Animal 19 (2025) 101541

Contents lists available at ScienceDirect

Animal

The international journal of animal biosciences

Effect of weaning and cow-calf contact on the physiological and clinical health, performance, and behaviour of dairy cows and their calves

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ARTICLE INFO

Article history:
Received 17 September 2024
Revised 28 April 2025
Accepted 29 April 2025
Available online 6 May 2025

Keywords:
Calf growth
Cow-calf separation
Dam-calf contact
Energy balance
Welfare

ABSTRACT

Cow-calf contact (**CCC**) systems have become increasingly popular calf–rearing systems to promote positive welfare; however, weaning and separation may cause distress. This preliminary study aimed to investigate the interaction between weaning and CCC on the physiological health, performance, and behaviour of dairy cows and calves. Three systems were compared: conventional, pasture-based Irish system (**CONV**; 18 pairs), cow and calf separated ≤ 2 h postbirth, cows milked twice-a-day, calves artificially reared indoors; full-time access system (**FT**; 14 pairs), dam and calf allowed constant, pasture-based, unrestricted access and cows milked twice-a-day; and part-time access system (**PT**; 18 pairs), unrestricted access at night indoors, cows grazed outdoors by day while calves remained indoors, cows milked once-a-day (0800 h). All calves were weaned at 8 weeks of age; FT and PT pairs underwent a 7 d gradual weaning and separation process (PT cows switched to twice-a-day milking) while CONV calves were gradually weaned over 12 d. Clinical health scores (2x/week), blood samples (1x/week; analysed for physiological markers of health and performance), BW (1x/week), body condition score (1x/week; cows only), and behaviour (1 d/week; scan sampling 3x/d; 24 total observations) were taken the week before (**preWS**) and after (**postWS**) the weaning and separation process. The PT cows had higher body condition scores (3.18 ± 0.034) than CONV (2.95; FT cows were similar to both, 3.05) and lower non-esterified fatty acids (**NEFA**; 0.40 ± 0.038 mmol/L) than the FT cows (0.58 mmol/L; CONV cows were similar to both, 0.48 mmol/L) across both time-points. All calves preWS had lower summed clinical health scores (0.91 vs 1.25 ± 0.131; P = 0.017), beta-hydroxybutyrate (0.07 vs 0.39 ± 0.023 mmol/L; P < 0.001), and globulin (12.0 vs 14.5 ± 0.929 g/L; P = 0.010) than postWS. After weaning and separation, the FT (0.36 mmol/L; P < 0.001) and PT (0.34 mmol/L; P = 0.001) calves had higher NEFA than CONV calves (0.13 mmol/L). Calf weekly average daily gain (**ADGw**) was similar preWS (0.9 ± 0.142 kg/d), but CONV calves had higher ADGw postWS than FT (0.42 kg/d) and PT calves (0.40 kg/d). All calves performed more (P = 0.009) positive behaviours preWS (4.6 ± 6.37%) compared to postWS (2.3 ± 3.38%). Our results suggest the applied CCC slightly worsened cow and calf health, performance, and behaviour around weaning and separation.

Implications

This explorative study investigated the interaction effect of weaning and cow-calf contact on the health, production, and behaviour of dairy cows and their calves. We compared three different calf rearing systems that differed in cow-calf contact within a seasonal-calving, pasture-based dairy system. This research suggests that cow-calf contact slightly worsened cow and calf health and performance around the weaning and separation period, but further research is required to fully understand the mechanisms. Future cow-calf contact work should focus on designing management routines to minimise the transition's impact on cows and calves, regardless of their housing system.

Introduction

Currently in conventional dairy calf–rearing systems practised globally, farmers separate the cow and calf soon after birth. This practice has been adopted for many reasons, including increasing the amount of saleable milk, reducing the risks to calf health, and preventing the formation of the maternal bond (thereby preventing future distress when they need to be separated); however, this separation soon after birth is viewed negatively by the public (Ventura et al., 2016; Hotzel et al., 2017; Meagher et al., 2019). As a

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https://doi.org/10.1016/j.animal.2025.101541
1751-7311/

result, there is an increased interest from consumers, farmers, and researchers for alternative calf–rearing methods, such as cow-calf contact (**CCC**) rearing systems, where the cow and calf can stay in contact for a prolonged period of time (Sirovnik et al., 2020). However, CCC systems come with challenges that need to be overcome for CCC to be viable on commercial farms (Neave et al., 2022), such as weaning and separation of the bonded cow-calf pair.

Weaning and separating a bonded cow-calf pair, especially if done together, has been shown to cause distress to both cow and calf (Johnsen et al., 2021b; Wenker et al., 2022a; Bertelsen and Jensen, 2023). This distress can result in changes in behaviour (e.g., increase in vocalisations, Stěhulová et al., 2008; Enríquez et al., 2010; Johnsen et al., 2015a; searching behaviour, Neave et al., 2024; pacing or standing with their head out of the pen, Wenker et al., 2022a, Bertelsen and Jensen, 2023) and performance (e.g., reduction in calf weight gain, Sinnott et al., 2024 and cow BW, Metz, 1987; Bar-Peled et al., 1995) for both cow and calf. Behaviour and performance are intrinsically linked with health, as cows and calves in poor health have altered behaviour (Dittrich et al., 2019; Belaid et al., 2020; Duthie et al., 2021) compared to healthy individuals. Although a review concluded that immediate separation after birth was not advantageous to calf health (Beaver et al., 2019), more recent research has found that full-time access CCC with weaning and separation around 8–9 weeks of age can lead to poorer calf health (i.e. increased use of antibiotics; Wenker et al., 2022b; Sinnott et al., 2024) and potentially lead to more difficulty transitioning onto their postweaning diet (Wenker et al., 2022b; Sinnott et al., 2024). Although clinical health was assessed in these cases, looking at physiological markers of health and performance in these animals may provide more information into sub-clinical health and physiological changes in performance that do not lead to alterations in growth and weight.

Blood serum biomarkers of inflammation (e.g., serum amyloid A, albumin, globulin, and total protein) may indicate underlying infections or inflammation that may not be obvious during clinical health scoring (Bobbo et al., 2017a; Trela et al., 2022). Both serum amyloid A and globulin are positive acute–phase proteins while albumin is a negative acute–phase protein. Therefore, if inflammation was present, we would expect these biomarkers to increase and decrease, respectively. Serum mineral concentrations, such as calcium, magnesium, and phosphorus, may identify underlying health issues (i.e., hypocalcaemia in recently calved cows) or differences in diet and intake (e.g., cow magnesium is highly dependent on diet and rumen uptake, as cows cannot store magnesium; Suttle, 2022; Khiaosa-Ard et al., 2023). Serum biomarkers of energy balance, such as beta-hydroxybutyrate (**BHB**) and non-esterified fatty acids (**NEFA**), in conjunction with changes in BW and body condition score, can help monitor energy requirements and feed intake of cows (LeBlanc, 2010; Benedet et al., 2019) and calves (Quigley III, 1996; Deelen et al., 2016; Steele et al., 2017). Negative energy balance in cows typically results in lower BW and body condition scores and higher levels of BHB and NEFA. In calves around weaning, BHB has been used as a proxy of rumen development (Deelen et al., 2016), as it increases with larger amounts of consumed concentrates, therefore increases after weaning. Calf NEFA typically decreases after weaning (Ferronato et al., 2022).

This explorative study aimed to measure the physiological health, performance, and behaviour of cows and calves within three systems (two CCC rearing systems and one conventional system) before and after weaning to estimate whether animals within the three investigated systems responded differently. More specifically, we wanted to see if there were any differences between the three systems in terms of cow and calf physiological (i.e., serum markers of inflammation and minerals) and clinical (i.e., clinical health scores and BW) health and performance (i.e., energy balance, weight gain, and cow production), as well as behaviour (i.e., proportion of time spent lying, ruminating, and eating solid feeds). This study was specifically focused on two time-points: immediately before weaning and separation (**preWS**) and immediately after the 7 d gradual weaning and separation process (**postWS**). We expected that CCC would slightly worsen cow and calf health and performance after the weaning and separation process, and that the behaviour of cows and calves in CCC systems would differ from conventionally kept cows and calves.

