. pearl millet (*Pennisetum glaucum*), sorghum (*Sorghum bicolor*) and oat (*Avena sativa*), along with rice-corn cooked flour, whole milk powder, whey protein concentrate and sugar. In the present study, the developed baby food formulation stored at different temperatures viz. 10, 25, 37 and 45 °C was subjected to sensory analysis by consumer and trained panels. The shelf life determined by sensory analysis was supported by microbiological analysis of the product, to make sure the product is safe for consumption.

Materials and methods

Sample preparation

The nutriceal based fermented baby food was manufactured using the method optimized by Rasane et al. (2013), germinated pearl millet (*P. glaucum*), oat (*A. sativa*) and sorghum (*S. bicolor*) flour, blended into a ratio of 40 % pearl millet: 50 % oat: 10 % sorghum fermented with *Lactobacillus plantarum* (NCDC 20), obtained from National Dairy Research Institute (NDRI), Karnal, India. The fermented slurry was dried using tray and vacuum drying techniques and then ground to fine powder. The optimized baby food formulation had 3.60 ± 0.10 % moisture, 58.70 ± 0.10 % carbohydrate, 23.10 ± 0.10 % protein, 12.00 ± 1.01 % fat and 2.60 ± 0.10 % ash. The proximate composition including moisture content was determined using standard American Official Analytical Chemists (AOAC) procedures (AOAC 2000). The titratable acidity was determined using the method used by Sanni et al. (1999).

Storage temperatures and sampling times

The baby food samples were packaged in metalized polyester thick laminate pouches. The packaging material was procured

from Siddarth Marketing, Bangalore, India. The packaged samples were stored at four different temperatures viz. 10, 25, 37 and 45 °C. Regular sampling at an interval of 30 days was done for sensory analysis for 180 days and microbial analysis for total of 360 days.

Sensory analysis

The nutriceal based fermented baby food samples were subjected to sensory evaluation using 9-point hedonic scale. The sensory evaluation was performed by a panel of 10 semi-trained judges (the panel members were given a pre idea about sensory attributes of the product such as color, body and texture, flavour and mouthfeel, using a standard branded product available in the market) (5 male and 5 female, aged 25–35 years) from the Centre of Food Science and Technology, Banaras Hindu University, Varanasi, India. The baby food samples were served in plastic odorless trays. Water was used for rinsing between samples. All the analyses were conducted in triplicate. The sensory evaluation was carried out at 25 °C and 60 % relative humidity.

Consumer studies

Fifty mothers, experienced in purchasing baby food, of age 25–45 years were recruited from in and out of the campus of Banaras Hindu University, Varanasi, India. The testing was carried out in five sessions. For each sample, consumers had to score for the color and appearance, body and texture, flavor and sweetness, mouthfeel and overall acceptability of the product using a 9 point hedonic scale, in which 9=like very much and 1 = dislike very much. They also had to answer the question “would you normally give this product to your child?” with a yes or a no. A total of 200 samples of the formulated baby food stored at 10, 25, 37 and 45 °C were analysed using the consumer analysis. The censoring of consumer data was performed as explained by Hough et al. (2003). If for example, a consumer accepted all the samples stored for less than 30 days and rejected all the samples stored for more than 90 days, his/her rejection time was interval censored between 30 and 90 days. If a consumer accepted all samples he/she tried, then his/her rejection time is larger than the last time observed and it is therefore right censored. If a consumer accepted the fresh sample and rejected all the other his/her rejection time was left censored. If a consumer rejected the fresh sample, their results were excluded from the study.

Microbial analysis

The samples stored at 10, 25, 37 and 45 °C was analysed for total plate count, yeast and mould count and coliform count using plate count agar (PCA), potato dextrose agar (PDA) and

violet red bile agar (VRBA), respectively. The presence or absence *Clostridium botulinum*, *Salmonella sp* and *Staphylococcus aureus* was confirmed using reinforced clostridial agar, bismulth sulphite agar and mannitol salt agar, respectively for standard plate count method. The medium used for the all the microbial tests were procured from Himedia Laboratories Private Ltd., Mumbai, India.

Statistical analysis

Duncan multiple range test was performed for data obtained from microbial analysis to measure the test of significance between samples stored at different temperatures by post hoc test using SPSS 16.0 software (SPSS Italia, Bologna, Italy).

