Research Article

Effect of Functional Feeding Strategies Based on Nano-Biological Additives on the Productive Performance of Laying Hens

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Abstract | This study aimed to evaluate the effects of functional feeding strategies based on nano-synbiotic supplement containing *Lactobacillus gasseri* and *Lactobacillus helveticus* loaded onto an inulin-rich plant carrier (Nano-*Taraxacum officinale*) on productive performance and economic efficiency in laying hens. A total of 96 Lohmann Brown laying hens at 48 weeks of age were used in an 8-week trial and randomly allocated into four dietary treatments: control (0), 1, 2, and 3 g nano-synbiotic/kg diet. Results demonstrated significant improvements ($P < 0.05$) in all productive performance parameters compared with the control group. The 2 g/kg treatment achieved the highest hen-day egg production (91.4%) compared with the control (87.4%). Egg weight was also significantly increased, reaching 67.3 g in the 2 g/kg group compared with 65.9 g in the control. Similarly, egg mass improved from 57.63 g/hen/day in the control to 61.49 g/hen/day at 2 g/kg supplementation. Feed conversion ratio was significantly improved, decreasing from 2.26 in the control group to 2.11 in the 2 g/kg treatment, indicating enhanced feed utilization efficiency. The 3 g/kg treatment showed comparable results without additional significant improvement, suggesting a functional biological saturation at the 2 g/kg inclusion level. Economic evaluation revealed a significant increase in economic feed efficiency, with the highest value observed in the 2 g/kg treatment (129.8%) compared with the control (121.5%), demonstrating that biological improvements translated directly into greater economic return. In conclusion, nano-synbiotic supplementation significantly enhances productive performance and economic efficiency in laying hens, with 2 g/kg identified as the optimal inclusion level for maximizing biological and economic benefits without additional gains at higher supplementation levels.

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Introduction

The laying hen industry is currently facing increasing challenges in maintaining optimal productive performance while ensuring flock health, egg quality, and sustainable production systems. These challenges have intensified following global restrictions on the use of antibiotic growth promoters due to concerns regarding antimicrobial resistance and food safety. Consequently, improving gastrointestinal efficiency and maintaining a balanced intestinal microbial ecosystem have become essential strategies for enhancing nutrient utilization, immune competence, and metabolic stability in laying hens. Efficient gut function plays a fundamental role in regulating productive performance, including egg production rate, egg weight, and egg quality parameters (Al Salhi and Al Shatty, 2023; Al Salhi, 2026).

Functional feeding strategies based on bioactive additives, such as probiotics, prebiotics, and synbiotics, have gained considerable attention as effective and safe alternatives to antibiotic growth promoters. These additives improve intestinal microbial balance, enhance digestive enzyme activity, and increase nutrient absorption efficiency, thereby promoting both productive performance and physiological stability in laying hens. Several studies have demonstrated that probiotic supplementation can significantly enhance egg production, egg weight, feed conversion efficiency, and overall production performance compared to conventional feeding systems (Xu *et al*., 2023; Alagawany *et al*., 2025).

Synbiotic systems, which combine probiotics with prebiotic carriers, provide additional benefits by improving probiotic survival, colonization, and metabolic activity within the gastrointestinal tract. Plant-derived prebiotics rich in inulin, such as *Taraxacum officinale*, serve as favorable substrates for probiotic growth and activity. Recent advances in nanotechnology have enabled the development of nano-scale bioactive carriers, which improve stability, delivery efficiency, and bioavailability of probiotic formulations. Such nano-biological delivery systems protect probiotics against environmental stress, enhance gut colonization efficiency, and maximize their functional effects (Liu *et al*., 2024).

Egg quality traits, including eggshell thickness, yolk pigmentation, and internal quality, are closely linked to digestive efficiency and nutrient absorption. Functional nano-synbiotic formulations have been shown to positively influence these traits, thereby increasing production efficiency, reducing economic losses, and supporting sustainable egg production systems (Gallazzi *et al*., 2024; Alagawany *et al*., 2025).