Material and methods

Animals, management, and experimental design

This study was conducted from 8 March to 31 May 2021 at Teagasc Moorepark Research Farm, County Cork, Ireland, as part of a larger research project investigating cow-calf contact systems and discussed in more detail in McPherson et al. (2024a) and Sinnott et al. (2024). The aim of the overall project was to investigate how CCC could feasibly be incorporated into an Irish spring-calving, pasture-based system and to determine its effects on cow and calf health, behaviour, production, and welfare. Three different dairy calf rearing systems were implemented and compared: the conventional, no contact system (**CONV**); a full-time access CCC system (**FT**); and a part-time access by night CCC system (**PT**). The CCC period lasted for 8 weeks, until the gradual weaning and separation process began. Before calving (January 2021), 54 cows were balanced between the three different calf rearing systems (18 cows/system) in a randomised complete block design by an individual independent to the study. Cows were balanced by (mean ± SD): breed (70% Holstein-Friesian and 30% Holstein-Friesian × Jersey crosses (> 25% Jersey)), parity (1.76 ± 0.46), previous 35-week cumulative milk yield (4 677 ± 1 047.4 kg; the dam's first lactation cumulative 35-week milk yield was used for primiparous cows), previous lactation average somatic cell score (log_{10} of somatic cell count; 4.9 ± 0.44; the dam's first lactation average somatic cell score was used for primiparous cows), expected calving date (11 February 2021 ± 16 d), Economic Breeding Index (Berry et al., 2005), and expected calf breed (breed of sire). Sample size calculations were completed based on findings from previous experimental studies, using cow daily machine milk yield and calf plasma immunoglobulin G levels from 24 h post-birth, and gave a group size of 18 cow-calf pairs per system. The multiparous cows had all previously been separated from their calves immediately after (≤ 2 h) birth; thus, CCC was novel to all experimental cows. Four pairs from the FT system had to be removed from trial soon after calving (one pair due to failure of cow to bond with calf; three pairs due to calf illness requiring intervention) and were not replaced due to lack of cow availability at the end of the calving season. One early calving PT pair was removed from the system due to calf illness, but was able to be replaced with a cow that had yet to calve. The final system groups used in this analysis were 18 CONV cow-calf pairs (13 female dairy calves and five male dairy-beef calves), 14 FT cow-calf pairs (10 female dairy calves, two female dairy-beef calves, and two male dairy-beef calves), and 18 PT cow-calf pairs (10 female dairy calves, one male dairy calf, two female dairy-beef calves, and five male dairy-beef calves). Dairy-beef calves refer to a calf with a dairy dam and a beef sire. Measurements were performed using each individual cow or calf as the experimental unit.

Management of different systems

All cows were milked using a mid-line, 30–unit side-by-side parlour (Dairymaster, Ireland). The automatic cluster removers had a milk flow rate cut-off point of 0.2 kg/min with a 3 s time delay. Unless the specific system required it (see below for more details), the standard farm milking times were 0700–0900 h (morning milking) and 1430–1630 h (afternoon milking). For a comprehensive overview of other management aspects of the three systems during the CCC period, please refer to McPherson et al. (2024a) and Sinnott et al. (2024).

Conventional system management

The CONV pairs were separated within 1 h postcalving. The CONV cows joined a grazing herd where they were managed following normal grazing practices at the Teagasc Moorepark facilities (i.e., Kennedy et al., 2021; Murphy et al., 2023; explained in more detail below). The CONV cows were offered a predominately grazed grass diet and were milked twice-a-day (0700 and 1500 h). Concentrates (1–4 kg/d) were provided during milking in the parlour, and the amount provided depended on grass availability (see below for more details).

The CONV calves were artificially reared following conventional Irish practices (Conneely et al., 2014). At 3 days old, calves were moved into a group pen where they were offered milk replacer (Heiferlac, Volac; 26% CP) through an automatic milk feeder (Volac Förster Technik Vario, Germany). Calves received 6–9 L/d of milk replacer, depending on their age; gradual weaning commenced at 48 d of age and lasted for 12 d (see Sinnott et al., 2024 for more details). Calves were gradually weaned by the automatic milk feeder gradually reducing the volume of milk replacer fed. In the group pen, calves were also offered *ad libitum* water (via a water bowl), forage (hay), and concentrates (18% CP, Kaf Gro, Prime Elite, Dairygold, Cork, Ireland).

Full-time access system management: preweaning

Until the start of the gradual weaning and separation process, the FT pairs had continuous, unrestricted access to each other (22 h/d), except during milking times (twice-a-day at 0800 and 1600 h). Calves always had access to *ad libitum* water (water trough), concentrates, and forage (indoor forage was hay; outdoor forage was grass). The FT pairs were primarily kept outdoors at pasture, but depending on weather or grass availability (see McPherson et al., 2024a), they were occasionally housed indoors in a housing identical and adjacent to the PT pairs (described below). When indoors, the FT pairs were fed identically to the PT pairs. The FT cows were offered concentrates (1–4 kg/d) in the milking parlour.

Part-time access system management: preweaning

During the CCC period, PT pairs had unrestricted access to each other by night (17 h/d; 1500 to 0800 h). During the nightly contact period, the PT cows were housed in an indoor cubicle area, which was connected to an adjacent straw pen (PT calf pen) via a creep with gates. The PT pairs were separated in the morning (0800 h), when the PT cows were brought to the parlour for milking; the PT calves remained in their straw pen, where they were provided with *ad libitum* access to water (via a water bowl), forage (hay), and concentrates. The PT cows were turned out to pasture after the morning milking and remained there until the afternoon milking time (1500 h), when they were not milked and were brought back indoors to be reunited with their calves. In the cubicle area, PT cows had access to *ad libitum* grass silage and water via water troughs. The PT cows were fed concentrates (1–4 kg/d, dependent on weather and grass availability) in the parlour.

Weaning management: conventional system

The CONV pairs were separated at birth; therefore, they did not undergo the same weaning and separation process as the FT and PT pairs. Instead, CONV cows remained at grass under normal management conditions. The CONV calves were weaned gradually by the automatic feeder over the course of 21 days, by gradually reducing the calves' allotted volume of milk replacer each day (see Sinnott et al., 2024 for more details). After weaning, CONV calves were moved and regrouped in an indoor straw-bedded pen with calves from all systems.

Weaning, separation, and postweaning management: cow-calf contact pairs

The FT and PT cow-calf pairs were weaned based on calf age (57 ± 1.9 d) over a 7–day period using a gradual, three-stage process which occurred indoors in a novel shed. The weaning process was initiated after the morning milking. Cows went to the parlour for milking as normal, but during milking, the calves were moved into a different, separate shed (Supplementary Fig. S1). After morning milking, cows were brought back to the shed into which the calves had been moved. In the weaning shed, the pairs were housed in adjacent, straw bedded pens, with gates that prevented suckling but pairs could still hear, see, and touch each other through the gaps in the gates. The FT and PT cows and calves were kept separately (Supplementary Fig. S1). All pens had access to *ad libitum* water through water bowls. In their respective pens, calves had access to *ad libitum* concentrates, grass silage, and forage (hay), while cows had access to *ad libitum* grass silage. To maintain consistency during the weaning and separation period, all FT and PT cows were fed 3 kg/d of concentrates in the parlour. Single FT and PT pairs did not undergo the weaning and separation process by themselves; a minimum of two pairs were weaned together to prevent unnecessary distress.

In the first stage of weaning and separation (days 1 to 3; start of weaning), cows and calves were allowed 1 h of unrestricted access to each other around 1 h after morning milking (approximately from 1030 to 1130 h) where calves could suckle freely. This 1 h period was chosen to be shortly after the morning milking so that the cows would have some milk for their calf, but would not have enough milk that the calf would be satiated. During the second stage (days 4 to 5; calves were weaned), the pairs were not allowed access to one another; however, pairs were still able to see, hear, and touch each other through the gates separating the pens. At the start of the third stage (days 6 to 7; start of separation), calves remained in the weaning pen while the FT and PT cows did not return to the shed. Instead, the FT and PT cows joined a general herd of cows at grass, which were managed identically to the CONV cows. The FT and PT cows remained with this herd for the rest of their lactation and had no further contact with any calves. After the end of the weaning and separation process, all calves, regardless of system, were grouped in an indoor straw–bedded pen (40 m^2; 2 m^2/calf) for 8 ± 1.7 d until they were moved to an outdoor paddock (at 71 ± 4.5 d old). Calves could not see mature cows in the indoor straw–bedded pen, but may have been able to see mature cows when in the outdoor paddock.

