The objective of this study was to compare the effects of rearing calves outdoors, with and without all-weather calf jackets, with calves reared indoors on calf immunity and animal performance. In February 1999, male Holstein calves (mean (s.e.) weight 55 (1.90) kg) were randomly assigned to one of three treatments (n=30 per treatment): 1) outdoors with jacket, (J; mean age 19 (s.e. 2.0) days); 2) outdoors without jacket (NJ; mean age 19 (s.e. 1.8) days), and 3) indoors on straw (I; mean age 19 (s.e. 1.0) days). Calves received an individual allowance of 25 kg of milk replacer dry matter during the first 42 days with ad libitum access to a concentrate ration from day 0 to 63. The jackets were removed from the calves on day 42. Live-weight gain from day 0 to day 63 of the study was not significantly different between treatments (J, 0.79; NJ, 0.80; I, 0.80 kg). Sixty percent of the J calves and 53% of the NJ calves required four or more antibiotic treatments for respiratory disease while corresponding treatments were required for 97% of the I calves. The incidence of diarrhoea was significantly higher in both outdoor treatments compared to the I treatment. There was no significant difference in white blood cell counts or in serum immunoglobulin concentrations between treatments on days 0, 21, 42 and 63 or in in vitro interferon-γ production on day 63. It is concluded that using calf jackets on calves reared outdoors had no beneficial effect on calf performance or immune status. The incidence of respiratory disease was higher and diarrhoea incidence was lower in calves reared indoors compared with calves reared outdoors. There was no significant difference in incidences of diarrhoea and respiratory disease between the two outdoor treatments.

Keywords: Calf health; calf welfare; calves; immune function; Interferon-γ

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Introduction

The successful management of artificially reared calves requires the provision of suitable housing and a reduction in exposure to environmental pathogens. Outbreaks of pneumonia and diarrhoea are common in enclosed, intensive calf-rearing units, where bovine respiratory disease and enteric disease are transferred from calf to calf. Diarrhoea and respiratory disorders are the main hazards to successful calf rearing and their occurrence before 90 days of age affects the performance of the animal later in life and is associated with a higher age at calving (Waltner-Toews et al., 1986a; Waltner-Toews, Martin and Meek, 1986b; Warnick, Erb and White, 1994).

A Canadian survey (Waltner-Toews et al., 1986a), in which the scale of operation ranged from 23 to 154 calvings per year, showed that the farm-level mortality rate per 6-month season (winter/summer) was 6% and that 20% of calves were treated for scours and 15% were treated for pneumonia before the age of weaning. Peters (1986) studied the effects of housing (indoors vs. outdoors), penning (individual vs. group), breed, time since purchase and season of purchase of calves. He concluded that pneumonia was the most common disease symptom (48.3% of calves) with diarrhoea the second most common (14.1%). Neither the type of penning or housing affected the incidence of pneumonia but diarrhoea was most common in individually penned calves. Limitations of space, diet, and social environment have been reported to impair the welfare of calves reared in intensive farm systems (LeNeindre, 1993). Andrighetto et al. (1999) observed a favourable effect of group rearing on growth performance in the late period of growth. Fisher et al. (1985) reported lower daily gains and less favourable feed efficiency for postweaned calves housed in narrow individual pens (0.66 m × 1.48 m) than for animals kept in wider individual pens (1.36 m × 1.48 m).

Sargeant et al. (1994) reported that of 4863 white-veal calves reared on six commercial white-veal farms in Ontario, and followed through production, 3.7% died, with pneumonia being the largest single cause of death. Peak death and cull losses occurred during the seventh and eighth week of production. In an effort to reduce the incidence of respiratory disease in calves some producers have used outdoor hutches. Housing mostly in the presence of adult cattle was a risk factor for pneumonia, whereas housing mostly alone in a hutch was protective (Virtala et al., 1999). An alternative to the use of calf hutches, is the use of an all-weather-jacket for calves reared outdoors. Data are not available to indicate the beneficial effects of rearing calves outdoors with jackets on immunity and performance.

