

**Study 2: An evaluation of underlying performance mechanisms in an experimental group of animals kept on the index farm from January 2007 to April 2008**

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## ABSTRACT

Long-term problems of cattle performance have been observed on a farm (the 'index farm') in Co. Kilkenny, Ireland. The objective of this study was to determine whether periods of poor weight gain or weight loss, if observed, were associated with changes in haematological parameters, essential element status, or exposure to heavy metals. Cattle (n = 10) sourced off farm were moved to the index farm in January 2007 (Day 0; 16/01/2007) and kept there until April 2008 (day 469; 29/04/2008). Blood-samples were collected by jugular venepuncture and cattle were weighed on Days 0, 112, 154 (blood-sample only), 203, 231, 259, 287, 315, 357, 386, 413, 442 and 469 relative to the start of the study. Weight gains were intermittent during the period of observation: very good ADGs were observed during the initial housing period (January to May 2007; 0.96 kg/day), very poor during summer grazing (May to October 2007, 0.13 kg/day) and good during the subsequent housing period (October 2007 to April 2008, between 0.55 and 1.2 kg/day). During the summer grazing period, the cattle experienced a coincident reduction in weight gain, which is suggestive of a single insult over a defined period of time. At this time, there was no overt evidence of disease in these cattle. Adequate ADGs during summer 2007 have been reported elsewhere in Ireland. Selenium concentrations declined during the study period and were considered to be deficient from October 2007 to January 2008. However, the concentrations improved on winter feeding and returned to normal by February 2008. Copper (Cu) concentrations declined after cattle were introduced onto the index farm, and were considered deficient from May 2007 until the end of the study. In this study, essential element deficiencies were again identified as potential contributors to the ongoing performance problems on the index farm. A single Cd peak was measured in cattle during this study, the significance of which is uncertain but subject to further study.

Keywords: cattle, growth rates, cadmium exposure, selenium deficiency, glutathione peroxidase, copper, deficiency

## INTRODUCTION

Significant shortfalls in cattle performance on a dairy farm in Castlecomer, Co. Kilkenny (the index farm) have been identified; growth rates of young cattle on the index farm are lower than expected, with animals achieving a significantly smaller stature and lower weights than expected for similar production systems.

Many factors have been proposed as contributors to the poor performance on the index farm. Disease, both clinical and sub-clinical, management and nutrition have been considered. Environmental pollution from a nearby brick factory has also been suggested as a possible contributor to the disease syndromes observed on the index farm. Production history of the index farm suggests that two distinct but potentially related syndromes have been observed in young-stock on the farm: 1) stunting of cattle growth, resultant in cattle of small stature, and 2) poor thrive, characterised by periods of low live weight gains or weight loss. Additionally, these periods of poor live weight gains and weight loss occur at clearly defined time-points (*Overview*, Appendix 2), at varying intervals, and in all groups of young growing cattle present on the farm at that particular point in time. When these downturn periods occur, the majority of cattle within these growing groups are similarly affected.

An intensive observational study during the winter housing season of 2007 (January to May 2007) was conducted on the index farm (*Study 1*). However, this study failed to detect a period of poor growth, and the cause of poor performance on the index farm remains uncertain. Acceptable live weight gains were achieved during the 18-week period of observation, equivalent to 0.66 and 1.00 kg/animal/day for index-farm sourced cattle and for off-farm sourced cattle, respectively (*Study 1*). Evidence of selenium (Se) deficiency was demonstrated in the cattle raised on the index farm, characterised by low whole blood concentrations of Se and glutathione peroxidase (GPx). The Se status of the off-farm sourced cattle, originating from a dairy farm in Co. Meath, was initially good, as expected in the Meath area (Fleming and Parle, 1987). Whole blood Se concentrations decreased by 35 % over this 18 week-study period, and GPx concentrations decreased by 27 % in the off-farm sourced cattle. Selenium responsive stunting of cattle and poor productivity has been reported in cattle in Se deficient areas worldwide, particularly in New Zealand and Canada. Despite their deficient Se status, over the winter trial period the index sourced cattle kept on the index farm gained 0.66 kg/day. There were haematological changes in cattle on the index farm during the intensive winter trial period: packed cell volume (PCV) was within the normal

range for cattle, the red blood cell count (RBC) was high and the mean cell volume (MCV) low, perhaps suggestive of alterations to the synthesis of new red blood cells on the index farm.

This study was conducted with ten Aberdeen Angus cross Friesian cattle (AAX) that had been introduced onto the index farm in January 2007. The aim of this study was to determine whether periods of poor weight gain or weight loss, if observed, were associated with changes in haematological parameters, essential element status, or exposure to heavy metals.

## **MATERIALS AND METHODS**

### **Immediately prior to the period of interest (December 2006)**

Aberdeen Angus crossbred Friesian cattle (n=10) were sourced off farm from a commercial farm in Co. Meath, located approximately 110 km from the index farm. These cattle formed part of a treatment group (off farm sourced cattle and kept on the index farm) for the work described in *Study 1*, and were kept on the index farm for a further 12 months after the end of *Study 1* to characterize changes in haematological parameters, essential element status, or exposure to heavy metals. For completeness, this study describes changes in these cattle from introduction onto the index farm in January 2007 until their sale in April 2008.

As part of the work described in *Study 1*, prior to the initiation of this study, all animals were vaccinated for bovine pneumonia complex with Bovipast RSV (Intervet, The Netherlands; 5 mls intra-muscularly (im); twice at appropriate intervals) and Bovilis IBR marker (Intervet, The Netherlands; 2 mls; intra-nasally). Additionally, at the same time, animals were vaccinated for salmonellosis (Bovivac S, Intervet, The Netherlands; 5 mls; im; twice at appropriate intervals). Serological evidence of infection with BHV-1, BVDV, *Salmonella dublin* and *S. typhimurium* and fluke infestation were determined, and a parasiticide and flukacide were administered to all animals.

### **During the observation period (January 2007 to April 2008)**

The study period was 469 days in duration. Observation began on the 16<sup>th</sup> January 2007 (Day 0) and ended on the 29<sup>th</sup> April 2008 (Day 469).

### *Animals*

Ten AAX beef cattle, aged 266 to 345 days and weighing 244 to 339 kgs, were introduced onto the index farm located in Castlecomer, Co. Kilkenny, on 14<sup>th</sup> January 2007. During the subsequent summer period (08/05/2007 to 02/11/2007) they were kept at pasture on the index farm. During each winter period (14/01/2007 to 08/05/2007 and 02/11/2007 to 29/04/2008) they were housed in a slatted floor unit and fed a diet of home produced grass silage and 2 kg of commercial beef ration per animal per day during their first winter and 2 kg/day during the second winter period, calculated to result in an average daily gain (ADG) of 0.75 kg/day. Animals were weighed on 12 occasions over the 16 months; 16/01/2007 (Day 0), 08/05/2007 (Day 112), 07/08/2007 (Day 203), 04/09/2007 (Day 231), 02/10/2007 (Day 259), 30/10/2007 (Day 287), 27/11/2007 (Day 315), 08/01/2008 (Day 357), 06/02/2008 (Day 386), 04/03/2008 (Day 413), and 02/04/2008 (Day 442) and 29/04/2008 (Day 469); using an electronic weighing scale system (Tru Test 2000 series, Auckland 1730, New Zealand) and ADG (kg/day) were calculated for each time period.

