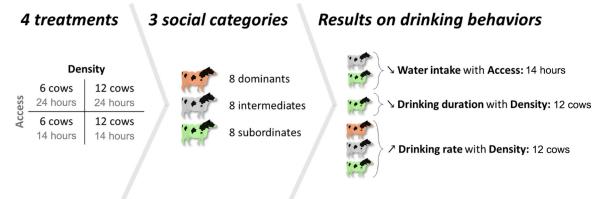
# Stocking density at feeders and drinkers and temporal feed restriction affects dairy cows' drinking behavior

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# **Graphical Abstract**



# **Summary**

The absence of thirst is key to good welfare, but little is known about how farm management can affect the drinking behavior of dairy cows. We studied how stocking density and limited feed-access times impacted cows' drinking behavior. We expected that the effects of competition would vary depending on social status, so effects on dominant, intermediate, and subordinate cows were assessed separately. Dominants were not impacted by treatments, but intermediates and subordinates drank less water when feed was available for just part of the day. Subordinates spent less time drinking when overstocked. Some cows delayed drinking when competition increased. We conclude that competition for access to feed and water impacts drinking behavior, especially for subordinate cows.

## **Highlights**

- Restricted feed access reduced water intake.
- · Overstocking induced faster drinking.
- Dominant cows modified drinking behavior in response to competition.
- Intermediate and subordinate cows drank less with restricted feed access.
- Subordinate cows spent less time drinking when overstocked at the feeder.



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# Stocking density at feeders and drinkers and temporal feed restriction affects dairy cows' drinking behavior

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Abstract: Water is an essential resource for dairy cows, and its consumption is closely linked to feed intake. Social competition can lead to changes in drinking behavior, especially for subordinate cows. We studied how changes in stocking density at the feeder (1 or 2 cows per feeder), and drinker (6 or 12 cows per drinker) and a temporal feed restriction (14- or 24-h access) affects drinking in 4 groups of 6 cows each, following a Latin square design. We recorded drinking behavior within each group of 6 focal cows, and 2 were identified as most dominant, 2 as most subordinate, and the 2 others as intermediate, based on success in agonistic interactions at the drinker. Cows drank less water when feed availability was restricted (on average  $5.3 \pm 3.4$  [SE] L/d less; 4.5% average difference). Subordinate cows also spent less time drinking when overstocked (9.3  $\pm$  6.7 min/d less; 24.3% difference). During treatments with more competition, cows were more likely to be observed drinking in the hours after the peak in drinking observed for the control treatment. A high level of competition among group-housed animals is considered a welfare problem and can lead to injuries and reduced production. Our findings provide evidence that feed restriction and higher stocking density of cows at the drinker and feeders changes the drinking behavior of cows, with subordinate animals experiencing more pronounced effects.

Sufficient access to fresh drinking water for dairy cows is essential to avoid negative effects on animal health and productivity (NRC, 2001). Daily water intake is strongly associated with DMI (Dado and Allen, 1994), and monitoring the water intake of cows might allow for improved farm management (Lukas et al., 2008).

There is a growing body of work indicating that social status of dairy cows can affect access to feed (DeVries et al., 2006). When the amount and quality of the forage is low (Barroso et al., 2000) or when cows are housed under confined housing conditions (Huzzey et al., 2006), dominant cows have priority access to the feed, causing subordinate cows to modify their feeding behavior. What remains unknown is how the social position of the individual cows affects drinking behavior, and how this is affected by competition for feed and water (Jensen and Vestergaard, 2021).

Competition increases between cows when temporal or spatial access to feed is restricted (Olofsson, 1999; Huzzey et al., 2006; Proudfoot et al., 2009) and when cows are more motivated to eat (e.g., after fresh feed delivery; DeVries and von Keyserlingk, 2006). Increased competition can result in physiological indicators of stress (Friend et al., 1977) and altered feeding behaviors, specifically reductions in the time spent feeding, and increased time spent standing close to the feeders (Collings et al., 2011; Crossley et al., 2017). Feed competition has been shown to increase the feeding rate in primiparous cows (Proudfoot et al., 2009), such that cows spend less time close to the resource when access is limited (Olofsson, 1999). Previous work identified 2 types of social competition between animals: direct ("interference," as accessing resources using agonistic interaction as displacements and replace-

ments) and indirect ("scramble," represented by ways to access resources avoiding the direct competition such as increasing feeding rate) competition (Proudfoot et al., 2009). High competition at the feeding area may thus result in abnormal intake and meal patterns (Shaver, 2002), which in turn may affect milk production and lying behavior (Crossley et al., 2017). Subordinate ruminants received more displacements during periods of increased competition for feed (Huzzey et al., 2006) and this social stress may lead to desynchronized feeding as animals attempt to avoid peaks when competition is greatest (Olofsson, 1999; Zobel et al., 2011). Given the correlation between feed and water intake, it follows that during times of higher stocking density, social instability and increased frequency of aggressive interactions at the feed bunk may influence drinking behavior despite water intake not changing throughout the day. To our knowledge, the association between increased competition at the feed bunk and drinking behavior has not been studied.

