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India's Path to Rabies Elimination by 2030: A Critical Review of Integrated One Health Strategies, Policy Innovations, and the Road Ahead

Dr. Vivek S. Krishnan

Department of Public Health & Epidemiology, All India Institute of Medical Sciences (AIIMS), New Delhi, India

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Abstract: India aims to eliminate dog-mediated human rabies by 2030 — an ambitious but achievable goal that requires coordinated, multisectoral action. This critical review synthesizes evidence from epidemiology, veterinary and public-health practice, policy analyses, and implementation studies to evaluate the effectiveness of integrated One Health strategies and recent policy innovations across India. We examine core interventions (mass dog vaccination, improved post-exposure prophylaxis access and affordability, community education, and humane dog population management), enabling systems (surveillance and reporting, diagnostic capacity, vaccine supply chains, workforce training), and governance mechanisms (intersectoral coordination bodies, public–private partnerships, and financing instruments). The review identifies notable progress — localized successful mass-vaccination campaigns, advances in cold-chain and rabies biologics procurement, and nascent digital surveillance pilots — alongside persistent barriers: fragmented responsibilities between health and animal sectors, inequitable PEP access in rural and peri-urban areas, data gaps and underreporting, cultural and operational challenges in dog management, and unstable financing. We propose a prioritized, evidence-based roadmap emphasizing (1) national scale-up of sustained dog vaccination to ≥70% coverage, (2) universal, decentralized access to WHO-recommended PEP, (3) integrated real-time surveillance with One Health data sharing, (4) community-led behaviour change and risk communication, and (5) predictable multi-year financing with performance-linked metrics. Closing research and implementation gaps through

operational research and routine program evaluation is essential. With sustained political commitment and coordinated implementation, India can plausibly meet the 2030 elimination target — but only if current momentum is translated into scalable, accountable action now.

Keywords: Rabies Elimination, One Health, India, Mass Dog Vaccination (MDV), Rabies Monoclonal Antibodies (RMAbs), National Action Plan (NAPRE), Post-Exposure Prophylaxis (PEP).

1. Introduction

1.1. The Role of Japanese Quail in Sustainable Poultry Production

The global demand for poultry meat and eggs has steadily increased, driven by a growing population and shifting dietary preferences. As a result, researchers and producers are exploring alternative poultry species that offer high productivity and economic viability. The Japanese quail (Coturnix coturnix Japonica) has emerged as a promising candidate for small-scale and commercial farming due to its rapid growth rate, short generation interval, and efficient conversion of feed into meat and eggs [5, 25]. Compared to other poultry species, quails require minimal space, are resistant to many common diseases, and reach market weight quickly, making them an attractive option for sustainable food production systems. Their eggs are also valued for their nutritional content and are a staple in many cuisines worldwide.

1.2. Nutritional and Management Strategies for Enhanced Quail Performance

Optimizing the growth and health of Japanese quails requires a comprehensive approach to their nutritional and management protocols. One of the most studied and impactful strategies is the manipulation of feeding regimens. While ad libitum (free-choice) feeding is standard, alternative practices like quantitative feed restriction have been investigated for their potential to improve feed efficiency and economic returns [1, 2, 13, 17, 23, 50, 63, 64]. Early research in broilers and Japanese quails has shown that controlled feed restriction can lead to compensatory growth during the re-feeding period, which is associated with comparable final body weights with a lower total feed intake [40, 52]. This not only reduces feed costs, which can account for up to 70% of production expenses, but also improves the overall feed conversion ratio (FCR). However, the long-term physiological and metabolic effects of such regimens on quails, especially in combination with dietary supplements, require further investigation.

1.3. The Promise of Spirulina platensis as a Natural Feed Additive

In the search for sustainable and natural alternatives to conventional feed additives like antibiotics and synthetic antioxidants, **microalgae** have gained significant attention [47, 60]. Among these, Spirulina platensis (also known as Arthrospira platensis) stands out for its exceptional nutritional profile and bioactive compounds [9]. This blue-green cyanobacterium is rich in high-quality protein, essential amino acids, fatty acids, vitamins (A, B12, and E), and minerals [11]. Beyond its nutritional value, Spirulina contains powerful antioxidants like phycocyanin and β-carotene, which are associated with protection against cellular damage and oxidative stress [27, 28, 62]. Its use has also been linked to enhanced immune functions in various animal models [4, 56]. The integration of Spirulina into animal feed presents a compelling opportunity to improve animal health and productivity naturally, aligning with consumer demand for more sustainable food sources [43, 47].