Despite extensive research on probiotics and synbiotics in poultry nutrition, there is limited information on nano-synbiotic formulations combining locally isolated Lactobacillus strains with nano-scale plant-derived prebiotic carriers such as *Taraxacum officinale*. The potential synergistic effects of these nano-synbiotic systems on productive performance and egg quality under practical production conditions remain largely unexplored.

Given the current restrictions on antibiotic growth promoters and the limitations of conventional probiotic and prebiotic systems in terms of stability and delivery efficiency, advanced nano-synbiotic delivery systems using host-adapted probiotic strains and plant-derived nano-prebiotics are required to enhance productivity and production efficiency.

Therefore, the present study aimed to evaluate the effects of dietary supplementation with nano-Taraxacum officinale and nano-synbiotic formulations containing locally isolated *Lactobacillus gasseri* and *Lactobacillus helveticus* on productive performance traits, including egg production, egg weight, in laying hens.

Materials and Methods

Source of nano-synbiotic product

The nano-synbiotic powder (Nano-*Taraxacum officinale*), consisting of lactic acid bacteria (*Lactobacillus gasseri* and *Lactobacillus helveticus*) loaded onto inulin-rich chicory root powder, was obtained as a ready-to-use product that had been previously developed and fully characterized. The product contained no less than10^9 CFU/g of viable lactic acid bacteria. Previous results showed its effectiveness in improving the physiological performance of laying hens before its development, as reported by Al-Salhi *et al*. (2026). The final form of the nano synbiotic product (*Taraxacum offic*inale) used in this experiment is shown in Figure 1 after its development.

Laying hens management

The present study was conducted on 48-week-old laying hens at a private poultry farm in Thi Qar Governorate, southern Iraq, which operated under a fully automated management system equipped with environmental monitoring sensors. The trial lasted for eight weeks, during which temperature, ventilation,