Current recommendations specify the number or size of drinkers on dairy farms to avoid excessive competition between cows (NRC, 2001) but do not take into account variations in feeding conditions. Competition at feeders may increase from either increased animal density or time constraints on access to feeder; for example, in situations where there is no feed available. When many cows attempt to feed at the same time, competition at the drinkers may also increase, given that feeding is often followed by drinking (Langhans et al., 1995). However, almost no work to date has investigated the effects of overstocking at the feeder and drinker, and that of temporal feed restriction on drinking behavior.

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A previous study (Collings et al., 2011) explored, on 8 groups of cows, how temporal restriction of feed access and stocking of cows at the feed bunk (i.e., of both feed and water bins) alters feeding behavior and increases the number of displacements at the feeder. At the outset of the experiment cows averaged  $204 \pm 40$  (mean  $\pm$  SD) DIM and produced  $36.5 \pm 2.6$  kg/d. When experiencing restricted temporal access and overstocking at the same time, cows spent less time feeding but consumed the feed at a faster rate compared with when they only had restricted access or were overstocked. Temporal restriction to feed resulted in fewer visits to the feeder, with the greatest effects noted during the first 2 h after the morning feed delivery. Data for water intake and drinking behavior were also available but were not analyzed by Collings et al. (2011). Thus, the current study aimed to use these data to investigate the effect of competition at the feed bunk and at the drinkers on drinking behaviors, agonistic competition for water (as indicated by replacements at the drinker), and scramble competition (as indicated by delayed visits to the drinker). We hypothesized that these responses would vary depending on the cow's social position within the group, with higher impact for the subordinate cows. Full methods are described in Collings et al. (2011) but are also briefly summarized below.

We used 4 groups of multiparous (parity ranging from 2 to 6), late-lactating Holstein cows; these groups were the last 4 of the 8 groups tested in Latin square design used by Collings et al. (2011). These 4 groups were chosen so as to minimize temperature-humidity index (THI) differences between treatments (THI =  $84.2 \pm 3.7$ ; range 78.4-91.4). Cows (n = 24) were housed in groups of 6 or 12 according to the treatments as described below, but only 6 cows per group (those receiving all treatments) were considered in the study. This study took place at The University of British Columbia Dairy Education and Research Centre (Agassiz, BC, Canada; animal care protocol A10-0162). Cows were fed a TMR twice daily at 0615 and 0930 h, were milked at 0600 h and 1700 h, and produced 36.5  $\pm$  2.6 kg/d (mean  $\pm$  SD; as reported by Collings et al., 2011). By design, the 12 cows in the group had ad libitum access to water from 1 electronic drinker (Insentec RIC, Marknesse, the Netherlands); this resulted in a slightly higher stocking rate than the 10 cows/ drinker maximum recommended by the NRC (2001), and TMR was provided in 6 electronic feeders (Insentec RIC, Marknesse, the Netherlands). Each group had access to 12 lying stalls.

Data were collected over 7 d from the 4 groups of 6 cows during 4 periods of 1 wk that varied by treatment (described below) between June 23 and July 19, 2010. The 6 cows were randomly assigned to each of the 4 groups. Each group was balanced for parity, projected 305-d milk production and DIM (Collings et al., 2011). All cows were from the main herd of the UBC Dairy Education and Research Centre, and as such all had experience living in a freestall barn. All cows used in the current study were given 7 d to habituate to the Insentec feeders and drinkers immediately before the start of the study. Each group was tested in each of the 4 treatments conditions, with treatment order assigned using a randomized 4 × 4 Latin square. The 4 treatment combinations were based upon 2 levels of cow stocking density at the feed bunk (100% vs. 200%; achieved by stocking either just the 6 experimental cows/pen, or by combining the 6 experimental cows with 6 filler cows), and 2 levels of feed access (ad libitum feed access for 24 h/d or temporally restricted access, with feeders open from 0600 h to 2000 h). The filler cows were provided 3 d of adaptation before data collection was initiated (as per Collings et al., 2011). Filler cows

were selected based on their parity, production, and lactation stage, and previous experience of consuming feed and water from the Insentec bins. All cows were gait scored (following by Flower and Weary, 2006) at the beginning of the study; one cow was identified as having a gait score >4 and was removed from the study and identified to the farm manager for treatment.

