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Full Length Research

Evaluation of some selected nutraceuticals (King-Herbs oral solution® Gumbo ND® and Grand Humi Vet®) with Gumboro vaccine for preventing experimental Gumboro disease in broilers

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ABSTRACT: Infectious bursal disease (IBD) is a highly contagious and immunosuppressive viral disease of chickens that causes significant economic losses in poultry production. This study assessed the prophylactic efficacy of three nutraceuticals—King-Herbs Oral Solution®, Gumbo ND®, and Grand HumiVet®—administered alone or in combination with an intermediate Gumboro vaccine in broiler chickens experimentally challenged with a very virulent IBD virus (vvIBDV). A total of 240-day-old Cobb500 broilers were randomly allocated into 12 groups (20 birds/group). Group A served as the negative control, while Group B was the positive control challenged with vvIBDV. Groups C1–C3 received individual nutraceuticals combined with vaccination and vvIBDV challenge. Groups D1–D3 received nutraceuticals without vaccination but were challenged. Groups E1–E3 received nutraceuticals only, while Group F was vaccinated and challenged without nutraceutical supplementation. Nutraceuticals were administered via drinking water from day 5 to day 28. The intermediate Gumboro vaccine was given on days 11 and 22, and the vvIBDV challenge was performed orally on day 29. Clinical signs, cloacal temperature, body weight, and mortality were monitored between 3 and 7 days post-infection (dpi). Bursa of Fabricius samples were collected at 21, 28, 35, and 42 days of age for gross and histopathological evaluation. Birds in the positive control group showed severe clinical signs, elevated cloacal temperatures, reduced body weight, and marked bursal lesions. Nutraceutical–vaccine combinations (C1–C3) provided the highest level of protection, evidenced by minimal clinical signs, improved growth performance, and significantly lower lesion scores. Nutraceutical-only groups showed partial protection, while vaccinated controls exhibited moderate resistance. Overall, supplementation with King-Herbs Oral Solution®, Gumbo ND®, or Grand HumiVet® enhanced vaccine efficacy and reduced the pathological effects of vvIBDV, suggesting their value as adjuncts in IBD control programs.

Keywords: Infectious bursal disease (IBD), Very virulent IBD virus (vvIBDV), Nutraceuticals, Gumboro vaccine and Broiler chickens.

INTRODUCTION

Infectious bursal disease (IBD), commonly known as Gumboro disease, is a highly contagious viral disease of chickens that causes severe immunosuppression, predisposing affected flocks to secondary infections and reduced vaccine responsiveness (Dey *et al*., 2019; Franciosini and Davidson, 2022). The disease is caused by infectious bursal disease virus (IBDV), a non-enveloped, double-stranded RNA virus of the genus *Avibirnavirus* in the family *Birnaviridae* (Ingrao *et al*., 2013). Although chickens are the primary hosts, subclinical infections have been reported in other avian species such as guinea fowls, ducks, quails, and ostriches (Wang *et al*., 2007).

IBDV was first identified in Delaware, United States, in 1962, and since then, very virulent strains (vvIBDV) have emerged globally, causing high morbidity and mortality in susceptible flocks (Eterradossi and Saif, 2013). The virus targets actively dividing B lymphocytes in the bursa of Fabricius (BF), leading to lymphoid depletion, impaired humoral immunity, and increased susceptibility to bacterial, viral, and parasitic diseases (Long *et al*., 2011). Two serotypes of IBDV exist (serotypes I and II), with serotype I containing classical, variant, and very virulent pathotypes capable of causing disease in chickens (Michel and Jackwood, 2017).

Despite the widespread use of live attenuated, intermediate, and immune-complex vaccines, outbreaks of IBD remain common in many poultry-producing countries, including Nigeria. Several factors contribute to vaccination failure, including improper vaccine handling, interference from maternally derived antibodies, antigenic drift of field strains, and suboptimal flock immunity (Ekiri *et al*., 2021; Courtillon *et al*., 2022). Consequently, there is growing interest in integrative approaches that improve vaccine efficacy and overall flock resilience.

Nutraceuticals, bioactive compounds derived from plants, minerals, amino acids, and other natural sources, have gained increasing attention in poultry health management. They are reported to enhance immune responses, improve antioxidant status, modulate gut microbiota, and reduce the severity of viral infections (Costagliola *et al*., 2021). Examples include herbal extracts, organic acids, humic substances, vitamins, and polysaccharides from medicinal plants. Unlike pharmaceuticals, nutraceuticals are generally regarded as safe, require no withdrawal period, and may potentiate vaccine-induced immunity through immunomodulatory mechanisms (Eladl *et al*., 2020).

King-Herbs®, Gumbo ND®, and Grand HumiVet® are commercially available nutraceutical formulations commonly used by poultry farmers in Nigeria for routine health support and disease prevention (Sharma *et al*., 2000). These products contain combinations of herbal extracts, immune stimulants, antioxidants, humic acids, trace minerals, and polysaccharides that may enhance both innate and adaptive immune responses. However, scientific data evaluating their efficacy against very virulent IBDV in broilers, particularly when used alone or in combination with vaccination, remain limited.

This study was therefore designed to assess the prophylactic effects of three nutraceuticals (King-Herbs®, Gumbo ND®, and Grand HumiVet®) administered singly or in combination with an intermediate Gumboro vaccine on clinicopathological changes in broilers experimentally infected with vvIBDV. The findings aim to provide evidence-based recommendations for optimising IBD prevention strategies in poultry production systems.

MATERIALS AND METHODS

Study location

The study was conducted at the College of Veterinary Medicine, Federal University of Agriculture Zuru. The facility is equipped for controlled experimental infections and adheres to standard poultry biosecurity protocols.

Ethical approval

Ethical approval for the use of animals in this study was obtained from the Committee on Animal Use and Care (CAUC), College of Veterinary Medicine, Federal University of Agriculture, Zuru (FUAZ) under the approval number: FUAZCAC/FUAZ/2025/002. All procedures complied with international guidelines for the ethical use of animals in research, National Research Council (2020).

Experimental broilers

The sample size was calculated using G*Power software as described by Hinks *et al*. (2022). A total of 240 day-old Cobb500 broilers were purchased from a reputable hatchery in Ibadan, Nigeria. Birds were randomly allocated into 12 groups of 20 broilers each. All broilers were housed on deep litter with a stocking density of 0.14 m² per bird, consistent with recommended guidelines (El-Radhi, 2008).

Housing and biosecurity

The research pen was divided into 12 independent compartments, each measuring 210 cm × 240 cm and equipped with wire-mesh and wooden framing to prevent inter-group contact. Prior to placement, all pens and equipment were disinfected using formalin fumigation following established protocols (Mustafa *et al*., 2021). Strict biosecurity, including sanitation, movement control,

and isolation, was maintained throughout the experimental period.

Feeding and watering

Broilers were fed *ad libitum* using galvanised feeders. Feed was procured from an accredited distributor of a commercial feed mill and met NRC nutritional requirements. Clean borehole water was supplied ad libitum using 4-L plastic drinkers (one drinker per 20 broilers).

Brooding and lighting

Charcoal brooder pots were used to maintain optimal brooding temperatures (28–34°C) for the first week of life. A 200-W incandescent bulb was installed in each compartment to supply adequate lighting and mild heating.

Challenge virus

A very virulent infectious bursal disease virus (vvIBDV) strain was obtained from the Department of Veterinary Medicine, Ahmadu Bello University, Zaria. The isolate originated from a previous natural outbreak in vaccinated commercial layers. When 30-day-old cockerels were experimentally inoculated with 50 µL bursal suspension, 75% mortality was recorded. The viral suspension contained $10^{9.76}$ CID_{50}/mL, as previously characterised by Gana *et al* (2019). This strain was confirmed to belong to the vvIBDV lineage using molecular and pathogenicity criteria (Mosad *et al.,* 2020).

Medications and nutraceuticals

***King-Herbs Oral Solution*®**

King-Herbs Oral Solution® is a greenish herbal formulation produced by Aether Centre Biology Co., Ltd. (Beijing). It is recognised for its multiple biological functions, including the induction of interferon production, enhancement of humoral immunity, and improvement of intestinal integrity alongside the promotion of probiotic growth. Additionally, it contributes to increased feed intake and overall growth performance. The recommended dosage is 1 mL per litre of drinking water, administered over a period spanning from day 5 to day 29 of age, covering a total duration of 24 days.

***Gumbo ND*®**

This formulation is a dark green solution composed of several active ingredients, including sal-ammoniac, boric acid, sodium 2-hydroxybenzoate, D-glucitol, L-ascorbic acid, methionine, bromhexine hydrochloride, and Astragalus polysaccharide, which serves as an immune stimulant. It is primarily used for the prevention and supportive treatment of viral infections, particularly Infectious Bursal Disease (IBD) and Newcastle Disease (ND). The recommended dosage is 1 mL per litre of drinking water, administered continuously from day 5 to day 29.

***Grand HumiVet*®**

Grand HumiVet® is an organic supplement formulated with humic acid (76%), organic matter (92%), sodium humate (14%), as well as trace minerals and amino acids. It is known for its multiple beneficial functions, including immune enhancement, antiviral activity, stress reduction, and improved nutrient absorption. The recommended dosage is 1 g per litre of drinking water, administered from day 5 to day 29.

Vaccines and vaccination procedure

The intermediate live Gumboro vaccine Izovac® (Winterfield 2512 strain, 10^3 EID_{50}) was purchased from a licensed distributor in Kaduna, Nigeria, and administered via drinking water reconstituted at 500 doses per 5 litres, with the first vaccination given on Day 11 and a booster dose on Day 22; subsequently, ND vaccination using the La Sota strain was carried out on Day 36, and all vaccination procedures adhered strictly to OIE guidelines (OIE, 2021).

Clinical observations

Broilers were monitored daily for clinical signs such as depression, ruffled feathers, diarrhoea, prostration, anorexia and huddling. Clinical signs were expressed as percentages of affected birds per group following Babiker and Tawfeeg (2008).

Gross pathology

Dead broilers were immediately necropsied. Two live birds from each group were humanely euthanised by jugular exsanguination at 21, 28, 35, and 42 days. Tissues collected included the bursa of Fabricius, thymus, spleen and caecal tonsils. Gross lesions were scored based on haemorrhages, atrophy, enlargement, congestion, necrotic foci, and oedema, using Ingrao *et al*. (2013) and recent international guidelines (Hussein *et al*., 2018).

