Long-term and carry-over effects of feeding level on performance and energy partitioning of Boran (Bos indicus) and Boran X Holstein dairy cattle

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LONG-TERM AND CARRY-OVER EFFECTS OF FEEDING LEVEL ON
PERFORMANCE AND ENERGY PARTITIONING OF BORAN (BOS
INDICUS) AND BORAN X HOLSTEIN DAIRY CATTLE

A dissertation submitted to the

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Summary

Forty-eight dairy cows examined over a period of 5 years permitted an investigation of the effect of under-nutrition and subsequent restoration of diet level or high feeding level on the metabolism and performance of tropical dairy cattle, represented by an indigenous and an upgraded genotype. In particular, the following aspects of the metabolism and performance of heifers and lactating tropical dairy cows were investigated:

a) Effects of different levels of feed supply on growth and feed conversion of Boran (Bos indicus) and crossbred heifers during growth, work and pregnancy.

b) Effects of different levels of feed supply on lactational performance of Boran and crossbred cows.

c) Energy partitioning in Boran and crossbred cows after long-term adaptation to low or high feed supply, and carry-over effects during adaption to another feeding level.

The experiment involved 24 Boran and 24 Holstein-Friesian × Boran crossbred cattle with continuously recorded datasets from birth until the start of the 4th reproduction cycle. Cows were assigned to feeding regimes of either 1.0, 1.2 or 1.4 times the level of their maintenance energy requirements, representing low, medium and high feeding levels from their first oestrus as heifers until the birth of their third calf. During their third reproduction cycle, half of the cows of each treatment received the respective opposite feeding level (low or high), aiming to measure the carry-over effects of the previous feeding level. Cows which previously received the medium feeding level were not considered in latter analysis. Besides continuously registered intake, milk, weight and reproductive records, also digestibility, digesta kinetics, body composition was recorded during experimental periods. Additionally in lactation 3, various levels and expressions of variables were analysed in blood and fat tissue (hump, tail head), indicative of metabolic changes in nutrient partitioning. Effects of under-nutrition were evaluated in working, but also during growth stages and pregnancy of heifers, in primiparous and multiparous cows (during mid and late lactation).

Boran cows responded to long-term feed fluctuations mainly by mobilising or restoring body fat reserves. Crossbred cows tended to spend most of the energy for milk
production with the highest possible level. However, previously undernourished crossbred cows did not recuperate their milk potential during realimentation in a subsequent lactation. Similarly, previously well-fed crossbred cows did not proportionally decrease milk production in periods of under-nutrition by the amount of feed restriction.

The efficiency of feed energy conversion into retained energy was similar between genotypes, and results showed lower requirements for crossbreds. However, when analysing genotypes separately, efficiency of conversion and, in consequence, energy requirements tended to be lower in Boran.

Differences in energy partitioning were found to be potentially related to the roles of insulin-like growth factor-1 (IGF-1), lipoprotein lipase (LPL) and tumor-necrosis factor-α (TNFα). Previously undernourished crossbreds expressed elevated tissue levels of TNFα, while IGF-1 and LPL expressions in plasma and adipose tissue were higher in Boran cows.

The experiments showed a different response pattern to different feeding levels of Boran compared to crossbreds. When exposed to a specific level of energy supply for their whole productive life, the milk yield of crossbred cows, but not of indigenous cows, was affected by the feeding level. Results from lactation 3 showed carry-over effects of long-term exposure to another level of energy supply in such a way that the response was weak, both, to improvements and to declines in feeding level. These results have implications for market-oriented production systems, as well as for rural areas with poor natural resources and with little market access where increasing milk yield is often not a priority for the livestock owner. Opportunities for effectively using additional feed resources are of high priority in peri-urban areas, where an increase in marketable milk production is feasible with improved feeds for crossbred cows. By contrast, indigenous breeds, better adapted to low water and feed supply, mostly are the option for remote tropical regions with unfavourable conditions. Breed choice and production goals, therefore, need to carefully account for feed fluctuations, in the development of strategies for smallholder livestock farmers.
Zusammenfassung


a) Einfluss unterschiedlichen Futterniveaus auf das Wachstum und Futterumsetzung von Boranfärsern (Bos indicus) und Kreuzungsrindern während der postpubertalen Wachstumsphase, einer Arbeitsverrichtung und der Trächtigkeit.

b) Einfluss unterschiedlichen Futterniveaus auf die Laktationsleistung von Boran (Bos indicus) und Kreuzungskühen.

c) Energiepartitionierung bei Boran (Bos indicus) und Kreuzungskühen nach einer Anpassung an ein niedriges oder hohes Futterniveau, und Nachwirkungen bei einer Futterumstellung.

Futtermittel für Kreuzungsriinder machbar ist, von Interesse. Im Gegensatz dazu ist der einheimische Genotyp, der sich durch höhere Anpassung an niedrige Wasser- und Futteraufnahme auszeichnet, eine Option in unwirtlichen und abgelegenen Regionen. In jedem Fall müssen bei der Wahl des Genotypen, sowie bei gesteckten Produktionszielen, die Auswirkungen von möglichen Futtermengenschwankungen bedacht werden.
I General Introduction

1.1 Problem definition

The human population growth, the increasing urbanisation and rising incomes are predicted to double the demand for livestock and livestock products in developing countries in the next 20 years (Steinfeld, 2002). Some of the forecasted trends include serious challenges for the rural traditional livestock sector since the driving forces are market-orientation, vertically integrated business, use of cereal-based feeds, increased productivity, industrialisation and urban production systems.

Livestock are an important element of the agro-ecosystems in Africa. At family level livestock act as the keystone in most rural livelihoods. Livestock also make a critical contribution to regional and national economies. Seventy six percent of the world’s cattle stocks (1.37 billion) are based in developing countries, out of those 20% in sub Saharan Africa, half of that in Eastern Africa (FAOSTAT, 2003). In addition to providing work power, manure, hides and skins, these animals are the source of approximately 18% of the dietary protein consumed by Africa’s 605 million inhabitants and are estimated to account for 35% of the continent’s agricultural gross domestic product (GDP) (Winrock International, 1992).

In many tropical regions livestock are characterized as being in poor condition, with further body weight loss during dry season, high mortality during the first year of life, low growth rates, low reproductive performance and low milk yields, feed availability and quality during the dry season. From these observations it is a logical progression that the root of the problem is poor nutritional status due to limited feed quantity and quality and it is concluded that nutrition is a major factor affecting livestock productivity.

Ruminants in tropical countries are commonly offered seasonally available forages or post-harvest products. Feed supply is often underlying long-term fluctuations depending on the intensity, duration and time of occurrence of the rainy season. Rural farmers are particularly affected by seasonal feed fluctuations and feed scarcity caused by extended droughts, since storage is seldom practiced, while smallholders in peri-urban areas often purchase feed or industrial by-products. The increased demand in animal products in less developed countries (Delgado et al., 1999), therefore, is covered
often from peri-urban and urban production systems or international trade, where health care, feed and water often can be secured and improved breeds guarantee a high productivity (McLeod et al., 2002, Upton and Otte, 2002). The rural sector, however, lacks strategies and is endangered to participate in the dynamic growth. There is a need for understanding the options for and constraints of tropical livestock feeding, especially for the rapidly growing dairy sector in developing countries.

Constraints are defined as the factors limiting the performance of the system. All other elements of the system (technical, social, or institutional) are associated in such a way that removal of the primary constraint will have a direct impact on system performance. An implication of Liebig’s ‘Law of the Minimum’ is, therefore, that if the deficient nutrient is supplied in large quantity, given adequate water, the supply of some other nutrient will begin to act as a new constraint to growth. In contrast to the ecological definition, in system theory a constraint is defined as a measure of the reduction of variety or reduction of freedom. The system is constrained, if the variety of elements that the system can exhibit is smaller than the conceivable variety. The system cannot fully use the freedom that would seem to be available to it, because some internal or external law, relation or control imposed by nature or by man, prohibits certain combinations of values for the variables (Wilke, 1978).

The realities in tropical livestock systems seem complex since the economic and social objectives are commonly closely linked to the biological outcomes, and in most cases livestock are kept in order to achieve multiple objectives: profitability, income smoothing, manure output, draught power, insurance, year-round milk availability, total herd size, or ability to re-stock quickly following drought induced destocking. Delgado et al. (1999) argued that population growth, urbanization, and increasing incomes in the developing world will result in an increased consumer demand for milk and meat products. The conclusion was that the increased demand must be met by small-scale producers, as the economic potential for, and desirability of, large-scale, industrial style production is probably limited. In this context Sumberg (2002) suggests to analyse livestock production in relation to different systems, situations and producers at a more disaggregated level. Livestock production systems in Africa have been classified by, amongst other characteristics, their objective (commercial or
subsistence), level of resource use (intensive or extensive), level of specialization
(specialized or mixed) and primary output (meat or milk). Wiggins and Proctor (2001)
proposed a framework which shows this diversity through a matrix whose axes are
proximity to urban areas (markets) and quality of natural resources, and this matrix is
then used to generate hypotheses concerning likely development trajectories:
a) Peri-urban zones. Good access to markets and a strong non-farm economy,
emphasis on micro-scale, high-value farming and livestock activities.
b) Rural zones with good natural resources. In terms of agricultural development,
zones will likely move toward specialized, market oriented arable farming and
livestock production. The most important intervention is improvement of marketing
channels.
c) Rural zones with poor natural resources. With low populations and few possibilities
for agricultural development, zones will likely remain in extensive farming and
livestock, and will only develop a limited non-farm economy.
d) Remote rural zones with good natural resources. Lack of strategies. They will likely
remain in subsistence farming.
e) Remote rural zones with poor natural resources. Lack of strategies. They will likely
remain in low productivity subsistence farming, generating very small or no
surpluses.
The latter characterisation of livestock production systems indicates that significant
intensification of agriculture and livestock production are expected only in the peri-
urban zones and in the rural zones with good natural resources.

