Applied Animal Behaviour Science 187 (2017) 8–14

Contents lists available at ScienceDirect

Applied Animal Behaviour Science

journal homepage: www.elsevier.com/locate/applanim

The effect of feeding enrichment in the milk-feeding stage on the cognition of dairy calves in a T-maze

Kelsey Horvath, Mariana Fernandez, Emily K. Miller-Cushon *

Department of Animal Sciences, P.O. Box 110910, Gainesville, FL 32611, USA

ARTICLE INFO

Article history:
Received 21 June 2016
Received in revised form 7 November 2016
Accepted 29 November 2016
Available online 2 December 2016

Keywords:
Dairy calf
Enrichment
Cognition
Learning
T-maze

ABSTRACT

In many species, environmental complexity is known to affect cognitive development, yet it is common to house dairy calves individually in restrictive environments. The hypothesis of this study was that providing calves with simple nutritional enrichments would improve their success in a cognitive task and reduce reactivity to novel stimuli. Individually-housed Holstein heifer calves were assigned at birth to conventional management (C; n = 10), with access to milk (6 L/d) via bucket and grain concentrate, or enriched feeding (E; n = 10), with access to milk via teat to allow natural suckling, and chopped hay alongside concentrate. At week 5 of age, calves were tested in a T-maze with a reward (0.2 L milk; provided according to familiar delivery method) placed in one arm to assess initial spatial learning, reversal learning where the reward location was changed to the opposite arm, and response to an intramaze change (novel object, a colored ball, was placed in the maze between the start position and reward). Calves received 5 sessions/d for 5 days or until criteria (moving directly to correct side in 3 consecutive sessions) was reached for initial and reversal learning. Time to complete the test, movement in maze, kicks, and non-nutritive licking/sniffing were recorded from video. In the initial learning stage, the number of sessions required to meet the learning criteria was similar between calves ($P = 0.12$). However, E calves took longer to complete the task in early sessions (treatment by session interaction; $P = 0.02$), due to increased time spent on the correct side of maze before obtaining the reward (26.72 vs. 7.48 s; SE = 3.4; $P = 0.005$). In the reversal learning stage, there was no overall difference in the number of sessions to meet the learning criteria ($P = 0.20$), but E calves completed the task faster (19.84 vs 27.22 s; SE = 2.10; $P = 0.03$), and C calves spent 1.5 x longer on the incorrect side of the maze than E calves ($P = 0.04$), suggesting that C calves struggled to relearn the task. In both initial and reversal learning, C calves kicked more frequently ($P < 0.04$). During the novel object session, E calves found the reward faster (6.11 vs 20.6 s; SE = 4.06; $P = 0.01$), whereas C calves spent longer in the middle of the maze where the novel object was located (2.08 vs 13.4s; SE = 5.33; $P = 0.04$). These results suggest that providing simple feeding enrichments during the milk feeding stage may alter calf cognition and influence responses to environmental changes.

1. Introduction

Dairy cattle need to learn how to interact with their environment in order to respond appropriately to the management and environmental changes they will face over their lifetime. As such, it is important to understand the cognitive development of calves and how environment influences that development. Results of previous studies conducted using various maze tests suggest that cattle possess accurate spatial memory (Bailey et al., 1989), can rapidly adapt to new learning situations (Hosoi et al., 1995), and maintain a memory of a location over several days (Arave et al., 1992). Additionally, younger calves are more flexible in learning and relearning, while older cows maintain memories of locations better (Kovalčik and Kovalčik, 1986). Further evidence suggests that cattle tend to resist changing their choice of a maze arm once an association between that arm and a reward has formed, which can inhibit behavioral flexibility and reversal learning (Grandin et al., 1994). Studies in deer mice suggest that the effect of early life enrichment persists later in life (Hadley et al., 2006).

Providing environmental enrichment, such as novel foods, social companions, and substrates to manipulate, has been shown to affect success in maze tests in pigs (Bolhuis et al., 2004; Sneddon

* Corresponding author.
E-mail address: emillerc@ufl.edu (E.K. Miller-Cushon).

http://dx.doi.org/10.1016/j.applanim.2016.11.016

et al., 2000), mice (Tanimura et al., 2008), and rats (Einon, 1980; Fernandez-Teruel et al., 1997). Environmental complexity can prevent abnormal repetitive behaviors in deer mice (Lewis, 2004) and is associated with changes in neural functioning which can cause altered cognition (Würbel, 2001). Additionally, isolated rats exhibit reduced behavioral inhibition and are less likely to abandon a previously successful strategy when tested in a maze (Morgan, 1973). Housing dairy calves individually early in life impairs cognitive development which may potentially be due to sensitive periods in the brain's development of areas affecting behavioral flexibility (Meagher et al., 2015). The initial learning of a simple discrimination task is similar for individually and pair housed calves, but pair housed calves are able to more easily adapt their behavior when the stimuli are reversed (Gaillard et al., 2014).

