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Effect of energy fortification and functional nutrient multicomponent supplementation in peripartum HFX and Deoni cows on post partum performance

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This study examined whether prepartum close-up period energy fortification (+10% dietary gross energy via glycerol, jaggery, or maize) combined with a functional nutrient multicomponent supplement influenced colostrum immunoglobulin G (IgG) concentration, calf birth weight, fetal membrane expulsion time (FMET) and early lactation milk yield in HF-crossbred (HFX) and indigenous Deoni cows. Thirty-two prepartum cows (16 HFX, 16 Deoni; 3rd–5th parity; 35 ± 5 d pre-calving) were assigned to 2 (breed) × 2 (FNMS ± supplement) × 4 (energy source) factorial arrangement. Colostrum IgG was estimated from colostrometer-derived specific gravity. Day-0 colostrum IgG showed significant (P = 0.001) Energy × Breed and Energy × FNMS interactions; FNMS-supplemented HFX cows averaged 61.81 mg/mL compared to 56.30 mg/mL in unsupplemented Deoni cows. IgG concentration declined steeply by day 1 (44%) and no treatment differences were detectable from days 1–4. HFX calves at birth were heavier (49%) than Deoni calves (P < 0.001) and FNMS supplementation improved (P < 0.001) overall calf birth weight by 8% (12% in HFX, 5% in Deoni;). FMET was 38 min shorter (P < 0.001) in Deoni than HFX cows, with FNMS showing a borderline tendency to hasten expulsion (P = 0.08). Milk yield was overwhelmingly governed by breed genetics. No metabolic disease or calf mortality was recorded. These results suggest that FNMS supplementation during the close-up period enhanced colostrum quality and calf vigour in crossbred dairy cows.

Keywords: Calf birth weight, Colostrum IgG, Functional nutrient multicomponent supplement, Prepartum nutrition, Transition cow.

Introduction

The transition period is three to four weeks before and after calving which is the most critical nutritional phase in dairy cattle management. As parturition approaches, dry matter intake (DMI) falls by 25–35%, precipitating negative energy balance (NEB) that predisposes cows to various metabolic conditions like ketosis, fatty liver, displaced abomasum, and retained placenta (Drackley et al., 2005). A concurrent biological imperative during this period is colostrogenesis, which is the immunoglobulin G (IgG)-enriched secretory process in the mammary gland, essentially completes by calving. Because calves are born immunologically naive owing to the synepitheliochorial placentation of cattle, first-milking colostrum IgG is a critical determinant of passive immunity transfer, neonatal survival and lifetime health trajectory (Godden, 2008; Barrington and Parish, 2001, Westhoff et al., 2024).

Augmenting close-up diet energy density with fermentable carbohydrate sources(glycerol, jaggery crude sugar or maize starch) has been proposed as a strategy to blunt NEB without demanding large increases in feed volume (McLeod and Baldwin, 2000; Trubenbach et al., 2019). These energy substrates differ markedly in their chemical nature: glycerol is a polyol requiring phosphorylation via the glpK or dhaK pathway before microbial utilization; jaggery provides rapidly fermentable monosaccharides (glucose and fructose); and maize starch is a complex polysaccharide with longer fermentation kinetics (Blötz and Stülke, 2017).

Complementary to energy fortification, a functional nutrient multicomponent supplement (FNMS) containing niacin, omega-3 fatty acids, selenium and key trace minerals addresses the periparturient immune shift through multiple converging mechanisms. Niacin modulates adipocyte lipolysis via G protein-coupled receptor 109A (GPR109A) activation; selenium and zinc underpin selenoprotein-mediated antioxidant defence, while long-chain omega-3 fatty acids from flaxseed attenuate pro-inflammatory eicosanoid synthesis, collectively reducing the oxidative and inflammatory burden at parturition (Bertoni et al., 2006; Moallem et al., 2013; Aragona et al., 2016, Hare et al., 2023).