### *Blood sampling*

Blood samples were collected, by jugular venepuncture, on 13 occasions; 16/01/2007 (Day 0), 08/05/2007 (Day 112), 19/06/2007 (Day 154), 07/08/2007 (Day 203), 04/09/2007 (Day 231), 02/10/2007 (Day 259), 30/10/2007 (Day 287), 27/11/2007 (Day 315), 08/01/2008 (Day 357), 06/02/2008 (Day 386), 04/03/2008 (Day 413), and 02/04/2008 (Day 442) and 29/04/2008 (Day 469) into commercially available plain (with activated clotting factors), lithium heparin, and EDTA blood tubes (BD Vacutainer Systems, Plymouth, UK). Duplicate sera samples were obtained at each sample point, and banked for possible future analysis. Sera were obtained after centrifugation of blood samples at 1,600 X g for 20 min, following storage at room temperature for approximately 5 h (to facilitate transport to the laboratory). Whole blood (EDTA) was submitted to the University Veterinary Hospital's (UVH) Clinical Pathology suite for routine haematology analysis and for determination of GPx concentrations within 8 h of collection. Whole blood (lithium heparin) samples were submitted to a commercial laboratory (Independent Analytical Services Ltd., (IAS), Bagenalstown, Co Carlow) within 6 h of collection for whole blood essential element and heavy metal analysis.

### **Laboratory Analyses**

#### *a) Complete blood counts (CBC)*

Whole bloods (EDTA) were analysed approximately once monthly for routine haematology parameters using the Abbott CELL-DYN® 3500R (Abbott Laboratories Ireland Ltd.,

Citywest Business Campus, Dublin 24, Ireland) veterinary package. The CELL-DYN® 3500R uses flow cytometry to count and identify blood cells (Feldman *et al.*, 2006). Daily quality control samples were run on the analyser to determine the performance of the machine.

*b) Essential element, GPx status and heavy metal status*

As an estimate of Se status, GPx was determined using a Randox Imola RX Clinical Analyser, (Randox Laboratories Ltd., Crumlin, Co. Antrim, Northern Ireland) by the Ransel method as per kit instructions. Whole blood digestion for mineral analysis was conducted at a commercial laboratory (Independent Analytical Services Ltd., (IAS), Bagenalstown, Co. Carlow, Ireland). Briefly, minerals in whole blood samples were dissolved in concentrated nitric acid during microwave digestion. The sample was digested in a Milestone ETHOS EZ microwave system and APCU-TR40 pressure vessels using the following programme: sample size: 3.0 ml, Acid: 7 ml concentrate HNO<sub>3</sub>, Ramp time: 15 min to 180 °C, hold time: 10 min at 180 °C, Cooling: cooled to 50 °C (circa 1.5 hours). Digested samples were then made up to 25 ml with de-ionised water and filtered through a Whatman 540 filter paper. Samples were then packaged and couriered to an accredited laboratory (INDIKATOR GmbH, D-42329 Wuppertal, Germany) for whole blood essential element and heavy metal analyses using an Elan Inductively Coupled Plasma with a Mass Spectrograph Detector (ICP-MS) using certified standards. Blood samples were analysed for Copper (Cu), Zinc (Zn), Se, Sulphur (S), Phosphorus (P) Iron (Fe) and Cadmium (Cd). Results were expressed as mg/kg for S, P and Fe, while results for Cu, Zn, Se, Cd were expressed in µg/kg. Determination boundaries for ICP-MS analyses were as follows: Cu: 1 µg/kg; Zn: 1 µg/kg, Se: 5 µg/kg, Cd: 0.2 µg/kg, S: mg/kg, P: 1 mg/kg, Fe: 0.02 mg/kg. In the case where outlier results were reported, a repeat run was conducted by the laboratory. Following completion of the laboratory analyses heavy metal analyses for whole blood cadmium concentrations were considered to be unusually high, a proportion (20 %) of the samples were re-assayed by individual animal in date order to ensure that the results were correct. Whole blood samples submitted to IAS in January and February 2008 are unaccounted for, hence these results are unavailable.

*Monitoring of disease in the herd*

A visual clinical assessment of disease status was conducted on each animal at each weighing point. Animals were carefully observed to determine presence of normal demeanour and behaviours. External signs of clinical disease including nasal and ocular discharge, evidence of scour, poor gut fill, lameness, unusual swellings or asymmetry were noted. Coughing or

increased respiratory effort was observed for on every occasion. Detailed hands-on clinical examinations (including auscultation and assessment of temperature) were not conducted.

### *Statistical analyses*

A repeated measures model using the MIXED procedure in SAS 9.1 (Sas Institute Inc., 2003) was developed for each outcome measure. The distance between each time period that the measurement was taken was not equally spaced. This was accounted for by selecting either a spatial correlation (power, Gaussian, linear or spherical), unstructured or compound symmetry structure to account for the correlation between measurements within the same animal. The appropriate correlation structure was chosen based on the AICC and BIC from the model. Trends over time, by farm, or source or both, were tested in a random coefficient model. When there was no significant trend a repeated measures model was used.

The independent variables considered in each model were: DAYS, SEX and BASE\_VARIABLE. The baseline measure (BASE\_VARIABLE) was the measurement taken on the first day of the study. The two-way interaction between SEX and DAYS was tested in the models. A backward selection procedure was used to eliminate terms from the model ( $P > 0.05$ ). Residual and influence plots were used to assess the overall fit of the final model and to identify potential outliers. Outcome measures were appropriately transformed (e.g. log transformation) if the residual plots indicated that this was necessary. Days were compared using a multiple comparison correction (the False Discovery Rate method; FDR).

## **RESULTS**

### *Weights and average daily gains*

The mean age of the ten AAX cattle was  $320 \pm 7.2$  days and the mean weight was  $279 \pm 10.0$  kg at introduction onto the index farm in January 2007 (Day 0). Assuming a birth weight of 40 kg, the average daily gain (ADG) prior to introduction to the index farm was  $0.75 \pm 0.04$  kg/day. The mean live weight gain of the ten AAX cattle between January 2007 and April 2008 (Days 0 and 469) was 271 kg, equivalent to an ADG of 0.58 kg/day over the 16 month period on the index farm. Mean weight increased ( $P < 0.01$ ) at every successive weighing point, except between Days 112 and 259 (May and October 2007) when the cattle did not gain weight ( $P < 0.05$ ). The ADG of the ten cattle was above target ( $0.96 \pm 0.05$  kg/day) during their first housing period on the index farm between Days 0 and 112 (Jan to May 2007), but

ADG decreased ( $P < 0.05$ ) to  $0.13 \pm 0.03$  kg/day during the summer period between Days 112 and 259 (June to October, 2007) while at pasture. Over the second winter housing period (November 2007 to April 2008) the ADG improved ( $P < 0.05$ ) to  $0.77 \pm 0.04$  kg/day. The mean weight and ADG ( $\pm$  SEM) of the ten cattle at each weighting point are presented in Figure 1. As demonstrated by the standard error bars; individual cattle weights did not exhibit much variation at each weighting point and the ADGs were not influenced by outliers. Once the study was terminated on Day 469 (29<sup>th</sup> April 2008), the cattle were sold at a commercial mart so further weight records were not available for analyses. The nearby brick factory was in operation for ten months of the 16 month trial period; production commenced on 20<sup>th</sup> March 2007 and ceased on 22<sup>nd</sup> of December 2008. Overall, the ADG for the ten AAX cattle while the nearby brick factory was open was  $0.21 \pm 0.03$  kg/day compared with  $0.78 \pm 0.04$  kg/day while the brick factory was closed.