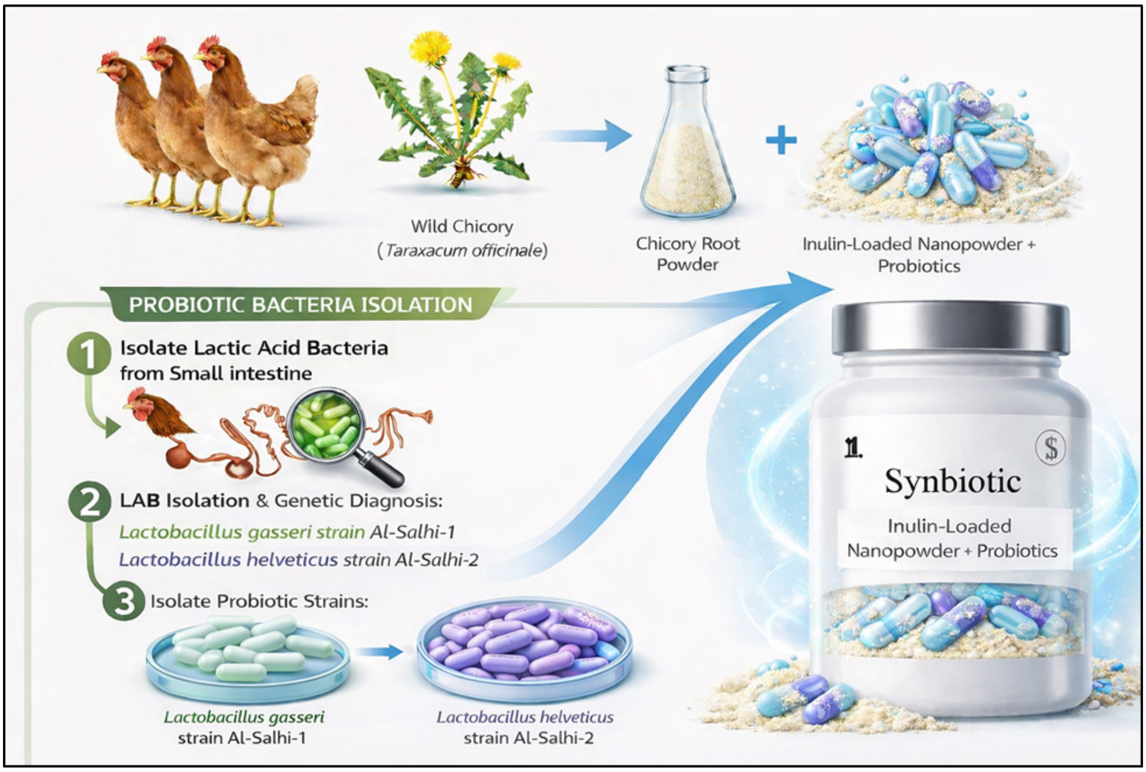

Figure 1: *Nano-Taraxacum officinale synbiotic powder*

and lighting were carefully controlled according to Lohmann Brown management guidelines to provide optimal conditions for egg production.

Birds were housed in cages measuring 1 × 1 m^2 with a height of 60 cm, with eight hens allocated per cage. Each cage was considered one experimental replicate. The basal diet was offered at 130 g/hen/day and divided into two equal meals at 06:00 a.m. and 04:00 p.m., using designated metal feeders for each replicate. The diet contained 17.5% crude protein and 2785 kcal/kg metabolizable energy, as presented in Table 1. Fresh drinking water was continuously provided via nipple drinkers throughout the experimental period.

The nano-synbiotic product, Nano-*Taraxacum officinale*, consisting of lactic acid bacteria loaded onto inulin-rich chicory root powder, was incorporated into the basal diet at graded supplementation levels across the experimental treatments. A total of 96 laying hens were equally distributed among four treatments, each with three replicates (eight hens per replicate), as follows:

T1: basal diet only (control). T2, T3, and T4 (1, 2, and 3 g nano-synbiotic/kg diet, respectively).

The treatments were randomly assigned to evaluate the effects of different inclusion levels of the nano-synbiotic product on the productive performance of laying hens under controlled conditions.

Table 1: *Ingredients and nutrient composition of the laying hen diet*

Ingredient	Inclusion (%)
Yellow maize (corn, ground)	45
Wheat (ground)	20
Soybean meal (48% CP)	20
Vitamin–mineral premix	2
Limestone ($CaCO_3$)	10
Vegetable oil (sunflower)	2.7
Salt (NaCl)	0.3
Total	100
Calculated Nutrients Composition	
Metabolizable Energy (kcal/kg)	2785
Crude Protein (%)	17.5
Energy : Protein Ratio	160
Crude Fiber (%)	3.7
Calcium (%)	4.3
Available Phosphorus (%)	0.47
Lysine (%)	0.84
Methionine (%)	0.37
Methionine + Cystine (%)	0.67
Metabolizable Energy (kcal/kg)	2785

The diet was formulated to provide a balanced nutrient profile suitable for 48-week-old laying hens, supporting consistent egg production and shell quality (NRC, 1994).

Measurement of production indicators

Production performance traits, including Hen-day Egg Production (HD %), Average Egg Weight (g), Daily Egg Mass (g/hen/day), Feed Conversion Ratio (FCR, g feed/g egg), and Economic Feed Efficiency (EE %), were calculated for each 14-day period according to the methods and equations described by Ogbuokiri (2018), Lochmann Tierzucht GmbH (2020).

$$HD\% = \frac{Number\ of\ eggs\ produced\ per\ day}{Number\ of\ hens} \times 100$$

$$Average\ Egg\ Weight\ (g) = \frac{Total\ weight\ of\ eggs\ produced}{Number\ of\ eggs\ produced}$$

$$Egg\ Mass\ (g/hen/day) = \frac{HD(\%) \times Average\ Egg\ Weight\ (g)}{100}$$

$$Feed\ Conversion\ Ratio(FCR) = \frac{Feed\ Intake\ (g/hen/day)}{Daily\ Egg\ Mass\ (g/hen/day)}$$

$$Economic\ Feed\ Efficiency(EE)\% = \frac{Daily\ Egg\ \text{Value (IQD)}}{Daily\ feed\ cost\ \text{(IQD)}} \times 100$$