Shelf life studies

Regression analysis between natural log values of storage period and natural log values of median rank ($\ln$ (storage period) vs $\ln$ ($1/1$ -median rank)) was performed to estimate the characteristic life and shape parameter. Median rank (MR) was estimated using the following Benard's approximation equation (Abernethy 2002):

$$MR = \left(\frac{R - 0.3}{n - 0.4} \right) \quad (1)$$

Here, R =rank given to the storage period according to consumer rejection, n =total number of ranks. Least rank was given to the storage period at which the predetermined value for overall acceptability of the product was reached at respective temperatures (10, 25, 37 and 45 °C). The highest rank was obtained by the storage period which gained the maximum overall acceptability value.

Survival analysis

Survival analysis was used to estimate the shelf life of the nutriceal based fermented baby food using the results obtained from consumers when asked if they would normally give the samples to their children (Hough et al. 2003; Gambaro et al. 2004, 2005, 2006). The methodology is primarily focused on the shelf life hazard on the consumer rejecting the product and not on the product deterioration. The survival analysis was carried out using MS-EXCEL 2010 software (Microsoft Corporation, Redmond, Washington, USA). Discrete statistical distribution (Weibull, lognormal, gamma and exponential) were fitted to the data obtained in the consumer test and the best fit (obtained by a visual inspection of the curves, since the data was discrete, thus the random variable t in (Eqs. 2 and 3) was never observed exactly, due to presence of censor) was used to express $F(t)$. For example, if a two parameter Weibull distribution is chosen for t , defining a

random variable ' t_r ' as the storage time at which the consumer rejects the sample, the cumulative distribution function $F(t)$ could be defined as the probability of a consumer rejecting a product before t , that is $F(t)=p(t_r < t)$. Choosing a Weibull distribution for t (Klein and Moeschberger 1997; Lindsay 1998), the rejecting function is given by Eq. 2.

$$F(t) = 1 - e^{-\left(\frac{t}{\beta}\right)^\alpha} \quad (2)$$

$$\ln \left[\ln \left(\frac{1}{1 - F(t)} \right) \right] = \alpha \ln t - \alpha \ln \beta \quad (3)$$

The Weibull shape parameter, called α indicates whether the rejection rate is increasing, constant or decreasing. The Weibull characteristic life, called β is a measure of the scale, or spread in the distribution of data.

The shelf life of the nutriceal based fermented baby food was obtained by substituting the parameters found in the previous fit followed by considering 25 and 50 % consumer rejection (Hough et al. 2003; Gambaro et al. 2006; Gimenez et al. 2007; Cruz et al. 2010).

Results and discussion

Microbial safety of the baby food samples

All the baby food samples stored at 10, 25, 37 and 45 °C, used for sensory analysis were subjected to microbial analysis. Table 1 depicts the data obtained from microbial analysis for nutriceal based fermented baby food for the storage period of 180 days. It can be seen that the total plate count and yeasts and molds count for samples stored at 10 °C showed the lowest microbial load as compared to the samples stored at 25, 37 and 45 °C for the total storage period of 180 days, however, there was no significant difference observed in the microbial quality. No coliforms and organisms such as *Clostridium botulinum*, *Salmonella spp.* and *Staphylococcus aureus* were detected in the samples. In view of these results, the ethical committee of Centre of Food Science and Technology, Banaras Hindu University, Varanasi, India, decided that all the samples were adequate for tests on humans.

Preliminary studies

The correlation between the sensory scores obtained from the semi-trained panel and the consumer panel. It correlation data suggests that there is high degree of positive correlation ($r^2=0.969$) between the scores obtained from semi-trained and

Table 1 Microbial analysis of nutriceal based fermented baby food during the test storage period

