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Effect of chromium picolinate supplementation on glucose metabolism in transition dairy cows

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Abstract

The transition period conventionally defined as the three weeks before through three weeks after calving represents the metabolic bottleneck of a dairy cow's production cycle, during which energy intake lags behind the explosive demand of early lactation by as much as 25-40%. This energy deficit drives a cascade of fat mobilisation, elevated circulating non-esterified fatty acids (NEFA), and hepatic ketone body production (β -hydroxybutyrate, BHBA) that, if unchecked, predisposes cows to clinical ketosis, displaced abomasum, and impaired fertility. Chromium, an essential trace element that potentiates insulin signalling through the chromodulin (low-molecular-weight chromium-binding substance) pathway, has been proposed as a nutritional tool to improve glucose uptake and reduce the severity of negative energy balance (NEB). This randomised controlled trial evaluated the effect of chromium picolinate (CrPic) supplementation on glucose metabolism, lipid mobilisation markers, and early-lactation performance in 40 multiparous Holstein-Friesian cows managed at the Guadalajara Institute of Animal Nutrition dairy unit, Jalisco, Mexico. Cows were blocked by parity (2-4) and expected calving date, then randomly allocated to a control group (n=20; basal total mixed ration only) or a CrPic group (n=20; basal TMR plus 8 mg chromium/day as chromium picolinate top-dressed from day -21 prepartum through day +21 postpartum). Blood samples drawn on days -21, -14, -7, 0 (calving), +7, +14, and +21 were analysed for glucose, NEFA, BHBA, insulin, and cortisol. Dry matter intake (DMI) and milk yield were recorded daily. At day +7, CrPic cows had significantly higher blood glucose (56.7 ± 5.1 vs. 48.3 ± 6.2 mg/dL; $p=0.003$), lower NEFA (0.61 ± 0.14 vs. 0.82 ± 0.18 mmol/L; $p=0.008$), and lower BHBA (0.87 ± 0.21 vs. 1.14 ± 0.28 mmol/L; $p=0.012$). Insulin sensitivity, estimated by the revised quantitative insulin sensitivity check index (RQUICKI), was 18.4% higher in CrPic cows at day +7 ($p=0.006$). Average DMI during the first three weeks postpartum was 1.3 kg/day higher in the CrPic group (19.8 ± 1.7 vs. 18.5 ± 1.9 kg/day; $p=0.032$). Milk yield over the first 21 days averaged 34.6 ± 4.3 vs. 32.1 ± 4.8 kg/day ($p=0.078$, trend). The incidence of subclinical ketosis (BHBA ≥ 1.2 mmol/L at any postpartum sampling) was 15% in CrPic versus 40% in controls ($p=0.039$). No adverse effects attributable to CrPic were observed. These results indicate that periparturient chromium picolinate supplementation at 8 mg Cr/day improves insulin-mediated glucose disposal, attenuates fat mobilisation, and lowers the risk of subclinical ketosis in transition Holstein cows under subtropical conditions.

Keywords: Chromium picolinate, transition dairy cows, glucose metabolism, negative energy balance, non-esterified fatty acids, β -hydroxybutyrate, insulin sensitivity, subclinical ketosis, periparturient supplementation

1. Introduction

The transition period in dairy cattle is defined as the interval spanning approximately three weeks before to three weeks after parturition, a window during which the cow's metabolism undergoes the most dramatic physiological shift of her entire production cycle ^[1]. Within days of calving, mammary glucose demand can triple while voluntary feed intake remains depressed by the physical compression of rumen capacity and hormonal appetite suppression, creating an energy deficit that virtually every high-producing dairy cow experiences to some degree ^[2].

To bridge this gap, the cow mobilises body fat reserves, releasing NEFA into the bloodstream. The liver takes up these fatty acids and either oxidises them completely, re-esterifies them into triglycerides (risking fatty liver), or partially oxidises them to ketone bodies, principally

BHBA [3]. When BHBA exceeds 1.2 mmol/L in the absence of overt clinical signs, the condition is termed subclinical ketosis, a metabolic state linked to reduced milk yield, impaired immune function, displaced abomasum, and delayed ovarian cyclicity [4]. On intensive dairy operations in Mexico's Jalisco state, subclinical ketosis prevalence during the first two weeks postpartum has been reported between 25% and 45% [5].