From the viewpoint of animal welfare, including animal health, it is important to develop a combination of management procedures which will minimise the adverse effects of respiratory disease on animal performance and health/welfare indicators. In the present study, the working hypothesis, using controlled comparisons was that, 1) calves reared outdoors using a calf jacket are less susceptible to respiratory disease than calves reared indoors and that 2) calves reared outdoors using calf jackets are more resistant to disease than calves reared outdoors without jackets. The overall objective of this study was to evaluate use of an all-weather calf jacket on immunocompetence and performance of calves artificially reared outdoors with that of calves reared indoors.
Materials and Methods

Animals and management
Ninety Holstein × Friesian calves (mean (s.e.) weight 55 (1.90) kg) were purchased directly from dairy farms in February 1999. On arrival at Grange Research Centre (day 0) the calves were weighed and allocated at random to one of the following treatments; 1) outdoors with an all-weather jacket (J; mean (s.e.) age 19 (2.0) days), 2) outdoors without an all-weather jacket (NJ; mean (s.e.) age 19 (1.8) days) and 3) indoors on straw (I; mean (s.e.) age 19 (1.0) days). Each calf received a dry matter allowance of 25 kg of milk replacer powder offered in water at 38 °C by bucket during the first 42 days and had ad libitum access to a concentrate ration consisting of (g/kg) rolled barley (775), soyabean meal (200), mineral/vitamin (25) from day 0 to day 63. The indoor group of calves were turned out to pasture on day 63. Clean fresh water was available at all times. Rectal temperature and faecal swabs were taken on arrival at the Research Centre and all calves were vaccinated with a Salmonella vaccine (Grovax, Hoechst Animal Health, Milton Keynes, Bucks, UK). For the 63-day experimental period three groups of 10 calves were housed on straw in a naturally ventilated Monopitch calf house (4.8 m × 10.0 m). The outdoor groups were accommodated in paddocks measuring 56 m × 16 m with a dry sand lying area (8 m × 5 m) at the rear of each paddock, with 10 calves per paddock. Shelter was provided against adverse weather conditions, such as wind and rain using tildenet (Warren Polytunnels, Ballivor, Co. Meath, Ireland) double layered fencing (1.2 m high) on three sides. A natural shelter (wooded area) was available on the fourth side of each plot. Jackets were removed from the calves on day 42 of the study and the calves were moved to fresh paddocks (16 m × 100 m) with a dry sheltered lying area (8 m × 5 m) and with shelter also provided against adverse weather conditions. Climatic conditions, including relative humidity and temperature of the calf house and outdoor paddocks, were recorded continuously throughout the course of the study using minidatal loggers (Radionics, Dublin, Ireland). All animals were weighed on day 0 and at 7-day intervals thereafter. Average daily feed intake (ADFI) were determined on a group basis over 7-day periods and total feed intake was calculated per management group from day 0 to day 63. Daily rectal temperature for all calves and frequency of antibiotic and electrolyte treatments were recorded. Calves with a rectal temperature of ≥40 °C and clinical signs of respiratory disease (moderate to severe respiratory distress on auscultation) were administered an antibiotic. All procedures were conducted under experimental license from the Irish Department of Health in accordance with the Cruelty to Animals Act (Ireland, 1876) and European Union Directive 86/609/EC.

Immunological variables
Calves were bled by jugular venipuncture on days 0, 21, 42 and 63 of the study. Serum immunoglobulin G1 was measured quantitatively by single radial immunodiffusion (sRID) and an internal Ig-standard (Mancini, Carbonara and Heremans, 1965). The zinc sulphate turbidity (IG) test was performed on all serum samples (McEwan et al., 1970).