1.2 Malnutrition in tropical livestock
As summarised before, major constraints that limit productivity improvements,
particularly for resource-poor dairy farmers, are the availability and quality of feed,
reproductive wastage, and the used genotype. Poor nutrition resulting from poor
quality feeds and fluctuating feed supply are often limiting the productivity of tropical
cattle. Feed production might often be limited due to the seasonal competition
between food crops with fodder production. Its harvest often is delayed until food crops
are harvested, resulting in low N and high fibre contents, and thus in low digestibility.
Consequently, nutrient availability is insufficient to maintain the stock and to support an increased production. The fluctuations in feed supply and feed quality are thus reflected in live-weights and body condition scores of animals (Kurtu et al., 1999; Mukasa-Mugerwa, 1989). Since these fluctuations are natural phenomena, ruminants have developed several adaptive mechanisms in their evolution to cope with undernutrition (Chilliard et al., 1995). Nevertheless, dairy animals spent crucial periods of their lives in states of malnourishment, being deprived of combinations of essential nutrients such as protein, energy, minerals, vitamins and water, may be susceptible in detrimental effects on their physiological development and productive life. Especially in the development of heifers this can seriously suppress the potential performance of the animal (Bell et al., 2000). Malnourishment during certain critical stages of lactation does not impair only reproduction and milk yield in the current lactation, but can also have negative effects on subsequent lactations.

Most smallholders in tropical and subtropical regions depend on indigenous cattle mainly of the *Bos indicus* genotype for multi-purpose use, with milk being generally the most important product. Since these cows are of low genetic merit for milk production, crossbreeding of the local *Bos indicus* types with *Bos taurus* breeds with high genetic merit for milk production is widespread. Both types are subjected to seasonal undernutrition prevailing in the smallholder production systems (Bebe et al., 2003).

The available feed potential to cope with undernutrition of dairy cows seems not to be fully utilised. A strategic use of limited feed resources is hypothesized to contribute significantly to an increased asset accumulation through dairy production resulting in improved human nutrition, food security, rural income, and poverty alleviation. However, to develop practical and cost-effective nutritional frameworks, it is important to have a better understanding of the effects of malnourishment in dairy animals.

In a lactating cow, confronted with a period of malnourishment, the first phase is characterised by an adaptive reaction to deficiency of about one week when a level in milk yield reduction is reached, which accounts for about half of the energy deficit (Kirchgessner et al., 1983). This is followed by the normal decline in milk yield as the lactation progresses. Additionally, heat energy expenditure for milk synthesis is reduced by energy deficiency (Kirchgessner et al., 1983). The phenomenon of an
incomplete reduction in milk yield in under-nutrition results from a homeorhetic mechanism, that supports the offspring even at the cost of the dam’s health. Such situations increase the risk of ketosis characterized by elevated concentrations of ketone bodies in blood plasma (Chilliard et al., 2000).

The response of cows to mild deficiency might differ from the reaction to a serious undernutrition with energy near or below maintenance requirements, in which in theory no energy remains for milk production. Consequently, opinions on energy metabolism and conversion efficiency of energy for maintenance in a period of severe under-nutrition are divided. Ortigues and Vermorel (1996) reported no adaptation of energy metabolism during a 7 week period of under-nutrition. The maintenance requirements for metabolisable energy (ME) and the efficiency with which the ME was utilized remained stable throughout the period of under-nutrition. Ortigues and Vermorel (1996) also found that heat production decreased rapidly within the first week of under-nutrition and more or less stabilised 2 weeks after the change in intake, which was twice the adaptation time found for mild deficiencies. In contrast, Margan and Graham (1988) reported a 7% increase in energy metabolism during under-nutrition, whereas Graham et al. (1974) found a 3.5% decrease in energy metabolism during under-nutrition at 50% of maintenance requirements.

Malnutrition resulted in a rapid response of the liver to a change in feeding level (Taylor and Murray, 1991) and a decrease of visceral tissues in relative proportion to body weight (Drouillard et al., 1991). Therefore, a decrease in the proportion of metabolically active visceral organs results in reduced total body energy expenditures during adaptation to nutrient restriction (Drouillard et al., 1991).

Responses in metabolism and performance of dairy cows to re-alimentation are similar when under-nutrition is related only to metabolizable protein deficiency (Kreuzer et al., 1985, 1991). Effects of under-nutrition on milk production were similar when both energy and protein supply were deficient (Windisch et al., 1989). Severely undernourished cows showed a similar metabolic response pattern when subsequently receiving a high feeding level (Coulon and Ollier, 1996).

Lactating cows seem to have only limited ability to compensate a depression in milk yield caused by deficient energy in subsequent periods of re-alimentation. Even when
the total amount of energy, which was not supplied in a period of deficiency, will be additionally supplied in a subsequent period, the milk yield will only slowly return to the value of animals fed constantly with sufficient energy (Kirchgessner and Windisch, 1989). When a level of re-alimentation, similar to the energy supply of never-undernourished cows was fed, the subsequent milk yield increase did not cover more than 50% of the difference to the control cows and the previously underfed cows.

Dairy cows in the tropics, do not always respond as predicted by nutrient response models to concentrate supplements (Kurtu et al., 1999). However, common response models are designed on the assumption that lactating cows are fed beyond maintenance level in their whole life.

Genotype selection seems to be a key issue to optimise the use of available feed resources. Indigenous cattle in most tropical countries are Zebu (Bos indicus). Upgraded dairy breeds (Bos taurus × Bos indicus) have been kept successfully in large commercial herds, and rarely in the rural smallholder sector, where feed and other inputs are not available. Though being adapted to tropical conditions, the potential of most B. indicus for milk production is low (Kurtu et al., 1999). Crossbreeding of Bos taurus and B. indicus genotypes produces an F1 with greater potential for milk yield than either one of their parents. Studies in Ethiopia (Kurtu et al., 1999) have shown that lactating crossbred cows can be used for work although longer calving intervals must be expected. Only the supplemented working cows had conceived for the third time without prolonged anoestrus (Zerbini et al., 1996). In order to be able to utilize crossbred cows for reproduction, the establishment of a composite genotype is necessary to avoid the loss of heterozygosis. Alternatively, though more costly, but avoiding the erosion in cattle diversity would be the selection within suitable B. indicus breeds for milk production. However, the optimum choice depends on the response of these alternatives to under-nutrition and re-alimentation.

Differences in partitioning and metabolic utilization of energy seem to be small between Bos taurus dairy breeds such as Holstein, Simmental and Jersey (Münger et al., 1996). Additional energy intake resulted in a higher milk yield. In contrast, Bos indicus as the indigenous breeds in tropical countries, when undernourished after calving, were
observed to initially replenish body reserves at the cost of milk production (Mukasa-Mugerwa et al., 1997).

The development of a framework for feeding strategies requires therefore an examination of how *Bos indicus* and crossbred (*Bos indicus* × *Bos taurus*) dairy cows can be most efficiently used for milk production in smallholder production systems. The type of adaptation to subsequent periods of under-nutrition is one of the critical tasks in dealing with under-nutrition since the adoption of costly strategies like feeding concentrates requires a yield response which is visible to the farmer.

1.3 Objectives

Therefore the following basic hypotheses to be tested in this work as derived from the considerations described above were:

- *Bos indicus* and *Bos indicus* × *Bos taurus* crossbreds respond differently in energy partitioning at low or high feeding levels.

- *Bos indicus* and *Bos indicus* × *Bos taurus* crossbreds when adapted to a specific feed supply for a long period of time, respond differently in partitioning the energy to milk and body tissue when feeding is changed to another level.

Knowledge of different responses between dairy cows of indigenous origin and crossbreds when receiving additional feed after serious under-nutrition ('re-alimentation') is essential for the development of feeding strategies for small livestock holders. It was investigated how far this response depends on the under-nutrition history of the cows by using animals underfed for their whole productive life, part-time underfed and never underfed. The major goal of this study was to test the above hypotheses, particularly to quantify the carry-over effects of under-nutrition in re-alimentation, by the use of parameters indicative for metabolic alterations caused by nutritional modifications, breed differences and different under-nutrition histories. Within the frame of the major goal, the specific objectives were:

- Determination and assessment of implications of effects of different levels of feed supply on growth and feed conversion of Boran (*Bos indicus*) and crossbred heifers during growth, pregnancy, and working
- Determination and assessment of implications of effects of different levels of feed supply on lactational performance of Boran (Bos indicus) and crossbred cows
- Characterization and quantification of differences in nutrient partitioning in Boran (Bos indicus) and crossbred cows after long-term adaptation to low or high feed supply, and carry-over effects when being adapted to a different feeding level
- Validation of the utility and viability of new feeding strategies derived from the project

1.4 Experimental background

The present project is based on one extensive experiment with cattle (from puberty to the end of the 3rd lactation) and the evaluation of long term production data of the experimental cows. The experiment was carried out at ILRI Debre Zeit (Ethiopia) using a herd of initially 48 female cattle being fed since 1997 the reported feeding levels. The animal numbers declined during the experiment mostly due to anoestrus caused by the extreme feeding treatment. The factors investigated were genotype (crossbred vs. zebu), nutritional history (well-fed vs. under-fed, i.e. 1.4 vs. 1.0 × maintenance energy requirement (M)) and the feeding level in the 3rd lactation (1.0 M vs. 1.4 M). The experimental design applied at first was based on six experimental groups with eight animals in each treatment. Half of this 8 animals were re-allocated in lactation 3, forming 8 groups of intended 4 animals. Daily energy requirements for the different animal groups and treatments were calculated according to MAFF (1987). Animals were fed grass hay (65%) as roughage and wheat bran (35%) as concentrate during all phases of the study. They were provided ad libitum with water and mineral blocks. Animals were housed in individual tie-stall pens in a well-ventilated barn having a concrete floor.

The experimental schedule generally consisted of periods with intensive and extensive data collection phases. There were intensive phases in the heifer period, in lactation 1 and in lactation 3 with a series of additional parameters determined in these periods. The extensive phases fell between the short intensive phases and aimed to obtain whole lactation performance data.
The number of parameters obtained depended on the phase of the experimental schedule. Continuous determinations included: individual milk yield, feed intake and body weight. In frequent periods, analysis of feed and milk samples were carried out. Determinations in the intensive phases of the experimental schedule included: balance measurements, nutrient digestibility, nutrient excretion via faeces and urine containing fertilizer nutrients. At the end of each intensive measurement period during the lactation 3 blood samples were collected before, while, and after feeding. Various blood traits related to energy metabolism were analysed in blood plasma or serum. Body composition was estimated with the body water space using the Deuterium dilution technique. Subcutaneous adipose tissue was collected by biopsies from the dorsal part of the animals, the tail head, and the mammary gland. cDNA transscripts of proteins related to energy metabolism on cellular level were quantified by Real-time PCR.