Table 1. Experimental design.

Group	Nutraceutical	Vaccine	Challenge
A	–	–	–
B	–	–	+
C1, C2, C3	KH, GND, GHV	+	+
D1, D2, D3	KH, GND, GHV	–	+
E1, E2, E3	KH, GND, GHV	–	–
F	–	+	+

Histopathology

Collected tissues were fixed in 10% neutral buffered formalin, processed using standard paraffin techniques, sectioned at 5 µm, and stained with hematoxylin and eosin. Bursal lesions were scored according to Follicular necrosis, Lymphocyte depletion, Interfollicular oedema, Cyst formation and Heterophil infiltration. Histological scoring followed updated criteria for IBDV studies.

Experimental design

The 240 Cobb500 broilers were randomly divided into 12 groups: a summary of the experimental design is presented in Table 1. On day 29, all challenge groups were inoculated orally with vvIBDV. Cloacal temperature was measured in 10 broilers per group from 3 to 7 dpi.

Data analysis

Data were analysed using GraphPad Prism version 5.03. The Shapiro–Wilk test was applied to assess the normality of the data distribution, while Levene's test was used to evaluate the homogeneity of variances. Group comparisons were performed using one-way analysis of variance (ANOVA), followed by Tukey's multiple comparison test for post hoc analysis. The results are presented as mean ± standard error of the mean (SEM), and statistical significance was considered at $p \leq 0.05$.

RESULTS

Changes in cloacal temperature

Cloacal temperature was monitored from 3 to 7 days post-infection (dpi) to assess febrile responses and potential prophylactic effects. The broilers receiving the nutraceuticals exhibited significant variations in cloacal temperature at early dpi, reflecting differential modulation of infection-induced febrile responses. At 3 dpi, several treated groups showed significantly higher cloacal temperatures compared to the control, whereas other groups exhibited lower temperatures. By 4–5 dpi, differences among groups were less pronounced, although specific nutraceutical-treated groups maintained significantly higher or lower temperatures relative to controls. At 6 dpi, groups E1, E2, and E3 consistently displayed elevated cloacal temperatures compared to other groups ($P < 0.05$–0.0001). By 7 dpi, cloacal temperatures among all groups were similar, with no significant differences observed. Overall, these results indicate that prophylactic administration of nutraceuticals, either alone or in combination with intermediate Gumboro vaccine, modulates cloacal temperature during the acute phase of Gumboro virus infection, suggesting a potential role in mitigating physiological responses associated with the disease (Table 2 and Figure 1).

Clinical signs observed

Broilers in the unmedicated, unchallenged control group (A) and those receiving nutraceuticals without vvIBDV challenge (E1, E2, E3) exhibited no clinical signs throughout the observation period (0%). Similarly, broilers administered nutraceuticals in combination with intermediate Gumboro vaccine and challenged with vvIBDV (C1, C2, C3) remained clinically normal, demonstrating complete protection.

Challenged broilers without nutraceutical prophylaxis (B, D1–D3, F) displayed variable clinical signs. Group B (challenged, no nutraceutical, no vaccine) exhibited 28.67% clinical signs at 3 dpi, which declined to 0.34% by 7 dpi. Group D1 (King-Herb Oral Solution, challenged, no vaccine) had 47.42% clinical signs at 3 dpi, resolving completely by 7 dpi. Group D2 (Gumbo ND, challenged, no vaccine) maintained high clinical signs (45.53%) across all dpi, indicating minimal protection. Group D3 (Grand HumiVet, challenged, no vaccine) exhibited 49.58% clinical signs at 3 dpi, which resolved by 6–7 dpi. Group F (vaccinated-only control) exhibited minimal clinical signs (0.34% at 3–5 dpi), confirming the efficacy of intermediate Gumboro vaccination. Overall, these data demonstrate that prophylactic administration of nutraceuticals in combination with intermediate Gumboro vaccine

Table 2. Cloacal temperature of broilers.

Group	Nutraceutical	Vaccine	vvIBDV Challenge	3 dpi	4 dpi	5 dpi	6 dpi	7 dpi
A	–	–	–	41.68 ± 0.14	41.92 ± 0.19	41.72 ± 0.16	41.54 ± 0.13	41.26 ± 0.12
B	–	–	+	42.30 ± 0.19^{a}	41.41 ± 0.15	40.70 ± 0.30^{a}	41.22 ± 0.07	41.41 ± 0.11
C1	King-Herbs®	+	+	41.73 ± 0.16	41.30 ± 0.08	41.46 ± 0.09^{b}	41.32 ± 0.06	41.44 ± 0.07
C2	Gumbo ND®	+	+	41.12 ± 0.08^{ab}	41.17 ± 0.06^{a}	41.38 ± 0.07^{b}	41.54 ± 0.06	41.55 ± 0.05
C3	Grand HumiVet®	+	+	40.98 ± 0.09^{ab}	41.35 ± 0.10	41.32 ± 0.06	41.60 ± 0.08	41.45 ± 0.08
D1	King-Herbs®	–	+	42.56 ± 0.12^{ac}	40.89 ± 0.74	41.03 ± 0.08^{a}	41.19 ± 0.08	41.35 ± 0.10
D2	Gumbo ND®	–	+	42.44 ± 0.15^{c}	41.12 ± 0.12^{a}	41.01 ± 0.11^{a}	41.41 ± 0.08	41.56 ± 0.11
D3	Grand HumiVet®	–	+	42.56 ± 0.20^{ac}	41.12 ± 0.41	41.08 ± 0.07^{a}	41.31 ± 0.06	41.40 ± 0.10
E1	King-Herbs®	–	–	42.06 ± 0.16	41.49 ± 0.11	41.98 ± 0.18^{bd}	41.81 ± 0.13^{bcd}	41.47 ± 0.10
E2	Gumbo ND®	–	–	41.39 ± 0.12^{bd}	42.07 ± 0.08^{bcd}	41.49 ± 0.11^{b}	41.71 ± 0.17^{b}	41.46 ± 0.14
E3	Grand HumiVet®	–	–	42.22 ± 0.09^{c}	42.03 ± 0.10^{d}	42.30 ± 0.10^{bcd}	41.84 ± 0.14^{bd}	41.53 ± 0.11
F	–	+	+	40.76 ± 0.08^{abcde}	41.25 ± 0.04	41.31 ± 0.05	41.41 ± 0.06^{e}	41.32 ± 0.07

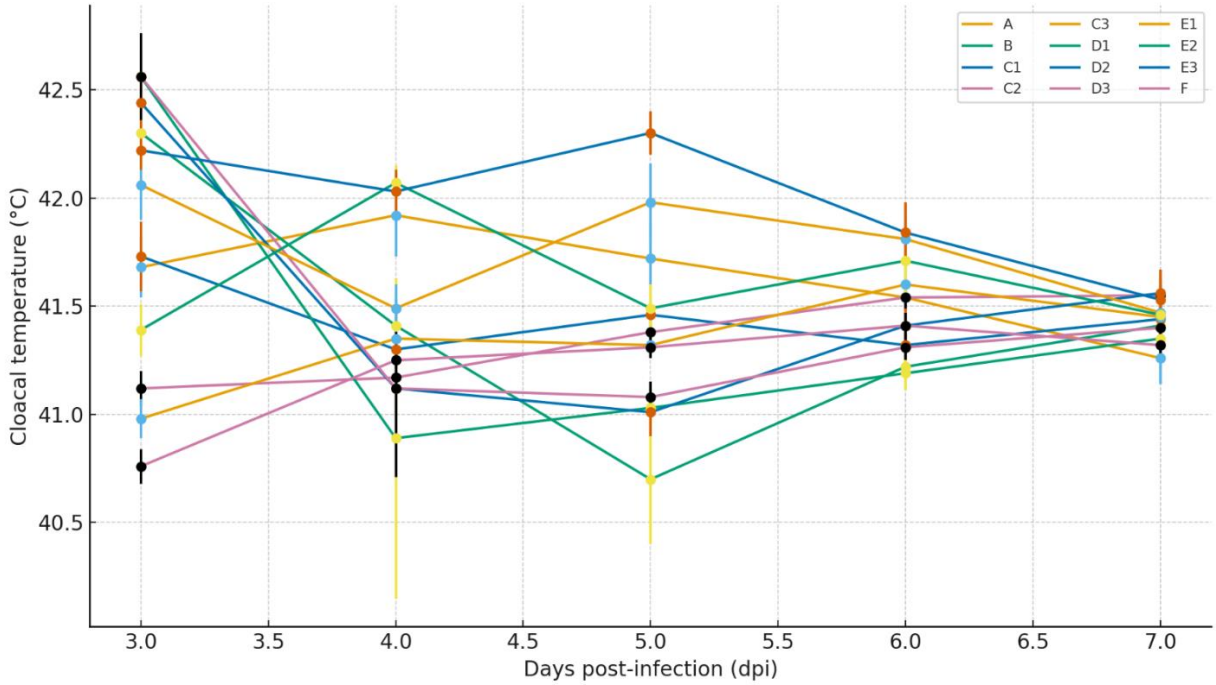

Figure 1. Different cloacal temperatures broilers in all treated and a control groups.

completely prevents clinical signs in challenged broilers, whereas nutraceuticals alone provide variable protection depending on the product (Table 3 and Figure 2).

Morbidity rates observed

Broilers administered King-Herb Oral Solution showed morbidity rates of 41.17% in groups B and D1 at 3 dpi. At 4 dpi, groups B, D1, and F recorded 25%, 57.14%, and 5.6%, respectively. By 5 dpi, morbidity rates were 25%, 23.08%, and 5.6% in the same groups. At 6 dpi, groups B and D1 had 6.67% and 7.69%, and at 7 dpi, 6.67% and 8.33%, respectively.

For Gumbo ND, morbidity rates at 3 dpi were 41.17% in group B and 21.43% in D2. At 4 dpi, groups B, D2, and F recorded 25%, 2.57%, and 5.6%, and at 5 dpi, 25%, 16.67%, and 5.6%, respectively. At both 6 and 7 dpi,

Table 3. Clinical signs.