The stimulated lymphocyte production of interferon-γ (IFN-γ) was determined following whole blood culture of heparinised plasma (Wood, Corner and Plackett, 1990) on day 63 of the study. Duplicate 1.48-ml aliquots of blood were cultured in 24-well culture plates (Costar Corporation, Cambridge, MA) with 20 μl
of PBS containing either 1 mg/ml of keyhole limpet haemocyanin (KLH) or 1 mg/ml of Concanavalin-A (Con-A) or no additive for 16 h at 37 °C and in an atmosphere of 5% CO₂ in air. The culture plates were then centrifuged and the supernatant harvested and frozen at –20 °C until it was assayed for IFN-γ production using an ELISA procedure (Rothel et al., 1990; CSL Biosciences, Parkville, Victoria, Australia). The in vitro KLH- and Con-A-stimulated IFN-γ production was calculated by subtracting the absorbance at 450 nm of wells that received PBS alone from the absorbance of wells that received either KLH or Con-A, respectively.

Haematological and physiological variables
The haematological variables (red blood cell number (RBC), haemoglobin (Hb), packed cell volume (PCV), mean cell volume (MCV), total white cell (TWC) count, percentage granulocytes, percentage monocytes, platelet number, percentage lymphocytes) were determined for unclotted (K₃-EDTA) whole blood samples using an electronic particle hematology analyser (Celltac MEK-610K, Nihon Kohden, Japan) on days 0 and 63. The physiological variables measured on day 0 and 63 were: blood copper (Cu²⁺), plasma glutathione peroxidase (GPX), serum non-esterified fatty acids (NEFA) and beta-hydroxybutyrate (BHB). Plasma glutathione peroxidase (GPX) was determined using the method of Paglia and Valentine (1967) adapted for the SPACE analyser (Schiapparelli Biosystems, Inc, USA). BHB and NEFA were determined using Randox assay procedures.

Statistical analyses
Data were analysed using SAS software (SAS, 1996). Live-weight data and IgG₃ concentrations were analysed using PROC MIXED. A maximum likelihood estimation method was used to estimate variance-covariance parameters of a model with treatment and treatment × day as fixed effects and random regression on day for each animal. An autoregressive structure was assumed for the covariance of the residual animal effects and this covariance structure was assumed to be heterogeneous across treatments.

Differences between treatments were tested using the LSMEANS statement. The effect of treatment on ADFI was analysed by one-way ANOVA on total feed intake for each management group. Haematological data, Cu, NEFA and BHB were analysed using one-way ANOVA. Differences among treatments were tested using the Tukey option in the MEANS statement. Differences between sampling times (day 0 and day 63) were tested using the paired t-test, the null hypothesis being that the mean difference between time points was equal to zero. ZST values were not normally distributed and were analysed using overall ranked data in the repeated measures option of PROC GLM and Kruskall-Wallis (treatment difference) and Friedman (day difference) post-hoc comparison tests were applied. GPX values were tested using Kruskall-Wallis and a Wilcoxon signed rank test to evaluate differences between day 0 and day 63 (Snedecor and Cochran, 1989). The incidences of respiratory disease and enteric disease were analysed using the chi-square test.

Results
The temperatures of the rearing environments ranged from –0.1 °C to 13.2 °C (average 6 °C) for the indoors, and from –2.1 °C to 11.1 °C (average 4 °C) for the outdoors, respectively. The relative humidity of the respective environments
ranged from 42 to 96% with an average 79% for indoors and from 47.6 to 99.9% with an average of 85% for the outdoors, respectively.

There was no significant difference between treatments with respect to mean live-weight (kg) from day 0 to day 63. Mean (s.e.) live weights (kg) at day 0 were 55.2 (2.00), 54.9 (1.92) and 55.1 (1.81) for treatments J, NJ and I, respectively. The corresponding values at day 63 were 105.7 (3.88), 106.0 (3.75) and 108.2 (3.42) kg, respectively. Similarly, there were no significant differences between treatments for mean total food intake per head from day zero to day 63 (means (s.e.) were – J: 28.5 (0.86) kg; NJ: 26.7 (1.01) kg; I: 30.1 (0.80) kg).