References


II Growth and feed conversion of Boran (*Bos indicus*) and Holstein × Boran heifers during three physiological states receiving different levels of a tropical diet

based on:

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Abstract

Forty-eight heifers of two genotypes, Boran (*Bos indicus*) and Holstein × Boran, were allocated to three feeding levels equivalent to 1.0, 1.2 and 1.4 times calculated metabolisable energy requirements for maintenance (ME\(_m\)). The experiment started at the onset of reproductive activity, when the Boran and crossbred heifers were 4.4 and 2.2 years old, respectively. The response to feeding level was investigated in three physiological states: (i) puberty & growth (87 days), (ii) work & growth (94 days, 51 days thereof with work), and (iii) pregnancy & recovery from work (115 days). Neither genotype nor feeding level had effects on nutrient digestibility, digesta passage rate and reproductive performance. Daily gains and feed conversion efficiency were higher in well-fed heifers (\(P < 0.001\) and \(P < 0.01\)). Estimated ME\(_m\) was 0.54 MJ/kg metabolic weight (W\(_{0.75}\)) in both genotypes during puberty & growth. Imposing work required additional 0.20 and 0.10 MJ ME/kg W\(_{0.75}\) in Boran and crossbreds, respectively. Early pregnancy, associated with recovery from work, apparently reduced ME\(_m\) to 0.30 MJ/kg W\(_{0.75}\). This could be explained as a result of compensatory growth. The efficiency of utilisation of metabolisable energy for growth (0.28) did not differ (\(P > 0.1\)) between genotypes and physiological states.

**Keywords:** Maintenance, Requirement, Draught, Reproduction, Compensatory growth, Zebu

II.1 Introduction

The average growth rate for milk production in the developing world has been 3.1% per annum from 1984 to 1993 and is forecasted to be further growing until 2020 at a rate of 3.3% per annum, this even more pronounced for Sub-Saharan Africa with expected rates of 3.8% (Delgado *et al.*, 1999). Important trends which reflect this change are intensification and more production-oriented animal keeping systems mainly in peri-
urban regions (Staal et al., 1997). However, smallholder livestock farming systems in Sub-Saharan Africa, particularly those in remote areas, are mainly based on widespread livestock activities, where livestock serves, beside production aspects, also as a source for draught, capital assets, insurance and cultural affiliations. So far, mainly oxen are employed for work (e.g. ploughing) and this on average for 51 days per annum with the cost of maintaining the animals exceeding the economic benefits of work, especially in a situation of increased industrialisation (Cole, 1997). Dairy heifers might be a better alternative although only in areas where there are no cultural barriers against the use of female animals for draught. The multipurpose function of livestock in remote areas is guaranteed best by indigenous, mostly Bos indicus, breeds which are well adapted to the specific environmental conditions (Pearson and Dijkman, 1994; Lawrence and Pearson, 2002). Market-oriented smallholder dairy farmers, by contrast, increasingly employ crossbred animals since their major purpose is to produce goods that can be sold (Bebe et al., 2003). In both, rural or peri-urban areas, feed scarcity is still a major problem. Efficient means to increase feed utilisation are demanded to be able to maintain or enhance livestock productivity, thus increasing the supply of valuable food to the population and alleviating poverty of the livestock keepers. Only few and contradictory information is available on growth performance and feed utilisation of dairy heifers of indigenous or crossbred origin under tropical conditions (Agnew and Yan, 2000). There is evidence that the efficiency of conversion of feed energy into weight gain is more variable than is accounted for in current net energy (NE) or metabolisable energy (ME) based systems (Nsahlai et al., 1997; Yan et al., 1997; NRC, 2001). Growth performance and metabolic energy utilisation may also change in different physiological states. For instance, after exposure to work, pregnant Holstein × Boran cows were found to restore proportionately 0.95 of their previous body weight (W) loss in a recovery period of 6 months and even 1.77 of the loss when receiving a ration of grass hay and 3 respective 5 kg/day of a supplement; by contrast non-pregnant lactating crossbreds restored proportionately only 0.12 (no supplement) and 0.59 (3 kg supplement) of the losses (Zerbini et al., 1996).

The basic hypothesis to be tested in the present study was that there may exist significant differences between purebred indigenous Bos indicus heifers and crossbreds
with *Bos taurus* in their response in growth, feed conversion and reproduction to long-term exposure with low, medium or high level of energy supply. Particular emphasis should also be put on the effect of working exercise as most previous controlled studies have investigated the influence of work with oxen and lactating cattle (e.g., Person and Lawrence, 1992; Zerbini et al., 1993). The approach chosen comprised about 300 days of experiment where genotypes were compared in three different physiological states including exposure to work and subsequent recovery.

**II.2 Materials and methods**

**II.2.1 Animals and research site**

Twenty-four East African Boran (*Bos indicus*; Tegegne et al. (1992) heifers and 24 Holstein Friesian × Boran (*Bos taurus × Bos indicus*) heifers were used in an experiment carried out at the ILRI Debre Zeit experimental farm located in the Rift Valley at 1920 m o.d. At the time when the experiment was conducted, the minimum and maximum ambient temperatures were 10 (±2) °C and 27 (±2) °C, respectively, and relative humidity was 59 (±12) % during the rainy season and 41 (±9) % during the dry season. Animals were tethered in a well-ventilated barn with pens sizes of 1.2 × 2.2 m and on concrete floor with a small slope. Boran and crossbred heifers were selected out of the ILRI station herds at ages of 44 (±0.2) and 2.2 (±0.3) years, and weighed 297 (±24) and 287 (±29) kg, respectively. Since the Boran cattle are typically late maturing, the difference in age between genotypes was deliberately chosen to facilitate the comparison between animals in similar physiological states. Similar weight and simultaneous start of the experiment for all animals were other decision criteria. The station Boran herd had its origin directly from the Southern Ethiopia Boran region. The experimental Boran heifers were bred through natural insemination of dams of this herd using eight station Boran bulls. The crossbreds had Holstein blood levels of 50% and 75%, and were sired by two Dutch Holstein Friesian bulls (Holland Genetics, Arnhem, The Netherlands). Calves of both genotype were reared on station, tethered in a calf barn and fed restricted amounts of milk in the first 2 months after birth, then for 6 month a diet consisting of grass hay *ad libitum* and a concentrate mix (1.5% of W) containing, per kg, 560 g wheat middlings, 240 g Noug cake, 185 g maize grain, 5 g salt and 10 g
bone meal. Afterwards the calves were moved to a feedlot receiving seasonal available grass hay until being allocated to the experimental groups.

11.2.2 Feeding treatments
Throughout the experiment the diet consisted always of the same proportion (0.65) of grass hay, mainly *Cynadon dactylon*, and wheat bran (0.35). Average analysed contents per kg of grass hay were 905±98 g dry matter (DM) and, in DM, 885±11 g organic matter (OM), 738±34 g neutral detergent fibre, 43±8 g crude protein, and 17.5±0.6 MJ gross energy (GE). The corresponding values for the wheat bran were 882±11 g, 953±4 g, 441±45 g, 137±28 g and 18.0±1.0 MJ. A priori, ME content of grass hay and wheat bran was assumed to be 7.5 and 10.1 MJ/kg (MAFF, 1987), and the conversion into ME was supposed to be 0.81×DE (AFRC, 1993). Feed was provided once daily, starting with wheat bran at 7 a.m. and followed by grass hay. Animals had *ad libitum* access to mineral licking blocks containing 120 g Ca, 39 g P and 1.3 g S per kg (The Awash Mineral Block Production Industry, Awash, Ethiopia). It was assumed that nutritional effects exclusively resulted from variations in feeding level since protein and mineral supply was estimated to be always sufficient in relation to energy supply, W and expected performance (Kearl, 1982; NRC, 2001). Heifers were blocked within genotypes by W. They were allocated to three feeding levels, equivalent to 1.0 (low), 1.2 (medium) and 1.4 times (high) maintenance allowance for metabolisable energy (ME$_m$) regarding the suggestions of MAFF (1987) for ME$_m$ as 8.3 + 0.091 × fasted W (kg).

11.2.3 Physiological states of the animals
The start of the experiment was on the same day for all 48 heifers. The effect of plane of nutrition was investigated in three periods throughout their heifer state which reflected different physiological states including non-dietary treatments such as insemination and working exercise. The first physiological state reflected puberty while animals were growing, and this period lasted for 87 days. Digestibility measurements were performed at the end of this period for 5 days in two batches (24 representative animals in each batch).
A second period of 94 days followed which simulated another physiological state by exposing the growing heifers to working exercise for a total of 51 days for always 5 days per week. For half of the animals of each treatment sub-group, work consisted of pulling sledges with a weight of 0.15 of W pulled on a 60 m path. The other subgroups walked on a 454 m path for 5h/day. The daily average distance covered was 16.1 km for heifers pulling sledges and 19.0 km for those walking. In the last 3 weeks of the second period, subsequent to the last working exercise, digestibility determinations were carried out for 5 days. This was followed by a period where in all heifers at the same time chromium-mordanted hay was administered and faeces were collected for later determination of digesta passage kinetics.

The final period comprised 115 days and simulated a third physiological state, namely early pregnancy (1st to 4th month) together with recovery from work. Again the last three weeks of this period were used for the rate of passage and digestibility measurements.

11.2.4 Experimental procedures

Oestrus behaviour and heat detection were monitored daily by a veterinarian, barn attendants and with the help of a teaser bull. After day 45 of the second experimental period (work) had passed, all heifers were artificially inseminated with Holstein Friesian semen as soon as detected to be on oestrus. Sixty days later, pregnancy diagnosis was performed by the determination of the presence or absence of the corpus luteum. In case of failure of pregnancy insemination was repeated. On average, heifers got pregnant 20 days after the start of the third experimental period.