Group	Nutraceutical	Vaccine	vvIBDV Challenge	3 dpi	4 dpi	5 dpi	6 dpi	7 dpi
A	–	–	–	0.00	0.00	0.00	0.00	0.00
B	–	–	+	28.67	26.10	9.36	3.91	0.34
C1	King-Herb Oral Solution	+	+	0.00	0.00	0.00	0.00	0.00
C2	Gumbo ND	+	+	0.00	0.00	0.00	0.00	0.00
C3	Grand HumiVet	+	+	0.00	0.00	0.00	0.00	0.00
D1	King-Herb Oral Solution	–	+	47.42	37.94	12.01	2.08	0.00
D2	Gumbo ND	–	+	45.53	45.53	45.53	45.53	45.53
D3	Grand HumiVet	–	+	49.58	40.17	9.38	0.00	0.00
E1	King-Herb Oral Solution	–	–	0.00	0.00	0.00	0.00	0.00
E2	Gumbo ND	–	–	0.00	0.00	0.00	0.00	0.00
E3	Grand HumiVet	–	–	0.00	0.00	0.00	0.00	0.00
F	–	+	+	0.34	0.34	0.34	0.00	0.00

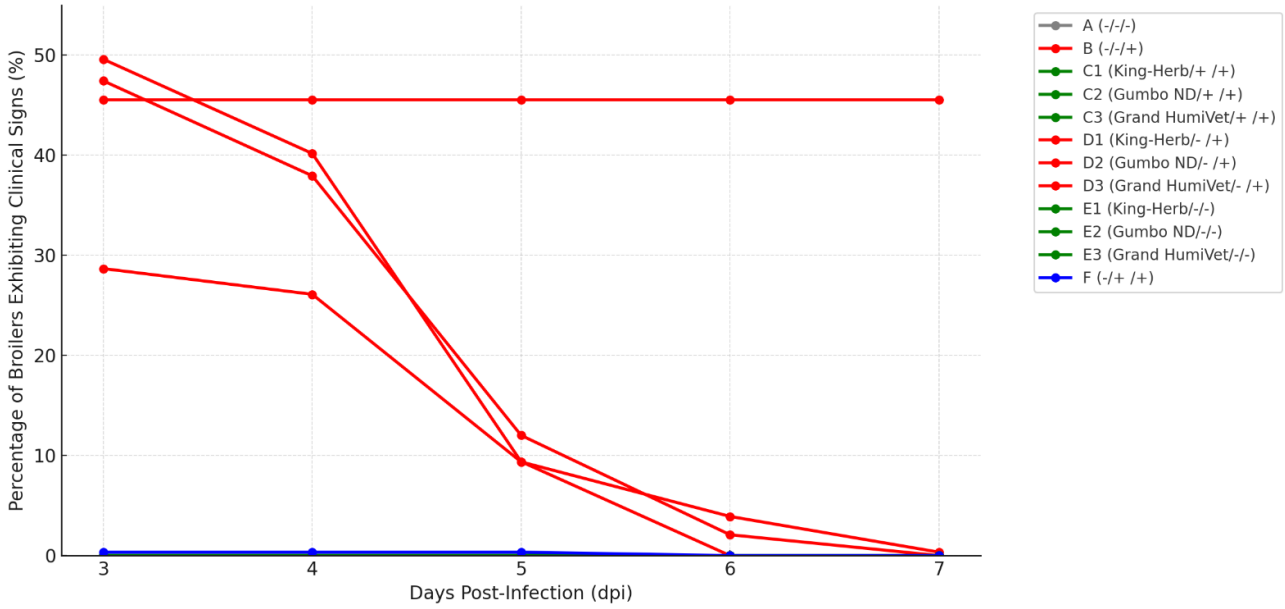

Figure 2. The percentage of birds showing clinical signs of Gumboro disease.

groups B and D2 showed 6.67% and 18.18% morbidity. Broilers administered Grand HumiVet had morbidity rates of 41.17% in group B and 33.33% in D3 at 3 dpi. At 4 dpi, groups B, D3, and F recorded 25%, 42.86%, and 5.6%, and at 5 dpi, 25%, 33.33%, and 5.6%, respectively. Only group B was recorded at 6 and 7 dpi with 6.67% morbidity (Table 4 and Figure 3).

Mortality rates observed

Mortality following vvIBDV challenge varied among the nutraceutical-treated groups. Broilers administered King-Herb oral solution (D1) showed mortality rates of 11.76% at 3 dpi, 7.14% at 4 dpi, 7.69% at 5 dpi, 8.33% at 6 dpi, and a peak of 25.00% at 7 dpi. Those given Gumbo ND (D2) recorded mortality of 14.29% at 4 dpi, 8.33% at 5 dpi, 9.09% at 6 dpi, and 28.57% at 7 dpi. Birds receiving Grand HumiVet (D3) exhibited 6.66% mortality at 3 dpi, 14.29% at 4 dpi, no mortality at 5 or 6 dpi, and 20.00% at 7 dpi. In the challenged, non-supplemented control group (B), mortality occurred at 4 dpi (5.88%), 6 dpi (6.25%), and 7 dpi (11.76%) (Table 5 and Figure 4).

Live body weight

Broilers administered the three nutraceuticals showed consistently higher live body weight (LBW) compared to

Table 4. Morbidity rates.

Group	Nutraceutical	Gumboro Vaccine	vvIBDV Challenge	Percent Morbidity				
				3 dpi	4 dpi	5 dpi	6 dpi	7 dpi
A	–	–	–	0.00	0.00	0.00	0.00	0.00
B	–	–	+	41.17	25.00	25.00	6.67	6.67
C1	King-Herb Oral Solution	+	+	0.00	0.00	0.00	0.00	0.00
C2	Gumbo ND	+	+	0.00	0.00	0.00	0.00	0.00
C3	Grand HumiVet	+	+	0.00	0.00	0.00	0.00	0.00
D1	King-Herb Oral Solution	–	+	41.17	57.14	23.08	7.69	8.33
D2	Gumbo ND	–	+	21.43	2.57	16.67	18.18	18.18
D3	Grand HumiVet	–	+	33.33	42.86	33.33	0.00	0.00
E1	King-Herb Oral Solution	–	–	0.00	0.00	0.00	0.00	0.00
E2	Gumbo ND	–	–	0.00	0.00	0.00	0.00	0.00
E3	Grand HumiVet	–	–	0.00	0.00	0.00	0.00	0.00
F	–	+	+	0.00	5.60	5.60	0.00	0.00

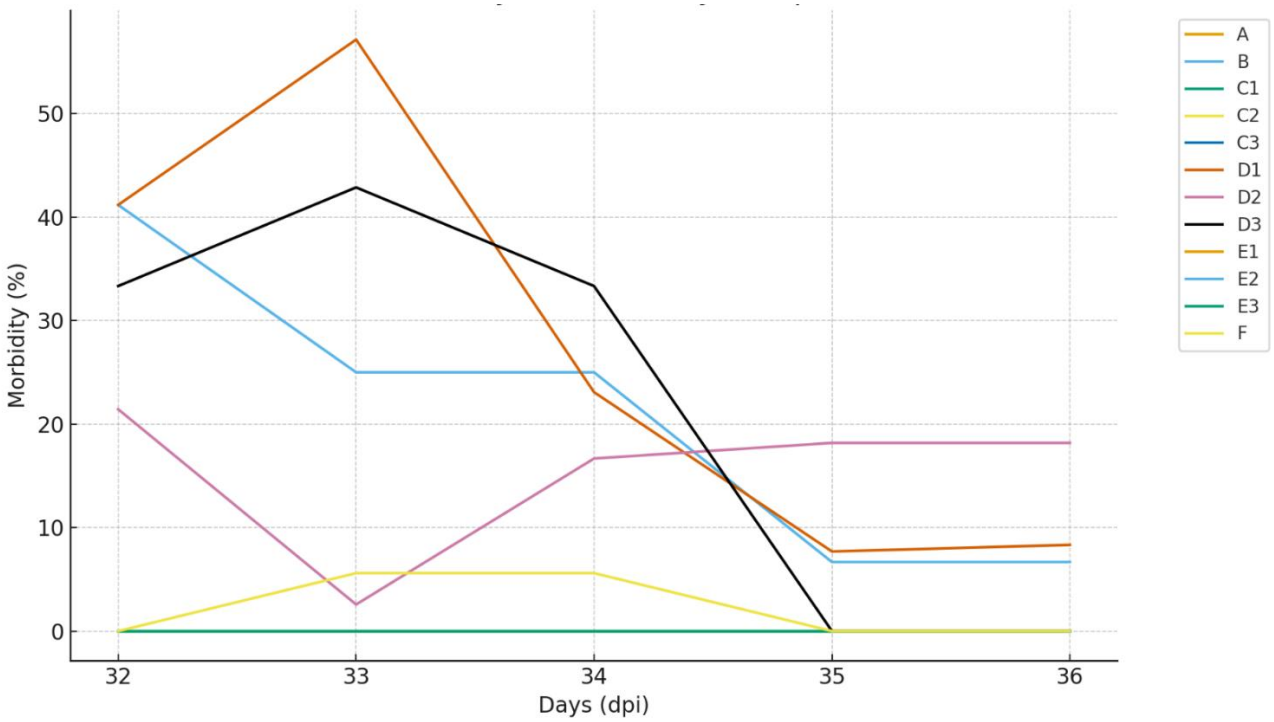

Figure 3. The graph of mortality rate after exposure.

the untreated control throughout the study. Among the treatments, the E groups (E1, E2, E3) exhibited the strongest protective effect, maintaining the highest LBW by 7 dpi. Broilers in group B, which received no nutraceuticals, consistently had the lowest LBW, reflecting the impact of infection. Overall, all nutraceuticals mitigated weight loss caused by the virulent IBD virus, with statistically significant differences observed between treated and control groups (Table 6 and Figure 5).

Gross pathology

Gross examination of broilers infected with infectious bursal disease virus (IBDV) revealed lesions characteristic of very virulent Gumboro disease. Congestion and haemorrhage were observed in the leg, thigh, and breast muscles, while the proventriculus showed haemorrhagic lesions. The bursa of Fabricius (BF) was the most severely affected organ, exhibiting enlargement, pallor, increased

Table 5. Mortality rates.