There was no significant difference in serum ZST units (Figure 1) or serum IgG1 concentrations (Figure 2) among treatments throughout the study period. On days 42 and 63 of the study, serum IgG1 concentration was significantly higher

**Figure 1:** Mean zinc sulphate turbidity (ZST) (units) of calves reared outdoors with jackets — ◆ —, and without jackets — ■ — and indoors — ▲ — on days 0 to 63 of the study (the vertical bar at each point represents the s.e.).

**Figure 2:** Mean IgG1 (mg/ml) of calves reared outdoors with jackets — ◆ —, and without jackets — ■ — and indoors — ▲ — on days 0 to 63 of the study (the vertical bar at each point represents the s.e.).
(day 42, P<0.05; day 63, P<0.05) in the J calves when values were compared with day 0 concentration. Serum IgG1 concentrations in NJ and I calves were not significantly different from the day 0 concentrations. There were no significant changes in granulocyte, monocyte, or lymphocyte percentages, haemoglobin or NEFA concentrations, platelet numbers or total white blood cell numbers, MCV and PCV between day 0 and day 63. On day 63 of the study the Treatment I calves had significantly lower (P<0.05) BHB concentrations (0.23 (s.e. 0.008) mmol/l) than calves in the outdoor treatments (0.28 (s.e. 0.01) mmol/l in J and NJ). Blood copper and GPX concentrations were similar across treatments on Day 0. There was no significant difference between treatments in either KLH- or Con-A-induced IFN-γ production from lymphocytes collected on day 63 of the study (KLH mean (s.e.) IFN-γ production for J, 0.12 (0.004); NJ, 0.14 (0.005) and I, 0.13 (0.004); CON-A mean (s.e.) IFNγ production for J, 0.66 (0.02), NJ, 0.69 (0.03) and I, 0.65 (0.015), respectively).

On days 21 and 42 of the study, red blood cell number, Hb concentration and haematocrit percentage were significantly lower in Treatment I compared with the two outdoor treatments (Table 1). The incidence of respiratory disease was higher in Treatment I while the incidence of diarrhoea was higher in the outdoor treatments, irrespective of calf jacket use (Table 2).

Table 1. Effect of rearing calves outdoors, with and without jackets, or indoors on red blood cell number, haematocrit percentage and haemoglobin concentration (mean ± s.e.) 21 and 42 days after initiation of treatment

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No. of red blood cells (×10¹²/l)</th>
<th>Haematocrit (%)</th>
<th>Haemoglobin (g/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Day 21</td>
<td>Day 42</td>
<td>Day 21</td>
</tr>
<tr>
<td>Jackets (n=30)</td>
<td>11.2 b±0.3</td>
<td>11.3 b±0.2</td>
<td>34.0 b±0.83</td>
</tr>
<tr>
<td>No jackets (n=30)</td>
<td>11.2 b±0.3</td>
<td>11.3 b±0.2</td>
<td>33.5 b±0.75</td>
</tr>
<tr>
<td>Indoors (n=30)</td>
<td>10.5 a±0.2</td>
<td>10.2 a±0.2</td>
<td>30.4 a±1.19</td>
</tr>
</tbody>
</table>

a,bWithin columns means not having a common superscript differ significantly (P<0.01).

Table 2. Incidence and treatment of respiratory disease and enteric disease in calves reared indoors or outdoors with or without jackets

<table>
<thead>
<tr>
<th>Disease and number of treatments given</th>
<th>Experimental group</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Outdoors</td>
</tr>
<tr>
<td></td>
<td>Jackets</td>
</tr>
<tr>
<td><strong>Respiratory disease</strong></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>1</td>
<td>8</td>
</tr>
<tr>
<td>≥2</td>
<td>18&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>Enteric disease</strong></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>13</td>
</tr>
<tr>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>≥2</td>
<td>12&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a,b</sup>Values, within rows, without a common superscript are significantly different (P<0.01).
Discussion

Calf performance traits, such as live-weight gain and ADFI, are useful indicators of the balance between the animal and its environment (Curtis, 1987). In the present study there were no significant differences between treatments with respect to live-weight gain or ADFI from day 0 to day 63 of the study. When calves are housed indoors the occurrence and spread of disease usually increases (Kelly et al., 1984). Arave, Mickelsen and Walters (1985) evaluated the effects of four rearing treatments using heifer calves that were assigned to the following treatments: groups of six, individual hutch, isolation, and isolation with handling. They concluded that there were no differences in weaning weight or average daily gain to weaning.