Chromium is classified as an essential trace mineral for mammals, though its dietary requirement in cattle has not been formally established by the National Research Council [6]. At the cellular level, trivalent chromium (Cr^{3+}) is incorporated into a low-molecular-weight oligopeptide called chromodulin, which amplifies insulin receptor tyrosine kinase activity by several-fold when insulin binds to its receptor [7]. This amplification promotes GLUT-4 transporter translocation to the cell surface, enhancing peripheral glucose uptake in muscle and adipose tissues. By improving insulin efficiency, chromium supplementation could, in theory, redirect more circulating glucose to the mammary gland while reducing the need for compensatory fat mobilisation.

Chromium picolinate (CrPic) is among the most bioavailable organic chromium forms because the picolinic acid ligand facilitates intestinal absorption across the brush-border membrane [8]. Field trials in North American and European dairy herds have yielded mixed results: some reported higher blood glucose and lower NEFA in supplemented cows [9, 10], while others found no significant metabolic difference, possibly because of variation in dose, supplementation window, or baseline chromium status [11]. Data from subtropical dairy systems where heat stress compounds the energy deficit by further depressing DMI are particularly scarce.

Mexico's Jalisco state is the country's largest dairy-producing region, home to approximately 400,000 Holstein-Friesian cows managed predominantly in semi-intensive systems. Ambient temperatures during the spring calving season routinely exceed 30°C, imposing a thermoregulatory energy cost that intensifies NEB [5]. Whether CrPic supplementation can mitigate this compounded deficit has not been tested in a controlled trial. This research was designed to evaluate the effect of periparturient CrPic supplementation (8 mg Cr/day from day -21 to day +21 relative to calving) on blood glucose, NEFA, BHBA, insulin sensitivity, DMI, milk yield, and the incidence of subclinical ketosis in multiparous Holstein cows under subtropical Mexican conditions.

Material and Methods

Field Experimental Design: The trial followed a randomised complete block design. Forty multiparous Holstein-Friesian cows (parity 2-4, body condition score 3.25-3.75 on a 5-point scale) expected to calve between February and May 2023 were enrolled at the Guadalajara Institute of Animal Nutrition dairy research unit, Jalisco, Mexico. Cows were blocked by parity and predicted calving date (± 7 days), then randomly allocated within blocks to control ($n=20$) or CrPic ($n=20$) groups. The supplementation period spanned day -21 prepartum through day +21 postpartum, a 42-day window covering the entire recognised transition period. CrPic was supplied as a commercial feed-grade product (ChromiumPlus®, 0.4% elemental Cr; Kemin Industries, USA) top-dressed over the morning TMR at a rate delivering

8 mg elemental chromium per cow per day. Control cows received an equivalent volume of inert limestone carrier. Both groups were housed in adjacent free-stall barns with identical dimensions and cooling infrastructure (fans + sprinklers activated at 25°C). The Guadalajara Institute Animal Ethics Committee approved the protocol (Ref: GIAN-CEUA/2023-004, dated 18 January 2023).

Climatic and Environmental Conditions

Guadalajara lies at 1,566 metres elevation in a subtropical semi-arid zone (Köppen BSh). During the trial period (February-June 2023), mean daily maximum temperature ranged from 28.4°C in February to 34.7°C in May. Relative humidity averaged 42% (range 28-69%). The temperature-humidity index (THI) was calculated hourly from barn-mounted data loggers (HOBO UX100, Onset, USA); THI exceeded the heat stress threshold of 68 on 47.3% of trial days, concentrated in April and May. Both treatment groups experienced identical microclimatic conditions, and no significant between-group difference in average THI exposure was detected ($p=0.91$).