Storage period (Days)	10 (°C)	25 (°C)	37 (°C)	45 (°C)
Total plate count (log CFU/g of sample)				
0	ND	ND	ND	ND
30	ND	ND	2.14±0.24 ^{aA}	2.04±0.15 ^{aA}
60	ND	ND	3.27±0.68 ^{aA}	3.54±0.35 ^{aA}
90	ND	2.11±0.0 ^{aA}	3.76±0.35 ^{aA}	3.97±0.27 ^{aA}
120	ND	3.12±0.11 ^{aA}	4.19±0.29 ^{aB}	4.59±0.21 ^{aC}
150	2.3±0.04 ^{aA}	3.46±0.02 ^{aA}	4.73±0.55 ^{aC}	4.92±0.09 ^{abB}
180	2.5±0.06 ^{aA}	4.31±0.03 ^{aB}	5.18±0.06 ^{aC}	5.47±0.11 ^{bC}
Yeast and mold count (log CFU/g of sample)				
0	ND	ND	ND	ND
30	ND	ND	1.87±0.19 ^{aA}	1.11±0.11 ^{aA}
60	ND	ND	2.49±0.27 ^{aA}	2.51±0.08 ^{aA}
90	ND	2.01±0.03 ^{aA}	3.21±0.39 ^{aB}	3.35±0.24 ^{aB}
120	ND	2.67±0.07 ^{aA}	3.69±0.08 ^{aB}	3.99±0.57 ^{aB}
150	1.71±0.02 ^{aA}	3.03±0.22 ^{aB}	4.17±0.13 ^{aC}	4.73±0.36 ^{aC}
180	1.96±0.07 ^{aA}	3.88±0.09 ^{aB}	4.91±0.74 ^{aC}	5.15±0.75 ^{aC}
Coliform count (log CFU/g of sample)				
0	ND	ND	ND	ND
30	ND	ND	ND	ND
60	ND	ND	ND	ND
90	ND	ND	ND	ND
120	ND	ND	ND	ND
150	ND	ND	ND	ND
180	ND	ND	ND	ND

ND not detected, CFU colony forming unit

Values are presented as mean± standard deviation ($n=3$)

Values with different small superscripts in a column differ significantly at $p<0.05$ for each test

Values with different capital superscripts in a row differ significantly at $p<0.05$ for each test

consumer panel. It can be seen that all the sensory attributes such as color and appearance, body and texture, flavor and sweetness, mouthfeel and overall acceptability of the product is positively correlated to each other. Thus, scores obtained from consumer panel can be used as a reliable estimate of the sensory quality of the nutriceal based fermented baby food. The overall acceptability score of the product are highly correlated to the other attributes that significantly changes with storage period. Therefore, overall acceptability scores of the product were used as the indicator of product deterioration causing consumer rejection.

Shelf life studies

Change in moisture content and titratable acidity during the storage period

Moisture content of a product is regarded as a critical parameter in determining the shelf life of a product with respect to its physico-chemical and microbial properties. Moisture content of any food material generally depends on its storage conditions such as temperature and packaging parameters. However, acceptable moisture content of a product will depend on the type of product under question. In dried food products such as

baby food, it is desirable to have low moisture content, to a predetermined extent. Conversely, it should not go above or below a critical value which may affect its physical properties such as reconstitution characteristics, wettability, dispersibility etc. and sensory attributes such as color and appearance, body and texture, flavor and sweetness and mouthfeel. These physical and sensory properties, if altered; affects the overall acceptability of the product by the consumers.

The initial moisture content of nutriceal based fermented baby food was $3.64\pm0.06\%$. Fig. 1 represents the change in the moisture content of the baby food stored at 10, 25, 37 and 45 °C, during the test storage period. It can be observed that the moisture content of the samples stored at 10, 25 and 37 °C, gradually gained moisture during the storage period of 180 days. The rate of gain of moisture by samples stored at 37 °C diminished after 120 days, as compared to the samples stored at 10 and 25 °C. On the other hand, the samples stored at 45 °C lost moisture during the storage time. This may be due to drying effect of the high temperature of storage, on the sample; however, the change in moisture was not significant ($p<0.05$).

The titratable acidity of the product varied slightly from the initial 0.28 ± 0.06 to 0.29 ± 0.02 , 0.29 ± 0.03 , 0.30 ± 0.01 and 0.30 ± 0.03 for samples stored at 10, 25, 37 and 45 °C,

Fig. 1 Changes in the moisture content during the test storage period

respectively. However, the change in titratable acidity with the progression of storage period and storage temperature was not statistically significant ($p > 0.05$). The correlation between sensory acceptability, titratable acidity and moisture content was not good and would not be of practical significance for the shelf life prediction of powdered products like nutriceal based fermented baby food. These results are in agreement with that obtained by Gambaro et al. (2004) while studying alfajor (chocolate coated individually wrapped cake).