Measurements

Physiological markers of health and performance

To be able to assess physiological markers, blood samples were taken from the cows and calves the week before weaning (preWS) and the week after weaning (postWS; Table 1; see below for more information on the date selection). Two tubes were collected at each time point: one 10–mL serum tube (BD Vacutainer Serum tube, no additive, silicone-coated interior) and one 10–mL plasma tube (BD Vacutainer Plasma tube, 158 USP units of sodium heparin (spray coated)). Cow and calf blood samples were taken using a 20G needle (BD Vacutainer PrecisionGlide Multiple Sample Blood Collection Needle – 20G × 1″ (0.9 × 25 mm)). Jugular venepuncture was used to obtain the calf blood samples, while cow blood samples were taken by coccygeal venepuncture. After blood sample

Table 1

Days relative to the start of the weaning and separation process that the two time-point samples (preWS = before weaning and separation; postWS = after weaning and separation process) were taken for each of the three systems (conventional (CONV; n = 18): no access to calf/dam, cow milked twice-a-day and kept at pasture, calf kept indoors; full-time access (FT; n = 14): full-time access to dam/calf outdoors at pasture and cow milked twice-a-day; part-time access (PT; n = 18): part-time access to dam/calf by night, calf kept indoors, and cow milked once-a-day) for the different measurements taken during the study.

Item		Cows			Calves		
		CONV	FT	PT	CONV	FT	PT
Number of animals		18	14	18	18	14	18
Days in milk/ weaning age		56 ± 0.0	57 ± 2.4	57 ± 1.6	57 ± 1.7	57 ± 2.4	57 ± 1.6
Days relative to the start of the weaning and separation process							
Behaviour							
	preWS	−7 ± 0.0	−5 ± 2.1	−6 ± 1.9	−4 ± 1.9	−4 ± 1.7	−6 ± 1.9
	postWS	7 ± 0.0	12 ± 2.3	8 ± 1.9	11 ± 1.9	11 ± 1.8	8 ± 1.9
Blood samples							
	preWS	−5 ± 1.7	−5 ± 3.4	−7 ± 2.1	−6 ± 2.1	−6 ± 2.0	−5 ± 1.4
	postWS	8 ± 0.0	12 ± 1.8	10 ± 2.3	11 ± 2.3	11 ± 4.1	11 ± 1.9
BW/body condition score							
	preWS	−5 ± 2.6	−3 ± 2.4	−5 ± 2.5	−6 ± 1.9	−6 ± 1.7	−5 ± 1.4
	postWS	15 ± 2.2	12 ± 3.5	14 ± 2.2	11 ± 2.3	11 ± 4.1	11 ± 1.9

Abbreviations: preWS = the week before the 7−day weaning and separation process; postWS = the week after the 7−d weaning and separation process.

collection, plasma samples were immediately centrifuged at 3 000 rpm for 15 min at 4 °C, the plasma decanted and frozen in duplicate at −20 °C. Serum samples were refrigerated immediately after collection for 24 h to allow for the blood to clot and then centrifuged at 3 000 rpm for 15 min at 4 °C, after which the serum was decanted and frozen in duplicate at −20 °C.

One duplicate of each of the serum samples was sent to a commercial laboratory for analysis (FarmLab Diagnostics, Emlagh, Elphin, Co. Roscommon, Ireland), where serum levels of albumin (g/L; Prestige 24i LQ Albumin, catalogue number: 4–238; PZ Cormay S.A., Warsaw, Poland), BHB (mmol/L; β-hydroxybutyrate (β-Hb) Liquid Reagent, catalogue number: GLC24019; GlenBio Ltd., Antrim, United Kingdom), calcium (mmol/L; Prestige 24i Calcium Arsenazo, catalogue number: 4–247; PZ Cormay S.A., Warsaw, Poland), cortisol (μg/mL; Veterinary Cortisol Biolis Liquid Reagent, catalogue number: GLC24851; GlenBio Ltd., Antrim, United Kingdom), globulin (g/L; calculated from total protein – albumin), magnesium (mmol/L; Prestige 24i LQ MG, catalogue number: 4–229; PZ Cormay S.A., Warsaw, Poland), NEFA (mmol/L; NEFA Biolis Liquid Reagent, catalogue number: GLC24083; GlenBio Ltd., Antrim, United Kingdom), phosphorus (mmol/L; Prestige 24i LQ Phosphorus, catalogue number: 4–243; PZ Cormay S.A., Warsaw, Poland), and total protein (g/L; Prestige 24i LQ Total Protein, catalogue number: 4–236; PZ Cormay S.A., Warsaw, Poland) were determined using a BIOLIS 30i instrument. The samples were analysed according to Farmlab Diagnostic's standard operating procedures and the manufacturer's specifications; there were no deviations from these procedures. The other serum duplicate was analysed for serum amyloid A (g/mL) using commercially available bovine serum amyloid A ELISA kits (Cow Serum Amyloid A ELISA, catalogue number: SAA-11; Life Diagnostics, Inc., West Chester, PA, USA) in the cow and calf serum samples, at the Teagasc lab. For these kits, the serum had to first be diluted. Cow and calf serum samples were tested at different dilutions: 400x and 600x for cows and 400x, 600x, and 1200x for calves. After testing the dilutions, all cows were tested at 400x and the majority of calves were tested at 600x; some calves were tested at 1200x if their concentration was too high at 600x (5 calves). All tests were conducted according to the manufacturer's instructions.

Clinical health scoring

Clinical health scoring was performed on cows and calves twice a week (Tuesdays and Fridays), starting from when cows and calves had entered their respective system herds, and ended once calves were 12 weeks of age and cows were in their 12th week of lactation. Ten aspects of calf health (demeanour, ocular discharge, ear position, nasal discharge, cough, dehydration, mobility, interest in surroundings, faecal hygiene, and naval score) and nine aspects of cow health (demeanour, ocular discharge, ear position, nasal discharge, cough, dehydration, mobility, interest in surroundings, faecal hygiene) were scored using the calf health scoring method from Barry et al. (2019) which had previously been used in cows by Crossley et al. (2021). Each aspect of health was scored on a scale, most from 0 to 3, based on severity (the clinical health scoring scale can be found in Supplementary Table S1). Health scoring was performed by two observers (inter-observer reliability: 89% agreement first attempt, 97% agreement second attempt). A summed clinical health score was calculated by summing all aspects of clinical health (10 for calves and nine for cows), where a higher score indicated a less healthy animal.

Somatic cell score

Milk samples were obtained weekly (one composite sample from a consecutive afternoon and morning milking) for each cow over their entire lactation. During each milking, samples were obtained using collection units that collected a small amount of milk continuously throughout the entire milking event. As the PT cows were not milked in the afternoon, their milk was only sampled during the morning milking collection period. The weekly milk sample was analysed for somatic cell count (Milkoscan FT6000, Foss Electric DK). The weekly somatic cell count was converted to somatic cell score by taking the $\log_{10}$ of the somatic cell count.

Body weight, condition, and average daily gain

Cow BW (kg) and body condition score (5−point scale with 0.25 increments, where 1 = emaciated and 5 = extremely fat; Edmonson et al., 1989) were recorded weekly for the first 12 weeks of the lactation. Following morning milking, cows would enter a crush that had an electronic, portable weigh-scale with Winweigh software package (Tru-test Limited) located at the end. Body condition score was assessed by a single observer (intra-observer reliability; weighted kappa = 0.9589) when the cow was on the scales (a body condition score from 1 = emaciated to 5 = extremely fat, with 0.25 increments was used; Edmonson et al., 1989).

Calf BW (kg) was recorded once a week using a weighing scale (TrueTest XR 3000, Tru-test Limited, Auckland, New Zealand). Weekly average daily gain (**ADGw**; kg/d) was calculated by subtracting the previous week's weight from the current week's weight, then dividing by the number of days between the weight measurements. Average daily gain from birth (**ADGb**; kg/d) was calculated by subtracting the birthweight from the current week's weight, then dividing by the calf's age in days.