Cross-breeding of Deoni cattle, an indigenous draft-cum-dairy breed native to the Deccan plateau with Holstein Friesian (HF) bulls to produce HFX (crossbreds) is a prevalent improvement strategy in peninsular India. These two genotypes differ considerably in body weight, milk production potential, metabolic resilience and presumably in the hormonal and immunological determinants of colostrogenesis, making them a biologically informative pair for studying prepartum nutritional interventions. The present study, therefore, was designed to evaluate the *in vivo* effects of close-up period energy fortification with or without FNMS on colostrum IgG, calf birth weight, fetal membrane expulsion time (FMET) and early lactation performance in HFX and Deoni cows.

Materials and Methods

2.1 Ethical Approval and Study Location

All experimental procedures were approved by the Institutional Animal Ethics Committee (CPCSEA/IAEC/LA/SRS-ICAR-NDRI-2019/No.10). The study was conducted at the Livestock Research

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Centre, ICAR-NDRI Southern Regional Station, Bengaluru (12.947°N, 77.608°E; elevation 921 m amsl) between November 2019 and March 2020. The site experiences a tropical savanna climate (Köppen–Geiger: Aw) with mean ambient temperatures of 17–28°C and relative humidity of 51–71% across the study period. Heat index values recorded during the experimental period ranged from the normal to alert category for most months.

2.2 Animals, Housing, and Experimental Design

A total of thirty-two prepartum cows, 16 HFX (HFX; *Bos taurus* × *Bos indicus*) and 16 Deoni (*Bos indicus*) were enrolled at 35 ± 5 days before expected calving. All animals were in their 3rd to 4th parity with average body weights 476.69±16.78 kg in HFX and 350.06±8.55 kg in Deoni. Animals were individually housed in ventilated 6 × 6 ft calving pens with evaporative cooling, twice-daily cleaning, daily bathing and unrestricted access to clean drinking water.

A 2 (breed: HFX vs. Deoni) × 2 (FNMS: with or without) × 4 (energy source: control, glycerolor jaggery or maize) factorial arrangement was employed, resulting in 16 treatment cells with two animals per cell (N = 32). The basal diet was formulated to meet nutrient requirements ICAR (2013) for late-gestation cows and comprised of mixed green fodder (Para grass : Hybrid Napier : Maize at 25:25:50), finger millet straw (*Eleusine coracana*), and a commercial concentrate mixture (Karnataka Milk Federation, Bengaluru; with principal ingredients such as maize 38%, deoiled oilseed cakes, rice bran and molasses). Green fodder, straw, and concentrate were offered in 50:20:30 ratio (roughage-to-concentrate ratio 70:30). Isocaloric energy fortification at +10% of total dietary gross energy was provided as glycerol (GE: 3,994 kcal/kg), jaggery (GE: 3,860 kcal/kg), or ground maize grain (GE: 4,094 kcal/kg). Experimental feeding ran from 35 days prepartum to calving.

2.3 Functional Nutrient Multicomponent Supplement (FNMS) Composition

The FNS was a precision blend formulated to supplement micronutrients after accounting for dietary supply relative to target requirements (ICAR, 2013). The composition of FNMS used in the present study consisted of copper (Cu) at 20 mg/kg DM, zinc (Zn) at 60 mg/kg DM, cobalt (Co) at 0.11 mg/kg DM, selenium (Se) at 0.3 ppm DM, iodine (I) at 0.4 mg/kg DM, chromium (Cr) at 5 mg/d, niacin at 12 g/d, and omega-3 fatty acids at 40 g/d. Omega-3 fatty acids were supplied through whole flaxseed (*Linum usitatissimum*; 35.6% ether extract, 53% of total fatty acids as alpha-linolenic acid). The FNMS was offered once daily at 08:00 AM mixed with the concentrate mixture.