Behavioral flexibility can help individuals adapt more quickly to changing environments which may provide long-term welfare benefits. Providing social enrichment in the form of pair-housing makes calves less fearful in novel social and environmental situations than individually-housed calves (Jensen and Larsen, 2014). Pair housed calves will also show habituation to a novel object when repeatedly exposed to the object, but individually housed calves do not show this apparent recognition of the object (Gaillard et al., 2014). Individually housed calves are more reactive, have increased defecations, more backing-off events, and spend more time running and exploring a test arena compared to pair housed calves (de Paula Vieira et al., 2012). Providing calves with complex environments through the use of social enrichment may help them cope with novel situations, stressful events, and increase their ability to learn.

Despite the benefits of social contact, many dairy calves are housed individually to prevent the spread of diseases and may benefit from other forms of enrichment. In addition to social contact, the feeding method used also plays an important role in a calf's early environment. Calves are often fed using a bucket on-farm, which does not satisfy their motivation to suck and can lead to increased non-nutritive oral behaviors (de Passillé, 2001). Providing enrichment in the form of a rubber teat reduces non-nutritive oral behaviors, and the action of sucking on the teat additionally elicits the release of hormones such as cholecystokinin and insulin which are involved in digestion (de Passillé, 2001). In addition to the rubber teat, hay may be a beneficial nutritional enrichment for calves since it influences rumen development, may improve rumen environment (Khan et al., 2011), and reduces non-nutritive sucking (Haley et al., 1998). Since some natural feeding behaviors can be satisfied through provision of a rubber teat and hay, they may be beneficial nutritional enrichments for dairy calves, but their effect on cognitive development is unknown.

This study examined the effects of hay provision and teat access on the ability of calves to learn and relearn a spatial discrimination task conducted in a T-maze. Further, we assessed the reaction of the calves to a novel intramaze change. It was hypothesized that calves provided simple nutritional enrichments during the milk-feeding stage would perform similarly during initial learning, but have improved performance measured as fewer sessions to reach criteria and quicker completion times during reversal learning tasks and exhibit reduced reactivity to novelty.

2. Material and methods

2.1. Animals and housing

A total of 16 Holstein heifer calves at the University of Florida Dairy Research Unit (Hague, FL, USA) were enrolled into the study at birth and were uniquely identified with RFID ear tags. Calves were housed at the Calf Unit of the University of Florida Dairy Unit in individual wire-mesh pens (0.9 × 1.8 m; width × depth) that permitted visual and auditory, but no tactile contact with other calves. Calves were managed according to the standard operating procedures for this facility. All pens were located in an open-sided barn, protecting calves from downward wind and rain. Pens were bedded with sand that was replaced weekly. Calves were fed 4 L of colostrum as a first meal. They then daily received six liters of pasteurized waste milk mixed with a powdered enhancer (Pasteurized Milk Balancer Protein-Blend, Purina Animal Nutrition LLC, Shoreview, MN, USA) was delivered at 0600 h and 1800 h. Calves had *ad libitum* access to calf starter (Ampli-Calf STR 20P R50 DBZ9.1 Medicated, Purina Animal Nutrition LLC, Shoreview, MN, USA) with supplement (SMI 3.5G AS700 CMB Medicated, Purina Animal Nutrition LLC, Shoreview, MN, USA) and water. As per standard operating procedure at the University of Florida Dairy Research Unit, all calves were disbudded by a University of Florida veterinarian during week 4 of life by hot iron and provided both local anesthesia and analgesia during the procedure. All study procedures have been reviewed and approved by the University of Florida Animal Care and Use Committee.

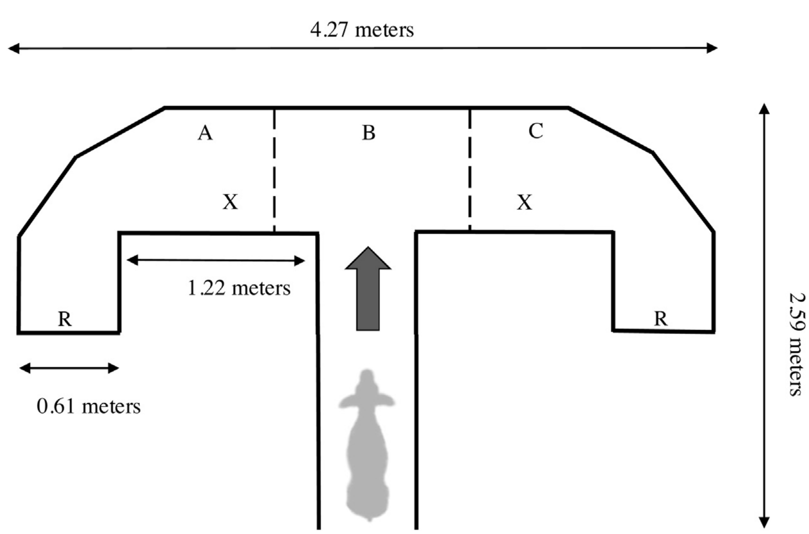

Fig. 1. Diagram of T maze. B denotes the middle zone of the maze, while A and C are the arm zones. The milk reward and empty bucket or bottle were placed in locations marked R. The X marks the position of the novel object, although it was only placed on the side with the reward during the novel object session. A panel was placed at the base of the T after the calf entered to prevent the calf from exiting.