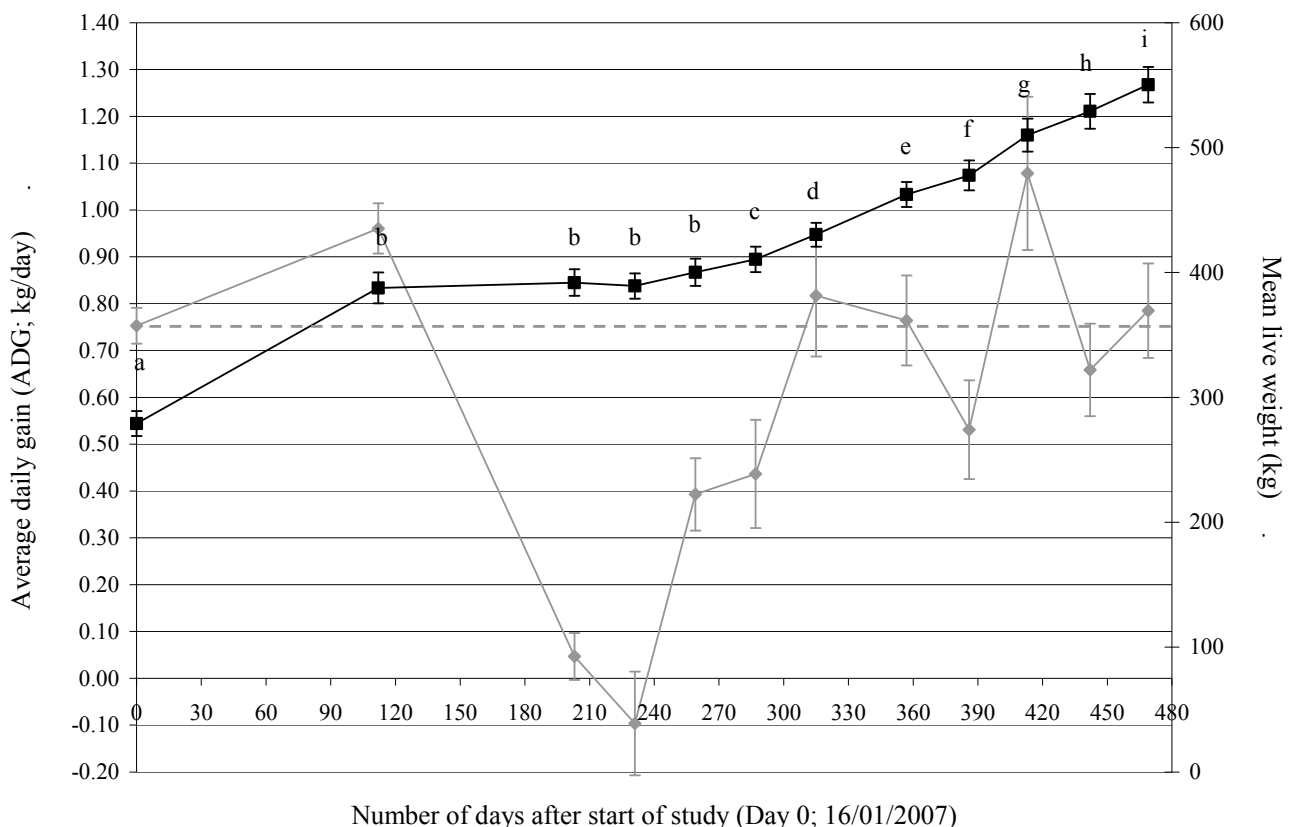


Figure 1. The mean weight  $\pm$  SEM of the ten Aberdeen Angus crossbred Friesian cattle increased ( $P < 0.0001$ ) from the start of the study period in January 2007 (Day 0) until the end of the study in April 2008 (Day 469; black line; ■); however, no significant live weight gains were observed between Days 112 and 259 (May and

October 2007). Mean average daily gains (ADG;  $\pm$  SEM; grey line;  $\blacklozenge$ ) of the animals was lower ( $P < 0.001$ ) between Days 112 and 231 (May and September 2007) compared with all other times. <sup>a,b,c,d,e,f,g,h,i</sup> Mean weights with different superscripts are different ( $P < 0.01$ ).

### *Animal Health*

No animal health concerns were observed during the study period.

### *Haematology*

The mean  $\pm$  SEM white blood cell count (WBC) for the ten AAX cattle varied between  $6.79 \pm 0.79$  and  $10.67 \pm 0.76 \times 10^9$  cells/l throughout the 16 month period and was always within the normal range for adult cattle. The mean WBC was increased ( $P < 0.05$ ) at the two sample points in June (Day 154) and August 2007 (Day 203) compared with all other sampling points, with the exception of the sampling point in May (Day 112) and the two sample points in October 2007 (Days 259 and 287). The proportion of neutrophils was not altered at any time point throughout the trial period. The proportion of eosinophils, expressed as a percentage of the total white cell count, increased ( $P < 0.001$ ) from  $1.16 \pm 0.40$  in May 2007 (Day 112) to a peak of  $11.43 \pm 2.01$  in September 2007 (Day 231), and subsequently decreased; this peak coincided with the end of the period characterised by poor live weight gains during the summer season at pasture (Figure 2).

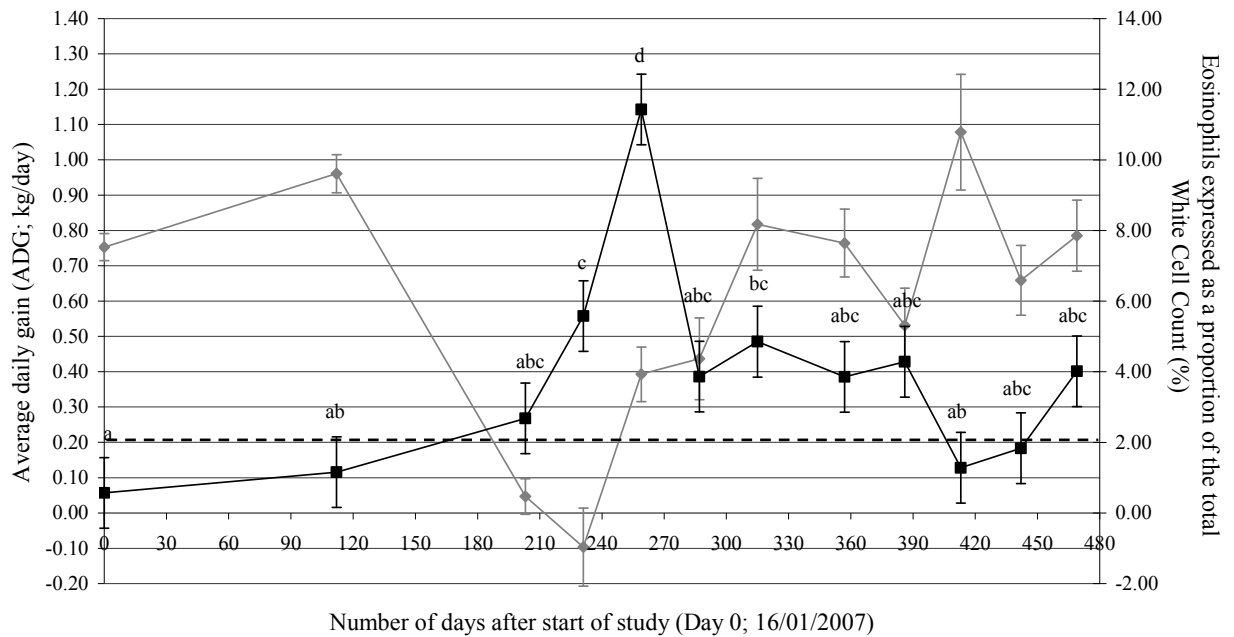


Figure 2. The proportion of eosinophils (expressed as a percentage of the total White Cell Count (WBC x 10<sup>9</sup>/l; %; black line; ■). The horizontal dashed line at the base of the figure represents the upper limit of the normal range for proportion of eosinophils in cattle blood. The mean average daily gains (ADG; grey line; ◆); of the animals between Days 0 and 469 (January 2007 and April 2008); <sup>a,b,c,d</sup>Means within line with different superscripts are different (P < 0.05)