2.4 Measurements and Analytical Methods

Proximate composition of dietary ingredients and refusals was determined using AOAC (2000) procedures. Cell wall constituents (NDF, ADF, ADL, hemicellulose, cellulose) were analysed per Van Soest et al. (1991). Gross energy was measured using an adiabatic bomb calorimeter. Colostrum was collected aseptically at each of the first five milkings (0–4 days of post-calving). IgG concentration (mg/mL) was estimated from colostrometer-measured specific gravity using the conversion equation: IgG = 958 × SG − 969 (Mechor et al., 1992). Calf birth weight (kg) was recorded within 2 h of birth. The fetal membrane expulsion time (FMET) was measured as the duration between the time of calf birth and the time of complete expulsion of placental membranes. Milk yield was recorded daily from day 5 post-calving and aggregated to weekly totals for eight weeks and average was calculated.

2.5 Statistical Analysis

The *in vivo* data were analysed using a 2 × 2 × 4 factorial ANOVA with breed, FNMS and energy source as fixed effects. For colostrum IgG (repeated over days 0–4) and weekly milk yield (repeated over 8 weeks), a mixed model with repeated measures was fitted (PROC MIXED) using an autoregressive AR(1) covariance structure. Least-squares means ± SEM are reported; treatment means were separated by Duncan's Multiple Range Test. Statistical significance was declared at $P \leq 0.05$ and tendencies at $P \leq 0.10$.

Results and Discussion

3.1 Colostrum IgG Concentration

Estimated colostrum IgG concentrations across days 0–4 are presented in Fig 1. All day-0 group means (range 55.10–62.77 mg/mL) exceeded the 50 mg/mL threshold associated with adequate colostrum quality (Godden, 2008), which is encouraging from a herd management standpoint. The overall day-0 mean was 58.76 mg/mL. *Error bars omitted for clarity, Broken lines indicate*

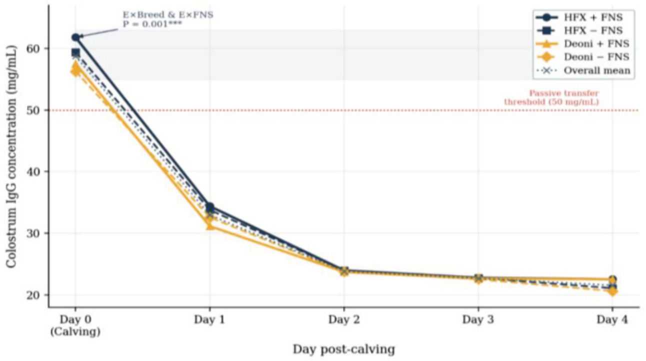

Figure 1. Estimated colostrum IgG concentration (mg/mL) across days 0–4 day post-calving by breed, energy and FNMS supplementation

without FNS and solid lines indicate with FNS. Dotted grey line is overall mean. Values are least-squares means from colostrometer-derived SG conversion. All day-0 group means exceeded the 50 mg/mL indicative threshold for adequate colostrum quality (broken red line). Treatment effects were confined to day 0; no significant differences were detected from days 1 to 4.

A significant (P = 0.001) Energy × Breed interaction was observed on day 0: HFX cows averaged 60.61 mg/mL as compared to 56.90 mg/mL in Deoni cows. The Energy × FNMS interaction was equally significant (P = 0.001), with FNMS groups averaging 59.66 mg/mL versus 57.86 mg/mL without supplementation. The highest (P=0.087) group mean was recorded in FNMS-supplemented HFX cows (61.81 mg/mL) and the lowest in unsupplemented Deoni cows (56.30 mg/mL). The three-way E × B × S interaction was non-significant (P = 0.310), suggesting that the FNMS benefit on day-0 IgG was consistent across energy substrates within the same breed.