2.2. Study design

Calves were randomly assigned into one of two treatments from birth: Control (C) and Enriched (E). Control calves had their milk placed in their pens and drank directly from the bucket. The bucket was removed after milk was consumed. For E calves, a teat was available at the front of the pen attached to a line fitted to a one-way valve running to the milk bucket outside the pen. The teat remained in place throughout the day, while the line and bucket were removed after the milk meal had been consumed. Additionally, E calves received *ad libitum* access to chopped timothy hay (2") by bucket. During the week of testing (week 5 of life), calves received 1 L of milk during the test as a reward, so 1 L of milk was removed from the second meal to maintain a total of 6 L/day.

2.3. Cognitive test

During week 5 of life, cognition was assessed through spatial discrimination tasks. The cognitive test was modeled after a similar study performed in pigs (Bolhuis et al., 2004). The test consisted of learning tasks in a T-maze (Fig. 1). The maze was constructed using 24 panels (0.61 × 1.22 m) of reconfigurable wire mesh pens (Oxgord Dog Animal Large Metal Wire Playpen 48 inches, OxGord Inc., Los Angeles, CA, USA). The wire mesh panels were covered with landscape fabric to create a visual barrier for the calves. The maze was supported on the outside by stacked bales of straw. Calves entered into the maze through the base of the "T" and exited through the arm containing the reward, which was allowed to open after the calves found their reward.

Each calf received a maximum of 5 sessions per day for 5 days or until criteria (moving directly to and begin consuming the reward in 3 consecutive sessions) was reached for all learning tasks. Criteria was selected based on preliminary tests showing that calves did not perform regressive errors after 3 correct completions and also to provide a cut-off that would fit within the limited time frame we were able to test the calves. Each session had a maximum time of 3 min to complete the task. A reward of 0.2 L milk was placed in one arm of the maze and was balanced between each side for each treatment to prevent an effect of laterality (Hosoi et al., 1995). Control calves received their milk reward via buckets hanging on the wall of the maze, while E calves received their milk via bottles attached to the wall to maintain similarity to their normal milk feeding method. The unrewarded side, therefore, had either an empty bucket or a bottle without the teat. Open containers of milk were placed outside each arm of the maze and out of sight of the calves to prevent calves from using olfactory cues to find the reward. Each calf was led through the maze on their first session to both arms and allowed to drink the milk reward. Calves were allowed 25 total sessions to meet criteria for initial learning, and calves unable to meet criteria were considered to not learn the task and were removed from further testing. However, all calves passed initial learning. The fewest sessions any individual calf completed was 4 to meet criteria, and the slowest initial learner required 18 sessions to meet initial learning criteria.

Once calves had met the learning criteria for the initial learning stage (moving directly to and begin consuming the reward in 3 consecutive sessions), the location of the reward was changed to the opposite arm and reversal learning began. Calves were allowed 25 sessions minus the number required to meet criteria for initial learning, and calves unable to meet criteria were considered to not learn the task and were removed from further testing. The fewest sessions any individual calf completed was 8 to meet criteria, and the slowest reversal learner required 20 sessions to meet reversal learning criteria.

Once calves met the criteria for the reversal learning task, they were given one session with an intramaze change. The intramaze change consisted of a novel object in the form of a multi-colored ball being placed in the middle section between the start and the reward (Fig. 1), with the reward maintained in the same position as during the reversal learning stage. Calves needed to pass the object to receive their reward and were provided enough room to move past the object without touching it.

Table 1
Ethogram of behaviors observed from video recorded during each test session of initial and reversal learning and the novel object test.

Behavior	Description
Total time	Duration of time from entering maze until beginning reward consumption
Middle duration	Duration spent in middle zone of maze
Correct duration	Duration spent in the arm with the reward
Incorrect duration	Duration spent in the arm without the reward
Crossing	Number of times entering a new zone
Licking/sniffing	Duration with mouth less than 5 cm from parts of maze other than reward
Kick/buck	Number of times legs were lifted from ground higher than a normal step

2.4. Behavioral observation

The behavior of all calves was recorded continuously during each session by video camera (GoPro Hero3, GoPro Inc., San Mateo, CA, USA). The latency to begin consuming the reward was measured by stopwatch. The video was reviewed using Behavioral Observation Research Interactive Software (Friard and Gamba, 2016) to measure additional behaviors as defined in Table 1. All video observations were performed by a single, trained observer. Pearson correlation coefficients were calculated (within PROC CORR of SAS v. 9.4) to assess intra-observer reliability: $R^2 = 0.99$ (n = 10, $P < 0.001$) for all behaviors except for the duration of time spent in the middle section of the maze ($R^2 = 0.84$ n = 10, $P = 0.002$).