The animals were weighed every second week between 07.00 and 09.00 h after 12 h of fasting in order to determine the fasting W. These data were used to adjust the individual feed allocation based on the ME\textsubscript{m} estimate. Along with W recordings, body condition scores (BCS) were assessed as outlined by Nicholson and Butterworth (1986) by two trained barn attendants. A nine-grade scale ranging from extremely meagre to extremely obese was applied. Feed offered and refused was recorded daily during the experiment. Samples of feed offered were taken every second week, while samples of feed refused were collected separately for individual animals. Both offer and refusals of
each heifer were pooled monthly. Animals had free access to fresh tap water provided in buckets. Faeces were collected in closed plastic buckets during 24 h per day over a period of 5 days. Proportionate (0.10) faeces samples were obtained after weighing, thoroughly mixed and frozen at -20 °C for subsequent chemical analysis. Passage rate was determined using the method of Uden et al. (1980). An amount of 280 g unchopped teff straw (Eragrostis tef) was prepared for each animal by soaking the straw in a solution of potassium dichromate in a galvanized pot at a concentration of 33% (w/w) of chromium. The material was then completely immersed, sealed with aluminum foil and heated in an oven at 105 °C for 24 h. After being thoroughly washed with tap water and ascorbic acid at a concentration of 1:2 (w/w) for 1 h, the mordanted teff straw was dried for 48 h at 65 °C (Uden et al., 1980). This mordanted feed was dosed to each animal at rate of 80 g and mixed and offered together with approximately 150 g wheat bran to enhance palatability. Rectal grab samples comprising approximately 100 g fresh faeces were collected during the following times after dosage: 4, 8, 12, 36, 40, 48, 60, 72, 96, 120 and 144 h. Faecal samples were dried at 105 °C for 24 h and ground through an 1-mm screen for later chromium determination. The experiment was approved retrospectively by the meanwhile constituted Institutional Animal Care and Use Committee (IACUC, Nairobi, Kenya). Animals were under permanent observation by a veterinarian.

II.2.5 Laboratory analyses
Feed and faeces were analysed for DM and OM according to AOAC (1990), and GE; only in two last experimental periods) by ballistic bomb calorimetry (CBB-330-010L, Gallenkamp, Loughborough, U.K.). The dried faecal samples were prepared for analysis of chromium by a method similar to that reported by Cheong and Salt (1968). Samples (0.5 g) were digested at 150 °C for 1 h and, subsequently, at 200 °C for 1 h both in a 5 ml mixture of 1:5:2:3 (v/v/v/v) ratio of concentrated H₂SO₄, 60% HClO₄ and concentrated HNO₃. After cooling, samples were diluted to 75 ml volume with distilled deionised water, covered with parafilm and shaken. Samples were stored over night for complete sedimentation of particles. Chromium concentration in the solution was determined by atomic absorption spectrophotometry (model 2380, Perkin Elmer, Norvalk, CT, USA)
with air-acetylene flame. Standards were prepared with the same acid mixture that had been used for digestion to ensure that all solutions had a common matrix background.

II.2.6 Calculations and statistical analysis

The calculation of digesta kinetic data was performed by the model of Grovum and Williams (1973). This model considers digesta flow as a multi-compartmental exponential process. In detail the variables considered are (i) passage rate \(k_1\), explained as the outflow rate constant of the reticulo-rumen, (ii) fast outflow rate \(k_2\), considered as the outflow rate constant of the caecum and proximal colon, (iii) mean retention time (MRT), and (iv) time of first marker appearance after administration, defined as transit time (TT) through omasum, small and large intestines (time of first marker appearance). Calculations were based on the following equation:

\[
y = Ae^{-k_1(t-TT)} - Ae^{-k_2(t-TT)} \text{ when } t \geq TT \text{ and } y = 0 \text{ when } t < TT
\]

where \(y\) is the estimated and \(A\) is the measured marker concentration in the faecal DM, \(t\) is the time of sampling and \(e\) is the anti-logarithm. All calculations based on this equations were performed as outlined by Grovum and Phillips (1973) using 36 h for the start of the descending slope.

The heifers' ME\(_m\) (MJ/day) was calculated according to AFRC (1993) as \([F+A]/k_m\), where \(F =\) fasting energy metabolism (MJ/day) calculated as 0.53\times[fasting W (kg)]\(^{0.67}\), \(A =\) activity allowance (MJ/day) for standing cattle calculated as 0.0071\times W (kg), \(k_m =\) efficiency of energy utilisation for maintenance calculated as 0.35\times q_m+0.503, \(q_m =\) metabolisability of gross energy (ME/GE; assumed to be 0.575 from diet analysis data). Retained ME (ME\(_r\), MJ/d) was calculated as \(\Delta W\times[EV_g]\), where \(\Delta W\) is W change and \(EV_g\) (energy value of gain) was derived from \((4.1+0.033\times W-9\times10^{-6}\times W^2)/(1-1\times0.1475\times\Delta W)\) given for growing cattle by AFRC (1993). The extra energy required for work activity was assumed to account for the difference between ME\(_m\) determined in the puberty & growth state and ME requirements for zero ADG in the work & growth state.

The data of the three physiological states were compared with a ‘two between-, and one within-subject factor analysis’ by analysis of variance and the statement sum of
square type 4 (SS4) by the general linear model of SAS version 8 for Windows (Collyer and Enns, 1986; SAS, 2001). Model 1 applied over all data was as follows:

\[ Y_{ijkl} = \mu + \alpha_i + \beta_j + \alpha\beta_{ij} + \gamma_{k(ij)} + \delta_l + \alpha\delta_{il} + \beta\delta_{jl} + \gamma\delta_{k(ijl)} + e_{ijkl} \]  

where \( Y_{ijkl} \) = individual observation, \( \mu \) = overall mean, \( \alpha_i \) = effect of feeding level, \( \beta_j \) = effect of genotype, \( \gamma_{k(ij)} \) = effect of individual heifer, \( \delta_l \) = effect of physiological state and \( e_{ijkl} \) = residual error. On occurrence of significant effects of the physiological state (\( P < 0.05 \)), multiple comparisons among the three overall means were performed by the least square means option of the general linear model of SAS (2001). Model 2 was applied for analysis of variance of data within each physiological state:

\[ Y_{ijk} = \alpha_i + \beta_j + \alpha\beta_{ij} + e_{ijk} \]  

In the case of significant feeding level \( \times \) genotype interactions (\( \alpha\beta; P < 0.05 \)), multiple comparisons of the means applying model 2 were carried out using the PDIFF option and the Bonferoni method. The tables give arithmetic means and standard errors (S.E.) for each individual physiological state as obtained with model 2.

Additionally, weekly data from the three physiological states were subjected to regression analysis (in a combination with analysis of variance) to predict average daily gain (ADG) for a given ME intake (ME) regarding genotype, ME, and the interaction of genotype \( \times \) ME. Intercepts of these regression equations represent ADG during fasting. The genotype effect represents the difference between genotypes in the amount of ME required at zero ADG or, when excluding ME \( \times \) genotype interactions, the differences in their ADG at any given level of ME. The regression coefficient associated with ME is an estimate of the efficiency of conversion of ME to ADG, and the ME \( \times \) genotype interaction represents the difference in feed efficiency between the two genotypes. The regression analysis was carried out by the general linear model of SAS (2001) using the SOLUTION statement and the sum of square type 4 (SS4).

11.3 Results

11.3.1 Intake, digestibility and performance

Intakes of dry matter (DM) and organic matter (OM) differed (\( P < 0.001 \)) among the three feeding treatment in both genotypes as expected (Table II-1). On average, heifers receiving medium and high feeding levels, respectively, consumed 22.0% and 43.6%
more DM, and 21.8% and 43.7% more OM than heifers with low feed supply across all physiological states. Nevertheless, differences among feeding levels were slightly smaller than expected since grass hay refusals by the Boran genotype increased with feed supply ($P < 0.001$). Amounts of hay refused declined ($P < 0.001$) with time. Across all physiological states, feed intake as expressed per kg of metabolic W ($W^{0.75}$) was lower in Boran heifers with 60.1, 71.2 and 75.6 g OM/day than in crossbred heifers with 60.4, 72.3 and 83.9 g OM/day, respectively, when exposed to low, medium and high level of feeding. Since the feeding levels affected $W$, differences among feed allocation levels were not always constant.

This is the reason why in the first physiological state no genotype differences were observed in absolute feed intake, whereas differences in $W$ and, therefore, in feed intake had developed in the two subsequent physiological states. The calculated ME intake ($ME$, and $ME/W^{0.75}$) was similarly affected by feeding level and genotype as OM. The level of $ME/W^{0.75}$ slightly but significantly increased from the puberty & growth state to the pregnancy & recovery state, mainly because of the declining levels of hay refusals. Digestibility of OM did not differ between genotypes and among feeding levels (Table II-2). However, an increase of 0.16 in OM digestibility (from 0.59 to 0.65) was observed from the first to the second period, and this level was maintained with 0.66 during the pregnancy & recovery state. Energy digestibility, which was measured only during the work & growth and the pregnancy & recovery states, amounted to 0.64 and was influenced neither by feeding level nor genotype. Effects of genotype and feeding level on digesta kinetics were generally small. An increasing feeding level resulted in a passage rate ($k_1$) faster by 54% ($P < 0.1$), but the fast outflow rate ($k_2$) was not correspondingly influenced.
Table II-1 Effect of feeding level, genotype and physiological state (P) on feed and energy intake of growing heifers (n=8 per treatment)