Changes in the consumer overall acceptability

The consumer overall acceptability results were subjected to statistical analysis; analysis of variance (ANOVA), to find the significance of variation in the data obtained. Based on this, it was found that the overall acceptability scores from the consumers significantly decreased with the progression of the tested storage period at all four storage temperatures ($p < 0.001$). A linear correlation ($r^2 = 0.98$) was found between the overall acceptability scores (obtained from the consumers panel) and the storage time. Hough et al. (2002) suggested determination of shelf life with identifying the first significant negative change in the overall acceptability of the product. As can be seen for Fig. 2 and evidenced by the correlation analysis, the overall acceptability significantly changed with progression of the storage period. Considering the predetermined overall acceptability of 5.0 in a 9-point hedonic scale as the quality limit for the food product, the shelf life of the nutriceal based fermented baby food could be estimated to be near about 120 days for sample stored at 10 and 25 °C and 90 days for samples stored at 37 and 45 °C. This storage time corresponds to the time when consumers first detected a significant change in the sensory characteristics of the product, compared to the fresh sample. However, at this point the consumer can still accept or even like the product. For this reason, the estimated shelf life (i.e. about 120 days for sample

stored at 10 and 25 °C and 90 days for samples stored at 37 and 45 °C) of the product is too short, owing to the stringent criteria. Nutriceal based fermented baby food is a dried, powdered, acidic and microbiologically stable product, therefore estimating such a low shelf life of this product seems too conservative.

Survival analysis

The shelf life of a food product is not just limited to physical or chemical deterioration of the food product, but is a composite function dependent on the interaction of the consumer with the food (Hough et al. 2003). Therefore, the fundamental idea of the survival analysis technique focuses on determining the shelf life hazard on the basis of consumer perception for either acceptance or rejection of the product, rather than on product deterioration.

The Weibull distribution was selected to model the rejection times as it showed the best fit to the present data set. Several previous studies in shelf life determination used Weibull distribution for shelf life modelling of various food products such as yoghurt (Karagul-Yuceer et al. 1999; Curia et al. 2005; Cruz et al. 2010), roasted and ground coffee (Cardelli and Labuza 2001), alfajor (Gambaro et al. 2004), lettuce (Kim et al. 2005; Araneda et al. 2008), apple baby food (Gambaro et al. 2006). Survival analysis methodology is a simple technique employing fifty or a hundred individuals to express their acceptance or rejection for different formulated samples and/or samples stored at different conditions such as temperature, pressure, humidity etc., sufficient to estimate shelf life or a definite formula. One of the advantages of this technique is that the test utilizes the actual data obtained from the end user of the product, which gives the actual scenario of the product according to consumer preference (Hough 2006). There is no statistical test to evaluate the goodness of fit of the various parametric models. Thus, the visual assessment of

Fig. 2 Consumer acceptability as a function of storage time

how the parametric models fit the non-parametric estimation is the common practice used to choose the appropriate model (Cruz et al. 2010).

Therefore, in the present study, the distributions gamma, Weibull, log-normal and exponential were fitted to the baby food acceptability data. The Weibull distribution adjusted best fit for the data obtained for the baby food samples. Thus, Weibull distribution being very flexible model for survival data was used to determine the shelf life of the nutriceal based fermented baby food.

The Weibull estimation parameters α and β (Eq. 1) were calculated (with their standard errors). The values of α and β for samples stored at 10, 25, 37 and 45 °C are given in Table 2. Using these parameters, the rejection function ($F(t)$) plot was determined as shown in Fig. 3. To predict a shelf life, the probability of a consumer rejecting the product i.e., $F(t)$, needs to be selected. Several studies on shelf life predication modelling used 25 % rejection (Gambaro et al. 2004, 2005), while some other used 50 % rejection to estimate the shelf life (Gacula and Singh 1984; Cardelli and Labuza 2001). Thus, over the time both 25 and 50 % rejection were considered in number of studies (Gambaro et al. 2006; Araneda et al. 2008; Cruz et al. 2010). Therefore, in the present study, the shelf life

of the formulated baby food samples was determined at 25 and 50 % consumer rejection.

Amongst the four storage temperatures, the samples stored at 45 °C were the first to be rejected by the consumers. The samples were rejected on the basis of the overall acceptability, when the consumers scored the sample lower than five on a 9-point hedonic scale compared to the control sample (fresh sample). The first rejection score was obtained at day 90 and the acceleration stage started after day 120 (1 month after the first rejection) when 50 % of the consumers rendered the samples unacceptable, as described by Labuza and Schmidl (1988). The highest rejection score was obtained on day 180 resulting in the end of the study, due to lack of new consumer panellists to be incorporated in the study. This data was summarised and cumulative hazard analysis was applied to it as shown in Table 3. The data can be satisfactorily fitted according to Weibull model by using the following equation:

$$\log(\Sigma H) = 3.5018 \log(t) - 5.6761 \quad (4)$$

Where, t is the time in days and H the hazard value. The criterion for the end-point of shelf life was set as the time when 50 % probability of panellists grade the samples as being unacceptable. This probability corresponds to a cumulative hazard (ΣH) of -0.693 or 69.3 % as described by Sidel and Stone (1976).