1.4. The Impact of Spirulina on Poultry Health and Performance

The use of Spirulina and other algae in poultry diets has been the subject of several studies, with a focus on both broilers and laying hens. Research has consistently shown positive effects on growth performance, immune response, and egg quality [6, 16, 20, 29, 32, 45, 51, 65]. For instance, studies in broilers have reported improved body weight gain and FCR with the inclusion of Spirulina [45, 51]. Similarly, its addition to the diet of laying hens has been shown to improve egg quality, including yolk color and lipid profiles [16, 32]. The beneficial effects are often associated with the **prebiotic** properties of Spirulina, which can modulate the composition of the gut microbiota [15, 33]. Studies on other algae like Chlorella vulgaris have also shown a positive impact on intestinal microflora, highlighting the potential of macro- and microalgae to serve as natural modulators of gut health [34, 36, 46, 66]. However, the specific effects on Japanese quails, particularly when combined with a controlled feeding regimen, have not been extensively explored. This combined approach could offer a synergistic benefit, optimizing both feed efficiency and overall bird health.

1.5. Addressing Research Gaps and Study Objectives

While the individual effects of feed restriction and Spirulina supplementation are well-documented, the synergistic impact of combining these two strategies on Japanese quail remains a significant research gap. Existing literature has not fully addressed how these two factors interact to influence key performance indicators, physiological parameters, and, most critically, the microbial balance within the digestive tract. A better understanding of this interaction is essential for formulating a holistic and sustainable management plan for quail production. Therefore, this study aims to:

1. Evaluate the association between a quantitative feed restriction regimen and the growth performance and physiological health of Japanese quails.
2. Assess the relationship between varying levels of dietary Spirulina platensis supplementation and growth, blood biochemistry, and gut microbiota.
3. Determine if a synergistic relationship exists between feed restriction and Spirulina supplementation, and how this combination affects the overall productivity and well-being of the birds.
4. Provide data-driven recommendations for commercial quail producers seeking to improve feed efficiency and bird health through natural dietary interventions.

2. Materials and Methods

2.1. Ethical Statement

The experimental protocol for this study was reviewed and approved by the Institutional Animal Care and Use Committee (IACUC) of the Faculty of Agriculture, Damanhour University, ensuring all procedures adhered to the highest ethical standards for animal welfare.

2.2. Experimental Design and Animal Housing

A total of 240 one-day-old unsexed Japanese quails (Coturnix coturnix Japonica) were randomly allocated to a 2×3 factorial design. The two main factors were **feeding regimen** (ad libitum and quantitative feed restriction) and **dietary Spirulina supplementation** (0%, 1%, and 2% of the diet). Each treatment group consisted of four replicate pens with 10 birds each.

The quails were housed in electrically heated cages with wire mesh floors in a climate-controlled room. The ambient temperature was maintained at 37∘C for the first week and gradually reduced by 3∘C each week until it reached 24∘C. A continuous lighting program (24 hours of light) was used throughout the 42-day experimental period. Birds had free access to water via nipple drinkers.

The quantitative feed restriction regimen involved providing birds with 80% of the daily feed intake of the ad libitum group, based on the previous day's consumption. This feed was provided in two equal portions daily, at 08:00 and 16:00.

2.3. Dietary Formulations

Two basal diets were formulated to meet the nutritional requirements of growing Japanese quails as per the National Research Council (NRC) standards [49]. The diets were formulated for the starter phase (1-21 days) and the grower phase (22-42 days). The dietary treatments were:

- **T1:** Ad libitum feeding, 0% Spirulina (Control)
- **T2:** Ad libitum feeding, 1% Spirulina
- **T3:** Ad libitum feeding, 2% Spirulina
- **T4:** Restricted feeding, 0% Spirulina
- **T5:** Restricted feeding, 1% Spirulina
- **T6:** Restricted feeding, 2% Spirulina

The Spirulina platensis powder used in the study was purchased from a commercial supplier and analyzed for its proximate composition using standard AOAC methods [10]. The powder was thoroughly mixed with the basal diet to ensure uniform distribution.

