

DEVELOPMENT AND STORAGE STABILITY OF SOYBEAN MEAL-BASED PET TREATS UNDER AEROBIC AND VACUUM PACKAGING

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Abstract: This study aimed to develop a pet treat utilizing soybean meal (SBM) as the primary protein source and to evaluate its shelf-life under aerobic and vacuum packaging over 90 days of storage. The formulated pet treats were analyzed for physicochemical parameters like pH water activity, Thiobarbituric acid reactive substances (TBARS) value, tyrosine value and microbiological studies of total viable count (TVC), *E. coli*, *Salmonella* and Yeast & Mould counts at regular intervals. Results indicated that vacuum packaging significantly ($p < 0.05$) preserved the pet treat quality by retarding moisture uptake, lipid oxidation and protein degradation compared to aerobic packaging. The SBM-based pet treats demonstrated excellent microbiological stability, with no pathogen detected throughout the study. It is concluded that soybean meal serves as an effective protein source for formulating shelf-stable pet treats, with vacuum packaging playing a crucial role in maintaining their sensory quality and chemical shelf-life.

Keywords: pet treat, soyabean meal, physicochemical, microbiological, vacuum and aerobic packaging.

Introduction

Pet ownership has seen a significant increase globally, fostering a corresponding growth in the pet food industry, with a particular focus on nutrition and palatability. This increasing demand highlights the necessity for innovative and sustainable ingredients that not only meets the nutritional requirements of companion animals but also addresses the concerns regarding cost-effectiveness and environmental impact (Wagoner *et al.*, 2022). Within this context, soybean meal, a co-product of soybean oil extraction, emerges as a promising ingredient due to its high protein content and widespread availability (Sørensen *et al.*, 2009). This research paper investigates the potential of incorporating soybean meal into pet treat formulations, by evaluating its impact on physico-chemical and microbiological quality.

Materials and Methods

Selection of Ingredients and Formulation

For the formulation of the pet treat, soybean meal served as the non-animal protein source. We obtained the required raw materials such as soybean meal, deoiled rice bran, calcium

carbonate, dicalcium phosphate, calcite powder, trace mineral mix and vitamin premix from the feed mill unit of the Veterinary College & Research Institute, Namakkal. Other key ingredients, such as refined wheat flour (Maida), salt, egg, rice flour, tapioca flour, jaggery, yeast, baking soda and almond gum were sourced locally from Namakkal.

The pet treat was formulated to contain soybean meal (SBM) as the primary protein source at 19.50%, with other ingredients including vegetable oils (14%), maida (10.50%), deoiled rice Bran (9.30%), salt (0.5%), rice flour (8.75%), tapioca flour (13%), Jaggery (5.5%), trace mineral mix (0.25%), yeast (0.05%), baking soda (0.5%), almond gum (0.4%), vitamin premix (0.25%), calcium carbonate (0.75%), dicalcium phosphate (1.25%), calcite powder and Egg (15.00%). The formulation was designed to be iso-nitrogenous and iso-caloric, targeting 18% protein and 4000 kcal/kg.

Preparation of pet treat:

The preparation of pet treats was carried out with careful selection and collection of high-quality raw materials, followed by precise weighing of all ingredients as per the formulated proportions. The weighed ingredients were then thoroughly mixed and finely ground to obtain uniform dough, which was subsequently proofed for 30–40 minutes to enhance its texture and consistency. After proofing, the dough was moulded into circular shapes weighing approximately 25 g each and baked in a preheated hot air oven at 150°C for 50 minutes, then the baked treats were cooled at ambient temperature for 15 minutes. Finally, the pet treats were packed in high-density polyethylene (HDPE) pouches using aerobic and vacuum packaging methods and stored at room temperature for a period of 90 days for further evaluation of their physico-chemical and microbiological stability.