Scan sampling behaviour scoring

Live behaviour scoring was performed on cows and calves in all systems 1 day per week (Monday) by two independent observers for 11 weeks (inter-observer reliability: first attempt = 85% agreement, second attempt = 98%), starting once they had entered their respective systems. Cows and calves were scored during three different time periods in a day: before the morning milking, when cows had not yet been moved to the milking parlour (range from 0600 to 0800 h); midday (range from 1030 to 1230 h), after cows had settled following the morning milking (while the PT pairs were separated); and, after the afternoon milking (range from 1600 to 1800 h). The 2 h window for each scoring time period was due to the number of system groups, which were often located in different sheds and paddocks across the farm. At each time period during the day, behaviour was scored for 16 min using scan sampling at a 2-min intervals (a total of eight observations were recorded for each animal during each time period). All cows and calves present (system dependent, i.e. all FT cows and calves were observed at the same time, while the CONV cows and calves never were) were scored at the same time. The different system groups were typically scored in the same order and by the same observer each week. The ethogram used to score behaviour can be found in Table 2. During scan sampling, the cow or calf's 'posture' (standing or lying down) was always recorded (24 total observations per day), while activity behaviours (i.e., ruminating, walking, or playing) were recorded only when observed (if the animal was idle or sleeping, no activity behaviour was recorded during that observation).

Data processing

Pre- and postweaning date selection

Since we did not allow individual cow-calf pairs to undergo the weaning and separation process by themselves (discussed in more

Table 2
Ethogram describing the different behaviours scored using live, in-person scan sampling. Cows and calves were behaviour scan sampled (2 min between each scan) during three sessions each day (15 min per session; before morning milking, midday, and after afternoon milking), 1 day per week. Behaviours are separated into two different categories: posture (standing and lying) and activity behaviours (i.e., ruminating or playing). Posture was always recorded, while activity behaviours were only recorded if observed during the scan. Activity behaviours with low prevalence were combined into behaviour categories during the analysis process. Behaviours that were specific to cows or calves are also noted.

Category	Behaviour	Animal	Description of behaviour
Posture			
Lying	Lying	Cow and calf	Animal is resting either sternally or laterally with all four legs hunched close to body either awake or asleep.
Standing	Standing	Cow and calf	Animal is in a static upright standing position with weight placed on all four legs
Activity behaviours			
Abnormal	Head out of pen	Cow and calf	Animal puts the tip of its nose/ head through openings in a fence or over the top of the fence. Animal puts its head through or over a gate/fence.
	Oral Manipulation of Pen Structure	Cow and calf	Animal licks, nibbles, sucks, or bites at the pen structure (barriers, walls, buckets, troughs etc.)
	Pacing	Cow and calf	Animal repeatedly walks back and forth the same area (i.e. in front of a gate) in an active manner
	Tongue Rolling	Cow and calf	Animal makes repeated movements with its tongue inside or outside its mouth
	Urine drinking / oral manipulate prepuce / cross sucking	Calf only	Animal drinks the urine of another calf / Animal attempts to suck the naval area of another animal / Animal attempts to suck any body part of another animal.
	Vocalisation	Cow and calf	Bellowing from animal
Drink milk	Calf nursing/ calf suckling	Cow / calf	Cow is allowing her calf or another calf to nurse from her udder. / Calf is suckling from her dam or another cow.
	Drinking Milk	Calf only	Calf is standing in the automatic milk feeder drinking milk, with their mouth on the nipple.
Drink water	Drinking Water	Cow and calf	Animal is stood at the water bowl or trough with their nose/mouth partially submerged in the water.
Eat solids	Eating concentrate	Calf only	Calf is standing with their head within the flaps of the concentrate feeder. / Calf is standing with their head just outside the concentrate feeder while making lateral motions of the jaw
	Eating forage	Calf only	Calf makes a lateral motion of the jaw while standing at the hay feeder. Calf is lying down on the straw and is making repeated lateral motions of the jaw with straw sticking out of their mouth. / Calf is lying down on the straw and is rooting their nose around in the bedding. / Calf is standing with their head near the ground making lateral motions of the jaw (inside on straw bed only).
	Eating grass silage	Cow and calf	Animal is stood at the feed bunk making a lateral motion of the jaw.
	Grazing	Cow and calf	Animal is standing with their head near the ground making lateral motions of the jaw. Active eating of grass is observed.
Maintenance	Defecation/ Urination	Cow and calf	Animal defecates or urinates
	Grooming	Cow and calf	Animal uses tongue to repeatedly lick own back, side, leg, tail areas
	Scratching / Rubbing / Stretching	Cow and calf	Animal scratches itself with one of their legs (generally hind legs)/ Animal rubs itself on pen structure/ Animal stretches itself
Positive	Play Behaviour	Cow and calf	Animal runs, jumps, changes direction suddenly, bucks, kicks hind legs, twists or rotates body / Animal mounts, or attempts to mount, a pen mate (calf only)/ Animal is engaged in head to head pushing with another animal (calf only) / Animal plays with an object in the pen or field.
	Social Interaction	Cow and calf	Animal licks another animal in the same area multiple times / Animal nudges another animal with its nose
Ruminating	Rumination	Cow and calf	Ruminating / chewing
Walking	Walking	Cow and calf	Animal is actively moving from one point in the pen to another in an active walking motion
Other	Other	Cow and calf	Cow or calf performs any other activity not mentioned

detail above), there was some variation between systems in the day of lactation/age that cows and calves were weaned (Table 1). Measurements were also only taken on specific days of the week. Therefore, to enable the comparison of before and after weaning, we selected the last measurement that was taken before weaning and separation for the before sample (preWS) and the next measurement that was taken after the 7-d weaning and separation process (postWS) as the after measurement. Each variable then had two observations: one preWS and one postWS. The average number of days the preWS and postWS measurements were taken before and after the start of the weaning and separation process is presented in Table 1. Due to their gradual weaning process, the CONV calves' preWS samples were collected during their weaning process.

Scan sampling behaviour proportions

Behaviour proportions were calculated independently for each cow and calf on each day of observation (preWS and postWS) and were calculated by summing the number of scans (occurrences) where that behaviour was observed and dividing by the total number of scans per day (24). Some behaviours were observed too rarely for analysis and hence were combined into behavioural categories (more details can be found in Table 2). As posture (standing or lying) was always recorded, only the results for lying are reported. All animals were retained in the analysis. Although proportions were used in the analysis, for ease of understanding, all proportions were converted to percentages for the results section.

Statistical analysis

All data were analysed using SAS (v9.4, SAS Institute). All data (other than behaviour proportion data) were first checked for normality using the Shapiro-Wilk test (PROC UNIVARIATE), where data were considered approximately normal if the W-statistic was > 0.85; the shape of the histogram was also checked. Mean, variance, SEM, and coefficients of variation for all measured study variables can be found in Supplementary Table S2. The Tukey-Kramer test was used for all posthoc pairwise comparison tests. The threshold for significance was $P < 0.05$, and tendencies were $P < 0.10$.

All physiological (cortisol, albumin, globulin, serum amyloid A, total protein, calcium, magnesium, phosphorus, BHB, NEFA) and clinical (summed clinical health scores, somatic cell score, BW, body condition score, ADGw, ADGb) markers of health and performance were analysed using linear mixed models in SAS (PROC MIXED). Cow and calf variables were analysed separately. For the cows, the model consisted of the fixed effects of system (CONV, FT, and PT), time-point (preWS or postWS), and parity (1, 2, or 3+). Although the cows were blocked for parity, the FT cows that were removed from trial were disproportionately of parity 3+ (see above and McPherson et al., 2024a for more details); when model fit was tested with and without parity, the Bayesian Information Criterion was lower with parity included as a fixed effect. For calves, the model consisted of the fixed effects of system (CONV, FT, and PT), time-point (preWS or postWS), calf breed (dairy or dairy-beef), and calf sex (male or female). The interaction between system and time-point was tested in all cow and calf models, and removed if not significant ($P \geq 0.05$). Time-point was used as a repeated measure, acting on the individual cow or calf, and several covariance structures were tested (compound symmetry, first-order autoregressive lag 1, and unstructured); whichever covariance structure gave the lowest Bayesian Information Criterion was used (only compound symmetry and first-order autoregressive lag 1 were used; for full details on which variance components were used in each model, please refer to Supplementary Table S3). Various covariates were used in the models, dependent on the variable being tested. In all analyses, each cow's days in milk or calf age on 1 June 2021 was used as a covariate to account for differences in calving date. Additional cow covariates included the health sub-index of their Economic Breeding Index (centred within parity) for all health variables (physiological and clinical) and cow BW and body condition score from the first week of their lactation for BW and body condition score, respectively. For the health and performance variables, model-generated least square means and SEM are presented. Although the fixed effects of parity (cows) and calf sex and breed (calves) were included in the models, we did not report them as we did not set out to estimate their effects. However, we have provided results for calf breed and calf sex in Supplementary Table S4.