The 3% improvement in day-0 IgG associated with FNMS is biologically plausible though more modest than the 18% gain reported by Aragona et al. (2016) using 48 g/d nicotinic acid. The FNMS in the present study incorporated only 12 g niacin/d, which may explain the attenuated response if dose-dependent GPR109A receptor activation is the primary mechanism. Selenium (0.3 ppm) and long-chain omega-3 fatty acids likely contributed via complementary antioxidant (selenoprotein activity) and anti-inflammatory (eicosanoid modulation) pathways that support lymphocyte function during colostrogenesis. Hough et al. (1990) and Westland et al. (2017) also reported similarly modest but directionally consistent improvements in colostrum IgG with nutritional supplementation. The colostrum quality on day 0 measured as specific gravity in either HFX or Deoni cows was in agreement with Vaz et al. (2004), who reported total immunoglobulin levels >50 mg/dL in HF cows.From day 1 onwards, IgG concentrations diminishes sharply across all groups (day-1 mean: 32.98 mg/mL, a 44% decline) and stabilised between 22 and 24 mg/mL through day 4. No treatment differences were detectable on days 1–4 (P > 0.107 for all terms), consistent with the established biology of mammary tight-junction closure at parturition which terminates immunoglobulin transport into milk regardless of circulating IgG levels (McGrath et al., 2016). The quality of colostrum decreased by approximately 50% on day 1 itself and by day 4 was only one-third of the day-0 value, which is in agreement with Puppel et al. (2019).

3.2 Calf Birth Weight

Calf birth weight data are presented in Table 2. HFX calves were on average 10 kg or 49% heavier (P<0.001) at birth than Deoni calves (30.16 vs. 20.29 kg;), reflecting the well-established direct proportionality between dam body size and offspring birth weight (Paputungan and Makarechian, 2000). The metabolic BW of HFX calves was approximately 26% greater, mirroring the 20% difference in metabolic BW between HFX and Deoni dams. These values are consistent with birth weight differentials of 8–12 kg between crossbred and indigenous calves reported by Singh and Srinivas (2020) for peninsular India.

Table 2. Calf birth weight (kg) and metabolic BW (kg0.75) by energy source, breed, and FNS supplementation

	Control	Glycerol	Jaggery	Maize	P-value
Calf BW (kg)					
Breed					
HFX	27.95	32.19	29.48	31.03	0.001***
Deoni	19.93	19.48	21.37	20.38	
FNMS					
+	24.28	27.91	26.11	26.86	0.001***
–	23.60	23.75	24.74	24.54	
HFX × FNMS					
+	29.75	35.13	30.28	32.13	0.027*
–	26.15	29.25	28.69	29.93	
Deoni × FNMS					
+	18.80	20.70	21.95	21.60	NS
–	21.05	18.25	20.79	19.16	
(E×B×S)					0.049*

*SEM: 0.685 (BW) and 0.227 (MBW). ***P ≤ 0.001; *P ≤ 0.05; NS = not significant.*

Table 3. Effect of energy source, breed and FNMS supplementation on Fetal membrane expulsion time (min)

	Control	Glycerol	Jaggery	Maize	P-value
FMET (min)					
Breed					
HFX	206	211	194	191	0.001***
Deoni	175	154	159	163	
FNMS					
+	188	176	174	173	0.08†
–	194	189	179	181	
HFX × FNMS					
+	195	210	188	188	NS
–	218	213	200	195	
Deoni × FNMS					
+	180	143	160	158	NS
–	170	165	158	168	
(E×B×S)					0.099†

****P ≤ 0.001; †P ≤ 0.10 (tendency); NS = Not significant (P > 0.05).*
FMET = Fetal Membrane Expulsion Time. E×B×S = Energy × Breed × Supplement interaction.
No animal in any group experienced retained placenta (≥ 720 min threshold).