<table>
<thead>
<tr>
<th>Genotype (G)</th>
<th>Boran</th>
<th>Holstein x Boran</th>
<th>Mean</th>
<th>S.E.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feeding level (F)</td>
<td>Low</td>
<td>Medium</td>
<td>High</td>
<td>Low</td>
<td>Medium</td>
</tr>
<tr>
<td><strong>Dry matter intake (kg/day)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
| Puberty & Growth | 4.62<sup>a</sup> | 5.47<sup>b</sup> | 5.75<sup>c</sup> | 4.45<sup>a</sup> | 5.33<sup>b</sup> | 6.46<sup>c</sup> | 5.35<sup>A</sup> | 0.111 | ns | **
| Work & Growth | 4.53<sup>a</sup> | 5.38<sup>b</sup> | 5.83<sup>c</sup> | 4.50<sup>a</sup> | 5.52<sup>b</sup> | 6.88<sup>c</sup> | 5.44<sup>A</sup> | 0.133 | ** | **
| Pregnancy & Recovery | 4.63<sup>a</sup> | 5.76<sup>b</sup> | 6.68<sup>c</sup> | 4.60<sup>a</sup> | 5.84<sup>b</sup> | 7.62<sup>c</sup> | 5.85<sup>B</sup> | 0.165 | ** | **
| Grass hay refusal (g DM/day) | | | | | | | | | | |
| Puberty & Growth | 44<sup>a</sup> | 147<sup>b</sup> | 926<sup>c</sup> | 3<sup>a</sup> | 14<sup>a</sup> | 105<sup>b</sup> | 206<sup>A</sup> | 50.2 | ** | **
| Work & Growth | 26<sup>a</sup> | 147<sup>b</sup> | 853<sup>c</sup> | 5<sup>a</sup> | 4<sup>a</sup> | 88<sup>b</sup> | 187<sup>A</sup> | 48.6 | ** | **
| Pregnancy & Recovery | 6<sup>a</sup> | 17<sup>a</sup> | 443<sup>b</sup> | 0<sup>a</sup> | 0<sup>a</sup> | 54<sup>a</sup> | 87<sup>b</sup> | 25.8 | ** | **
| **Organic matter intake (kg/day)** | | | | | | | | | | |
| Puberty & Growth | 4.36<sup>a</sup> | 5.17<sup>b</sup> | 5.45<sup>c</sup> | 4.20<sup>a</sup> | 5.03<sup>b</sup> | 6.10<sup>c</sup> | 5.05<sup>A</sup> | 0.106 | ns | **
| Work & Growth | 4.28<sup>a</sup> | 5.08<sup>b</sup> | 6.33<sup>c</sup> | 4.25<sup>a</sup> | 5.21<sup>b</sup> | 6.50<sup>c</sup> | 5.14<sup>A</sup> | 0.126 | ** | **
| Pregnancy & Recovery | 4.38<sup>a</sup> | 5.45<sup>b</sup> | 5.52<sup>c</sup> | 4.30<sup>a</sup> | 5.53<sup>b</sup> | 7.22<sup>c</sup> | 5.54<sup>B</sup> | 0.156 | ** | **
| Organic matter intake (g/kg W<sub>75</sub>/day) | | | | | | | | | | |
| Puberty & Growth | 60.2<sup>a</sup> | 71<sup>b</sup> | 73.6<sup>c</sup> | 60.6<sup>a</sup> | 72.8<sup>c</sup> | 83.8<sup>d</sup> | 70.3 | 1.20 | ** | **
| Work & Growth | 60.4<sup>a</sup> | 70.8<sup>b</sup> | 74.3<sup>c</sup> | 60.7<sup>a</sup> | 72.7<sup>b</sup> | 83.9<sup>c</sup> | 70.4 | 1.32 | ** | **
| Pregnancy & Recovery | 59.9<sup>a</sup> | 71.7<sup>b</sup> | 79.1<sup>c</sup> | 59.9<sup>a</sup> | 71.9<sup>b</sup> | 84.1<sup>d</sup> | 71.1 | 1.22 | ** | **
| **ME intake (MJ/day)** | | | | | | | | | | |
| Puberty & Growth | 41.2<sup>b</sup> | 47.9<sup>c</sup> | 50.5<sup>c</sup> | 38.7<sup>a</sup> | 45.7<sup>c</sup> | 57.9<sup>d</sup> | 47.0<sup>A</sup> | 1.07 | ns | **
| Work & Growth | 43.3<sup>a</sup> | 51.8<sup>b</sup> | 56.2<sup>c</sup> | 43.5<sup>a</sup> | 52.1<sup>b</sup> | 66.5<sup>c</sup> | 52.2<sup>B</sup> | 1.31 | ** | **
| Pregnancy & Recovery | 46.2<sup>a</sup> | 55.8<sup>b</sup> | 65.9<sup>c</sup> | 45.3<sup>a</sup> | 56.6<sup>b</sup> | 76.9<sup>g</sup> | 57.8<sup>c</sup> | 1.72 | ** | **
| **ME intake (kJ/kg W<sub>75</sub>/day)** | | | | | | | | | | |
| Puberty & Growth | 568<sup>a</sup> | 659<sup>b</sup> | 683<sup>c</sup> | 559<sup>a</sup> | 661<sup>b</sup> | 795<sup>c</sup> | 654<sup>A</sup> | 12.3 | ** | **
| Work & Growth | 611<sup>a</sup> | 722<sup>b</sup> | 756<sup>c</sup> | 623<sup>a</sup> | 727<sup>b</sup> | 868<sup>c</sup> | 716<sup>b</sup> | 13.1 | ** | **
| Pregnancy & Recovery | 631<sup>a</sup> | 734<sup>b</sup> | 822<sup>c</sup> | 623<sup>a</sup> | 736<sup>b</sup> | 895<sup>c</sup> | 740<sup>c</sup> | 14.8 | ** | **

<sup>a-c</sup> Differing capital letter superscripts denote significant differences between means of physiological state within columns within variable (P < 0.05) as determined by variance analysis model 1.
<sup>a</sup> Differing lower case superscripts denote significant differences between means within rows (P < 0.05) separate for each physiological state as determined by variance analysis model 2. P-value: * P<0.05, ** P<0.01, *** P<0.001, ns=non significant.
<sup>1</sup> Significant FxP interaction (P < 0.001). 1 Significant GxP interaction (P < 0.001). 1 Significant FxGxP interaction (P < 0.01).
Transit time was affected \((P < 0.05)\) by the feeding level, but effects were not consistent with respect to the three levels. When heifers were in the work & growth state, transit time was faster \((P < 0.05)\), rumen retention time was shorter \((P < 0.05)\) and mean retention time was shorter \((P < 0.1)\) than during pregnancy & recovery.

In the first measurement period W did not differ \((P > 0.1)\) either between genotypes or among feeding levels (Table II-3). The time was obviously too short to substantially differentiate the initially similar W of the animals during average of the 87 days of period 1. During the work phase, W did not significantly increase on average while animals showed an average gain of 30 kg during the pregnancy & recovery period. When subjected to work, only heifers receiving the high feeding level gained W (9.3%), whereas heifers of both genotypes maintained their W in the medium feeding level and lost weight at low feeding level. ADG in that period were higher by 93 g/day in the crossbreds than in the Boran. All animals gained weight during the puberty & growth phase and, particularly, during the pregnancy & recovery phase \((P < 0.05)\).

In both physiological states with consistent growth, i.e., the first and the third state, feeding level had a significant positive effect on ADG, and crossbreds were growing faster than Boran \((P < 0.1\) and \(P < 0.001)\). Table II-3 also gives the ADG converted into that amount of ME assumed to have been used for body energy retention.

This value ranged from -1.4 to +9.8 MJ/day. The BCS differed between genotypes \((P < 0.001)\), with the higher scores being observed in the Boran, and BCS markedly differed \((P < 0.001)\) among feeding levels. The overall level of BCS was similar in the first two periods of the experiment but increased \((P < 0.001)\) in the pregnancy & recovery state. This was almost exclusively the result of a corresponding increase in heifers receiving the high feeding level. Feed conversion efficiency (ADG per unit of digestible OM intake (DOM\(_{\text{r}}\)) increased \((P < 0.01)\) with increasing feeding level in the two physiological states with consistent weight gains (i.e., periods 1 and 3). From low to high feeding level (+40% feed) feed conversion ratio increased by 58%. Feed conversion ratio was higher \((P < 0.001)\) during the third than the first experimental period by a factor of more than two. There was a significant genotype effect on feed conversion ratio. Boran heifers converted DOM\(_{\text{r}}\) to ADG only at a level 0.72 times when compared with crossbred heifers. However, there were no genotype differences \((P > 0.1)\) when analysing physiological states separately.
Table II-2 Effect of feeding level, genotype and physiological state (P) on apparent digestibility (n=8 per treatment) and digesta passage variables (n=4 per treatment) in growing heifers

<table>
<thead>
<tr>
<th>Genotype (G)</th>
<th>Boran</th>
<th>Holstein x Boran</th>
<th>Mean</th>
<th>S.E.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feeding level (F)</td>
<td>Low</td>
<td>Medium</td>
<td>High</td>
<td>Low</td>
<td>Medium</td>
</tr>
<tr>
<td>Organic matter digestibility</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Puberty &amp; Growth</td>
<td>0.601</td>
<td>0.591</td>
<td>0.591</td>
<td>0.587</td>
<td>0.579</td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>0.645</td>
<td>0.649</td>
<td>0.649</td>
<td>0.654</td>
<td>0.638</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>0.671</td>
<td>0.653</td>
<td>0.662</td>
<td>0.663</td>
<td>0.652</td>
</tr>
<tr>
<td>Energy digestibility</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>0.653</td>
<td>0.641</td>
<td>0.616</td>
<td>0.654</td>
<td>0.636</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>0.654</td>
<td>0.631</td>
<td>0.654</td>
<td>0.638</td>
<td>0.621</td>
</tr>
<tr>
<td>Rate of passage (k, per h)</td>
<td>+</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>0.0201</td>
<td>0.0211</td>
<td>0.0487</td>
<td>0.0207</td>
<td>0.0335</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>0.0378</td>
<td>0.0319</td>
<td>0.0466</td>
<td>0.0414</td>
<td>0.0233</td>
</tr>
<tr>
<td>Fast outflow rate (k, per h)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>0.0865</td>
<td>0.0753</td>
<td>0.0875</td>
<td>0.0783</td>
<td>0.0876</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>0.0770</td>
<td>0.0756</td>
<td>0.0738</td>
<td>0.0877</td>
<td>0.0821</td>
</tr>
<tr>
<td>Mean retention time (h)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>74.4</td>
<td>70.1</td>
<td>65.0</td>
<td>81.8</td>
<td>66.3</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>57.4</td>
<td>57.9</td>
<td>52.7</td>
<td>53.5</td>
<td>65.2</td>
</tr>
<tr>
<td>Rumen retention time (h)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>37.1</td>
<td>34.0</td>
<td>29.9</td>
<td>41.2</td>
<td>31.6</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>23.7</td>
<td>24.8</td>
<td>20.1</td>
<td>22.2</td>
<td>29.8</td>
</tr>
<tr>
<td>Fast compartment retention time (h)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>8.18</td>
<td>9.41</td>
<td>9.36</td>
<td>9.14</td>
<td>8.77</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>9.05</td>
<td>9.37</td>
<td>9.94</td>
<td>8.53</td>
<td>8.81</td>
</tr>
<tr>
<td>Transit time (h)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>9.03</td>
<td>7.47</td>
<td>8.40</td>
<td>9.13</td>
<td>8.05</td>
</tr>
</tbody>
</table>

**Differing capital letter superscripts denote significant differences between means of physiological state within columns within variable (P < 0.05) as determined by variance analysis model. P-value: + P<0.1, * P<0.05, *** P<0.001, ns=non significant.**
Table 11-3 Effect of feeding level, genotype and physiological state (P) on body condition, gains and feed conversion of growing heifers (n=8 per treatment)