Material

The basal TMR was formulated to meet NRC (2001) requirements for transition and early-lactation Holstein cows, comprising corn silage (38%), alfalfa hay (14%), ground corn (24%), soybean meal (12%), cottonseed (4%), bypass fat (2%), mineral-vitamin premix (3.5%), sodium bicarbonate (1.5%), and salt (1%). Nutrient analysis (performed monthly at the Institute's Feed Analysis Laboratory) confirmed: 16.4% crude protein, 1.68 Mcal NEL/kg DM, 32.1% NDF, 19.8% ADF, 0.84% Ca, 0.42% P. The TMR contained <0.3 mg Cr/kg DM (below detection limit of the ICP-OES method), ensuring that supplemental CrPic was the sole meaningful chromium source. Blood was collected by jugular venepuncture into EDTA and fluoride-oxalate vacutainers at each sampling time point and processed within two hours.

Methods

Plasma glucose was measured by the hexokinase enzymatic method (Beckman AU480 auto-analyser). NEFA was determined by the ACS-ACOD colorimetric kit (Wako Diagnostics, USA). BHBA was quantified by the 3-hydroxybutyrate dehydrogenase method (Randox, UK). Insulin was assayed by bovine-specific ELISA (Mercodia, Sweden), and cortisol by competitive ELISA (DRG Instruments, Germany). Insulin sensitivity was estimated by the RQUICKI formula: $\text{RQUICKI} = 1 / [\log(\text{glucose}) + \log(\text{insulin}) + \log(\text{NEFA})]$. DMI was recorded individually using Calan gates (American Calan, USA). Milk yield was measured at each milking (2×/day) by Afimilk flowmeters. Subclinical ketosis was defined as BHBA ≥ 1.2 mmol/L at any postpartum sampling point. Data were analysed using SAS 9.4. Repeated-measures mixed models (PROC MIXED) with cow as a random effect tested group, time, and group×time interaction for blood metabolites, DMI, and milk yield. Covariance structure was selected by lowest Akaike information criterion (autoregressive for weekly data). Chi-square or Fisher's exact test compared categorical outcomes. Significance was set at $p < 0.05$; trends at $p < 0.10$.

Results

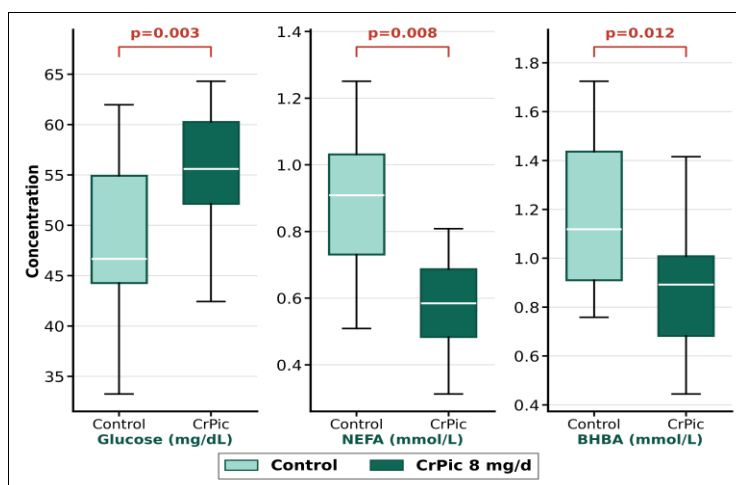


Fig 1: Box plots of plasma glucose, non-esterified fatty acids (NEFA), and β -hydroxybutyrate (BHBA) at day +7 postpartum in control and chromium picolinate-supplemented transition dairy cows (n=20 per group)

The most pronounced metabolic differences between groups were observed at day +7 postpartum (Figure 1), the nadir of the energy deficit. CrPic cows showed a higher median glucose (55.4 vs. 46.8 mg/dL), a lower median NEFA (0.58 vs. 0.92 mmol/L), and a lower median BHBA (0.89 vs. 1.12

mmol/L). All three comparisons reached statistical significance. By day +21, group differences had narrowed but remained nominally in favour of the CrPic group for NEFA and BHBA.