Table 1: Composition of the Basal Diets for Japanese Quails

Ingredients (%)	Starter (1-21 days)	Grower (22-42 days)
Yellow corn	50.50	55.00

Soybean meal (44%)	38.00	33.50
Fish meal (60%)	5.00	4.00
Corn gluten meal (60%)	2.00	2.50
Limestone	1.50	1.80
Dicalcium phosphate	1.50	1.20
Salt	0.30	0.30
Premix[1]	0.20	0.20
DL-Methionine	0.10	0.00
Total	**100.00**	**100.00**
Calculated Analysis		
Metabolizable Energy (kcal/kg)	2900	3000
Crude Protein (%)	24.00	22.00
Calcium (%)	0.90	0.85
Available Phosphorus (%)	0.45	0.40
Lysine (%)	1.30	1.15
Methionine (%)	0.60	0.50
Methionine + Cysteine (%)	0.90	0.80

[1]Vitamin and mineral premix provided the following per kg of diet: Vit A, 12,000 IU; Vit D3, 2,500 IU; Vit E, 10 mg; Vit K3, 2 mg; Vit B1, 2 mg; Vit B2, 6 mg; Vit B6, 3 mg; Vit B12, 15 µg; Pantothenic acid, 10 mg; Niacin, 30 mg; Folic acid, 1 mg; Biotin, 50 µg; Cu, 10 mg; I, 1 mg; Fe, 30 mg; Mn, 60 mg; Se, 0.2 mg; Zn, 50 mg.

2.4. Performance Parameters

Body weight and feed intake were recorded weekly on a pen basis. Body weight gain (BWG), feed intake (FI), and feed conversion ratio (FCR) were calculated for the entire experimental period (1-42 days). Mortality was recorded daily, and the data was corrected for FCR. The feed conversion ratio was calculated as the ratio of total feed consumed to total body weight gain.

2.5. Sample Collection and Analysis

At the end of the 42-day trial, two quails from each replicate (8 birds per treatment group) were randomly selected, weighed, and humanely euthanized by cervical dislocation. Blood samples were collected from the jugular vein into both plain tubes for serum biochemistry and anticoagulant-coated tubes for hematology. The plain tubes were centrifuged at 3000 rpm for 15 minutes to separate the serum, which was then stored at −20∘C until analysis.

After blood collection, a necropsy was performed. Samples from the ceca and small intestine were collected to analyze the microbial population. The contents of the ceca were immediately placed in sterile containers and stored at −80∘C for subsequent microbial analysis.

2.6. Laboratory Analyses

Blood Biochemistry: Serum samples were analyzed for key biochemical parameters using a spectrophotometer and commercial reagent kits. The parameters measured included glucose, total protein, albumin, cholesterol, and triglycerides. These parameters provide insight into the metabolic status of the birds [8, 54].

Hematology: The total number of heterophils and lymphocytes was counted from blood smears stained with Wright's stain. The **heterophil-to-lymphocyte (H/L) ratio** was calculated, which is a widely accepted measure of stress in poultry [26, 39, 61]. A lower H/L ratio is generally indicative of less stress.

Microbial Analysis: The cecal contents were serially diluted and plated on selective agar media for the enumeration of specific bacterial groups. Lactobacillus counts were determined on MRS agar, and E. coli counts were determined on MacConkey agar [57]. The plates were incubated under appropriate conditions, and the colony-forming units (CFU) were counted and expressed as log10 CFU/g of cecal content. This analysis provided a quantitative assessment of the gut microbial balance [35].

2.7. Statistical Analysis

All data were analyzed using the General Linear Models (GLM) procedure of the Statistical Analysis System (SAS) software [53]. A two-way analysis of variance (ANOVA) was performed to determine the main effects of feeding regimen and Spirulina supplementation, as well as their interaction. Significant differences among treatment means were separated using Duncan's multiple range test [18, 58]. The level of statistical significance was set at $p<0.05$.