The electronic bins (validated by Chapinal et al., 2007) were only accessible by one cow at a time and opened after reading the cow's radio frequency identification ear tag when it came within the read range of the reader positioned at the feed bin gate. For each visit, the software associated with the electronic bin system recorded the cow ID, date, start time, end time, and duration of the visit, and the weight of water in the bin at the beginning and the end of the visit. This weight difference was used to estimate water intake.

We used drinker visits recorded during the last 4 d of each treatment to estimate each cow's hourly water intake proportion (i.e., the proportion of the water drank during that hour relative to total daily water intake). The first 3 d of observation were excluded to avoid any carryover effects from the previous treatment.

We used an algorithm (Foris et al., 2019) to determine replacements at the drinker (i.e., aggressive contact from an actor cow that resulted in the receiver cow leaving the bin and the actor taking her place in a short time) and to identify the social hierarchy of cows. Using the normalized David's score (de Vries et al., 2006), we calculated one social hierarchy per group based on the last 6 d of each period (i.e., a total of 24 d of observations); this dataset was sufficiently large to obtain a robust hierarchy (i.e., a dataset containing at least 10 times more interactions than cows, as recommended by Sánchez-Tójar et al., 2018). Within each group of 6 cows, we identified 2 as the most dominant, 2 as most subordinate, and the 2 remaining cows as intermediate.

All 4 d of observations within each treatment were averaged to result in one value per cow per treatment. For group 4, during the treatment with 6 cows and 14 h of feed access, we experienced a technical issue with the water bins, resulting in unrealistic intake values (above 200 L/d per cow); data from this treatment for group 4 were not included in our analyses.

First, we conducted a preliminary analysis, using the group as the experimental unit, to check the consistency of our analyses of feeding behavior with those of Collings et al. (2011) and investigate drinking behavior with this model. After validating the consistency of our results and noting the effect of the treatment at the group level for drinking behavior, we worked at the individual cow level taking into account that each cow was part of a group and a social category in our statistical model.

Statistical analyses were performed using the mixed procedure of SAS version 9.4 (SAS, 2009). Water intake, drinking duration, drinking rate, and number of visits at the drinker per cow per day were specified as dependent variables. The statistical model tested the fixed effects of density (df = 1), feed restriction (df = 1), and social category (df = 2), as well as all 2-way interactions and the 3-way interaction, and included period (df = 3), group (df = 3), and the cow within group and social category (df = 12) as random effects. Error df were estimated using the containment method. The PDIFF statement was used to make pairwise comparisons, corrected with a Tukey adjustment. We graphically examined the residue normality and variance homogeneity across the dominance categories. We considered results as significant when  $P \le 0.05$  and

a tendency when  $0.05 < P \le 0.10$ . Interactions with P > 0.10 were excluded from the final model.

Cows drank on average  $120 \pm 15$  L/d (mean  $\pm$  SD), with an average drinking duration of  $25.5 \pm 17.8$  min with  $15.2 \pm 9.7$  min/d visits to the drinker, an average drinking rate of  $6.7 \pm 3.2$  L/min, and an average frequency of visits of  $15.2 \pm 8.8$ . Cows were replaced from the drinker  $3.3 \pm 3.6$  times/d (data not shown).

Findings relating to how treatments affected the drinking behavior of the cows in the 3 defined social categories are provided in Table 1. We found that in line with the original findings, this subset of the data shows no effect of treatment on DMI, even as function of social category. Cows drank less water during treatments when feed access was restricted to 14 h/d versus 24 h/d. The lack of difference in feed intake suggests that the reduced water intake was not simply due to reduced feed intake.

Drinking rate was affected by stocking density (Table 1); whereby, doubling the number of cows resulted in cows drinking faster. For time spent drinking (drinking duration) there was a stocking density × social category interaction, whereby subordinate cows spent about 25% less time daily at the drinker at the higher stocking density treatment compared with the lower stocking density treatment.

We also noted an interaction between the feed access restriction and social dominance on the daily number of visits to the drinker, but post hoc tests were not significant. We encourage future work to disentangle this interaction.