Daily Egg Value = Daily Egg Mass (kg) × Egg Price (IQD/kg)

Economic evaluation

Economic feed efficiency was calculated based on the prevailing market prices in Iraq during the experimental period (Ogbuokiri, 2018). The average market price of one egg carton (30 eggs) was 5,000 Iraqi dinars (IQD), while the average cost of one kilogram of layer feed was 1,000 IQD according to local market fluctuations. Egg value per kilogram was estimated based on the average egg weight recorded in the present study. Economic feed efficiency was calculated as the ratio between daily egg value and daily feed cost and expressed as a percentage.

Statistical analysis

All data were statistically analyzed using (SPSS, 2018). Differences among treatment means were assessed by one-way analysis of variance (ANOVA), and significant differences were further separated using Duncan's Multiple Range Test at a significance level of $p < 0.05$.

Results and Discussion

Effect of nano-synbiotic supplementation on hen-day egg production

The results presented in Table 2 clearly demonstrate a significant effect ($P < 0.05$) of nano-synbiotic supplementation on hen-day egg production (HD %) throughout the experimental period compared with the control group.

The third treatment (2 g/kg diet) consistently recorded the highest HD % values across all production phases (48–56 weeks of age), followed closely by the fourth treatment (3 g/kg diet), whereas the control group exhibited the lowest production percentages during all measurement intervals. This pattern indicates a positive dose-dependent response up to 2 g/kg, with minimal additional improvement observed at 3 g/kg, suggesting the attainment of a functional saturation level at the 2 g/kg inclusion rate.

Table 2: *Effect of nano-synbiotic supplementation on hen-day egg production (Mean ± SE)*

Treatment	(48–50 wk)	(50–52 wk)	(52–54 wk)	(54–56 wk)	Overall mean
T1 Control	88.4 ± 0.32 c	87.8 ± 0.35 c	87.0 ± 0.38 c	86.5 ± 0.41 c	87.4 ± 0.29 c
T2 (1 g/kg)	89.8 ± 0.30 b	89.4 ± 0.33 b	88.9 ± 0.36 b	88.5 ± 0.39 b	89.2 ± 0.27 b
T3 (2 g/kg)	91.6 ± 0.28 a	91.5 ± 0.30 a	91.3 ± 0.32 a	91.1 ± 0.34 a	91.4 ± 0.24 a
T4 (3 g/kg)	91.3 ± 0.29 a	91.2 ± 0.31 a	91.0 ± 0.34 a	90.8 ± 0.36 a	91.1 ± 0.25 a
Sig.	*	*	*	*	*

**Different letters within the same column indicate significant differences (P < 0.05).*

Table 3: *Effect of nano-synbiotic supplementation on Egg weight (Mean ± SE)*

Treatment	(48–50 wk)	(50–52 wk)	(52–54 wk)	(54–56 wk)	Overall Mean
T1 Control	65.4 ± 0.20 c	65.8 ± 0.21 c	66.1 ± 0.22 c	66.4 ± 0.23 c	65.9 ± 0.18 c
T2 (1 g/kg)	66.0 ± 0.19 b	66.4 ± 0.20 b	66.7 ± 0.21 b	67.0 ± 0.22 b	66.5 ± 0.17 b
T3 (2 g/kg)	66.8 ± 0.18 a	67.2 ± 0.17 a	67.5 ± 0.24 a	67.8 ± 0.25 a	67.3 ± 0.16 a
T4 (3 g/kg)	66.6 ± 0.16 a	67.0 ± 0.19 a	67.3 ± 0.20 a	67.6 ± 0.21 a	67.1 ± 0.15 a
Sig.	*	*	*	*	*

**Different letters within the same column indicate significant differences (P < 0.05).*

Effect of nano-synbiotic supplementation on average egg weight

Table 3 shows significant differences ($P < 0.05$) in average egg weight among treatments throughout the experimental period. Treatments T3 (2 g/kg) and T4 (3 g/kg) recorded higher egg weights compared with the control group, while T2 showed intermediate values.

