

Effect of Packaging Materials and Modified Atmosphere Packaging on the Shelf-life of Khoa

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Abstract

Preservation of freshly prepared Khoa was carried out at room temperature ($25\pm 1^\circ\text{C}$) and refrigerated temperature ($5\pm 1^\circ\text{C}$) by using different packaging materials such as LDPE, aluminum foil/PVC and three layer laminated (polyester/met. BOPP/LDPE) in different conditions viz. normal and shrink. All of the packaging materials on both conditions showed no significant difference except normal LDPE on mean sensory score and chemical parameters of Khoa during storage at $5\pm 1^\circ\text{C}$ whereas it showed highly significant difference at $25\pm 1^\circ\text{C}$ among three packaging materials. Three layer laminated sample showed significantly higher sensory quality, high pH, low acidity, peroxide value (PV), free fatty acids (FFA) and lower microbial count than other two packaging materials. Similarly, shrink packaged sample showed better quality in terms of sensory and microbial attributes of Khoa compared to normal packaging. No colonies of yeast and mold, coliform were observed during the storage period. Shrink packaged Khoa in three layer laminated increased storability to 27 days at $25\pm 1^\circ\text{C}$ against three days for unpacked ordinary Khoa.

Key words: Khoa preservation, packaging materials

Introduction

Khoa is an indigenous milk product which is also called *Kurauni* (in Nepali). It is produced in different regions for selling and domestic consumption. Khoa is a product of great commercial importance as it forms an important base material for the preparation of varieties of indigenous milk based sweets throughout the country (Rajorhia 2002). The Khoa is produced in small to large scale by using the milk of different locations. So there is a large variation in product quality (Solanki *et al.* 2002).

Khoa has a low shelf-life (three days in summer and six to seven days in winter) because of the unhygienic conditions at rural production centers, lack of low temperature chain, inherent high water activity and absence of protective packaging favor high microbial contamination. Thus, it is necessary to maintain a cool chain, preferably around -2°C during storage and shipping. For export, milk of excellent quality should be used for Khoa making and produced hygienically followed by instant chilling are considered essential (Rajorhia 2002).

It is essential to generate alternative ideas for utilizing the excess milk produced in the farm levels. Therefore, the main objective of this research is to generate ideas for the extension of shelf life of Khoa at ambient and refrigerated conditions with the combination of normal and shrink packaging also called as modified atmosphere packaging by using different conditions. This investigation was also undertaken to evaluate the sensory, biochemical and microbiological attributes of Khoa and improving its shelf-life by using different packaging materials in a different conditions.

Methodology

Khoa was collected from Nobel Dairy Pvt. Ltd., Pokharia, Biratnagar. The packaging materials were as follows: (1) Low-density polyethylene (LDPE) of gauge 65 micron which was collected from Kamdhenu Dairy Udhyog, Tarahara, Sunsari. (2) Double layer aluminium foil/PVC (Polyvinyl chloride) of gauge $250\ \mu\text{s}$ and $40\ \mu\text{s}$ respectively, was used. (3) Three-layer laminated polyester reverse print of gauge $12\ \mu/\text{metalized BOPP}$

of gauge 20 μ /LDPE of gauge 20 μ was used and it was received from Shiv Pharmaceutical Laboratory, Dharan, Sunsari.

Khoa was prepared by Solar Arks Khoa Machine following the standard method adopted by many dairies. Packaging was done in different sterile packaging materials as describe above. Khoa samples of about 100 g were packed in packaging pouches of total area 12.4 \times 11.4 cm² with a volume of 200 ml under two packaging conditions viz. normal and shrink. Under normal condition, required amount of Khoa was placed and sealed immediately. For shrink packaging, air inside the package was drawn out by using a vacuum pump.

Both the normal and shrink packaged samples were stored in a room having temperature (25 \pm 1 $^{\circ}$ C) and refrigerated temperature of 5 \pm 1 $^{\circ}$ C. For Khoa stored at room temperature, analysis was carried out at an interval of 4 days and for refrigerated temperature, analyses were carried out at an interval of 12 days. The effect of these packaging materials and shelf-life of Khoa were evaluated by sensory evaluation followed by physico-chemical analysis where pH, acidity, free fatty acid and peroxide value. The microbiological analysis such as yeast and mould, coliform and total plate counts were carried out.