The number of red blood cells (RBC) was increased above the normal range (4.5 to 7.0 x 10<sup>12</sup> cells/l) for adult cattle throughout the study period ranging from a mean of 6.77 ± 0.14 to 8.66 ± 0.18 (x 10<sup>12</sup> cells/l); while mean haemoglobin (Hgb) remained within the normal range (9.0 to 14.6 g/dl) throughout. However, after introduction onto the index farm the mean number of RBC in the AAX cattle decreased (P < 0.001) and remained lower throughout the period of the study, although values fluctuated (P < 0.05). The mean haemaglobin concentration varied throughout the study period, and for the most part, reflected changes in mean RBC concentration. The mean PCV for the ten AAX cattle fell within the normal range (0.25 to 0.36 l/l), apart from last bleed, Day 469, where mean PCV was just increased above the normal range (0.37 l/l). The mean value for mean cell volume (MCV; fl) was lower than the normal range when the ten cattle were introduced onto the farm. The mean cell volume increased (P < 0.001) after introduction and remained consistently at the low end of the

normal range (40 to 60 fl) between Day 112 and 259, after which mean cell volume increased ( $P < 0.05$ ) and continued to rise ( $P < 0.01$ ) until the end of the study on Day 469 (Figure 3). The same trends are observed for the mean cell haemoglobin (MCH pg; Figure 3). Mean cell haemoglobin content varied within the normal range throughout the study period until an increase ( $P < 0.001$ ) at the last three bleeding points between Day 413 and 469; all values were within the normal range (30 to 60 g/dl) apart from at the final bleed, Day 469, when it exceeded the normal range. The mean number of platelets did not differ significantly throughout the study period and were consistently at the low end of the normal range (100 to  $800 \times 10^9$  platelets/l).

#### *Essential elements, GPx and heavy metals*

The mean concentration of GPx (U/mlPCV) in the ten AAX cattle was 142.5 U/mlPCV on introduction onto the farm on Day 0, substantially above the threshold value of 40 U/mlPCV; indicative of normal Se status. Whole blood concentrations of GPx decreased ( $P < 0.0001$ ) over time from introduction at Day 0; reaching nadir concentrations of 33 U/ml/PVC on Day 315 (November 2007; Figure 4). Animals were considered to be Se deficient, based on GPx status, by Day 315. However, between Days 315 and 469 (November 2007 and April 2008), concentrations of GPx increased ( $P < 0.0001$ ; Figure 4). The same trend was evident in whole blood concentrations of selenium; mean whole blood Se concentrations were 194  $\mu\text{g}/\text{kg}$ , in the ten AAX cattle on Day 0, above the threshold value of 120  $\mu\text{g}/\text{kg}$ , indicative of normal Se status. Whole blood Se concentrations decreased ( $P < 0.0001$ ) between Days 0 and 259 (January and October 2007), reaching nadir concentrations in October 2007 (Day 259). Due to a technical issue in the laboratory, whole blood mineral analysis was not conducted in November 2007 and January 2008. Therefore, whether the value in October represents the actual nadir value cannot be determined. However, whole blood Se increased ( $P < 0.0001$ ) between Days 287 and 469; (Figure 4). Animals were considered to be Se deficient, based on Se status between Days 154 and 413 (May 2007 and March 2008).

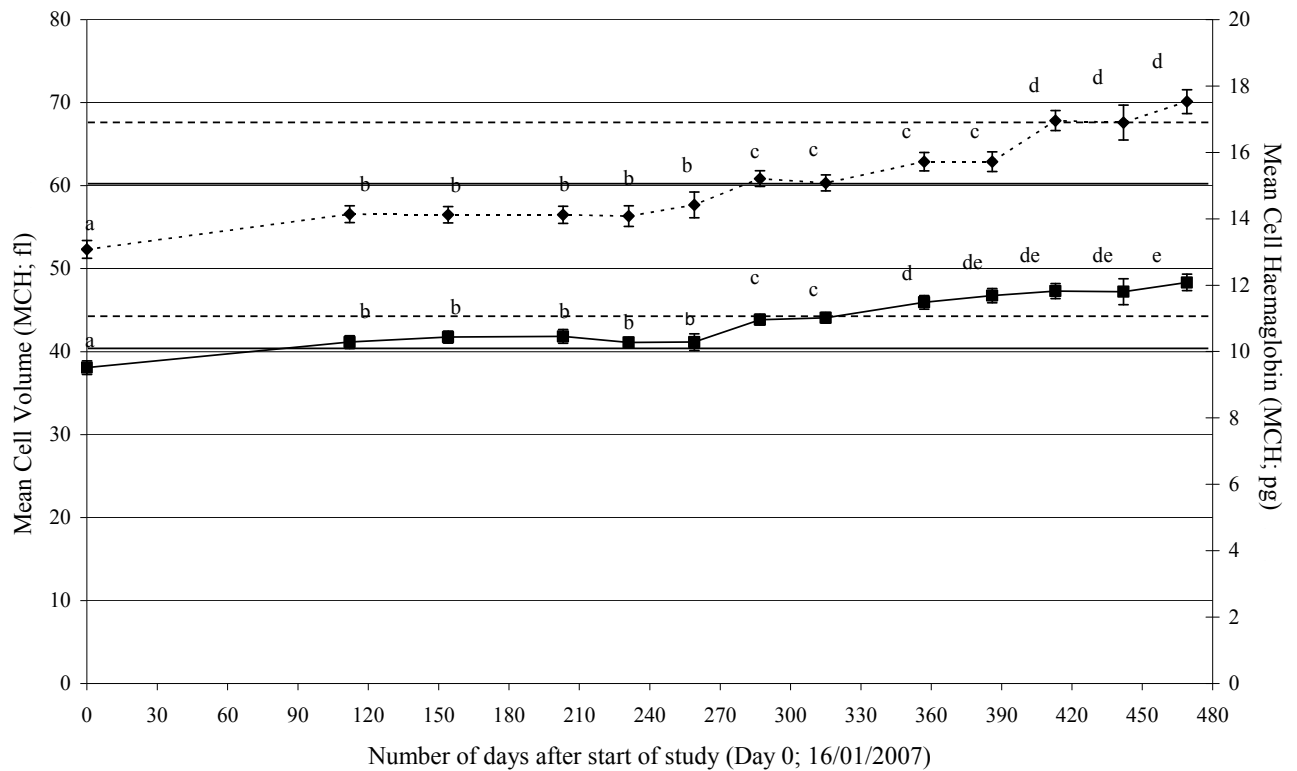


Figure 3. The mean  $\pm$  SEM mean cell volume (MCV; fl; solid line; ■) and the mean cell haemoglobin (MCH; pg, dashed line; ♦) for the ten Aberdeen Angus crossbred Friesian cattle kept on the index farm between Days 0 and 469 (January 2007 to April 2008). The normal range for the MCV is indicated by the solid black horizontal lines. The normal range for the MCH is given by the dashed black horizontal lines. <sup>a,b,c,d,e</sup>Means within line with different superscripts are different ( $P < 0.05$ )

Whole blood mineral analysis suggests that cattle had normal Cu status prior to introduction. Mean whole blood Cu concentrations decreased ( $P < 0.01$ ) in the ten AAX cattle between Days 0 and 112 (January and May 2007) and throughout the rest of the study period the ten cattle were considered Cu deficient, i.e. whole blood Cu concentrations were less than 1000  $\mu\text{g}/\text{kg}$  (Figure 5). Whole blood Cu concentrations continued to decrease ( $P < 0.001$ ) and reached nadir concentrations (mean concentrations 687  $\mu\text{g}/\text{kg}$ ) by Day 287 in October 2007. Copper concentrations remained low for the remainder of the study period.