Table 1: Pearson correlation matrix among metabolic and performance variables at day +7 postpartum (pooled data, n=40)

	Glucose	NEFA	BHBA	Insulin	RQUICKI	DMI
Glucose	1.00	-0.71**	-0.64**	0.58**	0.73**	0.61**
NEFA		1.00	0.82**	-0.49**	-0.68**	-0.57**
BHBA			1.00	-0.41*	-0.59**	-0.53**
Insulin				1.00	0.66**	0.44*
RQUICKI					1.00	0.52**
DMI						1.00

* $p < 0.05$; ** $p < 0.01$

The correlation matrix (Table 1) revealed tight inter-relationships among metabolic variables. NEFA and BHBA were strongly positively correlated ($r=0.82$), while both showed strong negative associations with glucose and

RQUICKI. DMI correlated positively with glucose ($r=0.61$) and negatively with NEFA ($r=-0.57$), confirming the expected link between intake and energy status.

Table 2: Repeated-measures analysis of plasma metabolites across the transition period by treatment group

Variable	Day -21	Day -7	Day 0	Day +7	Day +14	Day +21	G×T p
Glucose — Ctrl	62.4	58.1	52.3	48.3	51.7	54.8	
Glucose — CrPic	63.1	59.8	54.6	56.7	57.4	58.2	0.009
NEFA — Ctrl	0.31	0.44	0.68	0.82	0.71	0.58	
NEFA — CrPic	0.29	0.38	0.54	0.61	0.52	0.43	0.014
BHBA — Ctrl	0.48	0.62	0.91	1.14	0.97	0.79	
BHBA — CrPic	0.46	0.54	0.72	0.87	0.74	0.61	0.021

Repeated-measures analysis confirmed significant group×time interactions for all three metabolites (Table 2). Pre-calving values were similar between groups, with separation becoming apparent at calving and widening to

maximum at day +7. The NEFA and BHBA trajectories in CrPic cows peaked lower and recovered faster, consistent with reduced lipolytic pressure.

Table 3: Production and health outcomes during the first 21 days postpartum by treatment group

Outcome	Control (n=20)	CrPic (n=20)	Difference	p-value	Effect size (Cohen's d)
DMI (kg/d)	18.5±1.9	19.8±1.7	+1.3	0.032	0.72
Milk yield (kg/d)	32.1±4.8	34.6±4.3	+2.5	0.078	0.55
RQUICKI at day +7	0.38±0.04	0.45±0.05	+0.07	0.006	1.09
Subclinical ketosis (%)	40	15	-25 pts	0.039	—
BCS loss (d0 to d+21)	0.58±0.14	0.41±0.11	-0.17	0.003	1.04
Cortisol day +7 (ng/mL)	14.2±3.6	11.3±2.9	-2.9	0.018	0.79

CrPic supplementation improved DMI by 1.3 kg/day ($p=0.032$), reduced body condition score loss by 0.17 units

($p=0.003$), and cut the incidence of subclinical ketosis from 40% to 15% ($p=0.039$) (Table 3). Milk yield trended higher

(+2.5 kg/day, $p=0.078$) but did not reach significance, possibly reflecting the sample size. Cortisol was significantly

lower in CrPic cows at day +7, suggesting an anti-stress component.