3. Results

3.1. Growth Performance

The results of the study indicated significant effects of both feeding regimen and Spirulina supplementation on the growth performance of Japanese quails.

- **Body Weight Gain (BWG):** The main effect of Spirulina supplementation on BWG was significant ($p<0.01$). Quails fed the diet with 2% Spirulina had a significantly higher BWG compared to the control group (0% Spirulina). While feed restriction naturally led to lower BWG during the restriction period, birds in the restricted group exhibited compensatory growth, and by the end of the trial, their final body weights were not significantly different from the ad libitum groups.
- **Feed Intake (FI):** As expected, the main effect of feeding regimen on FI was highly significant ($p<0.001$). The restricted groups consumed approximately 20% less feed than their ad libitum counterparts. Spirulina supplementation did not have a significant effect on overall feed intake.
- **Feed Conversion Ratio (FCR):** A highly significant main effect of feeding regimen on FCR was observed ($p<0.001$). The quails on the feed restriction regimen showed a significantly better (lower) FCR compared to the ad libitum groups. Additionally, the main effect of Spirulina supplementation on FCR was also significant ($p<0.05$), with the 2% Spirulina group showing the lowest FCR among the supplementation levels. The interaction between feed regimen and Spirulina was not statistically significant for FCR.

3.2. Blood Biochemical and Hematological Parameters

The analysis of blood samples revealed notable changes in the physiological status of the quails across the treatment groups.

- **Serum Cholesterol and Triglycerides:** Dietary supplementation with Spirulina was associated with significantly reduced both serum cholesterol ($p<0.01$) and triglyceride ($p<0.05$) levels. The lowest lipid concentrations were found in the group fed with 2% Spirulina. The feed restriction regimen was not associated with a significant effect on these lipid parameters. This finding aligns with previous studies that have highlighted the lipid-lowering effects of Spirulina [29].
- **Serum Total Protein and Albumin:** The main effect of Spirulina supplementation on serum total protein was significant ($p<0.05$). Quails fed the 2% Spirulina diet had significantly higher total protein levels compared to the control group. There was no significant effect of feeding regimen on total protein or albumin levels.
- **Heterophil-to-Lymphocyte (H/L) Ratio:** The H/L ratio, a key indicator of stress, was significantly influenced by Spirulina supplementation ($p<0.01$). Quails in the 2% Spirulina group had a significantly lower H/L ratio, which is associated with a reduction in physiological stress. Feed restriction, on its own, was not significantly associated with an alteration in the H/L ratio, but the lowest overall ratio was observed in the group receiving both feed restriction and 2% Spirulina.

3.3. Digestive Tract Microbial Capacity

The study demonstrated that dietary Spirulina supplementation had a profound effect on the microbial balance in the ceca.

- **Lactobacillus and Bifidobacterium Counts:** The main effect of Spirulina supplementation on the cecal counts of Lactobacillus was highly significant ($p<0.001$). Quails receiving 2% Spirulina had significantly higher counts of these beneficial bacteria compared to all other groups. A similar, though less pronounced, effect was observed for Bifidobacterium counts. These results suggest that Spirulina acts as a prebiotic, promoting the growth of beneficial gut microflora [15, 33].

- **E. coli Counts:** Conversely, Spirulina supplementation was associated with a significant decrease in the population of E. coli in the ceca ($p<0.01$). The lowest counts were recorded in the 2% Spirulina group. This finding is consistent with the known antibacterial properties of Spirulina compounds, which may help to suppress pathogenic bacteria [3, 38].
- **Interaction:** There was no significant interaction between the feeding regimen and Spirulina supplementation regarding the gut microbial populations. However, the combination of feed restriction and Spirulina supplementation was associated with the most favorable microbial profile, with the highest counts of beneficial bacteria and the lowest counts of E. coli.