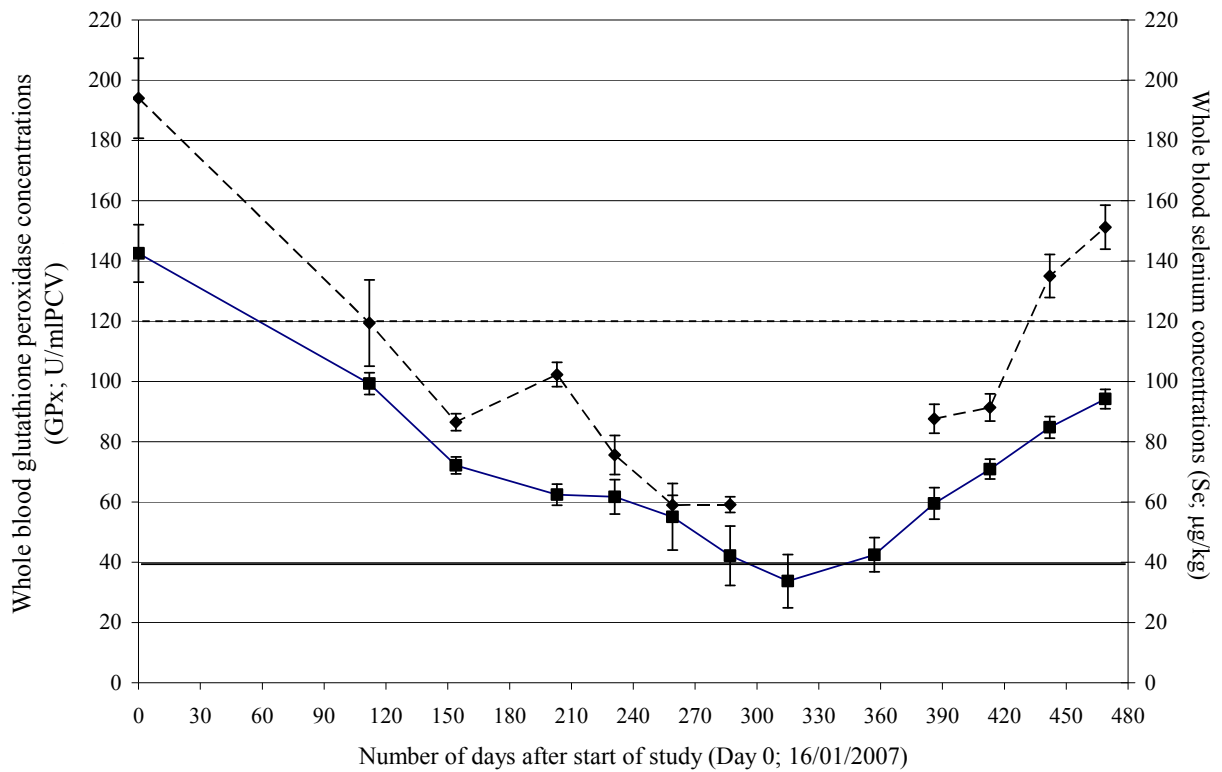


Figure 4. The mean  $\pm$  SEM whole blood concentrations of glutathione peroxidase (GPx) expressed relative to Packed Cell Volume (PCV; U/mlPCV; solid line; ■) and selenium (Se;  $\mu\text{g}/\text{kg}$ ; dashed line; ♦) in the ten Aberdeen Angus crossbred Friesian cattle kept on the index farm between Days 0 and 469 (January 2007 and April 2008). Selenium deficiency is suspected when Se concentrations are less than 120  $\mu\text{g}/\text{kg}$ ; indicated by the dashed horizontal line or when GPx relative to PCV is less than 40 U/ml PCV; indicated by the solid black horizontal line. Whole blood Se concentrations were not available for November 2007 (Day 315) and January 2008 (Day 357).

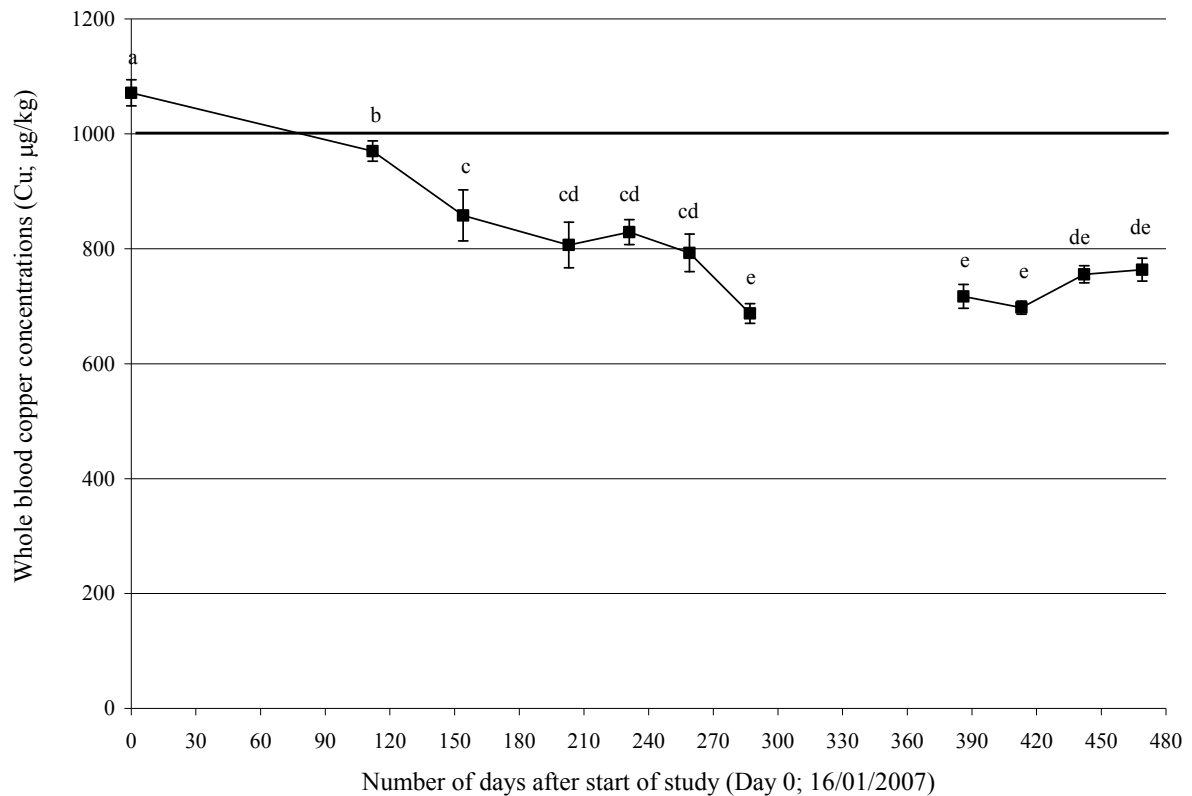
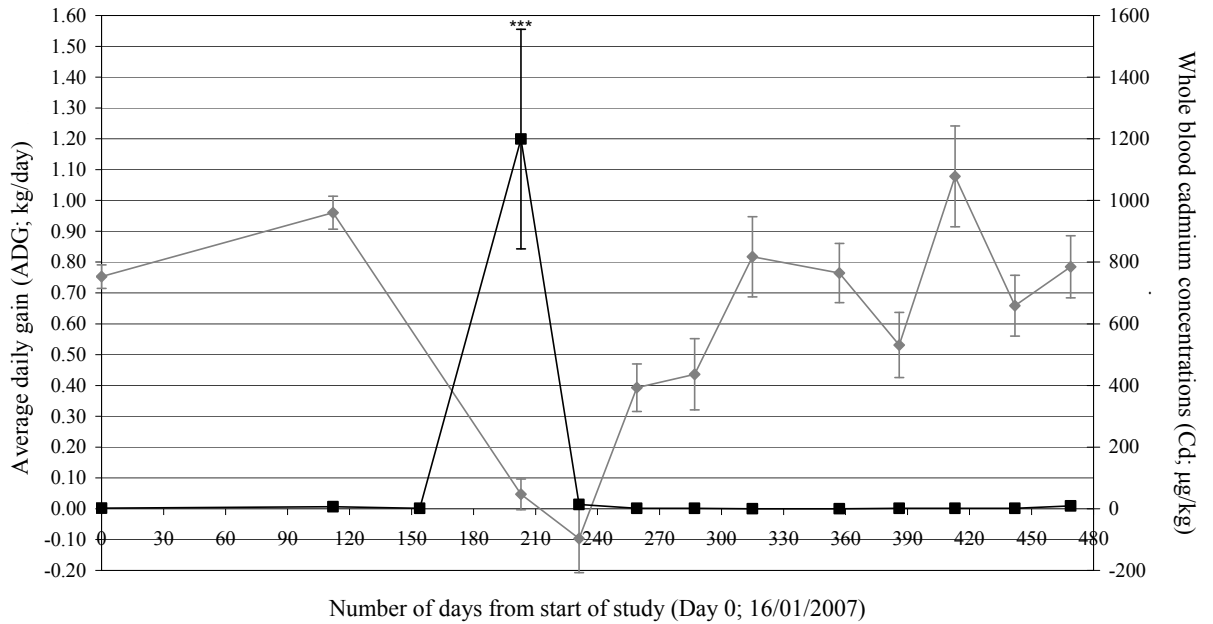