<table>
<thead>
<tr>
<th>Genotype (G)</th>
<th>Feeding level (F)</th>
<th>Boran</th>
<th>Holstein x Boran</th>
<th>Mean</th>
<th>S.E.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Low</td>
<td>Medium</td>
<td>High</td>
<td>Low</td>
<td>Medium</td>
<td>High</td>
</tr>
<tr>
<td>Body weight (kg)</td>
<td>302</td>
<td>304</td>
<td>311</td>
<td>285</td>
<td>284</td>
<td>304</td>
</tr>
<tr>
<td>Puberty &amp; Growth</td>
<td>293</td>
<td>299</td>
<td>312</td>
<td>289</td>
<td>298</td>
<td>331</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>306</td>
<td>322</td>
<td>346</td>
<td>305</td>
<td>327</td>
<td>380</td>
</tr>
<tr>
<td>Average daily gains (ADG) (g)</td>
<td>61^a</td>
<td>117^ab</td>
<td>124^b</td>
<td>20^a</td>
<td>156^ab</td>
<td>312^b</td>
</tr>
<tr>
<td>Puberty &amp; Growth</td>
<td>-106^a</td>
<td>-33^ab</td>
<td>109^c</td>
<td>-17^ab</td>
<td>92^bc</td>
<td>219^c</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>231^a</td>
<td>264^a</td>
<td>356^b</td>
<td>268^ab</td>
<td>411^b</td>
<td>542^c</td>
</tr>
<tr>
<td>ME used for body retention (MJ/day)</td>
<td>0.92^a</td>
<td>1.66^a</td>
<td>1.84^ab</td>
<td>0.26^a</td>
<td>2.17^a</td>
<td>4.79^ab</td>
</tr>
<tr>
<td>Puberty &amp; Growth</td>
<td>-1.44^a</td>
<td>-0.47^ab</td>
<td>1.63^bc</td>
<td>-0.21^ab</td>
<td>1.35^bc</td>
<td>3.46^b</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>3.46^a</td>
<td>4.06^ab</td>
<td>5.86^bc</td>
<td>4.08^abc</td>
<td>6.59^abc</td>
<td>9.82^c</td>
</tr>
<tr>
<td>Body condition score (points)</td>
<td>5.70</td>
<td>5.52</td>
<td>5.76</td>
<td>4.60</td>
<td>4.77</td>
<td>5.00</td>
</tr>
<tr>
<td>Puberty &amp; Growth</td>
<td>5.72</td>
<td>5.62</td>
<td>5.92</td>
<td>4.42</td>
<td>4.85</td>
<td>5.30</td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>5.67</td>
<td>5.82</td>
<td>6.30</td>
<td>4.35</td>
<td>4.91</td>
<td>5.59</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>21.9</td>
<td>39.5</td>
<td>38.8</td>
<td>9.3</td>
<td>54.1</td>
<td>82.5</td>
</tr>
<tr>
<td>Feed conversion ratio (g ADG/kg DOM)</td>
<td>77.9</td>
<td>74.5</td>
<td>85.4</td>
<td>91.7</td>
<td>113.9</td>
<td>111.4</td>
</tr>
</tbody>
</table>

^A-C Differing capital letter superscripts denote significant differences between means of physiological state within columns within variable (P < 0.05) as determined by variance analysis model 1.

^a-c Differing lower case superscripts denote significant differences between means within rows (P < 0.05) separate for each physiological state as determined by variance analysis model 2. P-value: + P<0.1, * P<0.05, ** P<0.01, *** P<0.001, ns=non significant.

^1 Significant FxP interaction (P < 0.001).

^2 Significant GxP interaction (P < 0.001).

^3 Body condition score ranks animals from 1=extremely meagre to 9=extremely fat condition.

^4 Estimate based on AFRC (1993). GxP interaction (P < 0.1).

^5 DOM= = intake of digestible organic matter. Not calculated for the 'work' period due to negative ADG values in some groups; significant FxP interaction (P < 0.05).
Boran and crossbred heifers showed the first oestrus at ages of 3.4 and 2.0 years, respectively. The age difference in the experimental design of 2.2 years therefore surpassed the actual difference in manifestation of oestrus activity by 8 months. Success of conception did not differ significantly between genotypes. On average the heifers were inseminated 1.92 (±0.127) times until they conceived. Although feeding level did not significantly influence the non-return rate, there was a weak trend for the crossbred heifers fed increasing amount of feed to conceive slightly earlier (2.3, 1.7 and 1.6 times inseminated with low, medium and high feeding level, respectively). Bos indicus heifers showed slightly shorter ($P < 0.1$) gestation lengths (275±0.7 days) when compared with crossbreds (277±0.4 days). All heifers were pregnant during the pregnancy & recovery period and were on average 2.1 (±0.21) months in gestation when the digestibility measurement was carried out.

### II.3.2 Estimated energy requirements

The results of the prediction of $\text{ME}_m$ are shown in Table II-4 and Fig. II-1. Non-significant components of the model ($\text{ME}_i$, genotype (G) or $\text{ME}_i \times G$) were removed until all remaining components were significant, while the calculation of the specific $\text{ME}_m$ was conducted using the overall model (i.e., including $\text{ME}_i$, G and $\text{ME}_i \times G$).

Energy requirements for maintaining $W$ during puberty & growth were not different between genotypes. The $\text{ME}_i$ per $W^{0.75}$ alone explained 0.42 of the variation in ADG during the puberty & growth stage ($P < 0.001$), while 0.44 of the variation was explained by the overall model including genotype and interaction ($P < 0.001$). Boran and crossbred heifers required similar amounts of 0.511 (±0.164) and 0.538 (±0.100) MJ/kg $W^{0.75}$. The $\text{ME}_m$ per kg of $W^{0.75}$ was 0.540 (±0.104) MJ when the genotype effect was excluded from the model. In the work & growth period, energy requirements for zero weight gain were 0.707 (±0.113) and 0.633 (±0.098) MJ/kg $W^{0.75}$ for Boran and crossbred heifers, respectively. Pulling sledges required 0.772 (±0.102) and 0.697 (±0.067) MJ/kg $W^{0.75}$ daily, while walking required only 0.689 (±0.115) and 0.528 (±0.124) MJ/kg $W^{0.75}$ for Boran and crossbreds, respectively. Assuming that $\text{ME}_m$ did not change relative to the previous state, the differences (on average of both types of work) of 0.196 and 0.095 MJ/kg $W^{0.75}$ were accounted for the energy required to perform work.
in Boran and crossbreds, respectively. Genotype effects were significant \((P < 0.05)\) when the interaction with feeding level was excluded from the regression model. The relationship between \(\text{ME}_i\) and \(\text{ADG}\) was positive \((P < 0.001)\) in both genotypes, and \(\text{ME}_i\) explained 0.53 of the variation in \(\text{ADG}\) similar to the overall model (0.53).

Figure II-1 Effect of daily intake of metabolisable energy (\(\text{ME}_i\)) on average daily weight gain (ADG) of Boran and Holstein \(\times\) Boran heifers in three physiological states (for equations cf. Table II-1).
Table II-4 Regression analysis of the conversion of the metabolisable energy consumed (ME) into average daily weight gain (ADG) during the different physiological states and estimates of metabolisable energy requirements for maintenance (ME\textsubscript{m}) and of the efficiency of metabolisable energy utilisation for growth (k\textsubscript{g})

<table>
<thead>
<tr>
<th>Physiological state</th>
<th>Equation constants</th>
<th>Estimates\textsuperscript{1}</th>
<th>R\textsuperscript{2}</th>
<th>C.V.</th>
<th>ME\textsubscript{m} of Boran (MJ/kg W\textsuperscript{0.75})</th>
<th>ME\textsubscript{m} of HxB\textsuperscript{1} (MJ/kg W\textsuperscript{0.75})</th>
<th>k\textsubscript{g}</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>intercept ME\textsubscript{E} (MJ/kg W\textsuperscript{0.75}) Genotype\textsuperscript{4} ME\textsubscript{E} x Genotype</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Puberty &amp; Growth</td>
<td>-0.408</td>
<td>0.798</td>
<td>-0.252</td>
<td>0.428</td>
<td>0.439 88.98</td>
<td>0.511 0.538</td>
<td>0.312</td>
</tr>
<tr>
<td>Work &amp; Growth</td>
<td>-0.674</td>
<td>0.954</td>
<td>-0.070</td>
<td>0.001</td>
<td>0.527 223.3</td>
<td>0.707 0.634</td>
<td>0.255</td>
</tr>
<tr>
<td>Pregnancy &amp; Recovery</td>
<td>-0.168</td>
<td>0.619</td>
<td>-0.144</td>
<td>0.335</td>
<td>0.645 23.81</td>
<td>0.271 0.326</td>
<td>0.269</td>
</tr>
</tbody>
</table>

\textsuperscript{1} The model used was \( y = a + b \text{ME} + c \text{genotype} + d \text{(ME x genotype)} \).

\textsuperscript{4} Non significant interaction. Factor accounts only for crossbred heifers, for Boran heifers constant is zero.

\textsuperscript{4} Non significant differences between genotypes. Factor accounts only for crossbred heifers, for Boran heifers constant is zero.

\textsuperscript{1} Holstein x Boran.
At early stage of pregnancy while recovering from work ME\textsubscript{m} (i.e., zero ADG) was apparently far lower than in all other physiological states accounting for 0.271 (±0.194) and 0.326 (±0.112) MJ/kg W\textsuperscript{0.75} for Boran and crossbred heifers, respectively. Together, ME\textsubscript{J} (P < 0.001) and genotype (P < 0.001) explained 0.63 of the variation in ADG, while the overall model accounted for 0.65 of the variation (P < 0.001). Efficiencies of ME utilisation for body energy retention (k\textsubscript{g}) were not different (P > 0.1) between genotypes. During puberty & growing, work & growth and pregnancy & recovery states k\textsubscript{g} ranged at 0.312 (±0.053), 0.255 (±0.039), and 0.269 (±0.042), respectively (Table 11-4).

11.4 Discussion

11.4.1 Feed intake and digestibility

Intake differences between feeding levels were always significant. When receiving the high feeding level, the intended level of increase in intake was smaller in Boran heifers in the two first physiological states because of refusals of 12 to 13% of the feed offered. Obviously, this amount of feed exceeded either their forage intake capacity or the threshold for physiological satiety. The first explanation seems more likely in the second experimental period where requirements were enhanced by working exercise. All noted interactions of feeding level and genotype in intake-related traits were caused by these refusals. In the pregnancy & recovery state, also Boran heifers offered the high feeding level had no significant refusals and consumed more than 75 g OM/kg W\textsuperscript{0.75}/day. Lipostatic regulation of intake is usually induced by the size of fat deposits; however, compensatory growth, which took place during the pregnancy & recovery period, is known to increase the point of satiety (Langhans and Scharrer, 2000).