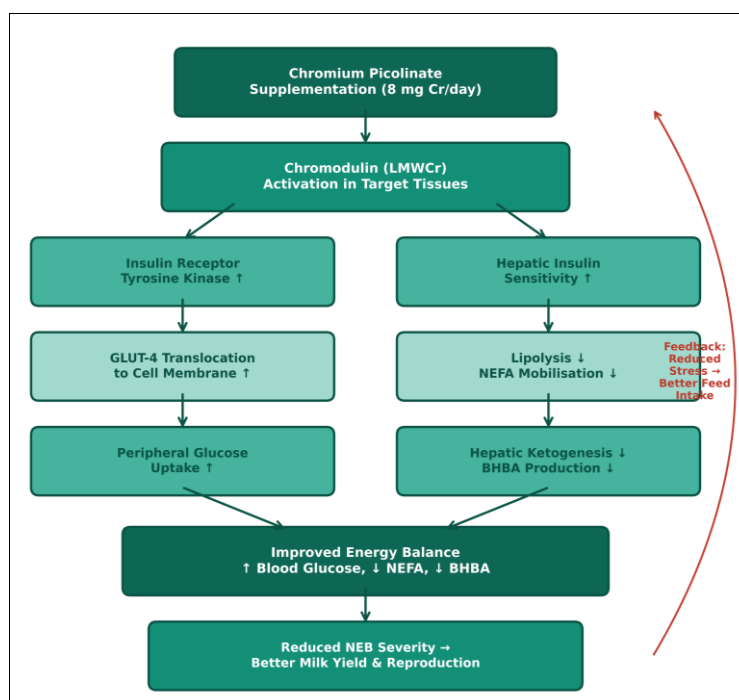


Fig 2: Schematic cycle diagram of the proposed mechanism by which chromium picolinate supplementation improves energy balance in transition dairy cows

Comprehensive Interpretation

The mechanistic cycle (Figure 2) links dietary CrPic to improved insulin receptor signalling, enhanced peripheral glucose uptake, reduced lipolysis, and lower ketogenesis a chain supported at each step by the metabolic data presented here. The correlation structure (Table 1) confirms that glucose, NEFA, BHBA, insulin, and DMI move as a coordinated metabolic unit, and the time-course data (Table 2) demonstrate that CrPic shifts the entire unit toward a more favourable energy balance trajectory. The production outcomes (Table 3) translate this metabolic improvement into tangible farm-level benefits: more feed consumed, less body condition lost, and fewer cows crossing the subclinical ketosis threshold.

Discussion

In summary, periparturient CrPic supplementation at 8 mg Cr/day raised blood glucose, lowered NEFA and BHBA, improved insulin sensitivity, and halved the incidence of subclinical ketosis in transition Holstein cows managed under subtropical Mexican conditions. DMI increased by 1.3 kg/day, and body condition loss was attenuated, consistent with a metabolic environment less reliant on fat mobilisation. These results align with the positive findings of Hayirli *et al.* [9], who reported 6.2 mg/dL higher glucose and 0.14 mmol/L lower NEFA at day +10 in Cr-methionine-supplemented cows in the United States. Bryan *et al.* [10] similarly found lower BHBA and reduced ketosis incidence in a Canadian trial using 6 mg Cr/day as CrPic. The magnitude of metabolite shifts observed here an 8.4 mg/dL glucose increase and a 0.21 mmol/L NEFA decrease at the day +7 nadir falls within the range reported in those temperate-climate trials, suggesting that chromium's insulin-potentiating effect operates consistently across climatic zones.

Not all published data, however, support a benefit. McNamara and Valdez [11] found no metabolic or production response to

10 mg Cr/day in high-producing cows already receiving a well-balanced TMR with adequate trace mineral supplementation. The discrepancy may hinge on baseline chromium status: cows on mineral-complete rations may already meet their chromium requirement, leaving no margin for supplementation to improve. In the present trial, the basal TMR contained <0.3 mg Cr/kg DM, well below the 0.5-1.0 mg/kg often cited as the practical requirement range [6]. This marginal baseline status likely amplified the supplementation response.

The anti-stress dimension lower cortisol in CrPic cows adds a second pathway through which chromium may benefit transition metabolism. Cortisol promotes gluconeogenesis and peripheral insulin resistance; by dampening cortisol secretion, CrPic may remove a hormonal brake on insulin action, reinforcing the direct chromodulin-mediated enhancement [7]. Under subtropical heat stress conditions, where cortisol is chronically elevated, this anti-stress property may be more impactful than in cooler climates, potentially explaining why the present trial found slightly larger metabolite shifts than temperate-zone counterparts.