Figure 5. The mean  $\pm$  SEM whole blood concentrations of copper (Cu;  $\mu\text{g}/\text{kg}$ ; solid line;  $\blacksquare$ ) in the ten Aberdeen Angus crossbred Friesian cattle kept on the index farm between Days 0 and 469 (January 2007 and April 2008). The suggested lower threshold for whole blood Cu concentrations is indicated by the solid black horizontal line. Whole blood Cu results are not available for Days 315 (November 2007) and 358 (January 2008). <sup>a,b,c,d,e</sup>Means with different superscripts are different ( $P < 0.05$ ).

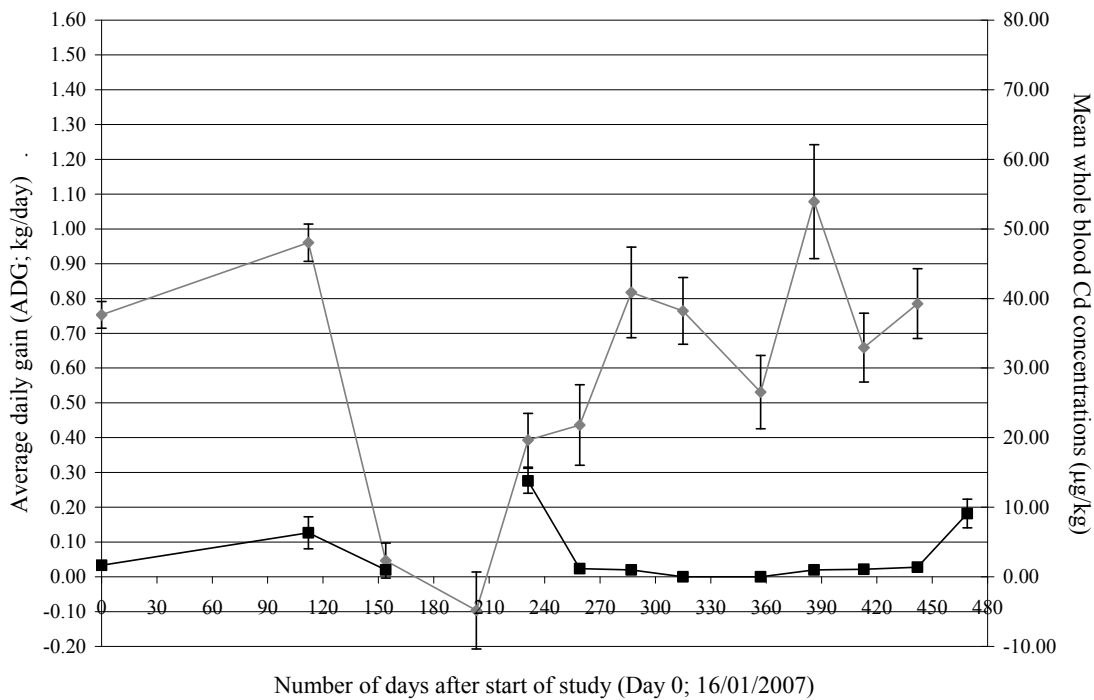
Mean whole blood Cd concentrations were 1.66  $\mu\text{g}/\text{kg}$  in the ten AAX cattle at introduction onto the index farm on Day 0. Animals with non detectable whole blood Cd concentrations were assigned a value of half the detection limit to allow for calculations of means. Mean Cd concentrations were not different between Days 0 and 154 (January and June 2007). Whole blood Cd concentrations peaked ( $P < 0.0001$ ) on Day 203 in August 2007 and were higher ( $P < 0.0001$ ) compared with all other time points (Figure 6). Cadmium concentrations decreased ( $P < 0.0001$ ) and remained lower ( $P < 0.0001$ ) throughout the rest of the study period. Whole blood concentrations of S increased ( $P < 0.02$ ) after animals were introduced onto the index

farm. The concentration of whole blood S peaked ( $P < 0.01$ ) on Day 203 in August 2007. Subsequently, whole blood concentrations of S decreased ( $P < 0.01$ ) and remained lower throughout the rest of the study period (Figure 6).

Panel a)



Panel b)



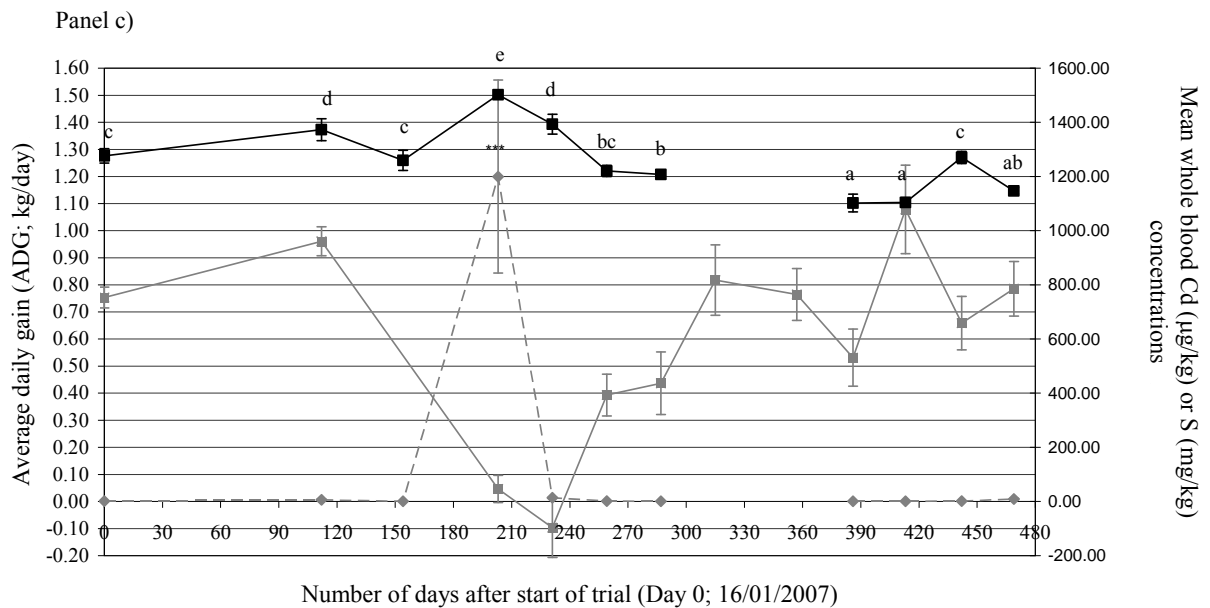


Figure 6. The mean  $\pm$  SEM concentrations of whole blood cadmium (Cd;  $\mu\text{g}/\text{kg}$ ; solid line; ■; panel a); mean  $\pm$  SEM concentrations of whole blood cadmium with peak concentrations removed (Cd;  $\mu\text{g}/\text{kg}$ ; solid line; ■; panel b) and whole blood sulphur (S) concentrations (mg/kg; solid line; ■) and Cd concentrations (dashed line; ◆; panel c) in the ten Aberdeen Angus crossbred Friesian cattle kept on the index farm between January 2007 and April 2008. Whole blood Cd and S results for November 2007 and January 2008 were not available. The mean average daily gains (ADG; grey columns) of the animals between Days 0 and 469 (January 2007 and April 2008). <sup>a,b,c,d,e</sup>Means within line with different superscripts are different ( $P < 0.05$ ). \*\*\*A peak ( $P < 0.0001$ ) in whole blood cadmium concentrations was detected on Day 203 in August 2007).