The FNMS supplementation was associated with an overall 8% improvement in calf birth weight (24.16 vs. 26.29 kg without and with FNMS, respectively; P < 0.001). The response was larger in HFX dams (12%; 28.50 vs. 31.82 kg) than in Deoni dams (5%; 19.81 vs. 20.76 kg). The three-way E × B × S interaction on birth weight reached significance (P = 0.049), driven primarily by calves born to dams receiving maize + FNMS being heavier than control calves. The approximate 2 kg absolute improvement in calf birth weight with FNMS (3 kg in HFX, 2 kg in Deoni) is biologically consistent with the functional roles of selenium in placental angiogenesis and cell proliferation, zinc in protein synthesis during the period of maximum fetal growth and omega-3 fatty acids in uteroplacental blood flow (Mee et al., 1995; ICAR, 2013). It is noteworthy that the last trimester accounts for more than 70% of calf birth weight, with approximately 30% accrued in the final month (ICAR, 2013), making the close-up nutritional window critically important for fetal development. Khan et al. (2004) reported 2.8 kg higher birth weight in cows fed 30% more than recommended energy during the last trimester, while Loerch (1996) reported 4.2 kg heavier calves from high-grain prepartum diets, contextualising the present 1–3 kg FNMS-associated improvement.

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3.3 Fetal membrane Expulsion Time

Results for FMET revealed that Deoni cows expelled fetal membranes significantly (P < 0.001) faster than HFX cows in Table 3. (163±5.4 vs 201±6.2 min). This breed difference most likely reflects differences in uterine musculature tone, oxytocin receptor density and the lower body weight of Deoni cows, which reduces mechanical resistance during placental separation. Importantly, all observed FMET values were well below the 720-min (12-h) threshold that defines retained placenta clinically, and no animal in any group experienced placental retention, a meaningful positive finding confirming the safety of the tested regimen.

FNMS supplementation showed a borderline tendency to hasten FMET (P = 0.08), possibly mediated by the selenium component (0.3 ppm): selenium is known to support prostaglandin F2α synthesis and myometrial contractility that facilitate placental separation and expulsion (Mee et al., 1995; Allison et al., 2000). Weiss and Gonzalo (2006) reported that Se supplementation at 0.3 ppm elevated plasma antioxidants and reduced immune-suppressive oxidative stress that is known to impair uterine contractility in the periparturient period. The Energy × Breed × FNMS interaction effect on FMET was significant at the 10% level (P = 0.099), though the absolute breed × FNMS difference was only 5–10 min, which has limited clinical significance.

3.4 Early Lactation Performance

The Table 4 presents weekly milk yield across the first eight weeks of lactation. The significant aspect of the data is the breed effect HFX cows produced between three and four times as much milk as Deoni cows every week without exception (P < 0.001 throughout), reflecting the fundamental difference in genetic milk production capacity between a Holstein-influenced crossbred and an indigenous draft-cum-dairy breed (Mohanavel and Srinivas 2016). These results show that neither prepartum energy source nor FNMS produced a statistically significant effect on weekly milk yield at any time point. In HFX cows, FNMS- animals produced numerically 5–7% more milk (Fig 2) in weeks 1 and 2 (18.04 vs. 15.47 kg/week in week 1 for the control group), but this difference was well within chance variation given the sample size (P = 0.64) (Fig 2). In Deoni cows, FNMS made no discernible difference at any point. The absence of a sustained milk yield response to prepartum FNS supplementation is consistent with DeFrain et al. (2004) and Osborne et al. (2009), and may simply reflect the relatively short carry-over duration of close-up nutritional interventions on lactation performance.

Table 4. Effect of energy source, breed and FNMS supplementation on early lactation (1–8 weeks) average milk yield (kg/cow/week)