2.5. Statistical analysis

Statistical analyses were performed using SAS v. 9.4 (SAS Institute Inc., Cary, NC, USA) with calf as the experimental unit. Significance was declared at $P < 0.05$, with trends reported if $0.05 < P \leq 0.10$. Data were checked for normality using the UNIVARIATE procedure of SAS. Results are reported as means and standard errors.

The number of sessions required to meet criteria were analyzed for the initial and reversal stage using a Wilcoxon rank-sum test (PROC NPAR1WAY), as data were not normally distributed. The effect of treatment on pass rate during the single novel object test session was analyzed using Fisher's Exact Test (PROC FREQ).

Behavioral data were analyzed separately by testing stage (initial, reversal, and novel object). For initial learning, behaviors were analyzed from sessions 1 through 3, because calves were led to the reward during session 0 as a training session and some calves had passed initial learning by the 4th session. For reversal learning, the behaviors were analyzed over the first 8 sessions of reversal learning since all calves received at least 8 sessions before meeting the learning criteria. Frequency of kicking was not normally distributed in any stage and was analyzed using the Wilcoxon rank-sum test. All other behavioral data were normally distributed and were analyzed by stage using PROC MIXED in a general linear mixed model with session as a repeated measure (for initial and reversal stages only, as the novel object test was performed only once).

Table 2
Behavior of calves exposed to either the control environment (C) treatment in their home pen or enriched environment treatment with a teat and hay provided (E) in their home pen during the first three test sessions after training in the initial learning stage in a T-maze. Data are reported as means for each treatment and standard errors (SE).

Behavior	Treatment (T)		SE	*P*-value[a]		
	C	E		T	S	T x S
Total time to find reward (s)	17.99	46.78	6.43	0.020	<0.001	0.022
Time spent in middle of T-maze (s)	13.11	7.32	6.56	0.53	0.037	0.28
Time spent in incorrect arm of T-maze (s)	5.71	16.32	6.19	0.23	0.034	0.17
Time spent in correct arm of T-maze (s)	7.48	26.72	3.40	0.0014	<0.001	0.0047
Frequency of crossing T-maze (no/test)	4.86	8.13	3.65	0.52	0.15	0.83
Duration of licking (s)	27.55	39.61	10.83	0.43	<0.001	0.011
Frequency of kicks (no/test)[b]	0.44	0.11	0.25	0.018	–	–

[a] Effects of treatment (T) and session number (S).
[b] Data were not normally distributed and were averaged across sessions and analyzed for effect of treatment using Wilcoxon Signed Rank test.

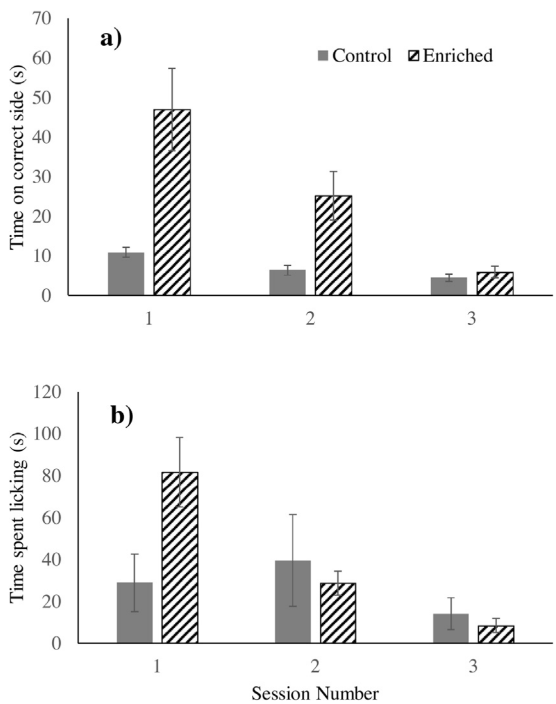

Fig. 2. Effects of enrichment (teat and hay) on (a) time spent in the correct arm of the T-maze (treatment by session interaction; *P* < 0.01), and (b) duration of time spent licking during the first 3 sessions of the initial learning test after training (treatment by session interaction; *P* = 0.01). Data are reported as means and error bars represent standard errors of each data point.

3. Results

In the initial learning stage, there was an effect of treatment on the behaviors performed during the first 3 sessions of the test after training (Table 2). Enriched calves took longer than C calves to complete the task in early sessions (treatment by session interaction; *P* = 0.02), due to increased time spent on the correct side of the maze before obtaining the reward (Fig. 2; treatment by session interaction; *P* < 0.01). The increased time spent on the correct side of the maze by E calves also corresponded with increased licking/sniffing in early sessions (Fig. 2; treatment by session interaction; *P* = 0.01). Control calves kicked more frequently during the test (*P* = 0.02). There was no effect of treatment on the number of times the calves crossed between areas of the maze, the amount of time spent in the middle of the maze, or the amount of time spent in the incorrect arm of the maze. There was no difference in the number of sessions required to meet criteria (moving directly to and begin consuming the reward in 3 consecutive sessions; 5.88 vs 8.13 session; C vs. E, SE = 1.10; *P* = 0.12). All 16 calves reached criteria during the maximum number of sessions allowed and proceeded to reversal learning.