The estimated shelf lives of the nutriceal based fermented baby food samples stored at 10, 25, 37 and 45 °C is shown in Table 4. It can be seen that the shelf life estimates using survival analysis for 25 % consumer rejection is nearly similar to that with estimated by using an acceptability score of 5.0 i.e. near about 90 days. Similar results were reported by Gambaro et al. (2006) for apple baby food. However, the shelf life estimates determined using 50 % rejection gave a relatively longer shelf life prediction of the product. The shelf life obtained at 25 and 50 % rejection probability varied

Table 2 α and β values of the Weibull distribution

Temperature	$\alpha \pm \text{standard error}$	$\beta \pm \text{standard error}$
10 (°C)	1.59 $\pm$ 0.11	123.05 $\pm$ 1.24
25 (°C)	1.62 $\pm$ 0.16	105.74 $\pm$ 1.68
37 (°C)	1.67 $\pm$ 0.10	88.40 $\pm$ 1.41
45 (°C)	1.73 $\pm$ 0.13	71.02 $\pm$ 1.22

α : shape parameter, β : characteristic life

Weibull distribution parameters α and β obtained from consumer data ($n=50$)

Fig. 3 Consumer rejection probability as a function of storage time

significantly ($p > 0.05$). It is also evident from the Table 4 that the predicted shelf life of the product also varied with variation in storage temperature. This may be the result of the changes in moisture content corresponding to the change in physical and sensory properties of the products such as colour and appearance, flavor and sweetness, body and texture and mouthfeel of the product, which ultimately affected the overall acceptability of the product. Although the variation in the acidity of the product was not significant statistically ($p < 0.05$), there is a significant negative correlation between overall acceptability of consumer ($r^2 = -0.85$) and semi-trained ($r^2 = -0.89$) panel considering the acidity of the product. The sensory perception of the acidity though chemically non significant, is therefore a significant criterion for deciding the consumer acceptability of the product. This suggested that the consumer's sensitivity to the acidity may be one of the factors responsible for

decrease in the consumer acceptability of the baby food samples over the storage period.

Development of shelf life model

Using the predicted shelf life obtained for samples stored at 10, 25, 37 and 45 °C, shown in Table 4, a shelf life model was obtained by adjusting these values to a shelf life plot (Fig. 4), by calculating the regression of the shelf life versus temperature to obtain the following equation (Villota et al. 1980; Labuza 1982):

$$\log \theta = 2.6 - 0.0075T \quad (5)$$

Where, T is the storage temperature (in °C) and θ is the shelf life (in days) of the nutriceal based fermented baby food. The equation obtained high correlation coefficient ($r^2 = 0.9523$), allowing estimation of effects of storage temperatures on shelf life of the product. This equation can be used to estimate the shelf life of the product at different storage conditions.

Table 3 Weibull hazard-ranking for sensory overall acceptability data in nutriceal based fermented baby food samples

Rank ^a	Days	H value ^b	$\sum H^c$
6	90	16.7	16.7
5	120	20.0	36.7
4	150	25.0	61.7
3	150	33.0	94.7
2	180	50.0	144.7
1	180	100.0	244.7

^a Rejections given by the panellists during the sensory evaluation are ranked according to the time (days of storage)

^b H = Hazard value = $100/\text{rank}$

^c $\sum H$ = Cumulative hazard = sum of all H values obtained for each rejection at that time

Table 4 Estimated shelf life for nutriceal based fermented baby food stored at 10, 25, 37 and 45 °C

Rejection (%)	Shelf life time (days)			
	10 (°C)	25 (°C)	37 (°C)	45 (°C)
25	98.0 ^{aA}	84.0 ^{bA}	71.0 ^{bB}	57.0 ^{bC}
50	321.0 ^{bA}	271.0 ^{aB}	221.0 ^{aC}	171.0 ^{aD}

Values with different small superscripts in a column differ significantly at $p < 0.05$ for each test

Values with different capital superscripts in a row differ significantly at $p < 0.05$ for each test