The highest overall mean was observed in T3, with values closely comparable to T4, indicating that supplementation at 2 g/kg achieved the optimal response without further significant improvement at the higher inclusion level.

The improvement in egg weight followed a similar pattern to hen-day egg production, reflecting a consistent enhancement in productive performance in the higher supplementation groups compared with the control.

Effect of nano-synbiotic supplementation on egg mass

Table 4 revealed significant differences ($P < 0.05$) in egg mass among treatments during all experimental periods. The highest egg mass values were recorded in T3 (2 g/kg), followed closely by T4 (3 g/kg), whereas the control group showed the lowest values throughout the study.

The overall mean confirmed that supplementation at 2 g/kg resulted in the greatest improvement in egg mass, with no substantial additional increase observed at the higher inclusion level. T2 showed moderate improvement compared with the control.

The increase in egg mass was consistent with the observed improvements in hen-day egg production and egg weight, indicating a clear positive response in productive performance among the supplemented groups.

Table 4: *Effect of nano-synbiotic supplementation on egg mass (Mean ± SE)*

Treatment	(48–50 wk)	(50–52 wk)	(52–54 wk)	(54–56 wk)	Overall Mean
T1 Control	57.81 ± 0.27 c	57.77 ± 0.23 c	57.51 ± 0.30 c	57.44 ± 0.32 c	57.63 ± 0.22 c
T2 (1 g/kg)	59.27 ± 0.26 b	59.36 ± 0.27 b	59.30 ± 0.29 b	59.30 ± 0.31 b	59.31 ± 0.21 b
T3 (2 g/kg)	61.19 ± 0.28 a	61.49 ± 0.26 a	61.63 ± 0.27 a	61.66 ± 0.26 a	61.49 ± 0.25 a
T4 (3 g/kg)	60.83 ± 0.25 a	61.10 ± 0.22 a	61.24 ± 0.25 a	61.34 ± 0.30 a	61.13 ± 0.20 a
Sig.	*	*	*	*	*

**Different letters within the same column indicate significant differences (P < 0.05).*

Effect of nano-synbiotic supplementation on conversion ratio (FCR)

As presented in Table 5, feed conversion ratio was significantly affected ($P < 0.05$) by the different supplementation levels. The control group exhibited the highest FCR values across all periods, indicating lower efficiency of feed utilization, whereas T3 (2 g/kg) recorded the lowest values. Both T3 and T4 showed clear improvements compared with the control and T2, with T3 achieving the most efficient feed utilization overall. The differences were consistent throughout the experimental periods, reflecting a stable improvement in conversion efficiency.

The reduction in FCR observed in the higher supplementation groups corresponds with the previously noted increases in egg production and egg mass, confirming a better productive response in these treatments.

Table 5: *Effect of nano-synbiotic supplementation on feed conversion ratio (Mean ± SE)*

Treatment	(48–50 wk)	(50–52 wk)	(52–54 wk)	(54–56 wk)	Overall Mean
T1 Control	2.25 ± 0.02 a	2.25 ± 0.01 a	2.26 ± 0.04 a	2.26 ± 0.02 a	2.26 ± 0.02 a
T2 (1 g/kg)	2.19 ± 0.01 b	2.19 ± 0.03 b	2.19 ± 0.01 b	2.19 ± 0.02 b	2.19 ± 0.02 b
T3 (2 g/kg)	2.13 ± 0.01 c	2.11 ± 0.02 c	2.11 ± 0.02 c	2.11 ± 0.01 c	2.11 ± 0.01 c
T4 (3 g/kg)	2.14 ± 0.03 c	2.13 ± 0.01 c	2.12 ± 0.02 c	2.12 ± 0.03 c	2.13 ± 0.02 c
Sig.	*	*	*	*	*