In the reversal learning stage, there again was an effect of treatment on the behaviors performed during the first 8 sessions of the test (Table 3). Enriched calves consistently took less time to complete the task (*P* = 0.03), and C calves spent longer on the incorrect side of the maze than E calves (Fig. 3; *P* = 0.04), suggesting that C calves struggled to relearn the task. Control calves again kicked more frequently during the test (*P* = 0.04). There was no overall difference between treatments in the time spent in the middle section of the maze, the number of crosses, or the duration of licking/sniffing. Again, there was no difference in the number of sessions to meet criteria (12.33 vs 10.0 session; SE = 1.56; *P* = 0.20). Twelve calves (6 of 8 from each treatment) met criteria for the reversal learning task within the maximum number of sessions allowed and proceeded to the novel object test.

During the single novel object session, E calves found the reward faster (*P* = 0.01), whereas C calves spent longer in the middle of the maze (*P* = 0.04) where the novel object was located (Table 4). Control calves also spent a greater amount of time licking/sniffing (*P* = 0.02) and tended to cross between areas of the maze more frequently during the test (*P* = 0.08). There was no difference between

Table 3
Behavior of calves exposed to either a control environment treatment (C) in their home pen or enriched environment treatment with a teat and hay provided (E) in their home pen during the first 8 sessions of the reversal learning stage in a T-maze. Data are reported as means for each treatment and standard errors (SE).

Item	Treatment (T)		SE	*P*-value[a]		
	C	E		T	S	T x S
Reversal learning						
Total time to find reward (s)	27.22	19.84	2.10	0.026	<0.001	0.22
Time spent in middle of T-maze (s)	2.08	1.83	0.15	0.25	<0.001	0.25
Time spent in incorrect arm of T-maze (s)	21.01	14.21	2.17	0.044	<0.001	0.17
Frequency of crossing T-maze (no/test)	3.61	3.61	0.32	0.99	<0.001	0.48
Duration of licking (s)	20.43	16.28	2.59	0.28	<0.001	0.87
Frequency of kicks (no/test)[b]	0.54	0.22	0.13	0.04	–	–

[a] Effects of treatment (T) and session number (S).
[b] Data were not normally distributed and were averaged across sessions and analyzed for effect of treatment using Wilcoxon Signed Rank test.

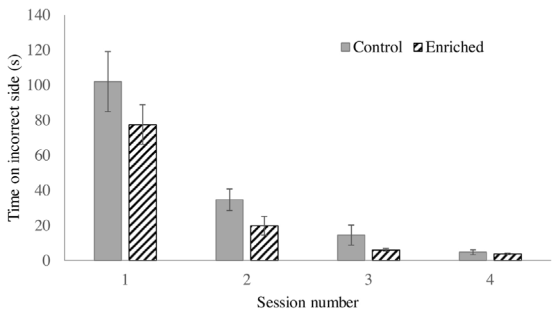

Fig. 3. Effects of enrichment (teat and hay) on duration of time spent in the incorrect arm of the T-maze during the first four consecutive test sessions of the reversal learning test (treatment; $P = 0.044$). Data are reported as means and error bars represent standard errors of each data point.

Table 4
Behavior of calves exposed to either a control environment treatment (C) in their home pen or enriched environment treatment with a teat and hay provided (E) in their home pen during the single novel object test session in a T-maze. Data are reported as means for each treatment and standard errors (SE).

Item	Treatment		SE	*P*-value
	C	E		
Pass rate[a] (% of calves)	33.3	83.3	–	0.24
Total time to find reward (s)	20.6	6.11	4.06	0.013
Time spent in middle of T-maze (s)	13.4	2.08	5.33	0.037
Time spent in incorrect arm of T-maze (s)	1.77	0.40	0.50	0.13
Frequency of crossing T-maze (no/test)	2.67	1.33	0.49	0.08
Duration of licking (s)	13.47	2.62	2.97	0.020
Frequency of kicks (no/test)[b]	0.67	0.5	0.44	0.75

[a] Criteria for pass defined as calf moving directly to correct side of T-maze, passing by novel object (2 of 6C calves vs 5 of 6 E calves passed).
[b] Based on Wilcoxon Signed Rank test.

treatments in the amount of time spent on the incorrect side of the maze or the number of kicks. There was no significant difference in the pass rate (2 vs. 5 of 6 calves; C vs. E., $P = 0.24$), defined as the calf moving directly to the reward without moving to the other arm of the maze, between treatments.