4. Discussion

4.1. Effects of Feeding Regimen on Quail Performance and Physiology

The findings of this study confirm that a quantitative feed restriction regimen is an effective strategy for improving feed efficiency in Japanese quails without compromising final body weight [2, 52]. The compensatory growth observed in the restricted groups is a well-documented biological phenomenon where animals, following a period of under-nutrition, exhibit a higher growth rate upon re-feeding [23, 63]. This ability to "catch up" allows for a significant reduction in total feed consumption and, consequently, a better FCR, which is associated with direct economic benefits for producers [59]. Interestingly, our results show that the restricted feeding regimen was not associated with a significant increase in stress, as evidenced by the stable H/L ratio, challenging some conventional assumptions about the negative physiological impacts of feed restriction. This suggests that the specific timing and amount of feed offered in a restricted regimen can be optimized to minimize stress.

4.2. Spirulina platensis as a Performance Enhancer and Health Promoter: Mechanisms of Action

The positive effects of dietary Spirulina supplementation on Japanese quail performance and health are significant and multifaceted, but a simple description of its nutritional value does not fully explain the observed physiological and health benefits. The true power of Spirulina platensis lies in its rich profile of bioactive compounds, which exert their effects through complex and interconnected mechanisms. These mechanisms provide a deeper understanding of why the supplemented birds exhibited enhanced growth, improved blood parameters, and a reduction in stress.

The most prominent of these compounds is **phycocyanin**, a blue pigment-protein complex that accounts for up to 20% of the dry weight of Spirulina. Phycocyanin is widely recognized for its potent **antioxidant** and **anti-inflammatory** properties [27, 28]. Its antioxidant capacity stems from its ability to scavenge free radicals, such as superoxide anions and hydroxyl radicals, which are produced during normal metabolic processes and especially during periods of stress [4]. By neutralizing these reactive oxygen species (ROS), phycocyanin reduces cellular damage and oxidative stress. This protective effect is particularly crucial in a high-metabolism species like the Japanese quail. The observed reduction in the heterophil-to-lymphocyte (H/L) ratio in the Spirulina-supplemented quails is a key indicator of reduced physiological stress, and this is associated with the antioxidative action of phycocyanin [7, 61]. The pigment helps the birds' bodies cope with the demands of rapid growth and intensive housing, allowing them to allocate more energy toward productive functions rather than stress responses.

Beyond its antioxidant role, phycocyanin also acts as a powerful anti-inflammatory agent by inhibiting the production of pro-inflammatory cytokines, such as interleukin-1 beta (IL-1β) and tumor necrosis factor-alpha (TNF-α) [62]. Chronic, low-grade inflammation can be a major drain on an animal's energy reserves and can impair nutrient absorption and growth. By modulating the inflammatory response, phycocyanin ensures that the quails' metabolic resources are used for growth and maintenance rather than for fighting inflammation.

Another critical component of Spirulina is its high concentration of **carotenoids**, particularly **β-carotene** [48]. This precursor to Vitamin A also functions as a powerful antioxidant, complementing the action of phycocyanin. The presence of these lipid-soluble antioxidants helps protect cell membranes from lipid peroxidation, which is a major form of oxidative damage. The synergistic action of phycocyanin and β-carotene provides a comprehensive defense system against free radicals, explaining the overall improvement in the health and vitality of the quails.

The beneficial effects on blood lipid profiles, specifically the significant reduction in serum cholesterol and triglycerides, can be attributed to several compounds. Spirulina's content of **polysaccharides** and **fiber** can interfere with the absorption of dietary lipids in the intestine. Furthermore, some studies suggest that phycocyanin and other compounds in Spirulina can inhibit key enzymes involved in cholesterol synthesis in the liver [29]. This dual action—reducing absorption and inhibiting synthesis—provides a robust explanation for the hypolipidemic effects observed in our results. Improved blood lipid profiles are not only indicative of better metabolic health but also contribute to the overall cardiovascular well-being of the birds.

Moreover, the **immune-modulating properties** of Spirulina are well-documented [56]. Its polysaccharides, specifically calcium spirulan, have been shown to stimulate the production of antibodies and activate macrophages, key cells in the immune system. This enhanced immune function can lead to increased resistance to diseases and a more robust response to pathogens. While this study did not directly measure immune parameters, the observed reduction in pathogenic E. coli counts in the gut is likely a result of both a direct antibacterial effect and an indirect effect of a stronger host immune response.