Quality assessment

Quality assessment of the pet treats was carried out at regular intervals of 0, 15, 30, 45, 60 and 90 days of storage. The evaluation consists of physicochemical and microbiological analyses to determine the stability and safety of the pet treat during the storage period. The physicochemical parameters analyzed included pH, water activity (aw), Thiobarbituric Acid Reactive Substances (TBARS), and Tyrosine Value (TV), which served as an indicator of chemical and oxidative changes during storage. Microbiological examination involved the enumeration of Total Viable Count (TVC), *Escherichia coli*, *Salmonella* spp., and Yeast and Mould counts to assess the microbial quality and shelf stability of the pet treats. All analyses were conducted following standard procedures as described in the respective reference methods.

Table 1: Comprehensive Quality Parameters of Soybean meal Based Pet Treats During 90-Day Storage

Parameters	Packaging	0 Days	15 Days	30 Days	45 Days	60 Days	75 Days	90 Days
pH	Aerobic	5.78 ± 0.02	5.64 ± 0.01	5.52 ± 0.02	5.39 ± 0.01	5.24 ± 0.01	5.15 ± 0.02	5.07 ± 0.01
	Vacuum	5.79 ± 0.02	5.72 ± 0.02	5.60 ± 0.01	5.40 ± 0.02	5.37 ± 0.02	5.30 ± 0.00	5.28 ± 0.01
WaterActivity (a_w)	Aerobic	0.23 ± 0.01	0.30 ± 0.01	0.36 ± 0.01	0.40 ± 0.01	0.43 ± 0.01	0.46 ± 0.01	0.50 ± 0.01
	Vacuum	0.23 ± 0.00	0.27 ± 0.00	0.33 ± 0.00	0.35 ± 0.00	0.38 ± 0.00	0.39 ± 0.00	0.41 ± 0.00
TBARS(mg MDA/kg)	Aerobic	1.15 ± 0.02	1.34 ± 0.02	1.69 ± 0.02	1.71 ± 0.02	1.83 ± 0.01	1.91 ± 0.02	1.93 ± 0.02
	Vacuum	1.14 ± 0.02	1.23 ± 0.02	1.49 ± 0.03	1.58 ± 0.02	1.70 ± 0.02	1.73 ± 0.03	1.79 ± 0.03
Tyrosine (mg/100)	Aerobic	55.27 ± 0.20	62.97 ± 0.32	70.53 ± 0.49	77.28 ± 0.39	80.13 ± 0.40	83.92 ± 0.57	88.97 ± 0.52
	Vacuum	55.45 ± 0.36	60.37 ± 0.34	64.23 ± 0.28	68.57 ± 0.36	71.22 ± 0.37	77.75 ± 0.34	76.68 ± 0.22
TVC (log CFU/g)	Aerobic	2.11 ± 0.03	2.40 ± 0.03	2.74 ± 0.05	3.04 ± 0.03	3.23 ± 0.04	3.36 ± 0.03	3.50 ± 0.03
	Vacuum	1.90 ± 0.01	2.08 ± 0.02	2.19 ± 0.02	2.38 ± 0.03	2.49 ± 0.03	2.65 ± 0.04	2.77 ± 0.04
Yeast & Mould (log CFU/g)	Aerobic	ND	ND	ND	0.03 ± 0.02	1.35 ± 0.04	1.98 ± 0.05	2.25 ± 0.04
	Vacuum	ND	ND	ND	0.02 ± 0.02	0.09 ± 0.03	1.10 ± 0.03	1.33 ± 0.02
<i>E. coli</i>	Both	Absent	Absent	Absent	Absent	Absent	Absent	Absent
<i>Salmonella</i>	Both	Absent	Absent	Absent	Absent	Absent	Absent	Absent

**Fig.1 Soybean meal based Pet treats**

Results and Discussion

The quality attributes of soybean meal based pet treats were evaluated during 90 days of storage under aerobic and vacuum packaging to assess their stability and shelf life. The results revealed that packaging type and storage duration significantly influenced the physicochemical and microbiological parameters of the product.

pH:

The pH gradually declined from 5.78 to 5.07 in aerobic and from 5.79 to 5.28 in vacuum-packaged samples during storage. This reduction may be due to organic acid formation by microbial activity. A similar observation was made by Dziezak (1986), who emphasized that pH values below 5.0 enhance storage stability by suppressing microbial growth through increased acidity, salinity, and low moisture. The slightly higher pH retention under vacuum indicates better stability.