**Different letters within the same column indicate significant differences (P < 0.05).*

Discussion of productive performance traits

The present findings demonstrated a significant improvement in productive performance following dietary inclusion of the nano-synbiotic additive. Hen-day egg production (HD %), average egg weight, egg mass, and feed conversion ratio (FCR) were all positively influenced compared with the control group. The 2 g/kg supplementation level (T3) achieved the highest numerical values across most performance parameters. Notably, the 3 g/kg level (T4) produced very similar results without statistically significant differences compared with T3, whereas the 1 g/kg level (T2) showed moderate but consistent improvement.

The comparable performance observed between 2 and 3 g/kg suggests the occurrence of biological functional saturation at 3 g/kg diet, where increasing the inclusion level beyond 2 g/kg did not yield additional significant productive gains. This plateau effect likely reflects the attainment of optimal microbial colonization capacity and metabolic interaction within the gastrointestinal ecosystem. Once gut microbial balance and epithelial interaction reach physiological equilibrium, further increases in additive concentration may not proportionally enhance nutrient utilization or production output.

The improvement in productive traits can be mechanistically attributed to enhanced gut microbial modulation. The nano-formulated lactic acid bacteria, delivered alongside an inulin-rich carrier, likely improved microbial adhesion, survivability, and colonization efficiency within the intestinal tract. Such stabilization of beneficial microbiota supports enhanced enzymatic activity, improved nutrient digestibility, and reduced pathogenic microbial pressure. Collectively, these effects contribute to improved energy and protein utilization, which are directly reflected in higher egg production rate and increased egg mass (Al Salhi *et al*., 2022; Zhang *et al*., 2025).

The parallel improvement observed in egg mass and FCR further indicates a coordinated enhancement in nutrient efficiency rather than isolated trait stimulation. This suggests that the nano-synbiotic did not merely stimulate oviposition frequency but improved overall metabolic efficiency. Similar dose-response plateau phenomena have been described in probiotic and synbiotic supplementation studies, where optimal responses are achieved within a specific inclusion range before reaching a biological ceiling effect (Al-Salhi *et al*., 2023).

Although variations among studies may arise due to differences in strain specificity, environmental conditions, diet composition, and hen age, the overall direction of the current results aligns with contemporary literature emphasizing the role of microbiome modulation in enhancing laying hen productivity (Al Salhi and Al Shatty, 2023; Xu *et al*., 2023; Gallazzi *et al*., 2024; Alagawany *et al*., 2025).

Effect of nano-synbiotic supplementation on economic feed efficiency

Table 6 indicates that economic feed efficiency was significantly influenced ($P < 0.05$) by the different supplementation levels. The control group recorded the lowest economic return throughout the experimental periods, whereas T3 (2 g/kg) achieved the highest values, followed closely by T4 (3 g/kg).

The overall means confirm that supplementation at 2 g/kg provided the greatest economic advantage, reflecting a superior balance between egg value and feed cost. Although T4 showed comparable performance, no substantial additional economic gain was observed beyond the 2 g/kg level. The progressive increase in economic efficiency from T1 to T3 demonstrates that improvements in productive performance were directly translated into higher financial returns under the prevailing market conditions.

Table 6: *Effect of nano-synbiotic supplementation on Economic Feed Efficiency (Mean ± SE)*

Treat-ment	(48–50 wk)	(50–52 wk)	(52–54 wk)	(54–56 wk)	Overall mean
T1 Control	121.9 ± 0.6 c	121.8 ± 0.5 c	121.3 ± 0.4 c	121.1 ± 0.7 c	121.5 ± 0.5 c
T2 (1 g/kg)	125.0 ± 0.5 b	125.3 ± 0.6 b	125.1 ± 0.7 b	125.0 ± 0.6 b	125.1 ± 0.6 b
T3 (2 g/kg)	129.2 ± 0.4 a	129.8 ± 0.5 a	130.1 ± 0.6 a	130.2 ± 0.8 a	129.8 ± 0.4 a
T4 (3 g/kg)	128.4 ± 0.7 a	129.0 ± 0.4 a	129.3 ± 0.7 a	129.5 ± 0.7 a	129.1 ± 0.6 a
Sig.	*	*	*	*	*