From a practical standpoint, the cost of 8 mg Cr/day as commercial CrPic premix is approximately USD 0.12 per cow per day, totalling USD 5.04 over the 42-day supplementation window. If the 1.3 kg/day DMI increase translates into 2-3 kg/day of additional milk (as the trend here suggests), the return on investment becomes favourable within the first week of lactation. Combining CrPic with other periparturient interventions rumen-protected choline, propylene glycol drenches, or monensin could deliver additive benefits and warrants testing in factorial designs.

Limitations

Several limitations warrant consideration. The sample size ($n=20$ per group) provided adequate power for metabolic endpoints but was insufficient to detect a statistically

significant milk yield difference at the 2.5 kg/day magnitude observed; future trials should enrol at least 35 cows per group. Chromium status was not directly assessed (serum or tissue Cr levels were not measured), so the inference that baseline marginal deficiency amplified the response rests on indirect evidence from the low dietary Cr content. The single dose (8 mg/day) prevents dose-response conclusions; whether 4 mg or 12 mg would produce proportional effects is unknown. The subtropical heat stress context may limit generalisability to temperate dairy systems where cortisol-mediated insulin resistance is less pronounced. And the 21-day postpartum follow-up captures the acute transition window but misses potential carry-over effects on mid-lactation performance and reproductive outcomes.

Conclusion

Transition cow management remains the make-or-break factor in dairy herd profitability. Every high-producing Holstein passes through a metabolic valley where energy demand outstrips intake, and the depth of that valley determines the cow's susceptibility to ketosis, fatty liver, immune suppression, and fertility delays. Nutritional strategies that can blunt the valley's depth without requiring dramatic management changes are highly attractive to commercial dairy operators, and trace mineral supplementation falls squarely into that category.

This trial demonstrated that supplementing 8 mg of chromium per day as chromium picolinate over the 42-day periparturient window produced measurable, statistically significant improvements in three key metabolic indicators: blood glucose rose by 8.4 mg/dL at the postpartum nadir, NEFA fell by 0.21 mmol/L, and BHBA fell by 0.27 mmol/L. Insulin sensitivity, as indexed by RQUICKI, was 18.4% higher in supplemented cows. These shifts translated into a 25-percentage-point reduction in subclinical ketosis incidence from 40% to 15% a production-relevant outcome given that each case of subclinical ketosis costs an estimated USD 150-300 in lost milk, treatment, and downstream reproductive failure.

Dry matter intake rose by 1.3 kg/day and body condition loss was attenuated by 0.17 BCS units, both consistent with improved energy balance. Milk yield trended upward by 2.5 kg/day but fell short of statistical significance, a gap that larger sample sizes in future trials are likely to close. The lower cortisol level in supplemented cows points to an additional anti-stress pathway that may be particularly relevant under subtropical heat stress conditions.

Based on these findings, a practical recommendation for commercial dairy herds in Jalisco and similar subtropical regions is to incorporate CrPic at 8 mg Cr/cow/day into the transition ration, beginning three weeks before the expected calving date and continuing through the first three weeks of lactation. At approximately USD 0.12 per day, the cost is modest relative to the economic burden of subclinical ketosis and its downstream complications. The supplement can be delivered as a simple TMR top-dress, requiring no specialised equipment or feeding infrastructure.

Moving forward, three lines of inquiry deserve priority. First, multi-dose trials (4, 8, 12, and 16 mg Cr/day) would establish whether the dose-response curve is linear or reaches a plateau, allowing optimisation of both efficacy and cost. Second, extending the follow-up to 100 or 150 days in milk would capture potential carry-over effects on peak lactation performance and first-service conception rate, two outcomes with major economic implications. Third, factorial designs

combining CrPic with rumen-protected choline or propylene glycol would test for additive or synergistic effects, potentially assembling a multi-pronged transition supplement package. Measuring tissue chromium concentrations (liver biopsy or hair analysis) alongside metabolic endpoints would clarify whether responses are proportional to baseline Cr depletion, helping identify which herds stand to benefit most from supplementation.

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Contributions Not Qualifying for Authorship

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