Our results confirm that subordinate cows are most affected during periods of increased competition for feed and water, as evidenced by the stronger effects of the stocking and feed restriction treatments on these cows. Other work has reported that certain individuals in a group can monopolize resource access (Sánchez-Tójar et al., 2018) during competition for feed (Zobel et al., 2011) or water (Nizzi et al., 2024).

In the current study, more dominant cows were higher in BW and parity compared with subordinate cows. Previous work has noted an association between social dominance and cow BW and age (Sołtysiak and Nogalski, 2010) with lower social rank for younger and smaller cows. Subordinate cows may be the recipient of more aggression because they were younger and thus less experienced in responding to aggressive social interactions (Proudfoot and Huzzey, 2022). Some authors have suggested that primiparous cows should be housed separately from multiparous groups to minimize negative effects on these younger animals (Grant and Albright, 2001).

When cows drank also varied with time of feed availability (Figure 1A). We observed 2 daily peaks in when cows were most likely to drink: following the morning feeding and milking and second following the afternoon milking. Cows had a higher water intake from 0800 h to 1000 h during the 14 h/d treatment versus when allowed access to feed 24 h/d. However, cows in the 14 h/d treatment drank less water from 2000 h to 2100 h, likely because at this time these cows no longer had access to feed. Globally, cows in the 14 h/d treatment drank a higher proportion of their daily water intake during the day hours (from 0700 h to 1900 h) and a lower proportion during the night hours (from 2000 h to 0400 h).

The effect of stocking on when cows drank also varied with time of the day (Figure 1A). From 0700 h to 0800 h, cows housed at 12 cows/group drank less water compared with when they were housed in groups of 6, but from 0800 h to 1000 h, this effect was

restriction, and social category (Soc. Cat.) on the daily water intake, drinking duration, drinking rate, and frequency of visits at the drinkers; reatments included 6 or 12 cows in the pen and ad libitum (24 h) or restricted (14 h) feed access from 0600 h to 2000  $m h^{1}$ Table 1. Effects of stocking density, temporal feed

		Don	Dominant			Intern	Intermediate			Subo	Subordinate						
Drinking behavior	6 cows, 24 h	6 cows, 14 h	6 cows, 6 cows, 12 cows, 12 cows, 24h 14h 24h	12 cows, 14 h	6 cows, 24 h	6 cows, 14 h	6 cows, 12 cows, 12 cows, 14h 24h 14h	12 cows, 14 h	6 cows, 24 h	6 cows, 14 h	6 cows, 12 cows, 12 cows, 14 h 24 h 14 h	12 cows, 14 h	SEM Density	ty Access	Soc. Cat.	Density: Soc. Cat.	Access: Soc. Cat.
Weight <sup>2</sup> (kg)			763			9	97			7	80.		17.8 x	×	0.040	×	×
Lactation number <sup>2</sup>		4	4.12			'n	.62			2	.38		0.52 ×	×	0.068	×	×
Water intake (L/d)	122	123	125	123	121	115	122	113	124	116	117	109	6.1 0.266		0.466	0.216	0.190
Drinking duration	22.9ª	25.3ª		22.6ª	18.7ª	17.7ª	7ª 23.6ª	15.6ª	39.0ª	37.4ª	4ª 30.3 <sup>b</sup>	27.7 <sup>b</sup>	7.0 <b>0.057</b>	0.421	0.105	0.049	0.292
Drinking rate (L/min)	7.55	7.03	7.55 7.03 7.98	8.03	7.20	7.78	7.12	8.48	4.48	4.53	5.99	5.47	1.2 0.012	0.567	0.112	0.423	0.144
Frequency of visits (no./d)	11.6	15.6	12.2	13.3	15.3	15.9	19.2	13.8	16.7	17.9	15.4	15.8	3.6 0.501		0.684	0.410	0.043

<sup>&</sup>lt;sup>1</sup>The 3 social categories of cows (i.e., dominant [n = 8], intermediate [n = 8], and subordinate [n = 8]) were obtained using a quantile method applied to the dominance score of cows. Bold italic values indicate a significant difference (P < 0.05) x = factors that were not taken into account in the statistical model. Different superscript letters within the same row indicate a significant post hoc pairwise difference within social categories when P-value of interactions was significant (P < 0.05)

<sup>&#</sup>x27;Weight and parity analyzed by linear model with 1 data point by cow and include only social category as fixed effect