Group	Nutraceutical	Gumboro vaccine	vvIBDV challenge	Mortality rates				
				3 dpi	4 dpi	5 dpi	6 dpi	7 dpi
A	–	–	–	0.00	0.00	0.00	0.00	0.00
B	–	–	+	0.00	5.88	0.00	6.25	11.76
C1	King-Herb Oral Solution	+	+	0.00	0.00	0.00	0.00	0.00
C2	Gumbo ND	+	+	0.00	0.00	0.00	0.00	0.00
C3	Grand HumiVet	+	+	0.00	0.00	0.00	0.00	0.00
D1	King-Herb Oral Solution	–	+	11.76	7.14	7.69	8.33	25.00
D2	Gumbo ND	–	+	0.00	14.29	8.33	9.09	28.57
D3	Grand HumiVet	–	+	6.67	14.29	0.00	0.00	20.00
E1	King-Herb Oral Solution	–	–	0.00	0.00	0.00	0.00	0.00
E2	Gumbo ND	–	–	0.00	0.00	0.00	0.00	0.00
E3	Grand HumiVet	–	–	0.00	0.00	0.00	0.00	0.00
F	–	+	+	0.00	0.00	0.00	0.00	0.00

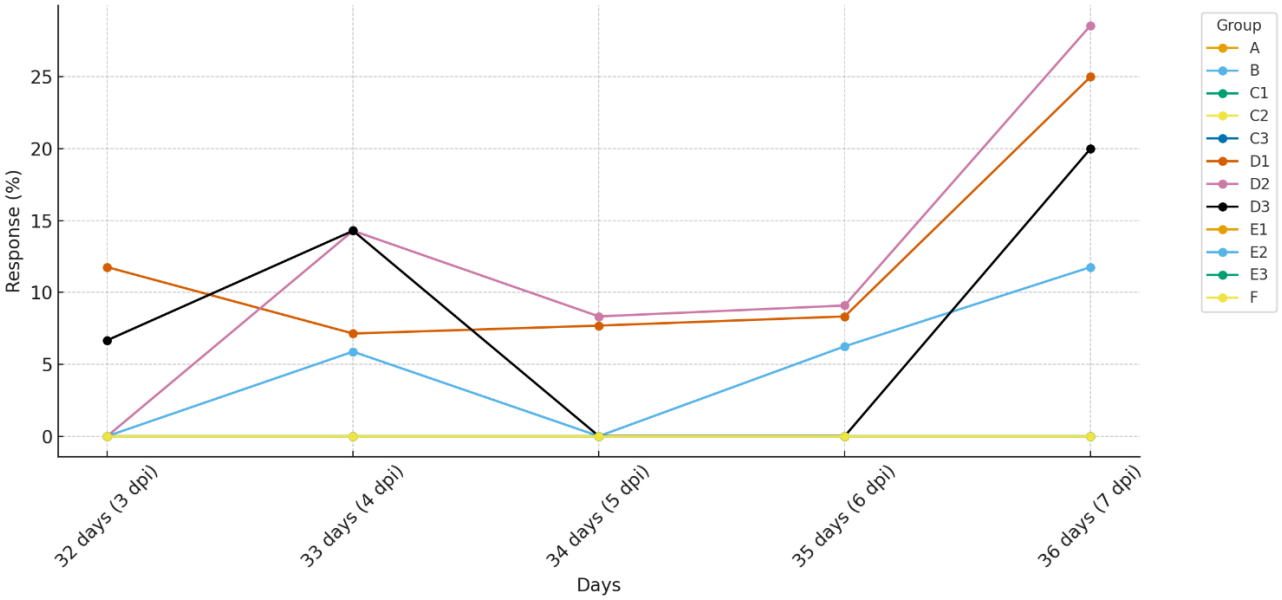

Figure 4. The responses of the broilers at different days during exposure.

turgidity, oedema, haemorrhage, multifocal necrosis, and varying degrees of atrophy. The spleen was enlarged with grey necrotic foci, haemorrhage, and atrophy, and the caecal tonsils showed haemorrhage and enlargement. Thymic lesions included congestion, haemorrhage, and turgidity, whereas mild congestion was noted in the lungs, trachea, pancreas, and liver, with the liver also appearing enlarged and pale. The cumulative gross lesion score for very virulent Gumboro infection in broilers was 120, and percentage lesion severity was calculated as the ratio of observed lesion scores to this value multiplied by 100.

In broilers administered King-Herb oral solution, percentage gross lesion scores at 3 days post-infection (dpi) were 0.00% (group A), 38.08% (group B), 2.50% (group C1), 24.17% (group D1), 3.33% (group E1), and 4.17% (group F), which declined at 7 dpi to 0.00%, 10.50%, 9.00%, 11.00%, 2.00%, and 9.50%, respectively. At 3 dpi, no BF lesions were observed in group A, whereas group B showed enlargement, haemorrhage, and congestion; groups C1 and F exhibited bursal atrophy with congestion; group D1 showed enlargement with haemorrhage and congestion; and group E1 showed mild congestion.

Similarly, broilers administered Gumbo ND showed percentage lesion scores at 3 dpi of 0.00% (group A), 38.08% (group B), 5.83% (group C2), 23.33% (group D2),

Table 6. Live body weight.

Group	Nutraceutical	Gumboro Vaccine	vvIBDV Challenge	Live body weight in grams		
				3 dpi	4 dpi	7 dpi
A	–	–	–	313.70 ± 14.82	374.00 ± 17.50	425.10 ± 12.17
B	–	–	+	257.10 ± 11.06^{a}	253.60 ± 12.25^{a}	328.50 ± 21.53^{a}
C1	King-Herb Oral Solution	+	+	337.00 ± 6.95^{b}	367.30 ± 12.28^{b}	429.70 ± 14.29^{b}
C2	Gumbo ND	+	+	314.70 ± 9.40^{b}	346.10 ± 9.48^{b}	395.90 ± 12.03^{b}
C3	Grand HumiVet	+	+	293.10 ± 6.91	327.00 ± 5.18^{ab}	365.80 ± 17.20^{a}
D1	King-Herb Oral Solution	–	+	288.60 ± 5.94^{c}	267.60 ± 4.47^{ac}	335.60 ± 10.62^{ac}
D2	Gumbo ND	–	+	332.90 ± 5.55^{b}	314.90 ± 5.75^{ab}	369.40 ± 10.14^{a}
D3	Grand HumiVet	–	+	266.10 ± 11.46^{a}	281.50 ± 7.35^{ac}	335.20 ± 12.81^{a}
E1	King-Herb Oral Solution	–	–	316.30 ± 10.66^{b}	367.00 ± 9.44^{bd}	457.60 ± 7.50^{bd}
E2	Gumbo ND	–	–	290.00 ± 3.16^{d}	320.70 ± 7.81^{ab}	395.80 ± 8.92^{b}
E3	Grand HumiVet	–	–	323.40 ± 8.40^{bd}	361.70 ± 6.62^{bd}	454.80 ± 7.36^{bcd}
F	–	+	+	299.50 ± 9.60	294.90 ± 9.10^{ace}	359.80 ± 7.76^{ace}

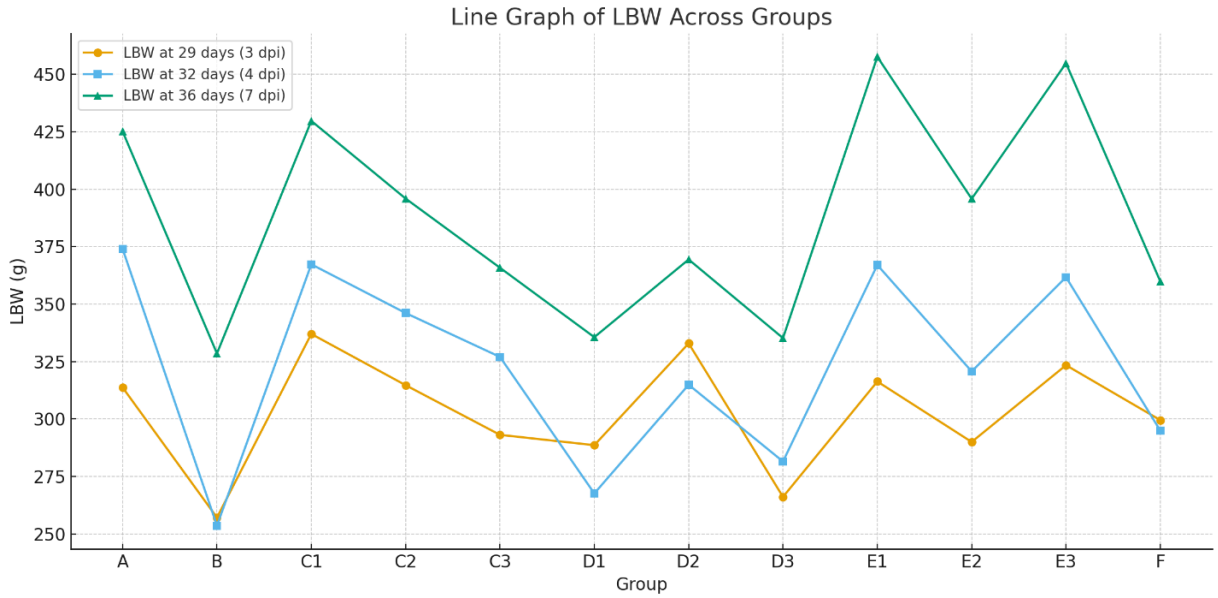

Figure 5. Live body weight of the broilers across groups.

2.92% (group E2), and 4.17% (group F), decreasing at 7 dpi to 0.00%, 10.50%, 9.00%, 13.00%, 7.00%, and 9.50%, respectively. Bursa of Fabricius lesions at 3 dpi were absent in group A; pronounced in group B (enlargement, haemorrhage, congestion); mild in group C2 (congestion); moderate in group D2 (enlargement and congestion); and characterised by congestion with haemorrhage in group E2, while group F showed bursal atrophy with congestion.

In broilers administered Grand HumiVet, percentage lesion scores at 3 dpi were 0.00% (group A), 38.08% (group B), 4.58% (group C3), 25.00% (group D3), 3.33% (group E3), and 4.17% (group F), with corresponding values at 7 dpi of 0.00%, 10.50%, 5.00%, 8.00%, 5.50%, and 9.50%. At 3 dpi, group A showed no BF lesion, group B exhibited severe enlargement with haemorrhage and congestion, groups C3 and D3 showed mild to moderate congestion with or without enlargement, group E3 showed congestion with haemorrhage, and group F showed bursal atrophy with congestion (Table 7 and Figure 6).