The sensory parameters were judged by 10 semi-trained panelists on a 9-point hedonic rating scale (Ranganna 2004). All the data obtained in this experiment were analyzed by Genstat program developed by Lawes Agricultural Trust (1995).

The fresh and stored samples were analyzed for chemical and physico-chemical parameters on the following ways. The total solid was determined by the procedure given in AOAC (1980). Fat by Roesse-Gottlieb, protein by Micro Kjeldahl method and moisture content, peroxide value and FFA (% oleic acid) were determined (Egan *et al.* 1981). The percentage of carbohydrate was determined by difference method. Ash and titratable acidity were determined by the standard procedure given in AOAC (1980). The pH of samples was determined as per the procedure given in Jul-Overlay (1986). The standard plate counts, yeast, mould and coliform counts were evaluated (counted) as per standard procedure (Jul-Overlay 1986).

Results and Discussion

Chemical composition of the fresh milk used for Khoa making

The chemical composition of milk used for Khoa making were evaluated. The water, fat, protein, lactose, ash, and total solids were found to be 85.4, 5.0, 3.3, 4.8, 0.7 and 13.7 %, respectively. The pH and acidity of fresh milk were found to be 6.7 and 0.18 %, respectively.

Chemical composition and microbiological quality of fresh Khoa

The freshly prepared Khoa was chemically analyzed. The moisture, fat, protein, carbohydrate, total solid and ash were found to be 24.8, 31.23, 20.2, 20.36, 75.2 and 3.4 %, respectively. The pH, acidity, free fatty acid and total plate count (CFU/g) were found to be 6.4, 0.2 %, 0.1% and 1.6, respectively. Whereas, in fresh Khoa samples peroxide value (PV), coliform, yeast and molds (Y & M) count were not recorded.

Optimization of packaging condition and storage period (days) of Khoa

The effect of packaging material with condition and storage period (days) on Khoa samples were analyzed using sensory parameters like texture, color, taste and smell, physico-chemical analyses such as pH, acidity, FFA, PV and microbiological analyses such as total plate count (TPC), yeast and mold count (Y&M) and coliform.

Sensory evaluation

The effect of packaging conditions on mean sensory score of Khoa has been presented in Fig. 1. The packaging conditions and materials differed significantly ($p < 0.05$) on sensory score of Khoa. Normal LDPE showed lower score for all parameters whereas shrink three layers laminated (polyester/met.BOPP/LDPE) packaged showed higher sensory score. Normal aluminium foil/PVC and shrink LDPE showed no significant difference. At normal packaging, three layers laminated package showed significantly higher sensory score than others.

The effect of storage period (days) on mean sensory score of Khoa stored at 5 \pm 1 $^{\circ}$ C has been presented in Fig. 2. The mean sensory score of texture, taste, color and smell did not vary with storage period (days). From the sensory analyses of Khoa stored for 12, 25, 40 and 53 days at refrigerated condition (5 \pm 1 $^{\circ}$ C), the samples

showed superior in terms of their acceptability. The effect of storage period (days) on mean sensory score of Khoa stored at $25\pm 1^\circ\text{C}$ is presented in Fig. 3. As increasing the storage period (days) all the mean sensory scores values showed to decreased significantly.

From the sensory analysis of Khoa stored for 4, 9, 14, 18, 23, 27 and 30 days at room temperature ($25\pm 1^\circ\text{C}$), Khoa was acceptable up to 9 days at normal LDPE, and became unacceptable on 14 days onwards. Normal 3-layer laminated (polyester/met. BOPP/LDPE) package had shown that Khoa was acceptable up to 18 days in terms of smell and taste; and became unacceptable on 23 days but in terms of color and texture it was accepted up to 23 days. Normal aluminium foil/PVC and shrink LDPE had shown that Khoa was accepted up to 18 days in terms of smell and taste and it had unacceptable on 18 days storage but in terms of color and texture it was acceptable up to 18 days. Shrink 3-layer laminated packaging showed Khoa was acceptable up to 27 days

in terms of all sensory parameters and had become unacceptable on 30 days.

Packaging caused a significant ($P < 0.05$) effect on taste and smell but insignificant ($P > 0.05$) effect on texture and color. However, the packaging had no significant effect on mean sensory score of Khoa stored under refrigerated temperature ($5\pm 1^\circ\text{C}$), but Khoa stored at room temperature ($25\pm 1^\circ\text{C}$) was significantly affected in all the packaging materials and conditions.