Other workers have reported varying incidences of enteric and respiratory disease in calves. Blom (1982) reported a 10.3% incidence of enteritis but a considerably higher incidence of respiratory disease (44.0%) in calves in Denmark. By contrast, Perez et al. (1990) and Wells, Garber and Hill (1996), reported a lower incidence of respiratory disease (5.8 to 8.4%), but a higher incidence of diarrhoea (24.6%) in dairy calves in The Netherlands and USA, respectively. Waltner-Toews et al. (1986b), Gardner et al. (1990), and Virtala et al. (1996) also reported incidences of diarrhoea (20.5%, 0.12 cases per calf-month at risk, and 28.8%) and respiratory disease (15.4%, 0.077 cases per calf-month at risk, and 25.6%) in calves from Ontario, Canada, California and New York, USA, respectively. Olsson et al. (1993) reported a very low incidence (0.8%) of respiratory disease; considerably lower than the percentages we found for calves treated, on two or more occasions, with antibiotics for respiratory disease (I, 96%; J, 60% and NJ, 53%). Low serum Ig concentrations due to the breakdown of passively acquired antibodies in calves over 1-month-old can predispose calves to respiratory disease. Corbeil et al. (1984) correlated Ig concentrations in serum and in nasal secretions of calves with pneumonia and diarrhoea during the first 12 weeks of life. The results showed that the peak onset of pneumonia occurred between 2 and 4 weeks of age when the serum IgG1, IgG2, and IgA concentrations were lowest. As IgG2 concentration increased, fewer calves developed pneumonia. The peak onset of pneumonia was also correlated with the lowest IgG and IgA concentrations in nasal secretions. Most calves developed pneumonia when serum concentrations of IgG1 were less than 1.5 g/dl, IgG2 less than 0.3 g/dl, IgA less than 0.1 g/dl, and IgM less than 0.2 g/dl and when the combined IgG and IgA values in nasal secretions were less than 0.2 mg of Ig/mg of protein. In the present study, outdoor rearing, irrespective of calf jacket, and indoor rearing of young dairy calves had no suppressive effect on the development of the humoral immune or cell mediated immune responses.

Chang et al. (1996) reported an enhanced humoral immune response in stressed calves following market transport, but its enhancement was antigen-dependent and variable. Assessment of immune function is a useful indicator of cattle welfare (Amadori et al., 1997), and the establishment of protective immunity depends critically on IFN-γ production (Arad, Nussinovich and Kaempfer, 1995; Johnson, 1997). IFN-γ is produced by activated T lymphocytes and natural killer (NK) cells in response to antigen. Most studies demonstrating reduced lymphocyte function in response to stress have used mitogens to produce a non-specific lymphocyte stimulation. Outdoor
and indoor rearing of calves in the current study did not impair the \textit{in vitro} IFN-\(\gamma\) production in response to both mitogen (CON-A) and the antigen KLH, indicating that immune response was not suppressed.

Jang (1986) reported that the average Hb concentrations in both beef and dairy calves (3 to 16 weeks old) was 11.3 \text{g/dl}. The lower concentrations of Hb detected in the indoor calves did not result in appetite suppression. Interference with synthesis or release of Hb, production or survival of RBC, or metabolism has the potential to cause disease.

It is concluded that, rearing calves outdoors using calf jackets had no beneficial effect on calf performance.

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\textbf{References}


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