The digestibility of feed can be influenced by work through increased body temperature, food particle residence in the gastro-intestinal tract and the efficiency of mastication of the breakdown of particles (Weston, 1985). However, effects on digestibility in working animals are variable as shown by Pearson and Lawrence (1992). These researchers found an increased OM digestibility in oxen fed roughage (one experiment) and a more concentrate-based diet (second experiment). However, a decrease in OM digestibility due to work occurred when roughage of poor quality was offered (second experiment). Fall \textit{et al.} (1997) found no changes in OM digestibility of
Zebu oxen when working for 0, 2.5 or 5 h/day. In the present study, OM digestibility was initially low (puberty & growth state) and increased in the work & growth period and was maintained at this level in the last period. This excludes the explanation of Pearson and Lawrence (1992) of a reduced digestibility due to the lack of time for rumination. The observed prolonged rumen retention time in the work & growth state relative to the pregnancy & recovery state instead supports the hypothesis of Weston (1985). Also others (Pearson and Lawrence, 1992; Fall et al., 1997; 5 h/day of work) found higher rumen retention times in working animals. However, for the present study it remains unexplained why the change in digesta kinetics from the work & growth to the pregnancy & recovery states was not associated with variations in digestibility of OM and energy. Generally, neither feeding level nor genotype had a significant influence on digestibility and digesta kinetics (except of effects of feeding level on transit time). This is in line with observations made by Doreau and Diawara (2003) describing only moderate changes in digestibility with varying feeding level, while Grimaud et al. (1998) and Atti et al. (2002) found decreases in digestibility in a situation of drastic underfeeding.

II.4.2 Changes in body weight and condition

In both genotypes there were significant increases in ADG with increasing feeding level, but the response to higher feeding levels in ADG seems to have been more pronounced in the crossbred heifers. This probably reflected the higher genetic growth potential of crossbreds compared to the Boran. Low responses in the Boran to high instead of medium feeding level, additionally illustrate that in the puberty & growth state the genetic potential of this genotype was already reached with medium feeding level. In the Boran heifers, BCS was higher with all feeding levels, including the group with low feed supply, than in the crossbred. This underlines the more pronounced genetic affinity of Boran towards a higher body fat retention, which additionally might explain the lower level of ADG, since weight gain consisting of fat tissue requires more dietary energy than that based on lean tissue.

As the feed allocation was not changed relative to W_{0.75} in the work period, heifers lost weight, except when receiving clearly more than the theoretical level of maintenance requirements for energy. Boran lost more W than the crossbreds during the work
period, presumably because of their higher body fat stores available at the start of the work period. Fat tissue is far easier to mobilise than body protein and can be more efficiently utilised for energetic purposes. However, Boran at low feeding level still had higher BCS in this experimental period than crossbreds with any feeding level. In the recovery period, where energy requirements for pregnancy were still low due to its early stage, compensatory growth was obviously occurring in both genotypes and with all feeding levels, with ADG being 1.6 times higher than those found during the puberty & growth state. Also Tolla et al. (2002) demonstrated the ability of Boran cattle (here oxen) for significant compensatory growth. Overall, this process was associated with a significant increase in BCS. In the Boran receiving the high feeding level, BCS could be considered excessive, and the BCS levels of the Boran group fed the low level was attained only by the crossbreds given the highest feed amount.

II.4.3 Reproductive performance
In the present study previous observations (Zerbini et al., 1993) were confirmed by showing that crossbred heifers tended to conceive better at high intake levels. Effects of feeding level on reproductive performance were also evaluated in lactating Boran crossbred cows with and without exposure to work (Zerbini et al., 1993, 1996; Zerbini and Wold, 1999). These researchers concluded that energy loss due to work affected reproductive performance of non-supplemented cows, and oestrus and conception were delayed until a specific energetic status was regained. Weight loss due to draught was also relatively fast regained. In the studies quoted above, pregnancy did not affect the ability to perform a similar work load as non-pregnant cattle, but re-storage of W losses in a subsequent recovery period was higher in non-supplemented, pregnant cows than in non-pregnant cows. This may have been at least partially the result of the W gain which has to be attributed to gains in reproductive organs.

The average gestation length of about 275 days was in the range of results reported by Mukasa-Mugerwa (1989) who also described that Bos indicus tended to have a longer gestation than Bos taurus cattle. However, this was not the case in the present study comparing crossbreds and purebred Boran. In farm practice, artificial insemination is typically less efficient in Boran compared to Bos taurus cattle since oestrus detection is difficult, as it is shorter, occurring often during night, and the social hierarchy in the
herd allows the teaser bull mounting only once per hour while crossbred cows allow 2.8 mounts/h (Mukasa-Mugerwa, 1989).

II.4.4 Feed utilisation and energy requirements

Feed utilisation. Feed conversion efficiency, here described as the amount of digestible OM required per unit of weight gain, was generally improved by increasing feeding level in both genotypes. This means that the increase in ADG exceeded that in energy intake because of the declining proportion of total energy intake spent for maintenance. At medium and high level of intake, feed conversion ratio was superior in the crossbreds compared to the Boran due to their high genetic growth potential. At the low feeding level, instead, under-nutrition had a severe adverse impact in the crossbreds thus reducing their comparative advantage. The occurrence of compensatory growth, finally, ensured a far higher feed utilisation in the pregnancy & recovery state compared to the puberty & growth state, and in the work period on average there was no noteworthy utilisation of feed for growth anyhow as the energy exceeding maintenance requirements was mainly spent to perform work. In the present investigation, there was no clear evidence from regression analysis that feeding levels and physiological states directly affected the ME amount required per unit of W gain. Assuming a fairly constant energy content of weight gain, this is also true for the efficiency of ME utilization for growth ($k_y$). The average level of $k_y$ of 0.28 favourably compares with range of $k_y$ found for roughage diets (compiled by NRC, 1984). Although the genotype effect was not significant, the result of the regression analysis suggests that there might be a slightly lower $k_y$ in Boran heifers in two of the three physiological states (puberty & growth and pregnancy & recovery), i.e. in situations with consistent weight gain. The latter observation, however, could have been an artefact due to the genetic limitations in growth which were lower in the Boran than in the crossbreds, thus apparently limiting $k_y$ at the highest plane of nutrition.

Maintenance energy requirements. The only physiological state, where feed conversion could be clearly attributed to only maintenance and growth, was puberty & growth, i.e., the first experimental period. The $ME_m$ estimated by regression during this period was quite similar between genotypes. The estimates of this study fit well into the range of
0.570 (±0.075) MJ/kg W⁰.⁷伍 described as the average of various studies (reviewed by Agnew and Yan, 2000). Although in the groups receiving the low plane of nutrition, energy was provided theoretically only at an amount which would not allow for growth, there was on average weight gain during puberty & growth suggesting that either energy requirement of the heifers was over-estimated or the energy content of the diets was under-estimated. Considering the actually measured diet contents of digestible OM (599 g/kg DM) and energy (12.35 MJ/kg DM), the latter is more likely. Accordingly, the predicted ME content (0.81×DE) of the diet was higher by 1.16 MJ/kg than the initially assumed value. Furthermore, the MAFF (1987) formula used when deciding on feed allocation includes a safety margin of proportionately 0.05. Together, this means that the treatment groups supposed to get only 1.0 x MEₘ actually received 1.1 to 1.2×MEₘ which corresponds to the estimate calculated from the regression analysis.

Energy requirements for work. Heifers of both genotypes were able to cope with the work load which was imposed on them. Also N'dama heifers in West Africa in which the onset of fatigue when ploughing was occurring only slightly earlier (proportionately 0.75 of time) than in oxen (Cole, 1997). Pulling sledges was found to cost 2.9 J/m while walking required only 0.9 J/m (P < 0.05). This is in the range of 1.0-2.2 J/m reported for walking, carrying loads or ploughing (Fall et al., 1997; Pearson et al., 1999). The estimates of ME requirements for work suggest that crossbreds apparently had only half of the requirements than the Boran (average predicted requirements were 2.4 and 1.2 J/m for Boran and crossbreds, respectively). The extra energy costs for work thus added up to proportionately 0.38 and 0.28 of MEₘ for Boran and crossbreds. This is comparable to values reported by Pearson et al. (1989) of +0.24 to +0.46 of MEₘ, but lower than the +0.67 estimated for Brahman walking for 5.5 h/day (Pearson and Archibald, 1989). An explanation for the higher W losses during work for Boran heifers (resulting in the apparently higher energy requirements for work) would be given when the crossbreds had mobilised body reserves for work in a way which was not reflected in corresponding W losses. Considering that kₐ apparently was similar to that calculated in the other physiological states there seems to have been no direct effect of
feeding level on energy requirement for work, an effect which could have been expected in excessively undernourished or obese animals.

**Maintenance energy requirements during the recovery stage.** Compensatory growth during the pregnancy & recovery state obviously did not change the metabolic energy utilisation for growth ($k_g$) relative to the other physiological states, but reduced the minimum ME$_r$ required to maintain W by more than a third. Reduced maintenance requirements during compensatory growth might be explained by persisting changes occurring in lipostatic signals and minimised energy turnover after a period of enhanced activity (work). Maintenance requirements are mainly accounting for maintaining functions in organs like liver (0.12), kidneys (about 0.07 of total) and other visceral organs. Due to enhanced activity and weight losses during the work period probably a reduction of visceral organs mass took place. Compensatory growth is known to last until protein and nutrients are replenished in visceral organs (Drouillard et al., 1991). Furthermore, the reduced ME$_m$ in the pregnancy & recovery state may reflect a shift of the organism toward a low metabolic rate which, however, is in contrast to the known very low efficiency of ME utilisation of 0.10-0.15 for pregnancy (Hoffmann, 1996). Obviously, the effect of compensatory growth was far higher than that of pregnancy. In the pre-work period, metabolic rates may have been higher as organs and animals were growing in size, while during the work period, where the animals did not gain much W, organs should have still become larger in size as a consequence of the body’s homeorhetic control. This explanation is supported of Freetly et al. (2002) who noted that metabolic rate declines with age and depends on breed. In mature cows, by contrast, Freetly and Nienaber (1998) found higher heat production when animals expressed high ADG, especially during compensatory growth.