Fig. 4 Shelf life plot of nutriceal based fermented baby food at 10, 25, 37 and 45 °C, θ_{10} , θ_{25} , θ_{37} and θ_{45} are the predicted shelf life (in days) at storage temperatures of 10, 25, 37 and 45 °C, respectively

Microbiological analysis to ensure product safety throughout the predicted shelf life

In order to ensure the microbiological safety of the baby food during the predicted shelf life, the baby food samples were subjected to microbial analysis for total plate count, yeasts and molds count, coliform count, *Clostridium botulinum*, *Salmonella spp.* and *Staphylococcus aureus*. Table 5 depicts the microbiological quality analysis of the baby food samples during the estimated shelf life for the samples stored at 10, 25, 37 and 45 °C. The product was found to be microbiologically stable for the entire storage period of 360 days. The change in microbial quality was not significant ($p < 0.05$). No coliform and organisms such as *Clostridium botulinum*, *Salmonella spp.* and *Staphylococcus aureus* were detected during

the storage period of 360 days. The samples stored at 45 °C showed reduced microbial growth as compared to the samples stored at 10, 25 and 37 °C, this may be due to the comparative drying effect of high storage temperature, lowering the moisture needed for microbial proliferation. Hence, the nutriceal based fermented baby food can be regarded as microbiologically safe even during the predicted shelf life of 322, 271, 221 and 171 days for samples stored at 10, 25, 37 and 45 °C, respectively.

Conclusions

In the present study, sensory evaluation of the nutriceal based fermented baby food, by semi-trained panel and

Table 5 Microbiological quality of the baby food samples during the estimated shelf life of the product

Storage period (Days)	10 (°C)	25 (°C)	37 (°C)	45 (°C)
Total plate count (log CFU/g of sample)				
210	2.73±0.03 ^{aA}	4.95±0.04 ^{aB}	5.99±0.02 ^{aB}	6.44±0.01 ^{bC}
240	3.00±0.07 ^{aA}	5.39±0.09 ^{aB}	6.26±0.02 ^{aC}	6.74±0.06 ^{bC}
300	3.15±0.05 ^{aA}	5.77±0.10 ^{aB}	6.83±0.04 ^{aC}	6.95±0.05 ^{bC}
330	3.61±0.03 ^{aA}	6.19±0.06 ^{aB}	7.11±0.07 ^{aC}	7.01±0.04 ^{bcD}
360	4.07±0.11 ^{aA}	6.86±0.03 ^{aB}	7.96±0.13 ^{aC}	7.14±0.07 ^{cd}
Yeast and mold count (log CFU/g of sample)				
210	2.12±0.01 ^{aA}	4.12±0.01 ^{aB}	5.17±0.11 ^{aB}	5.42±0.07 ^{aB}
240	2.84±0.05 ^{aA}	4.79±0.07 ^{aB}	5.87±0.16 ^{aC}	5.67±0.04 ^{abC}
300	3.03±0.04 ^{aA}	5.11±0.05 ^{aB}	6.11±0.09 ^{abC}	5.82±0.15 ^{abB}
330	3.27±0.12 ^{aA}	5.84±0.04 ^{abB}	6.59±0.12 ^{bC}	5.88±0.11 ^{bBC}
360	3.64±0.09 ^{aA}	6.14±0.06 ^{bB}	7.02±0.17 ^{bB}	5.95±0.08 ^{bB}

CFU colony forming unit

Values are presented as mean±standard deviation ($n=3$)

Values with different small superscripts in a column differ significantly at $p < 0.05$ for each test

Values with different capital superscripts in a row differ significantly at $p < 0.05$ for each test

untrained consumers was conducted to characterize the changes in the sensory properties of the product stored at 10, 25, 37 and 45 °C during the test storage period of 180 days. Survival analysis methodology was used to determine the shelf life of a nutriceal based fermented baby food on the basis of consumer rejection data using Weibull hazard analysis. Considering 25 and 50 % rejection probability, the shelf life of the baby food formulation was predicted to be 98 and 322 days, 84 and 271 days, 71 and 221 days and 58 and 171 days for the samples stored at 10, 25, 37 and 45 °C, respectively. Sensory analysis was found to be a better shelf life predictor as compared to the chemical data (moisture and acidity). Microbiological analysis was conducted to ensure the microbial safety of the baby food formulation during the predicted shelf life and no microbial hazard was detected during the entire shelf life study period of 360 days.

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