## DISCUSSION

### *Live weight gains*

The mean ADG of the ten AAX cattle introduced onto the index farm on Day 0 in January 2007 were very good (0.96 kg/day) during their first housing period on the index farm, between January and May 2007 (Days 0 and 112). These gains were as expected, noting that cattle had *ad libitum* access to a diet of grass silage and concentrate, calculated to ensure that

they received at least 2 kg of concentrate per head. The minimum recommended concentrate supplementation for a dairy to beef production system in an Irish context is 0.5 kg/day (Keane, 2001). Nonetheless, the period of summer grazing at pasture was associated with very poor ADG of 0.13 kg/day, between Days 112 and 287 (May to October, 2007). Indeed, after yearling turnout (8<sup>th</sup> May 2007; Day 112), a period of weight loss occurred during the first three-month period of the summer with cattle losing 0.05 kg/day, a period that is normally associated with excellent growth rates (Keane, 1998; 2001; 2002a; 2002b; 2008). The live weight gains reported in the present study are at odds with work conducted by Keane (2008) at the Teagasc Grange Research Centre, Co. Meath, Ireland, a centre that conducts research into commercial dairy to beef grass production systems in Ireland. Keane (2008) recorded ADG of 0.98 and 0.86 kg/day for the Friesian (FR) and AAX cattle groups during the first two months after turnout as yearlings, respectively. Some consideration must be given to the production potential of the index farm in comparison to that of the Grange research facility. Certainly, the gley soil type typical of the index farm, and throughout much of the Castlecomer plateau area, have poor physical conditions that make them unsuitable for cultivation or intensive grassland production (Gardiner and Radford, 1980). On the other hand, consideration must also be given to the fact that cattle on the index farm were housed and fed substantial levels of concentrates during the winter periods, thus lessening the effect of the lower quality farmland. An earlier study conducted at the same research centre reported ADG of 0.78 and 0.80 kg/day for FR and Charolais x Friesian (CHX) yearlings, with gains of 0.90 and 0.94 kg/day recorded in the same two groups during their two year old summer at pasture (Keane, 1998). Comparable research using CHX weanlings, demonstrated excellent ADGs (0.83 and 0.86 kg/day) for cattle fed silage and 2 kg concentrate indoor the previous winter, during a 218 day summer grazing season. These mean gains varied from 0.64 to 1.23 kg/day at different time points throughout the grazing season (Keane, 2002b). Indeed, CHX weanlings fed silage and 3 kg concentrate per day during a 122 day winter housing period, followed by turnout to pasture, gained a mean of 0.82 kg/day (varying from 0.51 to 1.18 kg at different periods) over the summer grazing period (Keane, 2002b); while similar work (Keane, 2002a) reported ADG of 0.95 and 0.88 kg/day over the second summer grazing season. Clearly, FR and Friesian crossbred (FRX) cattle in Irish dairy to beef production systems are capable of very good growth rates during the summer grazing period of their yearling year. The poor growth rates observed in the present study are not typical of those reported by the work conducted by Teagasc. However, based on the analysis of animal live weight data collected on the index farm between 2003 and 2006; these periods of poor live weight gains or periods of weight loss have occurred on a number of occasions on the index

farm (*Overview*, Appendix 2). In the present study, grazing yearlings were rotated between a number of different fields close to the farmyard. The grazing paddocks had a good cover of grass on all visits. The animals were monitored for evidence of clinical disease, by clinical observation and routine haematology throughout the period (at a minimum of fortnightly visits to the farm). Clinical endo-parasitic infestation was not evident. In addition, a generic ivermectin was administered parenterally on four occasions during the summer grazing period. It is noteworthy that all cattle in the group experienced the down turn in weight coincidentally, suggestive of a single insult at a defined period of time. Average daily gains improved after housing at the start of the winter period in early November 2007 when cattle were given *ad libitum* access to grass silage and fed 2 kg of concentrate. Management, nutrition, clinical disease, or parasitism, do not explain these poor live weight gains during the summer of 2007. Throughout their second winter on the index farm, the AAX cattle mean ADG varied between 0.55 and 1.2 kg/day, which is within the accepted range for cattle in similar production systems.

#### *Selenium and GPx*

Selenium is an essential for growth and fertility in animals. It is also essential for the prevention of a spectrum of diseases, which show a variable response to treatment with Se and Vitamin E. Selenium plays a role in many processes in the body. It is a component of twenty two known seleno-proteins, in the form of amino acid selenocysteine (Sec), of which GPx, an antioxidant, is the most recognised. Adverse effects on livestock growth and body condition (Drake *et al.*, 1969; Veling and Counotte, 1995), fertility (Hartley, 1963; Tasker *et al.*, 1987) and health have been demonstrated in Se deficient regions (Hartley 1967, Andrews *et al.*, 1968, McDonald 1975, Wichtel *et al.*, 2004). Indeed, a growth response to Se supplementation in normal lambs has been reported (Hartley and Grant 1961; McLean *et al.*, 1963). A review by Wichtel (1996) suggests that Se deficiencies are an important source of loss to New Zealand agriculture, a country where approximately 30 % of farmland is considered to be Se deficient. Ill thrift, decreased fertility, decreased milk yields and white muscle disease (WMD) have all been reported. Despite the low Se intake, however, the incidence of White Muscle Disease in New Zealand is low. Clinical occurrence of WMD has not been reported by the private veterinary practitioners on the index farm.

Selenium status of the index farm is clearly problematic. The ten AAX cattle had normal whole blood concentrations of Se and GPx prior to their introduction onto the index farm in January 2007 (Day 0). Selenium status (determined by both whole blood Se and GPx

concentrations) decreased between January and November, 2007, and the animals were considered Se deficient by June 2007, and remained so into March 2008. Despite their low Se status, whole blood GPx concentrations did not indicate that the cattle were GPx deficient until November 2007, GPx status improved after November and by the end of the study period mean whole blood GPx concentrations were 95 U/mlPCV. It is possible that the increase in GPx could be attributed to the change to the winter diet; cattle were housed in the beginning of November 2007, and were receiving approximately 2 kg concentrate daily, this should certainly improve GPx status. The difference in the response to winter-feeding during the two winter housing periods on the index farm must also be rationalised. In their first housing period on the index farm, during the intensive winter trial (January to May 2007), Se status of the ten AAX cattle decreased despite concentrate supplementation, however, during their second winter housing period GPx and Se concentrations improved. It is certainly possible that the response of blood concentrations of GPx and Se is dependent on the status of the animal at concentrate introduction. In this case, the AAX cattle introduced to the farm in January 2007 were of high Se status, so being fed a diet consisting of grass silage produced on a farm with low and deficient soil and herbage Se, would decrease Se status despite concentrate supplementation. However, during their second winter period on the index farm, these cattle were now classified as Se deficient, and hence it is likely that the concentrate feeding did contribute to their improved Se status by the end of the study period. Nadir Se concentrations were reached in October (Se) and November (GPx) 2007, and these animals were considered deficient until March 2008. Regardless, this was coincident with an improved ADG; therefore these animals were able gain weight while Se deficient. Heavy metal exposure may interfere with Se and GPx. Work conducted by Wasowicz *et al.* (2001) suggests that Cd exposure decreases the activity of antioxidant enzymes and is associated with lowered Se status.