**Different letters within the same column indicate significant differences (P < 0.05).*

Discussion of economic efficiency

The economic evaluation clearly demonstrated that the observed improvements in productive performance were directly translated into enhanced financial return. The increase in hen-day egg production and egg mass in the supplemented groups resulted in greater total egg output per replicate, thereby increasing gross revenue compared with the control treatment. Simultaneously, the improvement in feed conversion ratio (FCR) reduced feed cost per unit of egg mass produced, contributing to a higher net profit margin.

Although the inclusion of the nano-synbiotic additive increased feed cost, this additional expense was compensated by improved biological efficiency. The 2 g/kg supplementation level (T3) generated the highest economic return, as it achieved the most favorable balance between productivity enhancement and additive cost. In contrast, the 3 g/kg level (T4), despite yielding biologically comparable results, did not proportionally increase economic profitability. This outcome indicates that the biological functional saturation observed at the higher inclusion level was also reflected economically, where additional supplementation failed to generate sufficient incremental revenue to justify the higher cost.

These findings align with previous research indicating that improvements in feed efficiency and egg output through probiotic or synbiotic supplementation can significantly enhance economic return in laying hen production systems (Wang *et al*., 2024). Likewise, Abd El-Hack *et al*. (2023) emphasized that the economically optimal inclusion level of functional feed additives is not necessarily the highest biological dose, but rather the level at which productive gains outweigh the cost of supplementation. This nonlinear relationship between additive level and financial return supports the plateau effect observed in the present study.

Therefore, the current results confirm that nano-synbiotic supplementation improves economic efficiency when applied within the optimal dose range. Under the prevailing market conditions used in this study, 2 g/kg diet represented the most economically advantageous level, whereas increasing the inclusion to 3 g/kg achieved physiological stability without generating additional economic benefit. These findings reinforce the practical applicability of the additive in commercial production systems aiming to maximize productivity while maintaining cost-effectiveness.

Furthermore, a healthy environment plays a key role in the effectiveness of these bioactive or probiotic substances in preventing contamination, and recent studies have indicated that the use of detergents derived from natural organic waste (Al-Salhi *et al*., 2025; Naser *et al*., 2025) contributes to reducing the harmful microbial load in poultry farms.

Conclusions and Recommendations

Dietary supplementation with nano-synbiotic (Nano-*Taraxacum officinale*) at 2 g/kg diet significantly improved productive performance and economic efficiency in laying hens, achieving the highest egg production, egg weight, egg mass, and feed conversion efficiency. Increasing the supplementation to 3 g/kg diet did not provide additional benefits, indicating biological and economic saturation at 2 g/kg. Further studies are recommended to evaluate gut microbiota, egg quality parameters, and nutrient digestibility to better understand the mechanisms underlying these responses.

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Novelty Statement

This study evaluates a nano-synbiotic formulation based on *Lactobacillus gasseri* and *Lactobacillus helveticus* loaded onto an inulin-rich Taraxacum officinale carrier in laying hens. It highlights the optimal supplementation level that improves productive performance and economic efficiency under practical production conditions.

Author's Contribution

Nibras M. Abaas: Conceived and designed the study and wrote the manuscript.
Hiba Sh. Hussein: Conducted the experimental work and data collection.
Zainab A. Shakeer: Contributed to data analysis and manuscript preparation.
Saif Sh. Kamel: Performed statistical analysis and reviewed the final manuscript.

Generative AI and AI-assisted technology statement
The authors declare that no generative AI and AI assisted technology was used in the creation of this manuscript.

Conflict of interest
The authors have declared no conflict of interest.

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