4. Discussion

Dairy cattle need to develop behavioral flexibility in order to successfully cope with the stressors and changing environments they will encounter over their lifetime. It was hypothesized that calves provided nutritional enrichment in the form of hay and a teat would have improved performance in cognitive tests and reduced reactivity to novelty. Our results suggest that the provision of these simple nutritive enrichments during the milk feeding stage did alter the calves' ability to complete the cognitive tasks and reduced their reactivity to a novel object placed in the maze.

During the initial learning phase, E calves were significantly slower to find the reward than C calves, which is in contrast with our hypothesis that initial learning would not differ between treatments. In contrast to the present results, Gaillard et al. (2014) reported that the initial learning of a simple discrimination task was similar for calves provided differing degrees of social enrichment, but that calves raised in pairs were able to more easily adapt their behavior when the stimuli were reversed than calves housed individually. In the present study, however, the greater amount of time required to find the reward by E calves was due to significantly more time spent on the correct side of the maze before beginning to consume the reward, as opposed to greater time spent in other areas of the maze. This suggests that treatment groups performed equally well in the spatial discrimination task, but E calves were delayed in finding the teat. While both E and C calves were fed according to their familiar feeding method, C calves may have had an advantage in locating the reward during initial test sessions since they were able to visually see the surface of the milk in the bucket, whereas E calves had to locate the teat on the wall. The E calves spent more time licking/sniffing while in the correct side of the maze before finding the reward, especially in the first few testing sessions. Although the location of licking/sniffing within the maze wasn't recorded, increased licking/sniffing and increased time standing in the correct section indicates that they were searching for the teat rather than investigating the maze generally. Furthermore, the amount of time E calves took to complete a session and spent licking/sniffing decreased across sessions (Fig. 2), suggesting that they quickly learned the teat location. When repeating this experiment, it may be beneficial to increase the training period when the calf is lead to the reward to more than one session prior to the start of testing during which E calves would have the opportunity to better learn the teat location. We expect that this would result in no difference between treatments in time required to locate the teat or bucket during the initial stage.

During the reversal learning stage, the results supported our hypothesis that E calves would abandon a previously successful strategy sooner and would complete the maze faster than C calves. Control calves spent more time in the incorrect side of the maze compared to E calves suggesting that C calves took longer to alter their response to the change and had decreased behavioral flexibility. This agrees with studies in rodents where most errors occurred during the beginning of reversal learning in a T maze test, and unenriched mice had decreased performance and increased preservative errors compared to enriched mice (Tanimura et al., 2008). Additionally, unenriched rats made more total errors, made more errors in earlier sessions, learned slower, and had difficulty remembering long sequences of events in a spatial memory task than socially reared rats (Einon, 1980). Individually housed calves, in general, make more errors in a reversal learning task than calves housed in pairs or groups, which suggests that social isolation early in life impairs learning and potentially the development of neural structures related to learning (Gaillard et al., 2014; Meagher et al., 2015). The results of these previous studies with similar individual housing setups to the current study suggest that enrichment through socially housing calves results in animals that have more flexible responses to changes in their environment, whereas individually rearing calves results in cognitive impairments. Whereas these studies have identified the importance of social enrichment for cognitive development, the present results are the first to suggest that other forms of enrichment, such as nutritional enrichment, can also affect cognitive development in dairy calves.

The C calves kicked more per session in both the initial and reversal learning sessions than E calves, which indicates that they may be more reactive when placed into a novel environment. Insufficient environmental stimuli results in reduced behavioral flexibility (Gaillard et al., 2014) and increased reactivity when placed into a novel environment (de Paula Vieira et al., 2012) which suggests that these animals may have reduced abilities to learn a cognitive task since they are more reactive in unfamiliar situations. In the present study, however, the frequency of kicking was similar between treatments during the single novel object session. In agreement with this finding, mice housed in an enriched environment are quicker to habituate to novel environments, but are more reactive and alert when initially placed into a novel environment (van de Weerd et al., 1994). This suggests that E calves still reacted to novelty, but may have habituated to the environment more quickly than C calves during the initial and reversal test sessions. While we did see a difference in reactivity between treatments, we believe more research is needed to determine whether reactivity would be adaptive in either a natural or production setting and whether it is indicative of either a stress response or play behavior.