Due to the lack of sterilization facilities of the packaging materials, the product might be contaminated from the inner surface of the package. As the storage period (days) increased taste and smell had differed significantly in mean sensory score of Khoa. However, texture and color did not differ significantly ($P < 0.05$) as increased the storage period. The normal aluminum foil/PVC and shrink LDPE did not vary significantly. However, normal LDPE, normal 3-layer laminate and shrink 3-layer laminate packaging were significantly varied with each other.

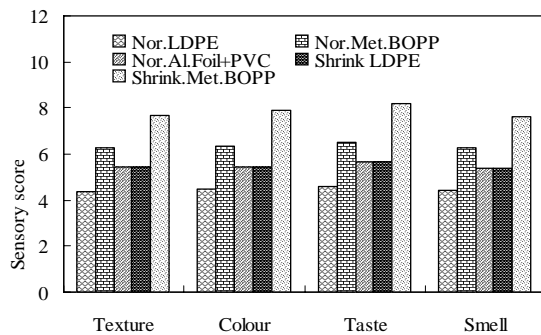


Fig. 1 Effect of packaging conditions on mean sensory score of khoa.

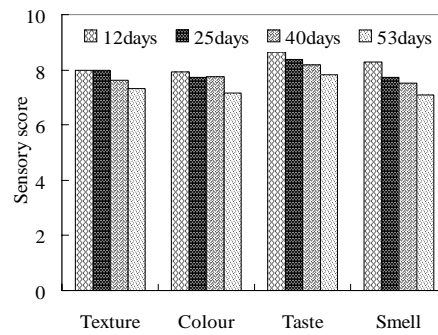


Fig. 2 Effect of storage period (days) on sensory score of khoa at $5\pm 1^\circ\text{C}$.

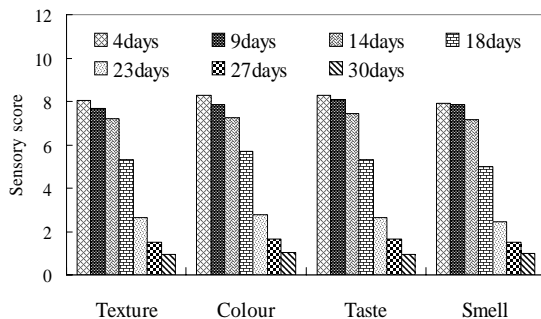


Fig. 3 Effect of storage period (days) on sensory score of khoa at $25\pm 1^\circ\text{C}$.

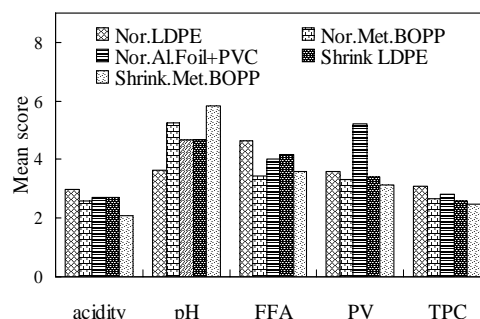


Fig. 4 Effect of packaging materials and condition on chemical and microbial parameters of Khoa.

Chemical analysis

The effects of different packaging material and condition on chemical parameters of Khoa are presented in Fig. 4. The different packaging had significant effect on chemical parameters of Khoa.

The shrink three layers laminated (polyester/met. BOPP/LDPE) packaged samples had higher pH than the other packaging. It might be due to lower microbial growth on the samples. The chemical parameter remained almost constant in both normal aluminium foil/PVC and shrink LDPE packaged Khoa.

The effect of storage periods on chemical parameters of the Khoa stored at $5\pm 1^\circ\text{C}$ and $25\pm 1^\circ\text{C}$ are presented in Fig.5 and 6. The chemical parameters viz. acidity, pH, free fatty acid (FFE), and peroxide value (PV) were found to be not significantly changed as it stored at

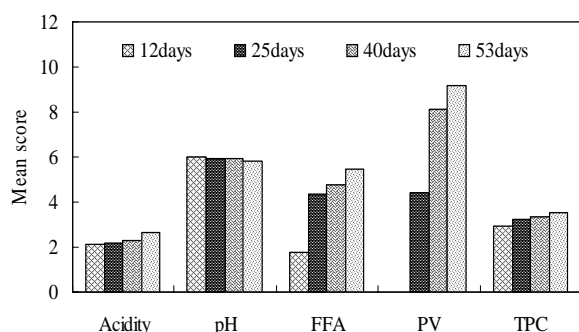


Fig. 5 Effect of storage period (days) on chemical and microbial parameters of *Khoa* stored at $5\pm 1^\circ\text{C}$.