Histopathological changes in the bursa of Fabricius of broilers

Histopathological examination of the bursa of Fabricius (BF) revealed that the negative control group (A; 0.00 ±

Table 7. Percentage gross lesion.

Nutraceutical Group	DPI	Percent gross lesion	Bursa of Fabricius Lesions
King-Herb Oral Solution	3	A: 0.00%B: 38.08%C1: 2.50%D1: 24.17%E1: 3.33%F: 4.17%	A: None B: Enlargement, hemorrhage, congestionC1: Atrophy, congestionD1: Enlargement, hemorrhage, congestionE1: CongestionF: Atrophy, congestion
	7	A: 0.00%B: 10.50%C1: 9.00%D1: 11.00%E1: 2.00%F: 9.50%	Same as above
Gumbo ND	3	A: 0.00%B: 38.08%C2: 5.83%D2: 23.33%E2: 2.92%F: 4.17%	A: None B: Enlargement, hemorrhage, congestionC2: CongestionD2: Enlargement, congestionE2: Congestion, hemorrhageF: Atrophy, congestion
	7	A: 0.00%B: 10.50%C2: 9.00%D2: 13.00%E2: 7.00%F: 9.50%	Same as above
Grand HumiVet	3	A: 0.00%B: 38.08%C3: 4.58%D3: 25.00%E3: 3.33%F: 4.17%	A: None B: Enlargement, hemorrhage, congestionC3: CongestionD3: Enlargement, congestionE3: Congestion, hemorrhageF: Atrophy, congestion
	7	A: 0.00%B: 10.50%C3: 5.00%D3: 8.00%E3: 5.50%F: 9.50%	Same as above

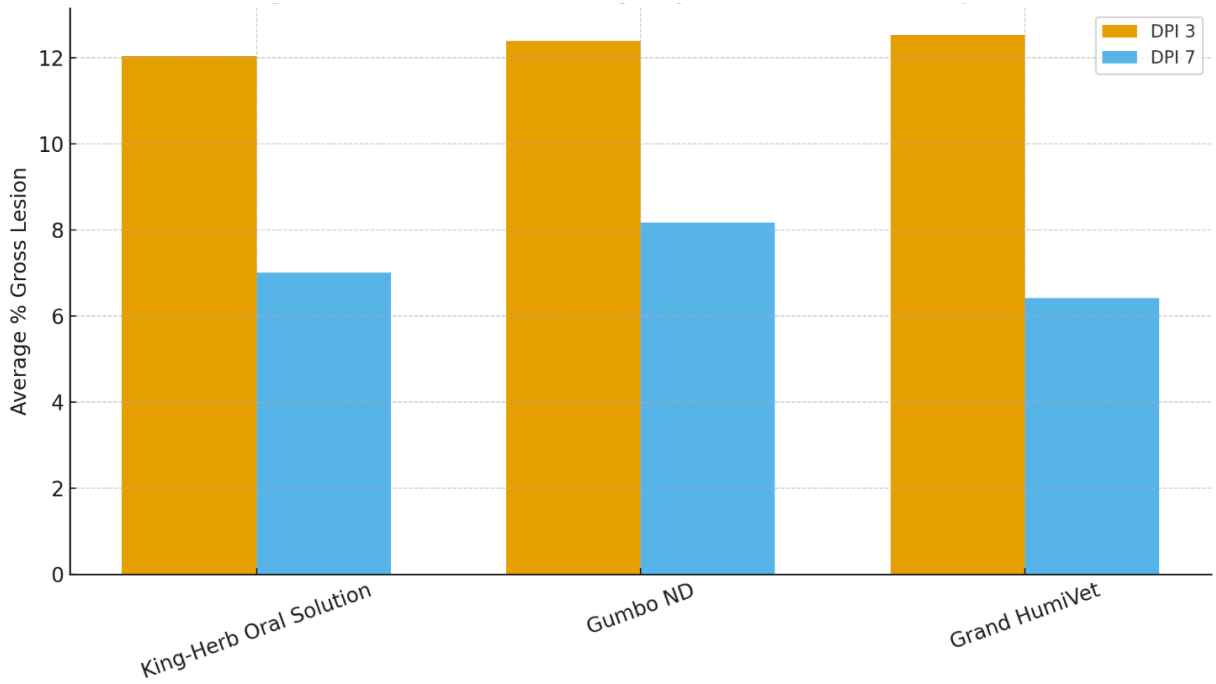

Figure 6. Percentage of gross lesion.

0.00) had significantly lower lesion scores than all infected groups across treatments ($p<0.001$). In broilers administered King-Herb oral solution, groups B (4.50 ± 0.17) and D1 (4.25 ± 0.05) showed significantly higher scores than groups C1 (3.45 ± 0.05), E1 (2.90 ± 0.10), and F (3.13 ± 0.13) (Table 7). Group B exhibited marked interfollicular oedema and follicular necrotic debris at 3 dpi, progressing to severe follicular necrosis, interfollicular thickening, atrophy, vacuolation, and cyst formation at 7 dpi. Group C1 showed mild vacuolation and follicular atrophy, whereas group D1 exhibited severe necrosis with pronounced interfollicular oedema. Minimal interfollicular

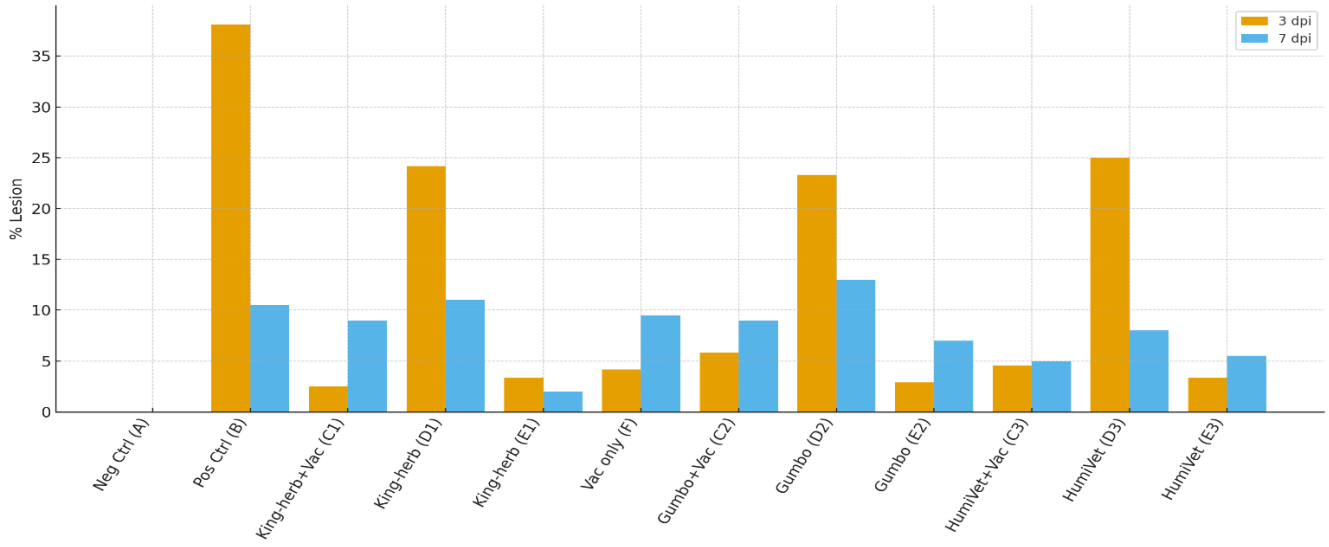

Figure 7. Percentage lesion.

Table 8. Gross and Histopathology**.**

Group / Treatment	3 dpi % Lesion	7 dpi % Lesion	Bursa of Fabricius Lesions
Negative Control (A)	0.00%	0.00%	None
Positive Control (B)	38.08%	10.50%	Enlargement, hemorrhage, congestion
King-herb + Vaccine (C1)	2.50%	9.00%	Atrophy, congestion
King-herb only (D1)	24.17%	11.00%	Enlargement, hemorrhage, congestion
King-herb only (E1)	3.33%	2.00%	Congestion
Vaccine only (F)	4.17%	9.50%	Atrophy, congestion
Gumbo ND + Vaccine (C2)	5.83%	9.00%	Congestion
Gumbo ND only (D2)	23.33%	13.00%	Enlargement, congestion
Gumbo ND only (E2)	2.92%	7.00%	Congestion, hemorrhage
Grand HumiVet + Vaccine (C3)	4.58%	5.00%	Congestion
Grand HumiVet only (D3)	25.00%	8.00%	Enlargement, congestion
Grand HumiVet only (E3)	3.33%	5.50%	Congestion, hemorrhage

thickening was observed in group E1, while group F showed vacuolation at 3 dpi and necrosis at 7 dpi (Plate I).

In broilers administered Gumbo ND, group B (4.50 ± 0.17) had significantly higher scores than group E2 (2.63 ± 0.63) ($p < 0.05$). Group B showed progressive follicular necrosis with interfollicular oedema and cyst formation. Group C2 exhibited diffuse cysts at 3 dpi and interfollicular thickening at 7 dpi, while group D2 showed follicular atrophy with mild follicular cell depletion. Groups E2 and F demonstrated follicular vacuolation at 3 dpi, progressing to atrophy or necrosis at 7 dpi (Plate II). Similarly, broilers administered Grand HumiVet showed significantly higher lesion scores in groups B (4.50 ± 0.17) and D3 (4.38 ± 0.13) compared with groups C3 (3.63 ± 0.13), E3 (2.90 ± 0.10), and F (3.13 ± 0.13). Group B showed severe progressive follicular necrosis with interfollicular oedema and cysts, while groups C3 and D3 exhibited follicular necrosis and depletion. Group E3 showed persistent follicular necrosis, and group F showed vacuolation progressing to necrosis at 7 dpi (Plate III) (Table 8, Figure 7 and Plates).