The analysis of cow and calf behaviour was performed separately. Analysis of each behaviour or behaviour category (Table 2) was done in SAS using generalised linear mixed models (PROC GLIMMIX) with a binomial distribution, a logit link function, and the Kenwood-Rogers method of determining df. For the cows, the model consisted of the fixed effects of system (CONV, FT, or PT), time-point (preWS or postWS), and parity (1, 2, or 3+). Each cow's days in milk on 1 June 2021 was used as a covariate to account for differences in calving date. For the calves, the model consisted of the fixed effects of system (CONV, FT, or PT), time-point (preWS or postWS), calf breed (dairy or dairy-beef), and sex (male or female). Each calf's age on 1 June 2021 was used as a covariate to account for differences in birth date throughout the spring. For both cow and calf models, the interaction between system and time-point was tested for each behaviour or behaviour category, and removed from the model if it was not significant ($P > 0.05$). The random effects for both cow and calf models were the individual animal (cow or calf) and the residual. Due to the nature of the logit function used in the behaviour models, only raw statistical means (± SD) are reported throughout for behaviour data.

Results

Cow physiological and clinical markers of health and performance

All estimates and test statistics for cow physiological and clinical markers of health and performance are presented in Table 3. The interaction between system and time-point was not significant for phosphorus, BHB, or NEFA, so only their individual effects will be presented below. There was no effect of system or time-point on the cow summed clinical health score, cortisol, serum amyloid A, or BHB.

During preWS, the FT cows had higher albumin than the PT cows, while CONV were similar to both; postWS all systems were similar. The FT cows had higher globulin preWS compared to postWS, while the CONV and PT cows had similar values preWS and postWS; there were no differences between the systems during preWS and during postWS. Despite the significant interaction between system and time-point for total protein, no posthoc comparisons differed. Serum calcium was higher in the FT cows preWS compared to postWS, while the CONV and PT cows had similar calcium preWS compared to postWS. The FT cows also had higher calcium than the PT cows preWS, while the CONV cows were similar to both. For magnesium, there was no difference preWS compared to postWS for the CONV and PT cows, while the FT cows had higher magnesium preWS compared to postWS. During preWS, all systems had similar magnesium, but postWS the CONV cows had higher magnesium than the PT cows, while the FT cows were similar to both. Phosphorus was affected by the system but not time point. The CONV cows had lower phosphorus (1.31 ± 0.064 mmol/L) compared to FT (1.56 mmol/L; $P = 0.024$) and PT (1.63 mmol/L;

Table 3
Estimates and SEM for physiological markers of health and performance for cows in three different calf rearing systems (conventional (CONV; n = 18): no access to calf and milked twice-a-day; full-time access (FT; n = 14): full-time access to calf and milked twice-a-day; part-time access (PT; n = 18): part-time access to calf and milked once-a-day) at two time-points (preWS: before weaning and separation; postWS: after weaning and separation). The interaction between system and time-point was tested in all models, but was removed if $P \geq 0.05$. 'NS' denotes a non-significant interaction. [†]Denotes that no comparisons were significant in the posthoc comparison test.

	preWS			postWS				*P*-value		
Variable	CONV	FT	PT	CONV	FT	PT	SEM	System	Time-point	System * Time-point
Clinical health score	1.2	1.0	0.8	1.4	1.2	1.1	0.14	0.065	0.052	NS
Cortisol, μg/mL	1.10	1.82	0.96	1.41	1.30	1.61	0.302	0.693	0.324	NS
Albumin, g/L	33.4[ab]	37.6[a]	27.9[b]	38.6[a]	34.1[ab]	34.5[ab]	1.85	0.032	0.063	**0.018**
Globulin, g/L	23.9[ab]	27.0[a]	23.6[ab]	23.7[ab]	19.7[b]	19.0[b]	1.60	0.374	<0.001	**0.026**
Log serum amyloid A	1.70	1.37	1.31	1.31	1.40	0.93	0.192	0.627	0.398	NS
Total protein, g/L	57.4	64.6	51.4	60.9	53.3	53.9	3.12	0.087	0.464	**0.033**[†]
Calcium, mmol/L	2.34[ab]	2.80[a]	2.14[b]	2.44[ab]	2.19[b]	2.24[ab]	0.128	0.089	0.199	**0.012**
Magnesium, mmol/L	0.98[abc]	1.13[a]	0.90[abc]	1.01[ab]	0.82[bc]	0.81[c]	0.053	0.037	0.004	**0.006**
Phosphorus, mmol/L	1.33[y]	1.65[x]	1.70[x]	1.29[y]	1.48[x]	1.56[x]	0.089	**0.002**	0.116	NS
BHB, mmol/L	0.35	0.44	0.43	0.35	0.33	0.44	0.040	0.126	0.350	NS
NEFA, mmol/L	0.44[xy h]	0.54[x h]	0.26[y h]	0.51[xy g]	0.61[x g]	0.54[y g]	0.056	**0.013**	**0.003**	NS

Abbreviations: preWS = the week before the 7–day weaning and separation process; postWS = the week after the 7–d weaning and separation process; BHB = beta-hydroxybutyrate; NEFA = Non-esterified fatty acids; NS = not significant.
[†] No comparisons were significant in the posthoc comparison
[a,b,c] Values within a row with different superscripts differ significantly at $P < 0.05$ for the interaction model.
[x,y] Values within a row with different superscripts differ significantly ($P < 0.05$) for system (no interaction in model).
[g,h] Values within a row with different superscripts differ significantly ($P < 0.05$) for time-point (no interaction in model).

P = 0.002) cows (FT and PT cows, *P* = 0.740). Serum NEFA was affected by both system and time point. The FT cows (0.58 ± 0.038 mmol/L) had higher (*P* = 0.009) NEFA than the PT cows (0.40 mmol/L), while the CONV cows (0.48 mmol/L) were similar to both. All cows had lower NEFA preWS (0.41 ± 0.032 mmol/L) compared to postWS (0.56 mmol/L; *P* = 0.003).

Calf physiological and clinical markers of health and performance

All estimates and test statistics for calf clinical and physiological markers of health and performance are presented in Table 4. The only variable with a significant interaction between system and time-point was NEFA; therefore, for the rest of the variables in this section, we will only present the fixed effects of system and time point.

There was no effect of system or time point on cortisol, albumin, total protein, and calcium. Calf health scores were higher postWS compared to preWS, but there was no effect of system. Globulin was not affected by system but was affected by time-point; calves had lower globulin levels preWS compared to postWS. Calf serum amyloid A was affected by time-point but not system. Calves had higher serum amyloid A preWS compared to postWS. Magnesium was affected by system and time point. The CONV calves had higher magnesium levels than PT calves, while the FT calves were similar to both. All calves had lower levels of magnesium preWS compared to postWS. Phosphorus was not affected by system but was affected by time-point; calves had lower levels of phosphorus preWS compared to postWS. Calf serum BHB was affected by time-point but not system; preWS BHB was lower than postWS. Calf NEFA was similar between the systems preWS (0.24, 0.36, and 0.32 ± 0.037 mmol/L for the CONV, FT, and PT calves, respectively); postWS the CONV calves had lower NEFA than the FT and PT calves (0.13, 0.43, and 0.34 ± 0.037 mmol/L for the CONV, FT, and PT calves, respectively). Within each system, NEFA did not differ between preWS and postWS.

Cow and calf performance variables

All estimates and test statistics for cow and calf performance variables are presented in Table 5. The interaction between system

Table 4
Estimates and SEM for physiological markers of health and performance for calves in three different calf rearing systems (conventional (CONV; n = 18): no access to dam and kept indoors; full-time access (FT; n = 14): full-time access to dam and kept outdoors at pasture; part-time access (PT; n = 18): part-time access to dam and kept indoors) at two time-points (preWS: before weaning and separation; postWS: after weaning and separation). The interaction between system and time-point was tested in all models, but was removed if $P \geq 0.05$. The NEFA model was the only calf health-related variable to contain the interaction; therefore, although the least-squares means values for the individual effects of system and time-point are presented here, they were not statistically analysed, so no *P*-values are presented.