The effect of both the storage period and different packaging on chemical parameters of Khoa stored at $5\pm 1^\circ\text{C}$ and $25\pm 1^\circ\text{C}$ are presented in Fig. 7 and Fig. 8, respectively. The both storage period and packaging, significantly affect on free fatty acids (FFA) and peroxide value (PV) of stored Khoa. As the storage period increased the FFA and PV increased significantly. At the end of storage period, different packaging had different PV in range about (0.8-1.1) meq/kg of Khoa stored at $5\pm 1^\circ\text{C}$, this value is far less than the value obtained in rancid fat (10 meq/kg of fat). The extent of lipolysis is measured by the level of FFA (Rossel 1989). FFA slightly increased in the case of stored at $5\pm 1^\circ\text{C}$ but significantly increased in the case of stored at $25\pm 1^\circ\text{C}$.

Joshi (1994), reported that hydrolysis of milk fat is catalyzed by lipase and results in formation of

$5\pm 1^\circ\text{C}$, whereas significantly changed as it stored at $25\pm 1^\circ\text{C}$. The acidity had shown to increased and pH had decreased as increasing the storage period. At refrigerated condition ($5\pm 1^\circ\text{C}$) pH of Khoa showed no any significant changes during storage. It might be due to use of low temperature storage. At room temperature ($25\pm 1^\circ\text{C}$) storage period (days) was affected significantly ($P < 0.05$) on all the chemical parameters. The acidity had shown to increase as the storage periods (days) increased.

At the end of storage period, different packaging had different PV in range about (0.8-1.1) meq/kg of Khoa stored at $5\pm 1^\circ\text{C}$, this value is far less than the value obtained in rancid fat (10meq/kg of fat). The extent of lipolysis is measured by the level of FFA (Rossel 1989). FFA slightly increased in the case of stored at $5\pm 1^\circ\text{C}$ but significantly increased in the case of stored at $25\pm 1^\circ\text{C}$.

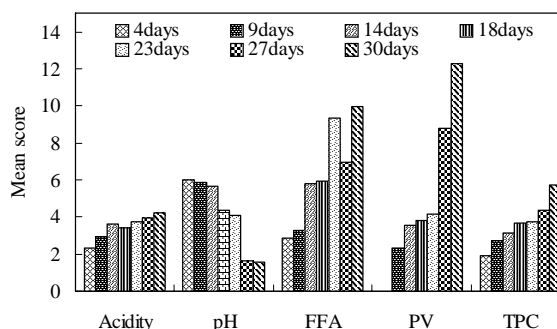


Fig. 6 Effect of storage period (days) on chemical and microbial parameters of *Khoa* stored at $25\pm 1^\circ\text{C}$.

glycerides and free fatty acids. The milk fat is only common fat that contains substantial amount of lower fatty acids. The principal fatty acid responsible for rancid flavor among the liberated fatty acids is the butyric acid. The perception threshold of the FFA is relatively high therefore, in case of dairy products; organoleptic test is more sensitive than chemical analysis (Kuzdzal Savoie & Trehin 1975).

The oxidative rancidity is more complex, unpredictable and more common in milk products than the lipolytic rancidity. However, it is less important for to spoilage in fresh milk (Allen. 1989). Peroxides are the primary products of oxidation, and hence, measurement of their concentration gives an idea about the extent of oxidation (Joshi 1994).

The method of assessing oxidation status is the hydro peroxide value. This is based on the fact that hydro

peroxides react with potassium iodide to liberate iodine which can be measured by its reaction with thiosulphite or electrochemically. The value represents mmol of O₂

per 2 kg of fat. Freshly refined material should have a peroxide value below 1. A fat is rancid at a peroxide level of about 10 (Gunstone 1999).