DISCUSSION

In this study, broilers inoculated with vvIBDV exhibited pyrexia in the early stage of infection (3 dpi), particularly in groups B, D1, D2, and D3. Fever following vvIBDV exposure has been attributed to viraemia and innate immune activation, which stimulates pyrogenic cytokines that act on hypothalamic thermoregulatory centres (Mota-Rojas *et al*., 2021; El-Radhi, 2008). Broilers possess well-

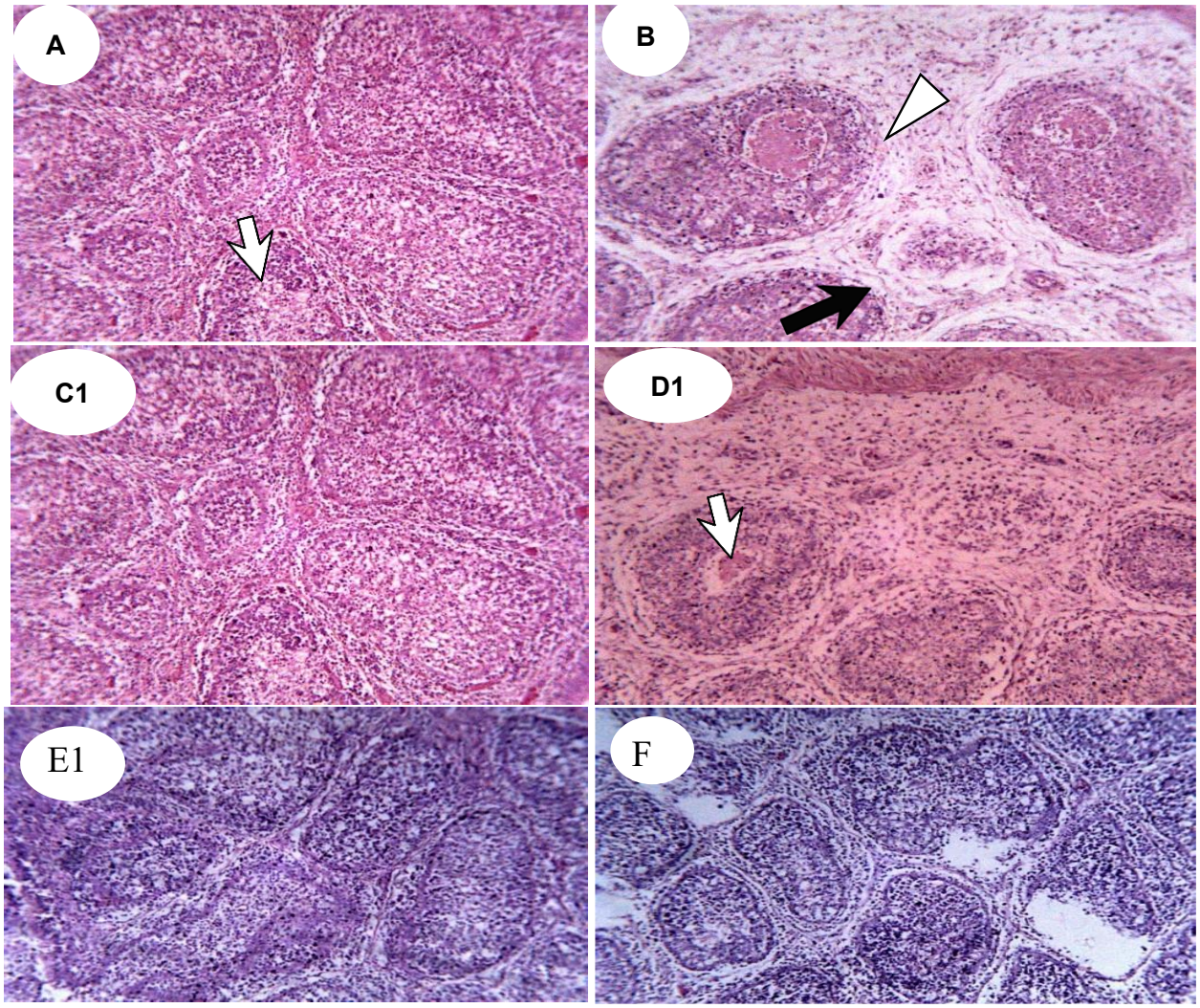

Plate I. (A) Photomicrographs of sections of bursae of Fabricius of broilers administered King-herb oral solution. (negative control) showing normal bursal architecture. (B) Positive control showing thickened interfollicular space filled with edema fluid and mononuclear cells (arrows), and necrotic debris in the follicles. (C1) group administered King herb oral solution, vaccine and inoculated with vvIBDV showing atrophy of the follicles and infiltration of mononuclear cells (arrows). (D1) group administered King herb oral solution and inoculated with vvIBDV showing necrosis of the follicles, widened interfollicular space filled with oedema fluid and mononuclear cells (arrows). (E1) group administered King herboral solutionoral solution showing depletion of lymphocytes and infiltration of mononuclear cells. (F) group vaccinated with Izovac vaccine and inoculated with vvIBDV showing necrosis and vaculation of follicles (arrows) and infiltration with mononuclear cells. H & E stain x 100 (At 3 days' post-inoculation).

developed Toll-like receptors and cytokine pathways that rapidly detect viral pathogen-associated molecular patterns, triggering IL-1, IL-6, and TNF-α production—potent mediators of febrile responses during IBDV infection (Mosad *et al.,* 2020). The absence of pyrexia in group E1 supports the conclusion that infection, rather than dietary or environmental factors, induced the febrile response.

Broilers in the vaccinated groups (C1, C2, C3, F) did not show significant fever, indicating that vaccination primed an anamnestic immune response that neutralised vvIBDV before substantial viral replication occurred. This aligns with recent reports showing that contemporary broiler strains mount rapid humoral responses to IBD vaccines when maternal antibody interference is minimised (Müller *et al*., 2012; Alkie and Rautenschlein, 2016). Groups A, E1, E2, and E3 maintained normal cloacal temperatures, highlighting the role of strict biosecurity—still the most effective preventive strategy against IBDV in broiler operations (Ngom *et al*., 2024).

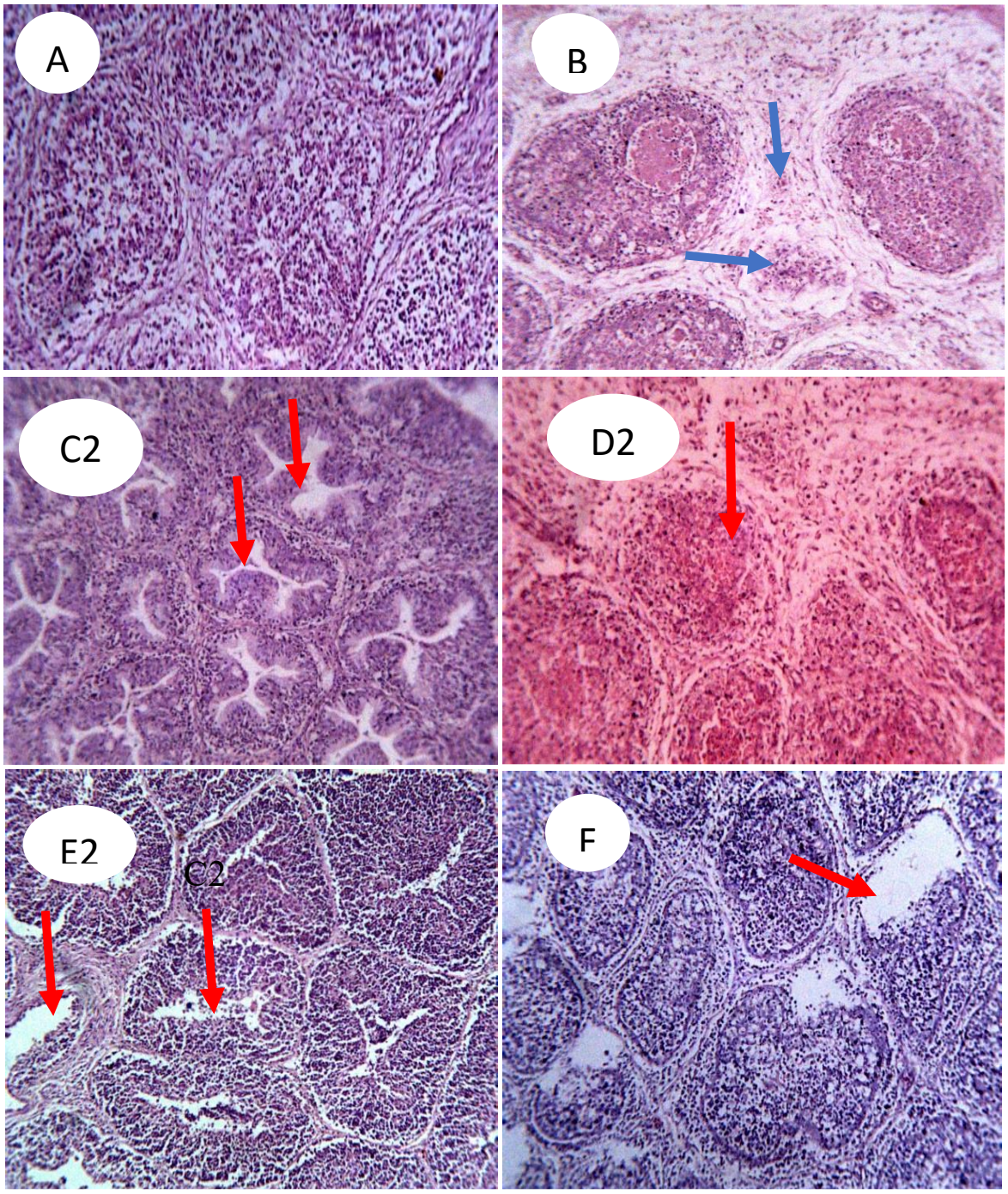

Plate II. Photomicrographs of sections of bursae of broilers administered Gumbo ND. (A) Negative control, normal follicles (B) Positive control, thickened interfollicular space filled with edema fluid (arrow), necrotic debris in the follicles. (C2) (Gumbo ND, vaccination and inoculated) diffused follicular cyst formation (arrows). (D2) (Gumbo ND and inoculated) (arrow) necrosis of the follicles, follicular atrophy(arrows). (E2) Gumbo ND necrosis andvaculation (arrows) (F) Vaccinated and inoculated, vaculation of the follicles (arrows). At 3 days' post-inoculation. H & E x 100.