	System					Time-point			
Variable	CONV	FT	PT	SEM	*P*-value	preWS	postWS	SEM	*P*-value
Clinical health score	1.0	1.0	1.2	0.16	0.572	0.9[b]	1.3[a]	0.13	0.017
Cortisol, μg/mL	0.75	0.78	0.61	0.130	0.547	0.61	0.82	0.104	0.056
Albumin, g/L	41.7	42.9	43.1	1.93	0.623	41.6	43.5	1.55	0.233
Globulin, g/L	14.9	13.0	11.9	1.18	0.127	12.1[b]	14.4[a]	0.92	**0.010**
Log serum amyloid A	1.01	1.17	1.12	0.089	0.363	1.29[a]	0.91[b]	0.070	**<0.001**
Total protein, g/L	56.6	56.3	55.1	2.81	0.896	53.7	58.3	2.30	0.061
Calcium, mmol/L	3.04	3.05	3.02	0.146	0.989	3.11	2.97	0.120	0.389
Magnesium, mmol/L	0.87[a]	0.75[ab]	0.73[b]	0.039	**0.012**	0.74[b]	0.83[a]	0.031	**0.016**
Phosphorus, mmol/L	2.55	2.63	2.53	0.123	0.794	2.45[b]	2.70[a]	0.099	**0.024**
BHB, mmol/L	0.21	0.25	0.24	0.027	0.451	0.07[b]	0.39[a]	0.023	**<0.001**
NEFA[*], mmol/L	0.18	0.39	0.33	0.028	–	0.30	0.30	0.024	–

Abbreviations: preWS = the week before the 7–day weaning and separation process; postWS = the week after the 7–d weaning and separation process; BHB = beta-hydroxybutyrate; NEFA = Non-esterified fatty acids.
[a,b] Values within a row with different superscripts differ significantly at $P < 0.05$.
[*] NEFA was analysed with the interaction of system and time-point; therefore, the *P*-values are not presented here.

Table 5
Estimates and SEM for performance variables for cows and calves in three different calf rearing systems (conventional (CONV; n = 18): no access to dam/calf and cow milked twice-a-day; full-time access (FT; n = 14): full-time access to calf/dam, cow milked twice-a-day, and calf kept outdoors; part-time access (PT; n = 18): part-time access to calf/dam, cow milked once-a-day, and calf kept indoors) at two time-points (preWS: before weaning and separation; postWS: after weaning and separation). The interaction between system and time-point was tested in all models, but was removed if $P \geq 0.05$.

Item	preWS			postWS			SEM	*P*-values		
	CONV	FT	PT	CONV	FT	PT		System	Time-point	System * Time-point
Cow performance variables										
Somatic cell score	4.65	4.37	4.98	4.43	4.55	4.68	0.200	0.292	0.184	NS
BW (kg)	476^{c}	486^{bc}	520^{a}	494^{abc}	477^{bc}	508^{ab}	7.3	0.001	0.819	**0.013**
Body condition score	2.95	3.02	3.17	2.95	3.09	3.19	0.042	**<0.001**	0.247	NS
Calf performance variables										
BW (kg)	68.1	78.8	80.2	80.1	90.0	89.7	2.24	**<0.001**	**<0.001**	NS
ADGb (kg/d)	0.61^{c}	0.83^{a}	0.84^{ab}	0.64^{c}	0.79^{ab}	0.78^{b}	0.035	<0.001	0.005	**<0.001**
ADGw (kg/d)	0.95^{ab}	1.01^{a}	0.94^{ab}	1.04^{a}	0.42^{bc}	0.40^{c}	0.142	0.043	0.002	**0.016**

Abbreviations: preWS = the week before the 7–day weaning and separation process; postWS = the week after the 7–d weaning and separation process; NS = not significant; ADGb = average daily gain from birth; ADGw = weekly average daily gain.
[a,b,c] Values within a row with different superscripts differ significantly at $P < 0.05$.

and time-point was not significant for somatic cell score, and neither were their fixed effects. There was an interaction between system and time point on cow BW. The PT cows were heavier than the CONV and FT cows preWS, but there was no difference between the systems postWS. Within each system, cow BW preWS and postWS did not differ. There was no interaction between system and time-point on cow body condition score or effect of time-point, but there was an effect of system. The PT cows (3.18 ± 0.036) were in a higher condition than the CONV (2.95; $P < 0.001$) while the FT cows (3.05) were similar to both (FT and PT, $P = 0.052$; CONV and FT, $P = 0.126$).

There was no interaction between system and time-point on calf BW (kg), but there were individual fixed effects of system and time-point. The CONV calves (74.1 ± 2.19 kg) weighed less than the FT (84.4 kg; $P = 0.003$) and PT (85.0 kg; $P = 0.001$) calves (FT vs PT, $P = 0.979$). Calves weighed more postWS (86.6 ± 1.51 kg) compared to preWS (75.7 kg; $P < 0.001$). There was an interaction between system and time point for ADGw. There was no difference between the systems preWS, but postWS, the CONV calves had higher ADGw than the FT and PT calves. The FT and PT calves had higher ADGw preWS compared to postWS, while the CONV calves were similar between the time-points. There was also an interaction between system and time point for ADGb. Although no system differed between the time-points, the FT and PT calves had similar and higher ADGb than the CONV, both preWS and postWS.

Cow and calf behaviour

The average prevalence (± SD) of each behaviour or behaviour category for each system, preWS and postWS can be found in Table 6 for both cows and calves.

There was an interaction between system and time-point for the proportion of time cows spent ruminating; the PT cows spent more time ruminating preWS compared to postWS, while the CONV and FT cows did not differ between the time-points. There

Table 6
Average prevalence (%) of cow and calf behaviours (mean ± SD) from live scan sampling observations before (preWS) and after weaning and separation (postWS) for cows and calves within three different calf rearing systems (conventional (CONV; n = 18): no access to calf/dam, cow milked twice-a-day and kept at pasture, calf kept indoors; full-time access (FT; n = 14): full-time access to dam/calf outdoors at pasture and cow milked twice-a-day; part-time access (PT; n = 18): part-time access to dam/calf by night, calf kept indoors, and cow milked once-a-day). The interaction between system and time-point was tested in all models, but was removed if $P \geq 0.05$. Behaviour categories with low prevalence were not statistically analysed, but were included in the table as raw results to add to the current knowledge base. †Denotes that no comparisons were significant in posthoc comparison tests.

Item	preWS			postWS			*P*-value		
	CONV	FT	PT	CONV	FT	PT	System	Time-point	System * Time-point
Cow behaviours									
Lying	27 ± 19.2	23 ± 20.2	16 ± 16.5	31 ± 20.4	9 ± 16.7	14 ± 19.0	**0.017**	0.354	NS
Ruminate	20 ± 21.7^{ab}	33 ± 29.7^{a}	37 ± 18.6^{a}	23 ± 17.4^{ab}	19 ± 15.6^{ab}	11 ± 16.2^{b}	0.660	0.003	**0.008**
Eat solids	44 ± 15.7	54 ± 35.0	52 ± 19.8	42 ± 18.9	49 ± 15.9	58 ± 24.8	0.088	0.954	NS
Abnormal	0 ± 1.0	0 ± 0.0	0.7 ± 2.1	0 ± 2.0	2 ± 4.5	1 ± 2.1	–	–	–
Positive	0 ± 0.0	1.5 ± 4.5	0.7 ± 2.1	1 ± 3.1	0 ± 0.0	0 ± 0.0	–	–	–
Maintenance	1 ± 1.9	2 ± 4.2	1 ± 1.9	2 ± 2.5	1 ± 2.1	1 ± 1.8	–	–	–
Calf behaviours									
Lying	51 ± 25.3	71 ± 21.4	66 ± 29.4	65 ± 30.7	62 ± 28.6	44 ± 21.8	0.348	0.301	**0.023**†
Rumination	29 ± 20.9	35 ± 36.6	11 ± 16.3	36 ± 22.8	32 ± 14.5	30 ± 20.3	0.052	0.051	NS
Eating solids	9 ± 9.1^{bc}	9 ± 11.5^{bc}	0 ± 1.3^{c}	13 ± 14.8^{b}	17 ± 15.2^{ab}	33 ± 14.7^{a}	0.200	<0.001	**0.002**
Maintenance	7 ± 8.1	5 ± 5.9	10 ± 10.4	5 ± 8.1	5 ± 5.7	6 ± 6.1	0.422	0.055	NS
Abnormal	6 ± 6.9	1 ± 2.2	7 ± 13.0	4 ± 6.5	5 ± 6.3	3 ± 5.9	0.294	0.440	**0.034**†
Positive	3 ± 3.8	5 ± 6.8	6 ± 7.7	2 ± 3.3	2 ± 3.5	3 ± 3.5	0.198	**0.009**	NS
Drinking milk	0 ± 0.0	2 ± 8.9	1.9 ± 7.9	0 ± 0.0	0 ± 0.0	0 ± 0.0	–	–	–
Drinking water	1 ± 2.7	0 ± 0.0	0 ± 1.3	1 ± 2.9	0 ± 1.1	1 ± 3.5	–	–	–
Walking	0 ± 1.0	1 ± 1.5	2 ± 3.2	1 ± 2.9	0 ± 0.0	0 ± 0.0	–	–	–

Abbreviations: preWS = the week before the 7–day weaning and separation process; postWS = the week after the 7–d weaning and separation process; NS = not significant.
† No comparisons were significant in the posthoc comparison.
[a,b,c] Values within a row with different superscripts differ significantly at $P < 0.05$ for the interaction model.

were no other significant interactions between system and time-point found in the cow behaviours. The proportion of time cows spent lying was affected by system but not time-point; CONV cows (29 ± 19.6%) spent more time lying than PT cows (15 ± 17.6%; $P = 0.034$) while FT were similar to both (16 ± 19.5%). There was no effect of system or time point on the proportion of time cows spent eating solids.