### *Copper*

The ten AAX cattle became Cu deficient following introduction onto the index farm, which may have contributed to poor animal performance. Copper deficiency in ruminants is associated with a number of diseases such as bone fragility, steely wool and depigmentation of wool and hair (Cunningham, 1950). Work conducted by Howell (1968) demonstrated an experimentally induced state of Cu deficiency in ewes resulted in decreased growth rates, lowered fertility and decreased haemopoiesis. However, the effect of Cu supplementation to improve performance in low Cu status animals under commercial conditions was less evident (Howell, 1968). The interactive effect of combinations of Se and Cu has been reported in a

number of studies (Hill, 1969; Thomson and Lawson, 1970). Early research in New Zealand, reported improved growth rates of lambs when receiving a combination of Se and Cu compared with Se alone (Hill, 1969). Heavy metal exposure may interfere with Cu status. Dietary Cd fed to sheep has been reported to decrease liver concentrations of Cu (Mills and Dalgarno, 1972; Doyle and Pfander, 1975). Campbell *et al.* (1979) suggests that the adverse effects of Cd exposure can be improved by supplementation with Cu.

### *Haematology*

Alterations in haematological parameters have also been characterised in the present study. Following their introduction onto the index farm, the red blood cell count decreased and remained lowered throughout the period of the study in the AAX cattle, although MCV and MCH increased sequentially as the time progressed. An eosinophilia was determined in the AAX cattle in early September 2007. Although most commonly associated with parasitism, in this study an injectable parasiticide (an ivermectin based product) was administered on four occasions throughout the season at pasture. This parasiticide was administered according to the manufacture's instructions and based on the animal's actual live weight, by personnel from CVERA on the first two occasions and by the farmer on the second two occasions. During fortnightly farm visits by CVERA staff throughout this summer period, no clinical signs of parasitism such as coughing or diarrhoea were apparent.. No change in live weight gain was attributable to the administration of the parasiticide on any of the four occasions. The cause of the eosinophilia in the AAX cattle is uncertain.

### *Cadmium*

A peak in whole blood Cd concentrations was identified in August 2007 in the ten AAX cattle. These results gave rise to further investigations, a detailed review of the scientific literature (in Appendix 7, accompanying the *Overview*) and consultations with international experts. A detailed discussion of these results is presented in the *Overview*, and is not considered further here.

## CONCLUSION

Weight gains were intermittent during the period of observation: very good ADGs were observed during the initial housing period (January to May 2007; 0.96 kg/day), very poor during summer grazing (May to October 2007, 0.13 kg/day) and good during the subsequent housing period (October 2007 to April 2008, between 0.55 and 1.2 kg/day). During the

summer grazing period, the cattle experienced a coincident reduction in weight gain, which is suggestive of a single insult over a defined period of time. At this time, there was no overt evidence of disease in these cattle. Adequate ADGs during summer 2007 have been reported elsewhere in Ireland. Selenium concentrations declined during the study period and were considered to be deficient from October 2007 to January 2008. However, the concentrations improved on winter feeding and returned to normal by February 2008. Copper (Cu) concentrations declined after cattle were introduced onto the index farm, and were considered deficient from May 2007 until the end of the study. In this study, essential element deficiencies were again identified as potential contributors to the ongoing performance problems on the index farm. A single Cd peak was measured in cattle during this study, the significance of which is uncertain but subject to further study.

## REFERENCES

- Andrews ED, Hartley WJ, Grant AB 1968 Selenium-responsive diseases of animals in New Zealand. *NZ Vet J* 16: 3-17
- Cunningham IJ 1950 Copper and molybdenum in relation to diseases in cattle and sheep in New Zealand. In: Copper Metabolism – a symposium on animal, plant and soil relationships WD McElroy, B Glass (Eds) John Hopkins Press, Baltimore, USA pp 246-273
- Doyle JJ, Phander WH 1975 Interactions of cadmium with copper, iron and manganese in ovine tissues *J Nutr* 105: 599-606
- Drake C, Grant AB, Hartley WJ 1969 Selenium and animal health Part 1. The effect of a tocopherol and selenium in the control of white muscle disease in lambs *NZ Vet J* 8:4-6
- Fleming GA, Parle PJ 1987 Trace elements in Irish soils – Content and distribution In: Trace elements and heavy metals in Irish soils McGrath D, Fleming GA, Culleton N (eds) Teagasc, Environment Research Centre, Johnstown Castle, Wexford, Ireland pp 1-15, 2008
- Gardiner M, Radford T 1980 Soil associations of Ireland and their land use potential. Soil Survey Bulletin No. 36. An Foras Taluntais
- Goyer RA 1995 Nutrition and Metal toxicity *Am J Clin Nutr* 61(Suppl): 646-650S
- Hartley WJ 1963 Selenium and ewe infertility *Proc NZ Soc Anim Prod* 23:20-27

- Hartley WJ 1967 Levels of selenium in animal tissues and methods of selenium administration. In: Selenium in biomedicine Muth OH (Ed) Westport, Connecticut; Avi Publishing Company pp 79-96
- Hartley WJ, Grant AB 1961 A review of selenium responsive diseases of New Zealand livestock *Fed Res* 20: 679-688
- Howell J 1968 The experimental copper deficiency on growth, reproduction and haemopoiesis in the sheep *Vet Rec* 83: 226-227
- Keane MG 2001 Responses in beef cattle to concentrate feeding in winter *Occ Ser No.3*, Grange research Centre, Teagasc, 40 pages
- Keane MG 2002a Development of an intensive dairy calf to beef system and associated grassland management *Beef Production Series No. 41* End of Project Report, Grange Research Centre, Project No. 4582, 36 pages
- Keane MG 2002b Concentrate supplements for weanling and finishing steers. *Beef Production Series No. 42* End of Project Report, Grange Research Centre, Project No. 4584, 36 pages
- Keane MG O'Riordan 1998 Development of a competitive forage based dairy calf to beef system *Beef Production Series No. 2* End of Project Report, Grange Research Centre, Project No. 4276, 24 pages
- Keane MG, Drennan MJ 2008 A comparison of Friesian, Aberdeen Angus x Friesian and Belgian Blue x Friesian steers finished at pasture or indoors *Livestock Sci* 115: 268-278
- McDonald JW 1975 Selenium-responsive unthriftiness of young Merino sheep in central Victoria. *Aust Vet J* 51: 536-543
- McLean JW, Thomson GG, Claxton BM 1963 A selenium responsive syndrome in lactating ewes *NZ Vet J* 11:59-60
- Mills CF, Dalgarno AC 1972 Copper and zinc status of ewes and lambs receiving increased dietary concentrations of cadmium *Nature* 239: 171-176
- Tasker JB, Bewick TD, Clark RG, Fraser AJ 1987 Selenium response in dairy cattle *NZ Vet J* 35: 139-140
- Thomson GG, Lawson BM 1970 Copper and selenium interaction in sheep *NZ Vet J* 18:79-82
- Veling J, Counotte GH. 1995 Selenium deficiency without clinical symptoms in young cattle on a dairy farm *Tijdschr Diergeneeskde* 120: 464-465

- Wasowicz W, Gromadzinska J, Rydzynski K 2001 Blood concentration of essential trace elements and heavy metals in workers exposed to lead and cadmium *Int J Occup Med Environ Health* 14: 223-229
- Wichtel JJ, Craigie AL, Freeman DA, Varela-Alvarez H, Williamson NB 1996 Effect of selenium and iodine supplementation on growth rate, thyroid and somatotrophic function in dairy calves at pasture *J Dairy Sci* 79:1865-1872
- Wichtel JJ, Keefe GP, Van Leeuwen JA, Spangler E, McNiven MA, Ogilvie TH 2004 The selenium status of dairy herds in Prince Edward Island *Can Vet J* 45: 124-132