Week	Breed	Control	Glycerol	Jaggery	Maize	P-value
I	HFX	15.19	14.38	14.82	14.71	0.001***
	Deoni	3.70	3.34	3.66	4.46	
II	HFX	18.29	17.50	15.96	15.91	0.001***
	Deoni	4.14	4.06	4.61	4.20	
III	HFX	18.48	16.66	16.32	16.30	0.001***
	Deoni	4.17	4.15	4.30	4.43	
IV	HFX	16.82	15.76	16.38	16.38	0.001***
	Deoni	4.52	4.05	4.51	4.53	
V	HFX	18.06	15.77	16.16	15.77	0.001***
	Deoni	4.60	4.07	4.61	4.65	
	VIHFX	18.70	16.21	15.95	15.77	0.001***
	Deoni	4.83	4.43	4.46	4.62	
VII	HFX	18.92	16.04	15.38	15.88	0.001***
	Deoni	4.88	4.56	4.19	4.56	
VIII	HFX	19.29	15.41	15.75	14.70	0.001***
	Deoni	4.66	4.50	4.49	4.56	
Avg	HFX	17.08	15.15	15.04	14.87	0.001***
	Deoni	4.22	3.93	4.14	4.28	

*SEM range: 0.885–1.252. Energy × FNMS and E × B × S interactions non-significant at all weeks (P = 0.161–0.978). ***P ≤ 0.001 for Energy × Breed at every weekly interval.*

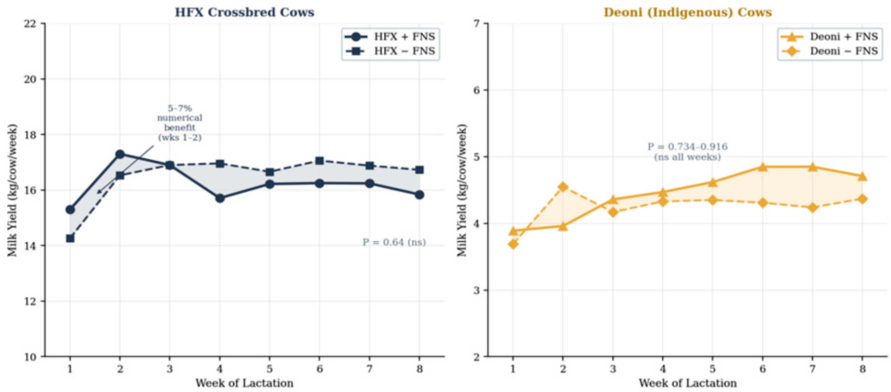

Figure 2: Early lactation weekly milk yield (weeks 1-8) with or without FNMS

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Prepartum FNMS supplementation showed a non-significant (P=0.64) 5-7% advantage in HFX cows during weeks 1-2 only dissipating from week 3. Deoni cows showed no FNMS-related milk yield difference at any week. HFX produced 3.75X more than Deoni (P<0.001) throughout.

3.5 Animal Health and Welfare

There were no cases of milk fever, clinical ketosis, displaced abomasum, retained placenta, calf respiratory disease or calf mortality were recorded across all 32 dam-calf pairs through out the observation period. While the absence of metabolic disease is encouraging and confirms the effectiveness and safety of all tested supplementation levels, the clinical observation window was extended only up to 30 days of post-calving.

Conclusion

The FNMS supplementation during the close-up period significantly improved day-0 colostrum IgG in HFX cows (Energy × Breed, P = 0.001) and improved calf birth weight across both breeds (8%; P < 0.001), with a numerically larger response in HFX (12%) than Deoni (5%) dams. A borderline tendency for FNMS to hasten fetal membrane expulsion (P = 0.08) was observed, consistent with selenium-mediated enhancement of myometrial contractility. Early lactation milk yield was overwhelmingly determined by breed genetics and was not significantly influenced by prepartum nutritional interventions. The absence of metabolic disease or calf mortality across all groups confirms the safety of the tested close-up supplementation regime. These findings support the inclusion of a multicomponent FNMS in close-up prepartum diets, particularly in HF-crossbred herds under Southern Indian conditions, to improve colostrum quality and calf birth vigour. Larger, adequately powered trials with direct IgG measurement by radial immunodiffusion or ELISA and comprehensive metabolic monitoring are warranted to confirm and extend these observations.

Conflict of Interest

The authors declare no competing interests. This investigation formed part of a doctoral dissertation conducted at ICAR-NDRI, Southern Regional Station, Bengaluru. No external funding with conditions attached to outcomes was received.

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