There were interactions between system and time-point for the proportion of time calves spent lying, eating solids, and performing abnormal behaviours; for all other behaviours, only the fixed effects of system and time-point are reported. There was an interaction between system and time-point in the proportion of time calves spent lying; however, no posthoc comparisons differed. The proportion of time calves spent ruminating was not affected by system or time-point. There was an interaction between system and time-point in proportion of time calves spent eating solid feed. There was no difference between the systems preWS, but postWS, the PT calves spent longer eating solid feed than the CONV calves, while the FT calves were similar to both. The PT calves spent less time eating solid feed preWS compared to postWS, while the CONV and FT calves did not differ between time-points. The time calves spent performing maintenance behaviours was not affected by system or time point. The proportion of time calves spent performing positive behaviours was affected by time-point but not system; calves spent more time ($P = 0.009$) performing positive behaviours preWS (5 ± 6.4%) compared to postWS (2 ± 3.4%). There was an interaction between system and time-point in the proportion of time calves spent performing abnormal behaviour; however, no posthoc comparisons differed.

Discussion

The aim of this exploratory paper was to investigate the effect of cow-calf contact rearing systems on the health, performance, and behaviour of cows and calves before (preWS) and after weaning (postWS). We compared three systems: the conventional, no –contact system (CONV), a full-time access CCC system (FT), and a part-time access by night CCC system (PT). These systems differed in several management aspects, including diet (cows: access to grass silage by night vs no grass silage access; calves: milk from dam vs milk replacer from an automatic milk feeder), housing (indoors in a shed vs outdoors on pasture), and milking frequency (once-a-day vs twice-a-day). We acknowledge that these differences made comparing the systems more difficult; however, these systems were chosen to investigate their viability within the Irish pasture-based, seasonal-calving system. Please refer to McPherson et al. (2024a) for a more detailed description on why these specific CCC systems were chosen.

Effect of calf rearing system on cow physiological health, performance, and behaviour before and after weaning

Overall, all cows in this experiment appeared to be in good health, as indicated by their summed clinical health scores. However, we observed differences preWS between the FT and PT cows in several physiological health parameters, indicating that the FT and PT cows may not have been as healthy as they outwardly appeared.

Weaning and separation from their calf, combined with the change in milking frequency (from once-a-day to twice-a-day parlour milking), appear to have influenced the PT cows' energy balance. The PT cows appeared to be in a more positive energy balance preWS compared to the CONV and FT cows, as suggested by their higher BW and body condition score and lower serum NEFA levels. Both higher BW and body condition scores (McNamara et al., 2008; Kennedy et al., 2021) as well as lower NEFA (Patton et al., 2006) have been observed previously in pasture-based dairy cows milked once-a-day in early lactation compared to cows milked twice- or thrice-a-day on the same diet.

Cows that are milked once a day often have higher somatic cell counts or somatic cell score compared to cows milked twice-a-day (Clark et al., 2006; Kennedy et al., 2021; Murphy et al., 2023), so we were expecting the PT cows to have higher somatic cell score than the CONV cows preWS. However, we did not observe a difference in somatic cell score preWS or postWS between our three systems. This may have been due to the effect of suckling on somatic cell score; previous CCC studies have shown that calf nursing can lower somatic cell score and mastitis incidence (Johnsen et al., 2016; Beaver et al., 2019; McPherson et al., 2024a). In this study, the PT cows had lower serum albumin than the FT cows preWS (CONV were similar to both), and this difference disappeared postWS. Albumin levels have been shown to decrease with increasing somatic cell score (Bobbo et al., 2017a, 2017b), but the difference observed here may have also been caused by another source of inflammation.

In contrast to Wenker et al. (2022b), who found no difference in serum calcium between the full-time CCC system they applied and their control, we found that preWS the FT cows had higher serum calcium than the PT cows (CONV were similar to both) and postWS there was no difference between the cows on the different systems. The reason for this is unclear; it may have to do with CCC, but we would expect to see a similar response in the PT cows preWS if this were true. The difference is also likely not caused by the difference in milking frequency, as cows milked once-a-day have been reported to have higher serum calcium than cows milked twice-a-day at the start of lactation (Loiselle et al., 2009). We suggest that the difference observed in serum calcium may be due to the smaller sample size of the FT cows (n = 14) compared to the CONV (n = 18) and PT (n = 18) cows. As a result of dropping four multiparous cows from the experiment due to issues with their calves' health, the FT cows were of younger parity than the CONV and PT cows. Younger lactating cows have been shown to have higher concentration of calcium and magnesium in their blood serum (McAdam and O'Dell, 1982), as more mature cows have a higher metabolic demand for calcium due to their increased milk production and decreased ability to mobilise bone calcium and absorb calcium from their intestines (Horst et al., 2005). However, we did not observe the same difference postWS, which suggests that some other mechanism is occurring, potentially related to their CCC system.

Serum magnesium was higher in the FT cows preWS compared to postWS, while the CONV and PT did not differ between time-points. Serum magnesium is typically considered to be highly reliant on cows' diets. Cows do not have the capacity to store magnesium in their bodies; the majority of magnesium absorption occurs in the rumen, where high levels of potassium can impair magnesium absorption (Suttle, 2022; Khiaosa-Ard et al., 2023). However, the FT cows were being provided an identical diet to the CONV cows preWS, so we should not have observed a difference in magnesium due to diet. It is possible that the FT cows did not consume the extent of their offered feed, potentially because they had a different daily routine than the CONV due to having their calf with them. Although we indirectly measured intake through the proportion of time spent eating solids, we did not find any difference in eating solids time between systems; however, intake per bite can vary and we only observed the cows eating for a small proportion of the day. In the future, direct measurements of cow and calf intake would be useful to provide more insight into why we saw these differences. A potential difference in intake may also explain why we observed a system difference in serum phosphorus (FT and

PT cows had higher phosphorus than the CONV cows). As serum phosphorus has been shown to be lower in dairy cows at peak lactation and during drought conditions (Betteridge, 1986), this may indicate that the FT and PT cows were not as metabolically stressed due to producing milk as the CONV cows both preWS and postWS.

For the cow's behaviour measurements, we only had sufficient observations to analyse three behaviour categories. This was likely due to our sampling method; scan sampling is more useful for frequently observed behaviours (i.e., lying or ruminating) than it is for less frequent behaviours (i.e., abnormal behaviours or play; Lehner, 1992). We observed that the CONV cows spent more time lying down than the PT cows, suggesting that they had a different daily routine than the CCC cows. This opposes previous research on lying time in CCC cows. Although Wegner and Ternman (2023) found that CCC did not affect cow lying time, Johnsen et al. (2021b) found that cow lying time increased when cows progressed from their CCC phase to being separated from their calf.

Although on the surface the cows in the CCC systems appeared to be in good health, preWS the FT and PT cows differed from each other in several physiological health parameters pertaining to energy balance (PT cows) and diet (FT cows). We also observed that the PT cows spent less time preWS lying than the CONV or FT cows, which may have negative implications on their welfare. Some of these differences may have been caused by diet or management of the FT and PT systems, but there appear to be other mechanisms at play. Here, we suggest that the distress caused by weaning and separation from their calf may have caused these differences, but this will require more investigation. In addition, different weaning and separation strategies, such as extending the process or changing the location of weaning and separation to pasture, may help ameliorate any differences observed in this study.