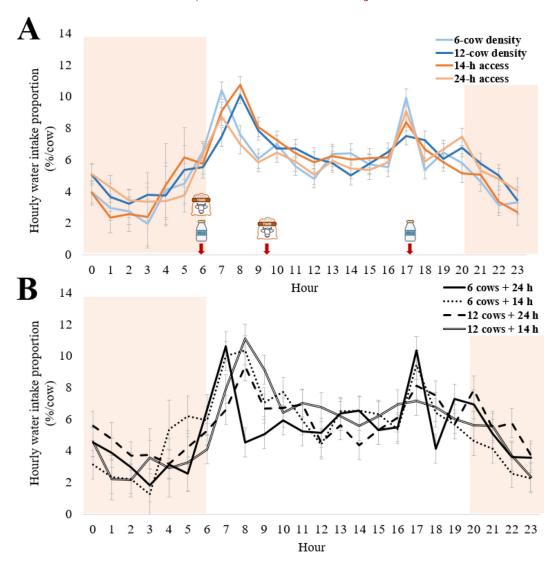


Figure 1. Hourly proportion of daily water intake over the course of a day by cows in each modality of treatment (A) and in each treatment (B). The light orange background indicates times of feed restriction. In panel A, light and dark orange lines indicate the 14- and 24-h of the feed access modality, respectively, and light and dark blue lines indicate the 6 and 12 cows of the density modality, respectively. In panel B, each treatment is represented by a line type. In the 14-h feed access treatment, cows were able to feed from 0600 h to 2000 h.

reversed. During the afternoon peak, from 1700 h to 1800 h, cows in low-density treatment drank more water than high-density treatment, but this effect was reverse in the following hour, from 1800 h to 1900 h. Finally, cows housed at 12 cows/group drank a higher proportion of their daily water intake during the night hours (from 2000 h to 0500 h).

Peak drinking times differed across treatments (Figure 1B). When 6 cows were provided 24-h access to feed, a drinking peak occurred within an hour of milking, but waned rapidly in the hours following. In contrast, when 12 cows were provided 14-h access to feed, only one morning peak occurred 2 h after milking and stayed elevated until 1000 h. The proportion of water drank at night was low during all treatments but especially so for the cows in the 14 h/d treatment that did not have access to feed at this time. The low drinking activity at night is thus likely related to the low feed intake at this time.

When 12 cows were present, management factors such as increased time away from the pen (as more cows needed to be milked), or increased competition in the first hour after return from milking could exacerbate the effects of social rank on feeding behavior. The feeding and drinking areas were highly competitive at this time, as shown by the high rate of displacements in Collings et al. (2011), such that some cows shifted their feeding and drinking behaviors to quieter times, explaining the lag in peak drinking times observed in the current study. Previous work investigating the effects of intense competition for water caused by high THI reported that cows with low social rank shifted their drinking times to avoid peak of competition (McDonald et al., 2020). Thus, collectively, a higher cow density during a competition time may explain the delay in postprandial drinking of the less dominant cows. The treatment with 12 cows/drinker was above the current NRC (2001) recommendation of 10 cows/drinker, perhaps accentuating the temporal avoidance of the competition by subordinate cows (as expected under scramble competition; Proudfoot et al., 2009). When cows were subjected to a time restriction on access to the feeder, we observed a greater impact on drinking behavior, resulting in a shift in drinking by an additional hour. Collings et al. (2011) found that cows in the 14 h/d treatment showed increased DMI in the 2 h following the delivery of fresh feed, likely explaining the drinking peak we observed. Feed restriction has also been shown to encourage drinking behavior synchronization in grouphoused pigs (Turner et al., 1999).

In this study, we followed 4 groups of cows in a Latin square design. Because of the THI difference, we could not use the other 4 groups of cows from the previous experiment (Collings et al., 2011), reducing the generalizability of our findings. The pen layout may also have introduced a bias. Drinkers were placed next to the feeder with the same automatic gates. During higher feed competition, some cows may have been struggling to find a place at feeders and ended up with their heads in the drinker instead, causing some incorrect replacements recorded (i.e., replacement having the purpose of eating and not drinking).

Temporally restricted feed access and higher stocking levels at feeders and drinkers induced changes in drinking behaviors, especially for more socially subordinate cows. We suggest that cows respond differently to increased competition, depending on their social rank within the group. Changes in hourly drinking patterns also occurred, with cows shifting their peak water consumption according to time of feed access and cow density. For subordinate cows, temporal feed restriction resulted in decreased water intake when the stocking density was above the recommended maximum of 10 cows per one drinker.

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## Notes

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**Nonstandard abbreviations used:** Soc. Cat. = social category; THI = temperature-humidity index.