The onset of clinical signs at 2 dpi agrees with current descriptions of vvIBDV pathogenesis in broilers, characterised by rapid viral replication in the bursa followed by systemic inflammatory and metabolic disturbances (Huang *et al*., 2021; Eterradossi and Saif, 2013). The absence of clinical signs in nutraceutical-

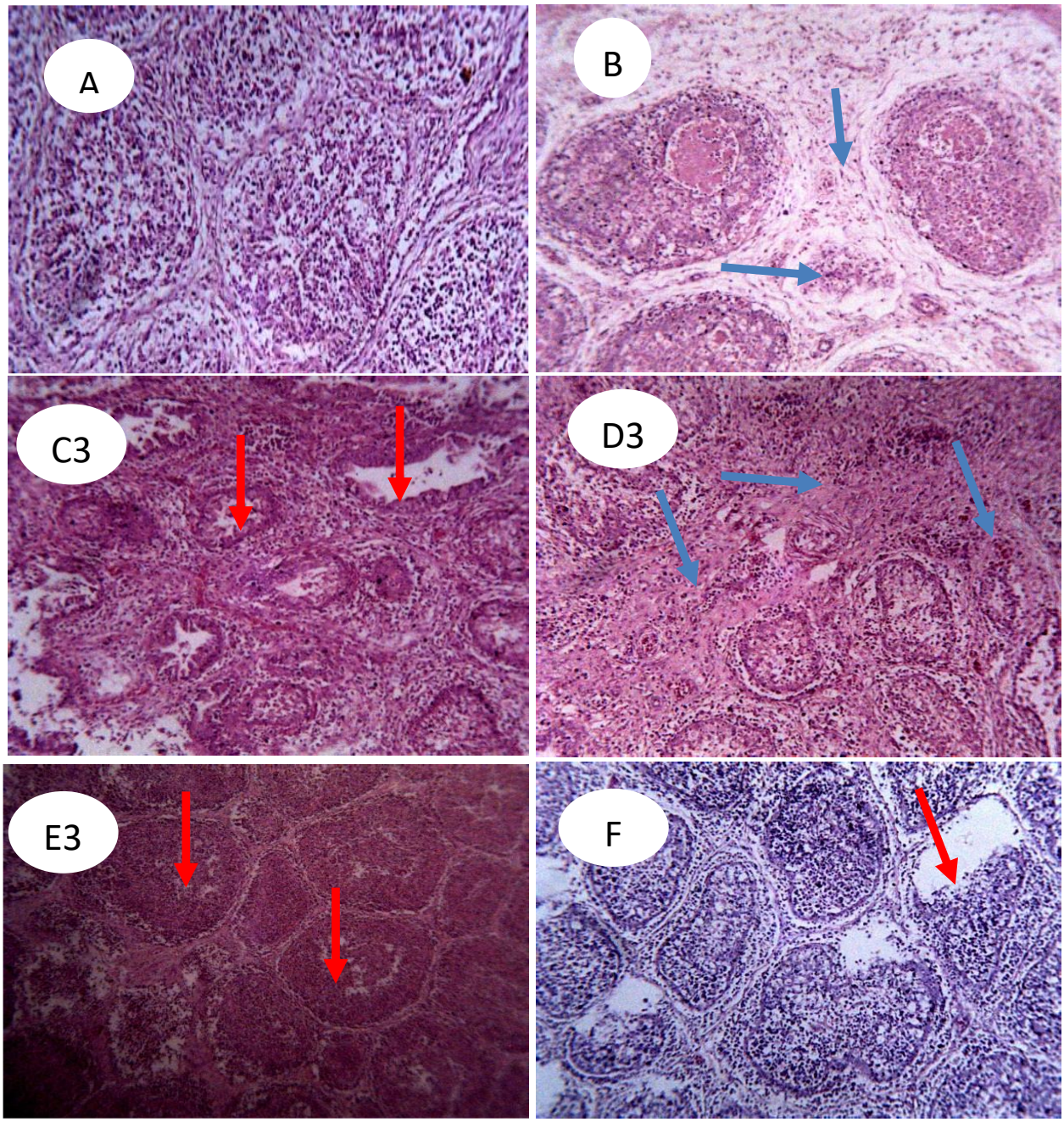

Plate III. Photomicrographs of sections of bursae of broilers administered Grand humi vet. (A) Negative control, normal follicles (B) Positive control, thickened interfollicular space filled with edema fluid (arrows), necrotic debris in the follicles. (C3) (Grand humi vet, vaccination and inoculated) vacuolation and necrosis (arrows). (D3) (Grand humi vet and inoculated) (arrow) severe necrosis of follicles (arrows) and thickened interfollicular space. (E3) Grand humi vet necrosis and depletion of follicles (arrows) (F) Vaccinated and inoculated, vaculation of the follicles (arrows). At 3 days' post-inoculation. H & E x 100.

supplemented and vaccinated groups (C1, C2, C3) suggests synergistic immunomodulatory effects. Several recent studies demonstrate that phytogenic and vitamin-based nutraceuticals enhance broiler immune function by reducing oxidative stress and modulating cytokine activity, thereby improving resistance to IBDV (Wati *et al*., 2015; Phillips *et al*., 2023).

Broilers in groups B, D1, D2, and D3 exhibited marked

clinical disease because they were not vaccinated against vvIBDV, supporting earlier observations that non-immune broilers are highly susceptible to acute IBD outbreaks (Dey *et al*., 2019; Sharma *et al*., 2000). Interestingly, group D2, which received Gumbo ND, showed comparatively lower clinical scores at 3 dpi, possibly attributable to the antioxidant properties of vitamin C and herbal constituents known to reduce lipid peroxidation and tissue oxidative damage during vvIBDV infection (Surai, 2016; Phillips *et al*., 2023).

Clinical signs such as ruffled feathers, diarrhoea, depression, and somnolence were consistent with classical IBD manifestations in broilers, which arise from necrosis of gut-associated lymphoid tissues and systemic inflammation (Mosad *et al.,* 2020). Mild clinical signs in vaccinated group F may be due to uneven vaccine take or suboptimal antibody titers, a well-recognised challenge in broiler flocks with variable maternal antibody levels (Alkie and Rautenschlein, 2016; Müller *et al*., 2012).

Morbidity rates were zero in non-inoculated and vaccinated groups (A, E1, E2, E3, C1, C2, C3), consistent with successful immunoprotection and biosecurity. Conversely, group B exhibited high morbidity due to vvIBDV exposure without vaccination or nutraceutical support. Elevated morbidity in groups D1, D2, and D3 suggests that nutraceuticals alone were insufficient to elicit rapid protective immunity. While nutraceuticals enhance immune resilience, they typically cannot completely substitute for vaccination during acute vvIBDV challenge (Huang *et al*., 2021; Phillips *et al*., 2023).

Mortality patterns followed similar trends. No mortality occurred in vaccinated groups, aligning with evidence that modern IBD vaccines effectively prevent death in broilers, even under vvIBDV pressure (Dey *et al*., 2019; Eterradossi and Saif, 2013). Mortality in D1 and D3 reflected uncontrolled viral replication in non-vaccinated broilers. Notably, these groups also had larger bursae compared to group B, indicating a higher population of target B-lymphocytes susceptible to vvIBDV replication, thereby exacerbating disease severity.

Live body weight was markedly improved in nutraceutical-supplemented broilers, especially in group C1. Phytogenic additives are known to enhance feed efficiency via improved nutrient absorption and antioxidative support (Wati *et al*., 2015). Broilers in groups B, D1, D2, D3, and F experienced reduced weight gain, consistent with IBD-associated anorexia, metabolic stress, and cytokine-mediated catabolism (Kogut & Arsenault, 2017; Huang *et al*., 2021). These observations reinforce that vvIBDV infection has significant economic implications on broiler growth performance.

Gross lesions in the bursa, spleen, thymus, and muscles correspond with classical vvIBDV pathology and match recent outbreak descriptions worldwide (Fan *et al*. 2020). Dehydration and emaciation were likely secondary to diarrhoea and reduced feed intake. Muscle haemorrhages observed in affected groups align with current findings linking IBDV to coagulopathy via endothelial damage and disruption of clotting pathways (Huang *et al*., 2021). The bursa lesions reflect the virus's tropism for IgM^+ B cells and subsequent cytokine-mediated inflammation (Mahgoub, 2012; Mosad *et al.,* 2020).

Histopathological observations, including interfollicular edema, lymphoid depletion, cystic degeneration, and heterophilic infiltration, are characteristic of vvIBDV infection in broilers and consistent with recent histopathologic studies (Sharma *et al*., 2000; Zhao *et al*., 2016). The persistence of lesions in nutraceutical-supplemented and vaccinated groups (C2, C3, F) suggests that although clinical protection occurred, some degree of subclinical viral replication or immune activation remained, a phenomenon widely recognised in IBDV-immunised broilers (Dey *et al*., 2019). Moreover, certain herbal additives may themselves induce mild histological alterations in lymphoid tissues (Jackwood, 2017), possibly contributing to observed microscopic changes.

Overall, this study demonstrates that biosecurity, vaccination, and nutraceutical supplementation synergistically reduced clinical disease, morbidity, and mortality in vvIBDV-challenged broilers**,** although nutraceuticals alone were insufficient to prevent severe outcomes without vaccination.

Conclusion and practical implications

The findings of this study demonstrate that supplementation with King-herb oral solution, Gumbo ND, or Grand HumiVet provided substantial protection to broilers challenged with very virulent infectious bursal disease virus (vvIBDV). Broilers receiving any of the nutraceuticals in combination with the Gumboro vaccine exhibited no clinical signs, no mortality, reduced cloacal temperatures, lower lesion scores, and superior body weight compared with untreated–inoculated controls. These results indicate that the evaluated nutraceuticals offered measurable immunomodulatory and performance-supporting effects during vvIBDV infection.

Overall, the data suggest that these products helped maintain immune competence and physiological stability, likely through their phytogenic, antioxidant, or antiviral components—mechanisms that have also been reported in recent broiler studies investigating herbal immunomodulatory and antiviral botanicals.

Given the continued economic losses associated with vvIBDV in broiler production, integrating such nutraceuticals alongside routine vaccination could strengthen flock resilience, reduce production losses, and enhance vaccine responsiveness. Their use may be particularly beneficial in high-challenge environments, farms with vaccine breaks, or operations aiming to reduce antibiotic reliance.