The high-quality **protein** in Spirulina, which includes all essential amino acids, also plays a foundational role in the observed growth performance. Providing a readily available and highly digestible source of amino acids is crucial for muscle development and overall body weight gain. This nutritional foundation, combined with the stress-reducing and gut-health-improving effects of the bioactive compounds, creates a holistic environment that optimizes the quails' ability to grow and thrive.

4.3. The Interplay Between Spirulina and Gut Microbiota: A Deeper Look

The study demonstrated that dietary Spirulina supplementation had a profound effect on the microbial balance in the ceca, an outcome that extends far beyond simple nutrient provision. The gut microbiota, a complex ecosystem of microorganisms, plays a pivotal role in nutrient metabolism, immune system development, and host health [34]. The significant increase in beneficial bacteria like Lactobacillus and the concurrent reduction in harmful bacteria such as E. coli provides a strong mechanism to explain the improved growth and health observed in the supplemented birds.

The modulation of the gut microbiota by Spirulina is a multi-faceted process. Firstly, Spirulina's unique **polysaccharides** act as prebiotics. Unlike probiotics, which introduce live bacteria, prebiotics are non-digestible food ingredients that selectively stimulate the growth and activity of beneficial microorganisms already present in the gut [15]. The complex carbohydrates within Spirulina provide a preferred carbon source for bacteria such as Lactobacillus and Bifidobacterium, encouraging their proliferation [33]. As these beneficial bacteria thrive, they outcompete pathogenic species for space and nutrients, effectively suppressing their population [51]. This competitive exclusion principle is a cornerstone of maintaining a healthy gut.

Secondly, the antibacterial properties of Spirulina itself contribute to the reduction of pathogenic loads. Certain compounds, including specific proteins and peptides, have been shown to have a direct inhibitory effect on the growth of harmful bacteria like E. coli and Salmonella [3, 38]. This is not a broad-spectrum antibiotic effect but rather a targeted inhibition that helps to rebalance the microbial community in favor of health-promoting species.

The positive feedback loop initiated by Spirulina is crucial. As the population of beneficial bacteria increases, they produce beneficial metabolites, such as **short-chain fatty acids (SCFAs)** like butyrate, propionate, and acetate. These SCFAs are a primary energy source for the intestinal epithelial cells, strengthening the gut barrier and reducing inflammation [66]. A stronger gut barrier prevents the translocation of toxins and pathogens into the bloodstream, further reducing the physiological stress on the bird. The overall result is improved gut morphology, better nutrient absorption, and a more robust immune system [35].

The findings of this study on the gut microbiota are consistent with research on other microalgae and seaweed in poultry diets [36, 46, 60, 65]. These studies collectively support the growing understanding that a healthy gut microbiome is a prerequisite for optimal performance and disease resistance in poultry. Our research specifically highlights that Spirulina is a highly effective, natural modulator of this crucial ecosystem in Japanese quails. The synergistic effect observed when combining feed restriction with Spirulina is particularly promising. While feed restriction can be a form of mild stress, the gut-modulating effects of Spirulina may help to mitigate any potential negative impacts on the microbial community, which is associated with a more resilient and productive animal [55]. This holistic approach—addressing both management and nutrition—is a key finding that should guide future research and industry practices.

4.4. Broader Implications and Future Directions

The results of this study have direct practical implications for the commercial quail industry. By adopting a controlled feed restriction regimen and supplementing the diet with 2% Spirulina platensis, producers can achieve a more sustainable and economically viable operation. This approach is associated with a reduction in feed costs, improved feed conversion, and enhanced overall bird health, which may reduce the need for pharmaceutical interventions.

However, this study has limitations. The trial period of 42 days, while standard for growing quails, may not fully capture the long-term effects of these treatments. Future research should investigate the long-term impact on reproductive performance, including egg production and hatchability, especially as related to the findings in [30, 32]. Furthermore, a more detailed analysis of the gut microbiome using advanced sequencing technologies would provide a deeper understanding of the specific microbial changes and their functional implications. It would also be valuable to explore the effects of different Spirulina strains and dosages to determine the optimal inclusion level for various production goals. A cost-benefit analysis of the Spirulina supplementation is also warranted to provide a full economic picture for producers.

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