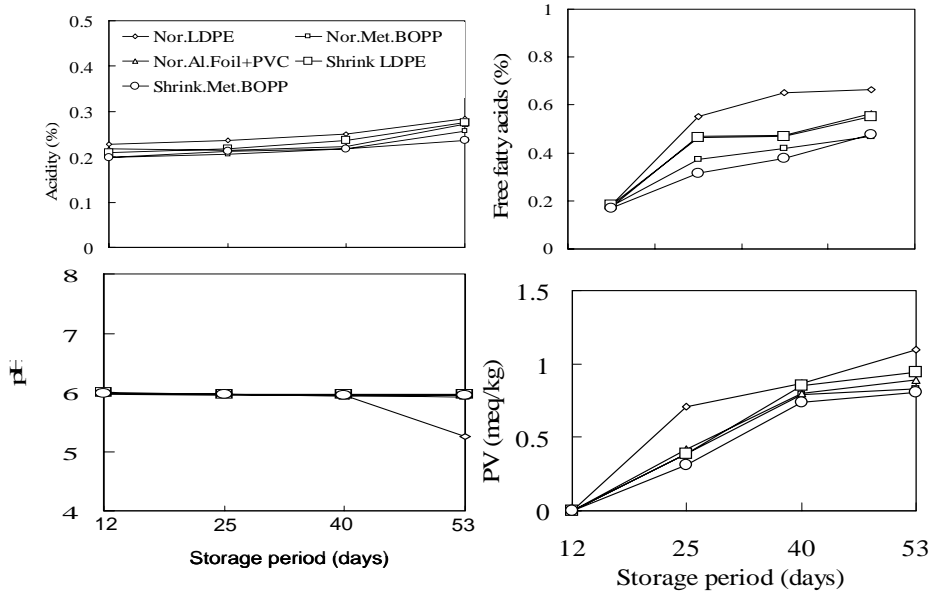


Fig. 7 Effect of storage period (days) on chemical parameter of khoa stored at 5±1°C.

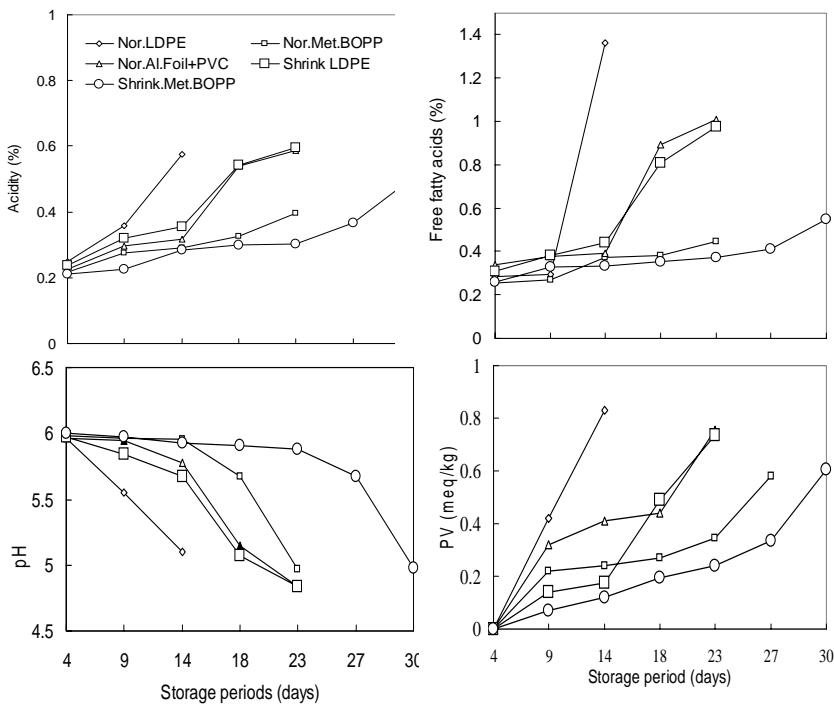


Fig. 8 Effect of storage days on chemical parameter of Khoa stored at 25±1°C.
(Note: PV = Peroxide value)

Microbiological analysis of stored Khoa

The total plate count (TPC), yeast and molds (Y&M), and *coliform* counts of stored Khoa were analyzed. There was no growth of yeast and molds and coliforms during the storage periods. The yeast and molds are aerobic and could not able to form colonies in packages of anaerobic environment. Likewise, the *coliforms* formed no colonies. They were destroyed during the cooking of Khoa before packaging. The microbiological quality of Khoa depends mainly upon the condition of post production handling, packaging and storage of the product. Gupta and Ghodekar (1985) reported that, fresh Khoa prepared under controlled sanitary conditions does not contain organisms capable of producing diseases to human beings.