Limitations of the study

Several limitations should be acknowledged when interpreting the findings of this study. Although the experiment was conducted under strict biosecurity and controlled conditions, such settings may not accurately reflect commercial poultry production systems where multiple stressors, co-infections, environmental fluctuations, and management variability can influence disease dynamics and treatment outcomes. Therefore, extrapolation of these findings to field conditions should be made cautiously.

Only a single very virulent infectious bursal disease virus (vvIBDV) isolate was used for experimental challenge. Given the documented genetic and antigenic diversity among circulating IBDV strains, the observed protective effects may not be uniformly reproducible against heterologous field variants.

The study primarily assessed clinical signs, lesion scores, mortality, and growth performance. Comprehensive immunological investigations, including quantitative antibody titers, cytokine expression profiling, viral load determination, and lymphocyte subset analysis, were not performed. Consequently, mechanistic conclusions regarding immunomodulatory activity remain inferential rather than directly demonstrated.

Post-challenge monitoring was limited to the acute phase of infection, and longer-term effects on immune competence, production indices, and response to subsequent vaccinations were not evaluated. Furthermore, only manufacturer-recommended doses of each nutraceutical were tested, and dose–response relationships were not explored.

Finally, the use of a single broiler genotype (Cobb500) limits generalizability across other commercial strains.

Future multi-strain challenge studies, field-based trials, and mechanistic immunological investigations are warranted to strengthen and validate these findings.

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REFERENCES

Alkie, T. N., & Rautenschlein, S. (2016). Infectious bursal disease virus in poultry: current status and future prospects. *Veterinary Medicine: Research and Reports*, *7*, 9-18.

Babiker, M. A. A., & Tawfeeg, E. (2008). Role of administration routes of anti-infectious bursal disease virus (Gumboro) vaccine on immunization of chicken. *International Journal of Poultry Science*, *7*(3), 279-282.

Costagliola, G., Nuzzi, G., Spada, E., Comberiati, P., Verduci, E., & Peroni, D. G. (2021). Nutraceuticals in viral infections: an overview of the immunomodulating properties. *Nutrients, 13*(7), 2410.

Courtillon, C., Allée, C., Amelot, M., Keita, A., Bougeard, S., Härtle, S., Rouby J-C., Eterradossi, N.,& Soubies, S. M. (2022). Blood B cell depletion reflects immunosuppression induced by live-attenuated infectious bursal disease vaccines. *Frontiers in Veterinary Science*, *9*, 871549. https://doi.org/10.3389/fvets.2022.871549

Dey, S., Pathak, D. C., Ramamurthy, N., Maity, H. K., & Chellappa, M. M. (2019). Infectious bursal disease virus in chickens: prevalence, impact, and management strategies. *Veterinary Medicine: Research and Reports*, 85-97.

Ekiri, A. B., Armson, B., Adebowale, K., Endacott, I., Galipo, E., Alafiatayo, R., Horton, D. L., Ogwuche, A., Bankole, O.N., Galal, H. M., & Cook, A. J. (2021). Evaluating disease threats to sustainable poultry production in Africa: Newcastle disease, infectious bursal disease, and avian infectious bronchitis in commercial poultry flocks in Kano and Oyo States, Nigeria. *Frontiers in veterinary science*, *8*, 730159.

Eladl, A. H., Mosad, S. M., El-Shafei, R. A., Saleh, R. M., Ali, H. S., Badawy, B. M., & Elshal, M. F. (2020). Immunostimulant effect of a mixed herbal extract on infectious bursal disease virus (IBDV) vaccinated chickens in the context of a co-infection model of avian influenza virus H9N2 and IBDV. *Comparative Immunology, Microbiology and Infectious Diseases*, *72*, 101505.

El-Radhi, A. S. M. (2008). Why is the evidence not affecting the practice of fever management? *Archives of Disease in Childhood*, *93*(11), 918-920.

Eterradossi, N., & Saif, Y. M. (2013). Infectious bursal disease. In Swayne D. E. (ed.), *Diseases of poultry* (14th edition. Wiley-Blackwell.

Fan, L., Wu, T., Wang, Y., Hussain, A., Jiang, N., Gao, L., Li, K., Gao, Y., Liu, C., Cui, H., & Qi, X. (2020). Novel variants of infectious bursal disease virus can severely damage the bursa of fabricius of immunized chickens. *Veterinary Microbiology*, *240*, 108507.

Franciosini, M. P., & Davidson, I. (2022). A walk through Gumboro disease. *Poultry*, *1*(4), 229-242.

Gana, B. A., Yidawe, P. J., Johnson, S., Bassey, E. A., & Ayuba, A. P. (2019). Antibody response to Newcastle disease vaccine of cockerels challenged with virulent infectious bursal disease virus and administered some complementary and alternative therapies. *Journal of Applied Sciences*, *19*(5), 466-472.

Hinks, A., Jacob, K., Mashouri, P., Medak, K. D., Franchi, M. V., Wright, D. C., ... & Power, G. A. (2022). An increase in serial sarcomere number induced via weighted downhill running improves work loop performance in the rat soleus. *bioRxiv*, 2022-02.

Huang, X., Liu, W., Zhang, J., Liu, Z., Wang, M., Wang, L., Zhou, H., Jiang, Y., Cui, W., Qiao, X., & Tang, L. (2021). Very virulent infectious bursal disease virus-induced immune injury is involved in inflammation, apoptosis, and inflammatory cytokines imbalance in the bursa of fabricius. *Developmental & Comparative Immunology*, *114*, 103839.

Hussein, E. A., Hair-Bejo, M., Adamu, L., Omar, A. R., Arshad, S. S., Awad, E. A., & Aini, I. (2018). Scoring system for lesions induced by different strains of Newcastle disease virus in chicken. *Veterinary Medicine International*, *Volume 2018,* Article ID 9296520, 9 pages.

Ingrao, F., Rauw, F., Lambrecht, B., & van den Berg, T. (2013).

Infectious bursal disease: a complex host–pathogen interaction. *Developmental & Comparative Immunology, 41*(3), 429-438.

Jackwood, D. J. (2017). Advances in vaccine research against economically important viral diseases of food animals: Infectious bursal disease virus. *Veterinary Microbiology*, *206*, 121-125.

Kogut, M. H., & Arsenault, R. J. (2017). Immunometabolic phenotype alterations associated with the induction of disease tolerance and persistent asymptomatic infection of Salmonella in the chicken intestine. *Frontiers in immunology*, *8*, 372.

Long, F. Y., Guo, Y. M., Wang, Z., Liu, D., Zhang, B. K., & Yang, X. (2011). Conjugated linoleic acids alleviate infectious bursal disease virus-induced immunosuppression in broiler chickens. *Poultry science*, *90*(9), 1926-1933.

Mahgoub, H. A. (2012). An overview of infectious bursal disease. *Archives of virology*, *157*(11), 2047-2057.

Michel, L. O., & Jackwood, D. J. (2017). Classification of infectious bursal disease virus into genogroups. *Archives of Virology*, *162*(12), 3661-3670.

Mosad, S. M., Eladl, A. H., El-Tholoth, M., Ali, H. S., & Hamed, M. F. (2020). Molecular characterization and pathogenicity of very virulent infectious bursal disease virus isolated from naturally infected turkey poults in Egypt. *Tropical animal health and production*, *52*(6), 3819-3831.

Mota-Rojas, D., Wang, D., Titto, C. G., Gómez-Prado, J., Carvajal-De la Fuente, V., Ghezzi, M., Boscato-Funes, L., Barrios-García, H., Torres-Bernal, F., Casas-Alvarado, A., & Martínez-Burnes, J. (2021). Pathophysiology of fever and application of infrared thermography (IRT) in the detection of sick domestic animals: Recent advances. *Animals*, *11*(8), 2316.

Müller, H., Mundt, E., Eterradossi, N., & Islam, M. R. (2012). Current status of vaccines against infectious bursal disease. *Avian pathology*, *41*(2), 133-139.

Mustafa, G., Ali, M. A., Smith, D. L., Masood, S., Qayyum, M. F., Ahmed, N., Rehman, A.U., Ahmad, S., Hussain, S., Arshad, M., & Schwinghamer, T. D. (2021). Formalin fumigation and steaming of various composts differentially influence the nutrient release, growth and yield of muskmelon (*Cucumis melo* L.). *Scientific reports*, *11*(1), 21057.

National Research Council (2011). *Guide for the care and use of laboratory animals* (8th edition). National Academies Press.

Ngom, R. V., Ayissi, G. J., Akoussa, A. M., Laconi, A., Jajere, S. M., Zangue, H. A., & Piccirillo, A. (2024). A systematic review and meta-analysis of the efficacy of biosecurity in disease prevention and control in livestock farms in Africa. *Transboundary and Emerging Diseases*, *Volume 2024,* Article ID 8683715, 14 pages.

Phillips, C. J., Hosseintabar-Ghasemabad, B., Gorlov, I. F., Slozhenkina, M. I., Mosolov, A. A., & Seidavi, A. (2023). Immunomodulatory effects of natural feed additives for meat chickens. *Life*, *13*(6), 1287.

Sharma, J. M., Kim, I. J., Rautenschlein, S., & Yeh, H. Y. (2000). Infectious bursal disease virus of chickens: pathogenesis and immunosuppression. *Developmental & Comparative Immunology*, *24*(2-3), 223-235.

Surai, P. F. (2016). Antioxidant systems in poultry biology: superoxide dismutase. *Journal of Animal Research and Nutrition*, *1*, 8.

Wang, Y. S., Wang, Z. C., Tang, Y. D., Shi, Z. L., He, K. W., Li, Y., Hou, J.B., Yao, H.C., Fan, H. J., & Lu, C. P. (2007). Comparison of four infectious bursal disease viruses isolated from different bird species. *Archives of virology*, *152*(10), 1787-1797.

Wati, T., Ghosh, T. K., Syed, B., & Haldar, S. (2015). Comparative efficacy of a phytogenic feed additive and an antibiotic growth promoter on production performance, caecal microbial population and humoral immune response of broiler chickens inoculated with enteric pathogens. *Animal Nutrition*, *1*(3), 213-219.

Zhao, S., Jia, Y., Han, D., Ma, H., Shah, S. Z. A., Ma, Y., & Teng, K. (2016). Influence of the structural development of bursa on the susceptibility of chickens to infectious bursal disease virus. *Poultry science*, *95*(12), 2786-2794.