Water activity:

Water activity increased slightly from 0.23 to 0.50 in aerobic and from 0.23 to 0.41 in vacuum-packaged samples, possibly due to moisture absorption. The gradual increase could be attributed to moisture migration from the surrounding environment. According to Ozgur and Sakine (2017), extruded dog foods generally possess a_w values between 0.30 and 0.54, which are low enough to prevent microbial spoilage. Likewise, Leiva *et al.* (2019) categorized dry pet foods as low-moisture and low- a_w products, ensuring minimal microbial activity. The a_w values of the present study (0.23–0.50) fall within this stable range, suggesting that both packaging systems effectively preserved product stability.

Thiobarbituric acid reactive substances (TBARS):

TBARS values, indicating lipid oxidation, increased from 1.15 to 1.93 in aerobic and 1.14 to 1.79 in vacuum conditions. Higher oxidation in aerobic storage may be due to oxygen exposure. Kappus (1991) reported that lipid oxidation in pet foods leads to the formation of aldehydes, ketones, and other volatile compounds that cause nutrient loss and off-odours. Rajkumar *et al.* (2005) similarly observed a steady increase in TBA values in meat products under different packaging conditions, with the highest values recorded toward the end of storage. The relatively lower TBARS values in vacuum-packed samples from the present study demonstrate that the exclusion of oxygen successfully minimized lipid oxidation and improved oxidative stability.

Tyrosine value:

Tyrosine values increased from 55.27 to 88.97 in aerobic and from 55.45 to 76.68 in vacuum samples, reflecting protein degradation. Kuleswan *et al.* (2017) reported tyrosine increases in stored pet kibbles, ranging from 28.89 to 104.92 mg/100 g. The slower increase in vacuum-packed treats indicates reduced proteolysis and better protein stability.

Microbial count:

TVC increased from 2.11 to 3.50 in aerobic and from 1.90 to 2.77 in vacuum-packed samples, showing microbial proliferation over time. Rajendra *et al.* (2011) observed a similar trend in dry dog foods where microbial total viable counts increased significantly during storage. Despite the gradual rise, the microbial load in vacuum-packed samples remained considerably lower, indicating the effectiveness of vacuum packaging in controlling aerobic microbial proliferation and extending shelf life.

Yeast and mould were not detected up to 30 days but increased to 2.25 in aerobic and 1.33 in vacuum samples by day 90. Witaszak *et al.* (2020) emphasized monitoring fungal growth in dry pet foods due to potential mycotoxin contamination. The lower fungal counts in vacuum packaging indicate reduced oxygen availability and better fungal control. *E. coli* and *Salmonella* were absent in all samples throughout storage, confirming hygienic processing and safety. Karthikeyan (2004) similarly reported the absence of *Coliforms* and *Salmonella* in dry dog foods stored aerobically and under vacuum. The present findings affirm that both packaging systems effectively inhibited pathogenic contamination.

Conclusion

The present study revealed that soybean meal-based pet treats exhibited satisfactory physicochemical and microbiological stability during 90 days of storage under both aerobic and vacuum packaging conditions. Although gradual increases in oxidative and proteolytic parameters were observed over time, all values remained within acceptable limits. Vacuum packaging was more effective than aerobic storage in reducing lipid oxidation, protein degradation, and microbial growth. The consistent absence of *E. coli* and *Salmonella* confirmed the hygienic safety of the product. Overall, vacuum packaging proved to be a better preservation method for maintaining product quality and extending the shelf life of soybean meal-based pet treats.

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