During the novel object test, the treatment groups showed distinct differences in reactions to the novel object and the behaviors expressed. Control calves required significantly more time to find the reward than E calves due to more time in the middle segment of the maze rather than passing the object to reach the reward. The increased time spent in the middle section was likely due to observing the ball, as we anecdotally noticed that the increased duration of licking/sniffing was directed at the ball. These results suggest that C calves were more reluctant to approach the object and opted to move to the incorrect arm of the maze to avoid the novel object, as indicated by an increased frequency of crossing between maze sections, rather than moving directly to the arm they learned contained the reward. On the other hand, E calves appeared to be less reactive to the novel object, as they crossed between maze sections a fewer number of times, spent less time licking/sniffing, and proceeded to the reward more quickly. In agreement with the present results, 28 week old pigs enriched with straw, peat, and tree bark displayed a more passive reaction and less distraction, as assessed by reduced time in contact with a white strip of tape that the pigs had to cross over, less time to reach the food reward, and fewer maze arms entered, compared to unenriched pigs (Mendl et al., 1997). However, in contrast to the current results, 8 week old pigs enriched with hay tended to have the opposite reaction to a novel object in the form of an upside down aluminum pan in a maze and were more reactive and distracted, as indicated by taking longer to complete the task and performing more errors by going to the incorrect arm (Bolhuis et al., 2004). In addition to age differences between these studies, differences in response have been attributed to different coping strategies (Bolhuis et al., 2004), motivation (Mendl et al., 1997), and emotionality (Fernandez-Teruel et al., 1997). In dairy calves, providing social contact has been shown to reduce fear in novel social and environmental situations (Jensen and Larsen, 2014), reduce reactivity and reluctance to approach an unfamiliar calf in a social novelty test (de Paula Vieira et al., 2012), and increase habituation to a novel object in the form of a red plastic bin (Gaillard et al., 2014). Additionally, environmental enrichment in mice decreases anxiety-like behavior and increases habituation to novel environments (Meshi et al., 2006). In agreement with previous studies, our results suggested that enrichment reduced reactivity to novelty during the intramaze change.

There was no overall effect of treatment on the number of sessions required to pass initial and reversal learning, suggesting that behavior is overall a more sensitive indicator of treatment effects than the number of sessions required to pass the cognitive task. This is in agreement with another study that found the number of sessions required to meet criteria was not different between enriched and unenriched pigs (Bolhuis et al., 2004), whereas the behaviors performed within each session did differ between treatments.

The forms of nutritional enrichment provided in this study were selected with the goal of increasing the natural feeding behaviors available to the calf and providing access to enrichment items that the calf is motivated to obtain. It is well established that calves are motivated to suck (de Passillé, 2001) and calves voluntarily select forage early in life (Miller-Cushon and DeVries, 2015). The combined effects of providing these nutritional enrichments on cognition suggest that the opportunity to exercise a more diverse range of feeding behaviors may be important for learning. It should be noted that these nutritional enrichments have different effects on the calf; for example, suckling behavior influences release of digestive hormones (de Passillé, 2001) and hay provision influences rumen development (Khan et al., 2011). It is possible that the improved cognition observed in calves provided access to these nutritional enrichments was primarily due to only one of these enrichments, or alternatively, they may have interactive effects. While we did not design the present study to specifically assess the independent effects of teat provision and hay access on cognition, this would be valuable to measure in future work.

Behavioral flexibility and the ability to learn is important for dairy cattle, because these animals will experience stressful environmental and management changes such as grouping (Bøe and Færevik, 2003) and novel experiences such as the milking parlor and automatic feeders. As cattle move through the stages of life, their ability to cope with stressors depends on their ability to learn about and adapt to new and changing environments. The results of this study suggest that early environmental enrichment may support early cognitive development, and therefore has the potential to improve welfare. We encourage further work to assess the longer-term influence of early life enrichment on cognition in dairy calves and how enrichment affects their ability to cope with stressors as they move into adulthood.

5. Conclusion

These results suggest that providing simple feeding enrichments in the form of a teat and hay during the milk feeding stage may alter calf cognition and influence responses to environmental changes. Calves provided enrichment took less time to find the reward during the reversal learning stage of the spatial discrimination task and were less reactive when introduced to a novel object. These results suggest that calves housed in unenriched environments may have reduced flexibility in learning cognitive tasks and increased reactivity in response to novel situations. Providing enrichment to dairy calves, including simple enrichments such as hay and artificial teats that allow them to engage in natural behaviors, may promote cognitive development.

Acknowledgements

We thank the staff and students at the University of Florida Dairy Unit. In particular, we thank Amber DeWitt, James Gross, and Kristen Solari for their technical assistance. KH was supported by a Matching Assistantship through the Department of Animal Sciences, University of Florida. This project was supported through departmental funding (Department of Animal Sciences, University of Florida).

References

Arave, C.W., Lamb, R.C., Arambel, M.J., Purcell, D., Walters, J.L., 1992. Behavior and maze learning ability of dairy calves as influenced by housing sex and sire. Appl. Anim. Behav. Sci. 33, 149–163.

Bøe, K.E., Færevik, G., 2003. Grouping and social preferences in calves: heifers and cows. Appl. Anim. Behav. Sci. 80, 175–190.

Bailey, D.W., Rittenhouse, L.R., Hart, R.H., Richards, R.W., 1989. Characteristics of spatial memory in cattle. Appl. Anim. Behav. Sci. 23, 331–340.