Generally, the post process contamination occurs in Khoa from the inner surface of packaging materials

due to the lack of its proper sterilization. Shrink 3-layer laminated packaging (polyester/met.BOPP/LDPE) had less number of organisms than other packaging mentioned above. Khoa stored at refrigerated temperature ($5\pm 1^\circ\text{C}$) did not show any significant increase in the total plate count.

The effect of storage period on total plate count in Khoa stored in different packages at $5\pm 1^\circ\text{C}$ and $25\pm 2^\circ\text{C}$ is presented in Fig. 9 and Fig. 10, respectively. Storage period (days) caused significantly increase on total plate count (TPC) of Khoa stored at $25\pm 2^\circ\text{C}$. The vacuum flushed Khoa sample had significantly lower TPC. In Fig. 9, it had been shown that microbial growths in all three packages have the same growth of microorganisms in terms of total plate count (TPC). TPC in normal aluminium foil/ PVC and shrink LDPE samples did not differ significantly.

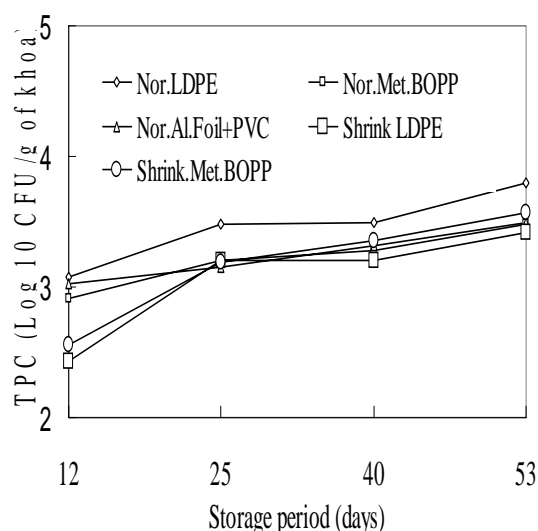


Fig. 9 Effect of storage period (days) on total plate count of *khoa* stored at $5\pm 1^\circ\text{C}$.

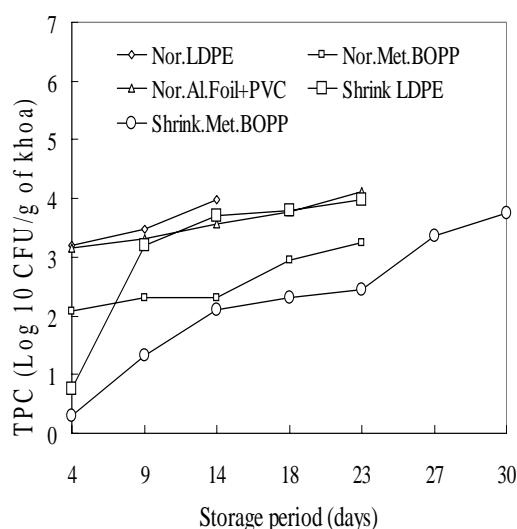


Fig. 10 Effect of storage days on total plate count of *khoa* stored at $25\pm 1^\circ\text{C}$.

At refrigerated condition, after 25 days, TPC flattened and remain constant until 40 days. It may be due to the exhaustion of nutrients and/or accumulation of metabolite especially on the surface of Khoa samples. Gupta and Ghodekar (1985) reported that, fresh Khoa prepared under controlled sanitary condition does not contain organism capable of producing diseases in human beings. In Fig. 10 it had shown that colony of TPC in all package sample differ significantly. In Fig. 10 and 5, at room temperature, shows normal LDPE

had higher TPC and normal met. BOPP and shrink met. BOPP (3-layers laminated) had lower TPC. However, colony of TPC in normal Aluminium Foil/PVC and shrink LDPE did not differ significantly.

Generally, the post process contamination occurs in Khoa from the inner surface of packaging material due to the lack of its proper sterilization. Shrink packaging had less number of organisms than the other packaging of Khoa mentioned above. All the packaging except

LDPE did not showed any significant difference in colony of total plate count.

Storage periods (days) caused significantly increase on total plate count (TPC) of Khoa. The shrink three layer laminated (polyester/met. BOPP/LDP) sample had significantly lower total plate count (TPC) was recorded.

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