Effect of calf rearing system on calf physiological health, performance, and behaviour before and after weaning

We did not find many differences in calf physiological health linked to their rearing system; instead, many health-related variables were only affected by the weaning process. All calves, regardless of system, had higher summed clinical health scores postWS, indicating that weaning left them in worse health. Serum globulin was also higher postWS, indicating that calves had more inflammation at that time. This matches previous research, which found that serum globulin was higher in artificially reared calves postweaning compared to preweaning (Kim et al., 2011). In contrast to this, we found that serum amyloid A, an α-globulin and thus another marker of inflammation (Eckersall and Bell, 2010; Bobbo et al., 2017a; Trela et al., 2022), was higher in preWS, indicating that our calves had more inflammation preWS. We do not have a suitable explanation for why we observed these opposing results; perhaps other positive acute phase proteins also classified as α-globulins responded differently to serum amyloid A in our calves, thus causing the difference. This warrants further investigation, especially since little research has documented calf serum amyloid A around weaning.

In addition, preWS calves expressed more positive behaviours (play and social interactions), indicating that regardless of their system, weaning may have had a negative impact on their affective states (Ahloy-Dallaire et al., 2018). The large observed response to weaning may indicate that weaning occurred too early in our calves. Although all calves were weaned at 8 weeks, a typical age in Ireland (Barry et al., 2020) and elsewhere in Europe (Johnsen et al., 2021c; Mahendran et al., 2022; European Food Security Authority Panel on Animal Health and Animal Welfare, 2023), 8 weeks may still be too young for calves to have completely adapted to a reliance on solid feed.

A gradual weaning strategy is thought to be the best for calves, both in terms of their welfare and their performance (i.e., postweaning growth; Steele et al., 2017). Calves typically start ruminating around 2–3 weeks of age (Swanson and Harris, 1958; Wang et al., 2022; McPherson et al., 2024b), and the proportion of time calves spend ruminating slowly increases during the preweaning period (McPherson et al., 2024b). Although gradual weaning will increase the time calves spend ruminating and eating solid feed, they may be too young at 8 weeks to have physiologically fully developed into ruminants. In CCC systems, several studies have investigated alternative methods of weaning and separating bonded cow-calf pairs, including physical suckling deterrents such as nose-flaps (Loberg et al., 2008; Wenker et al., 2022a) and udder nets (Johnsen et al., 2018), along with fence-line contact (Johnsen et al., 2015b; Wenker et al., 2022a; Vogt et al., 2024). These methods are used to help disentangle the processes of weaning and separation, thus reducing the potential distress experienced by both cow and calf. Although we utilised fence-line contact in this study, it was only used for 5 days; based on our personal observations of the calves' behaviour during this period, we believe that 5 days was not gradual enough. Perhaps a longer duration of fence-line contact, potentially combined with another physical method to prevent calf suckling, may have made the calves' transition less distressing. Most of the differences we observed between the systems had to do with calf performance and behaviour, especially around weaning. As expected, all calves had higher BW postWS compared to preWS, and the FT and PT calves had higher BW than the CONV calves across both time-points. This was most likely due to the FT and PT calves consuming a greater milk allowance during the preweaning period (CONV calves had a maximum allocation of 9 L/d of milk replacer while the FT and PT calves had essentially *ad libitum* access to milk) and is a common result in CCC research (Johnsen et al., 2021a; Wenker et al., 2022b; Sinnott et al., 2024). We also observed that the FT and PT calves had a decrease in ADGw postWS, while the CONV calves had a consistent ADGw, which was also an expected result of weaning and separation in CCC systems (Johnsen et al., 2021b; Wenker et al., 2022b; Sinnott et al., 2024). This may have been due to the combination of their more abrupt weaning process (resulting in a larger change in diet composition compared to the CONV calves) and potentially the distress caused by the separation from their dam. The CCC calves also experienced a much more abrupt weaning off of milk than the CONV calves (3 d vs 12 d), and the CCC calves had their milk intake schedule abruptly changed compared to the gradual reduction experienced by the CONV calves. However, we do not know the volume of milk consumed by the CCC calves during the first stage of the weaning process. Although previous research has shown that dam-calf contact time (i.e., full-time access vs part-time access) does not affect calf suckling time (Jensen et al., 2024), the total volume of milk available to the calf was likely low in this study, as the unrestricted contact period during the first stage of weaning occurred approximately 1 h after milking and suckling time was not always independent of contact time.

The proportion of time calves spent eating solids also increased postWS, with the PT calves increasing their time eating solids from 0.5% preWS to 33.1% postWS. Both the CONV and FT calves were observed eating solids around 9% preWS and did not have as big of a jump postWS. It is possible that due to the timing of the behaviour observation periods, we missed the times that the PT calves were eating solids preWS. Calf feeding periods have been shown to be easily missed with less frequent scan sampling behaviour scoring (Miller-Cushon and DeVries, 2011), CCC calves with part-time access to their dam have been recorded spending more time eating solids than calves with full-time access (Bertelsen and Jensen, 2023). To fully capture the feeding behaviour of calves in CCC systems, it may be beneficial to use a method that continuously measures their behaviour. This may be especially important in systems that have very specific daily routines, like the PT pairs did here.

The serum NEFA for the FT and PT calves also reflects their difficulties adapting postWS. Serum NEFA typically decreases in calves after weaning (Ferronato et al., 2022), which appears to be what we observed numerically in the CONV calves. The FT and PT calves had numerically higher serum NEFA than CONV calves postWS, which may be an indication that they struggled more nutritionally postweaning and likely reflects their lower ADGw. Previous work has found higher NEFA in abruptly weaned beef calves compared to more gradually weaned calves (Gonzalez et al., 2023). All calves increased BHB postWS, which was expected as higher BHB is often associated with larger solid feed intakes (Quigley III, 1996; Deelen et al., 2016), particularly concentrate intake, and thus, BHB is often used as a proxy for rumination.

In this study, our CCC calves' had a relatively short weaning and separation process. Our results suggest that weaning was stressful to all of our calves, regardless of their system. Future research should investigate whether a more gradual weaning and separation process would improve calf health and performance, and also potentially decrease the amount of distress caused, which would be beneficial for both the animals and the farmer. As the distress experienced by calves and cows during weaning and separation is one of the main issues that CCC farmers have with the system (Neave et al., 2022; Hansen et al., 2023), a solution is required for CCC systems to become widespread.

Conclusion

In this explorative study, in which we compared three different dairy calf rearing systems, we found evidence to suggest that the combination of weaning and separation in two different CCC systems influenced some cow and calf physiological health, performance, and behaviour indicators. Despite having no difference in their summed clinical health scores, the FT and PT cows were worse than the CONV cows in several physiological health parameters pertaining to energy balance (PT cows) and diet (FT cows). Differences in cow performance stemmed from both the different systems and the weaning process. Diet and management of CCC cows thus may play a large role in the transition from being suckled by their calf to no contact with their calf; future CCC research should focus on designing a management routine to minimise the impact that the transition appears to have had on the cows. Overall, weaning appeared to have more of an effect on our calves' health and performance than the system that they were reared in. However, calves in the CCC systems appeared to struggle more in the postweaning transition, as suggested by their lower ADGw and higher NEFA, despite consuming more solid feeds than the CONV calves postWS. As cows and calves in CCC systems need to adapt to the conventional dairy systems after separation, future work in all types of dairy housing and management systems should focus on which factors are most important on easing this transitional period. Overall, management of CCC systems may be the key to their success.

Supplementary material

Supplementary Material for this article (https://doi.org/10.1016/j.animal.2025.101541) can be found at the foot of the online page, in the Appendix section.

Ethics approval

Ethical approval for this study was received from the Teagasc Animal Ethics Committee (TAEC2020-290), and procedure authorization was granted by the Irish Health Products Regulatory Committee (AE19132/P124). Experiments were performed in accordance with the European Union (Protection of Animals Used for Scientific Purpose) Regulations 2012 (S.I. No. 543 of 2012).

Data and model availability statement

None of the data were deposited in an official repository, but are available from the corresponding author upon reasonable request.

Declaration of Generative AI and AI-assisted technologies in the writing process

During the preparation of this work the author(s) did not use any AI and AI-assisted technologies.

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Declaration of interest

None.

Acknowledgements

The authors wish to thank and acknowledge the Moorepark farm staff for their care of the animals and their assistance and patience with this experiment. We would also like to thank Aoife Jones for her assistance with running the serum amyloid A analysis.

Financial support statement

This research is funded by Science Foundation Ireland and the Department of Agriculture, Food, and Marine on behalf of the Government of Ireland under Grant Number [16/RC/3835] – VistaMilk.

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