Bolhuis, J.E., Schouten, W.G.P., Leeuw, J.A.D., Schrama, J.W., Wiegant, V.M., 2004. Individual coping characteristics, rearing conditions and behavioural flexibility in pigs. Behav. Brain Res. 152, 351–360.

de Passillé, A.M., 2001. Sucking motivation and related problems in calves. Appl. Anim. Behav. Sci. 72, 175–187.

de Paula Vieira, A., de Passillé, A.M., Weary, D.M., 2012. Effects of the early social environment on behavioral responses of dairy calves to novel events. J. Dairy Sci. 95, 5149–5155, http://dx.doi.org/10.3168/jds.2011-5073.

Einon, D., 1980. Spatial memory and response strategies in rats: age, sex and rearing differences in performance. Q. J. Exp. Psychol. 32, 473–489.

Fernandez-Teruel, A., Escorihuela, R.M., Castellano, B., Gonzalez, B., Tobena, A., 1997. Neonatal handling and environmental enrichment effects on emotionality, novelty/reward seeking, and age-related cognitive and hippocampal impairments: focus on the roman rat lines. Behav. Genet. 27, 513–526.

Friard, O., Gamba, M., 2016. BORIS: a free, versatile open-source event-logging software for video/audio coding and live observations. Methods Ecol. Evol.

Gaillard, C., Meagher, R.K., von Keyserlingk, M.A.G., Weary, D.M., 2014. Social housing improves dairy calves' performance in two cognitive tests. PLoS One, 9.

Grandin, T., Odde, K.G., Schutz, D.N., Behrns, L.M., 1994. The reluctance of cattle to change a learned choice may confound preference tests. Appl. Anim. Behav. Sci. 39, 21–28.
Hadley, C., Hadley, B., Ephraim, S., Yang, M., Lewis, M.H., 2006. Spontaneous stereotypy and environmental enrichment in deer mice (Peromyscus maniculatus): Reversibility of experience. Appl. Anim. Behav. Sci. 97, 312–322.
Haley, D.B., Rushen, J., Duncan, I.J., Widowski, T.M., De Passillé, A.M., 1998. Effects of resistance to milk flow and the provision of hay on nonnutritive sucking by dairy calves. J. Dairy Sci. 81, 2165–2172.
Hosoi, E., Rittenhouse, L., Swift, D.M., Richards, R.W., 1995. Foraging strategies of cattle in a Y-maze: influence of food availability. Appl. Anim. Behav. Sci. 43, 189–196.
Jensen, M.B., Larsen, L.E., 2014. Effects of level of social contact on dairy calf behavior and health. J. Dairy Sci. 97, 1–10.
Khan, M.A., Weary, D.M., von Keyserlingk, M.A.G., 2011. Hay intake improves performance and rumen development of calves fed higher quantities of milk. J. Dairy Sci. 94, 3547–3553.
Kovalčik, K., Kovalčik, M., 1986. Learning ability and memory testing in cattle of different ages. Appl. Anim. Behav. Sci. 15, 27–29.
Lewis, M.H., 2004. Environmental complexity and central nervous system development and function. Ment. Retard. Dev. Disabil. Res. Rev. 10, 91–95.
Meagher, R.K., Daros, R.R., Costa, J.H.C., von Keyserlingk, M.A.G., Hötzel, M.J., Weary, D.M., 2015. Effects of degree and timing of social housing on reversal learning and response to novel objects in dairy calves. PLoS One 10, e0132828.
Mendl, M., Erhard, H.W., Haskell, M., Wemelsfelder, F., Lawrence, A.B., Mendl, M., Erhard, H.W., Haskellt, M., Wemelsfelder, F.O., Lawrence, A.B., 1997. Experience in substrate-enriched and substrate-impoverished environments affects behaviour of pigs in a T-Maze task. Behaviour 134, 643–659.
Meshi, D., Drew, M.R., Saxe, M., Ansorge, M.S., David, D., Santarelli, L., Malapani, C., Moore, H., Hen, R., 2006. Hippocampal neurogenesis is not required for behavioral effects of environmental enrichment. Nat. Neurosci. 9, 729–731.
Miller-Cushon, E.K., DeVries, T.J., 2015. Invited review: development and expression of dairy calf feeding behaviour. Can. J. Anim. Sci. 95, 341–350.
Morgan, M.J., 1973. Effects of post-weaning environment on learning in the rat. Anim. Behav. 21, 429–442.
Sneddon, I.A., Beattie, V.E., Dunne, L., Neil, W., 2000. The effect of environmental enrichment on learning in pigs. Anim. Welf. 9, 373–383.
Tanimura, Y., Yang, M.C., Lewis, M.H., 2008. Procedural learning and cognitive flexibility in a mouse model of restricted repetitive behaviour. Behav. Brain Res. 189, 250–256.
Würbel, H., 2001. Ideal homes? Housing effects on rodent brain and behaviour. Trends Neurosci. 24, 207–211.
van de Weerd, H.A., Baumans, V., Koolhaas, J.M., van Zutphen, L.F., 1994. Strain specific behavioural response to environmental enrichment in the mouse. J. Exp. Anim. Sci. 36, 117–127.