**Pre-weaning management of calves on commercial dairy farms and its influence on calf welfare and mortality**

J. Barry, E.A.M. Bokkers, I.J.M. de Boer and E. Kennedy

*Animal*

**Materials & Methods**

Feeding Equipment Hygiene

In each visit, trained personnel physically assessed hygiene practices to determine the effectiveness of such routines while also verifying answers given during the interview. Hygiene of two feeding implements, for example, esophageal tube feeders and group feeders, was assessed using 3M Clean-Trace surface protein plus test kits (3M, Minnesota, USA). This is a semi-quantitative assay which involves collecting a swab sample from the implement of interest. The swab is then placed in a chemical solution, which creates a chemical reaction resulting in color formation. Color formation is related to the amount of proteins present, such as milk residues or biological contaminants (e.g., bacteria, fungi), which is measured on a four-point reference scale

Behaviour Observations

Indirect behavioural observations were made from 60 min video recordings taken during the farm visit. Recordings of two group pens, of contrasting composition (i.e. male and female calves), or if male and female calves were grouped together, then a group of older calves (>3 weeks old) and a younger group (<3 weeks old) were assessed, to provide a reflection of the situation on that particular farm. To ensure consistency across behavioural observation conditions, recordings were made during daylight hours. Once the groups were selected, video cameras (GoPro Inc., California, USA) were set-up to capture a clear and unobstructed image of the entire pen area, within which the activities of calves could be identified. Prior to recording, any lighting available was switched on to ensure maximum visibility in the recordings. Recordings were scored at a later date using the ethogram as outlined in Table S1, which is adapted from De Wilt (1985). Behaviours were observed by scan sampling at five minute interval for the duration of the 60 minute recording. At the 5 minute intervals, the behaviour of each calf was classified using the ethogram, e.g. resting, feeding etc. To ensure the behaviour could be correctly identified a period of ±30 seconds was used around the 5 minute time point. Frequency of behaviour was calculated by expressing the number of times a specific behaviour was observed relative to that of other behaviours, throughout the sixty minute observation period.

**Table S1.** Ethogram, adapted from de Wilt (1985), which categorises and defines various behaviours, used for bovine calf behavioural observations in the welfare protocol.

|  |  |  |
| --- | --- | --- |
| Category of behaviour | Behaviour | Definition |
| Posture | Standing | Calf is standing |
|  | Lying | Calf is lying/resting |
| General | Walking | Calf is walking |
|  | Not visible | Behaviour of the calf is not visible |
|  | Other | Events not reflective of welfare status (E.g. defecates or urinates) |
| Feeding behaviour | Drinking milk | Calf is drinking from a bucket, trough, teat or automatic feeding station. |
|  | Drinking water | Calf is drinking water |
|  | Eating | Calf eats concentrates or roughage, or other solid feed (proximity of head to feed) |
|  | Ruminating/chewing | Calf is chewing |
| Comfort behaviour | Grooming | Calf licks itself, including snout/nose licking. |
|  | Scratching/Rubbing/Stretching | Calf scratches itself with one of the legs (generally hind legs).  Calf rubs itself on pen structure  Calf stretches itself |
| Abnormal behaviour | Tongue playing/rolling | Calf makes repeated movements with its tongue inside or outside its mouth |
|  | Urine drinking / oral manipulate prepuce | Calf drinks the urine of another calf  Calf attempts to suck the naval area of another calf. |
|  | Orally manipulating pen structure | Calf licks, nibbles, sucks, or bites at the pen structure (barriers, walls, buckets, troughs etc.) |
| Play behaviour | Play behaviour/Mounting/ Head butting | Calf runs, jumps, changes direction suddenly, bucks, kicks hind legs, twists or rotates body.  Calf mounts, or attempts to mount, a pen mate.  Calf is engaged in head to head pushing with another calf. |
| Social behaviour | Social interaction | Calf licks another calf in the same area multiple times  Calf nibbles, sucks or bites at another calf |

*Behaviour analyses*

The effect of group size, space allowance and calf sex on group behaviour frequencies was estimated using a linear model in PROC MIXED (SAS version 9.4; SAS Institute Inc., Cary, NC, USA), with farm included as a repeated measure.

The dependent variables investigated included; lying, playing, standing, eating, grooming, social licking, walking, ruminating and rubbing/scratching behaviour in the following model:

*Yijkl= μ + fi + gj + Ak + Sl+ ejkl*

Where:

*Y* = observation;

*μ* = mean;

*Fi* = farm effect (*i* = 1…47)

*Gj* = Group size category (*j* = 1…4)

*Ak*= Space allowance category (*k* = 1…4)

*Sl* = Sex (*l* = male, female, mixed)

*eijkl*= residual error term

*Calf mortality*

The effect of transition milk feeding, number of transition milk feeds, source of transition milk, group feeding method and volume provided on calf mortality was estimated using a linear model in PROC MIXED.

The dependent variables investigated included; 28 day mortality rate, 3 month mortality rate, 6 month mortality rate and twelve month mortality rate in the following model:

*Yijklm = μ + ti + nj + sk + gl* + *vm* + *eijklm*

Where:

*Y* = observation;

*μ* = mean;

ti = Transition milk feeding (*i* = 1 or 2*)*;

*nj* = Number of transition milk feeds (*j = 1…10)*;

*sk* =Source of transition milk (*k = 1…3)*;

*gl* = Group feeding method (*l* = 1…4);

*vm* = Volume category (*m* = 1…4);

*eijklm* = residual error term.

For 12 month mortality rate, the effect of bedding depth category, hygiene score, scour vaccine use and pneumonia vaccine use estimated using the following linear mixed model in PROC MIXED:

*Yijkl* = *μ* + *Bi + Hj* + *Sk* + *Pl*+ *e*ijkl

Where:

*Y* = observation;

*μ* = mean;

*Bi = Bedding depth category (I = 1…4);*

*Hj* = Hygiene score (*j* = 1…4);

*Sk* = Scour vaccine use (*k* = 1 or 2);

*Pl*= Pneumonia vaccine use (*l* = 1 or 2*);*

*eijkl* = residual error term.

**References**

De Wilt J 1985. Behaviour and welfare of veal calves in relation to husbandry systems. PhD Thesis, Wageningen University, Wageningen, the Netherlands.