A major advantage of *Bos indicus* to adapt to tropical conditions is the ability to reduce rectal temperature (an indicator of thermal balance which is related to growth; Kadzere et al., 2002) under heat stress, an ability which purebred *Bos taurus* cattle lacks (Finch, 1986). This could explain the slightly lower estimated ME$_m$ of the Boran cattle compared to the crossbreds in the pregnancy & recovery state.
II.5 Conclusions

In the present study, type and the low and medium amount of feed provided were corresponding to those available to smallholder farming systems in Sub-saharan Africa. Results showed that crossbred heifers respond well to higher feed intake in any of the physiological states. Boran heifers, with a genetically lower growth potential, responded particularly when switched from low to medium feeding level. This would suggest that extra feed resources get more efficiently utilised by crossbred. On the other hand, under practical conditions, stress related to diseases and feed fluctuation could be detrimental to poorly fed or undernourished crossbred animals, which still performed well under controlled conditions in the present study. Efficiency of utilisation of ME for growth was similar in both genotypes and in the different stages investigated, and was unfavourably low accounting for 0.26 to 0.31 due to the poor quality of the diet. This underlines a clear potential for improving metabolic and overall energy utilisation, at least in the crossbred heifers, when using diets of higher digestibility. The large differences found in ME requirements between physiological states indicate the need for review of the current energy standards in the tropics. Long-term studies, like the one performed with same animals but during the first two lactations (Jenet et al., 2004), will assist to evaluate the viability of smallholder dairy systems based on the income generation from milk production in order to be able replace traditional systems using draught oxen.

Acknowledgements

The authors are grateful to Olga Karenfil, Bekele Andarge, Worku Getachew, Wana Abaguchi and Arage Bekele for handling the experimental animals. Assistance in the construction of a walking path and harness for single cow by the technical staff of the Animal Feed and Nutrition Unit and the workshop of Debre Zeit is appreciated. Further thanks are portrayed to the biometric team of Mamadou Diedhiou, and Aklilu Bogale, Amare Atale for data handling. This work has been partially supported by the Swiss Agency for Development and Corporation (SDC) through the Swiss Centre for International Agriculture (ZIL), ETH Zurich.
References


Long-term response to feeding level in lactational performance of Boran (Bos indicus) and Boran × Holstein cows

based on:

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Abstract

In an experiment lasting from 1.5 years before first calving until third parturition, 24 purebred indigenous Bos indicus (Boran) cows and 24 Boran crosses with Holstein (proportionately 0.50 and 0.75 Holstein blood level) were fed a diet composed of bermuda grass hay and wheat bran (2:1) offered at low, medium and high level. This level was adapted in amount to actual body weight every second week corresponding to assumed 1.0, 1.2 and 1.4 times of maintenance energy requirements. Cows were subjected to working exercise before and after first parturition. Body weight differences (lower with low and medium feeding level) developed mostly before calving for the first time and showed the typical decline at the start of lactation and increase in the dry period. Additionally, Boran cows fed the high level increased their body weight from the first to the second reproduction cycle. Body condition scores were higher in the Boran cows and, in both genotypes, with high feeding level. Independent of feeding level and genotype, calving intervals in cycles 1 and 2 were 530 and 421 days, respectively. Lactation length was considerably shorter in the Boran cows than in the crossbred cows. Milk yield, calculated over the first 13 weeks of lactation and over the whole lactation, was 2.06 and 3.06 times higher in the crossbreds than in the Boran. Milk of Boran cows had 1.24, 1.13 and 1.17 times higher contents of fat, total solids and protein. In the first 13 weeks of lactation, milk yield of crossbreds with high feeding level (8.7 kg/day) was higher (P < 0.05) than that of the crossbreds with medium (6.5 kg/day) and low feeding level (5.4 kg/day), respectively. Boran cows never showed a significant response in milk yield to feeding level. Accordingly, the amount of organic matter intake required per kg milk increased with feeding level in the Boran cows while it remained unaffected in the crossbreds. Organic matter digestibility, as measured in reproduction cycle 1, was higher (P < 0.05) in Boran. Response to high feeding level and estimated maintenance requirements of crossbred cows corresponded with current assumptions, but crossbreds subjected to medium feeding level expressed an unexpectedly low difference to those receiving the low level. It seems that current recommendations are not generally applicable to indigenous tropical breeds as these responded differently from crossbreds to feeding level.

Keywords: dairy cows, breed, zebu, under-nutrition, tropics
III.1 Introduction

Low availability and quality of feed are major constraints in increasing livestock productivity in the tropics. This often results in life-time under-nutrition of livestock. Restrictions may also be given by unimproved genotypes, as these are adapted to harsh conditions in a way that, when under-nourished, they do not substantially increase milk yield with improved feed supply (Olaloku and Oyenuga, 1974), but replenish body reserves first (Mukasa-Mugerwa et al., 1997). For decades, improved Bos taurus dairy breeds have been introduced in tropical countries in order to achieve higher productivity per animal, and this has resulted in noticeably higher incomes for smallholder milk producers (Bebe et al., 2003). Because of their low resistance against tropical diseases, imported genotypes are typically used to generate crossbreds with indigenous genotypes either as F1 or F2 (proportionately 0.75 blood level of one of the breeds). Different from under-nourished cows of indigenous breeds, there is at least a moderate response in milk yield of crossbreds to additional feed under tropical conditions (e.g. Gemeda et al., 1995). However, it is still unknown if these attitudes are different when this level is kept unchanged from the time before the first lactation starts. Additionally, there may be differences in maintenance requirements of Bos indicus and Bos taurus (e.g. Reid et al., 1991) and in requirements for milk production, the latter because of the differing milk composition. Direct comparisons over a long period are still lacking. The objective of the present study was, therefore, to investigate the lactational performance of the entire first two reproductive cycles (lactation and dry period) in indigenous and crossbred dairy cows already reared on always the same low, medium and high feeding levels from 1.5 years before their first parturition. This should show whether long-term abilities to adapt to feeding level exist in the two genotypes and whether or not these are similar. The study also should help to refine existing recommendations for the nutrient and energy supply of dairy cows (e.g. MAFF, 1987; AFRC, 1993; NRC, 2001) often developed from data of Bos taurus cows kept in moderate climates and thus being uncertain for Bos indicus and crossbreds kept in tropical environments (Kearl, 1982).
III.2 Materials and methods

III.2.1 Animals and nutrition

In the present study, either an East African *Bos indicus* landrace (Boran) or crossbreds of Boran and *Bos taurus* (Holstein Friesian) were used. In the crossbreds, proportionate Holstein blood levels were 0.50 and 0.75 (1:1) as the two major options for tropical smallholders (F1 and F2 crosses). Crossbreds were sired by eight Dutch Holstein Friesian bulls using imported semen (Holland Genetics, Arnhem, The Netherlands). All animals were subjected to the same nutritional regime from birth to the start of treatment feeding in the heifer stage approximately at their onset of cyclic activity. Calves were bred and reared on ILRI station, tethered in a calf barn and fed restricted amounts of milk in the first 2 months after birth, and *ad libitum* grass hay and a concentrate mix (1.5% of body weight (BW)) consisting of wheat middling (0.56 of total), noug cake (0.24), maize grain (0.185), salt sodium chloride (0.005) and bone meal (0.01) for 6 months. Then calves were moved to a feedlot receiving seasonally available grass hay until being allocated to the experimental groups. To ensure that the first parity will take place at the same time and that treatment feeding could start at the same date for all animals, the assumed first manifestation of oestrus activity was chosen besides BW as target physiological state for selection out of the station herds. In the literature, comparisons of onset of puberty and age at first calving show marked differences between crossbreds and Boran (e.g., Haile-Mariam and Kassa-Mersha, 1994). Accordingly a total $2 \times 24$ Boran and crossbred heifers were selected at ages of 4.4 (s.d. 0.19) and 2.2 (s.d. 0.27) years, respectively; however, heifers showed first oestrus at mean ages of 3.4 and 2.0 years with a high individual variation, so that the age difference in the experimental design of 2.2 years surpassed the actual difference in manifestation of oestrus activity by 8 months on average. At start of treatment feeding, the average BW were similar with 297 (s.d. 24.0) kg for Boran and 287 (s.d. 28.9) kg for Boran x Holstein cows. Animals were blocked within genotypes by BW and age and, in the crossbreds, by blood level, and were randomly allocated in a $2 \times 3$ factorial arrangement to three feeding levels. These were equivalent to 1.0, 1.2 and 1.4 times of maintenance requirements for metabolisable energy (ME$m$) calculated as $8.3 + 0.091 \times$ fasting BW (kg) as suggested by MAFF (1987). The feeding levels are further on referred
to as low, medium and high feeding level. Throughout the experiment, animals were weighed every second week between 7 and 9 a.m. after 12 h of fasting. This was the basis for adjustments of the individual feed allocation to the actual theoretical ME\textsubscript{m}. The diet always consisted of bermuda grass (*Cynodon dactylon* L.) hay (proportionately 0.65 of the diet) and wheat bran (0.35), which were allocated in amounts according to the treatment schedule applying estimates of MAFF (1987) of 7.5 and 10.1 MJ ME/kg for the bermuda grass hay and the wheat bran, respectively. Additionally, animals got *ad libitum* access to mineral licking blocks containing 120 g Ca, 39 g P and 1.3 g S per kg (The Awash Mineral Block Production Industry, Awash, Ethiopia). Crude protein and minerals supplied through feed and mineral blocks were calculated to cover more than maintenance requirements (Kearl, 1982; NRC, 2000), thus leaving energy supply as the first limiting trait. However, in periods of higher milk production protein may have been co-limiting while minerals were estimated to have been always sufficient in relation to energy supply. This guaranteed that all nutritional effects could be attributed to feeding level. The analysed composition of the Bermuda grass (*Cynodon dactylon*) and of the wheat bran is given in Table III-1. Data show a relatively small variation among monthly drawn samples.

Artificial insemination was performed from the start of treatment feeding on the first occurrence of oestrus. For that purpose, oestrus activity was monitored by daily observation of a veterinarian, of barn attendants and with the help of a teaser bull freely walking in the barn. After heifers had entered the experiment for 8 months they were palpated rectally for confirmation of oestrus and the presence of a corpus luteum. Animals with confirmed presence of corpus luteum were inseminated with Holstein Friesian semen on detected oestrus over a 5~day period. Following artificial insemination, pregnancy diagnosis was conducted after 60 days. On average, Boran heifers calved first at an age of 5.9 years (s.d. 0.24) and crossbred heifers with 3.7 years (s.d. 0.31), i.e. after 1.5 years of treatment feeding. This was the time when the intensive measurements described here started which comprised the entire first two reproduction cycles. As exposition to work is common for livestock in tropical smallholdings, animals were subjected two times for 51 days to working exercise at first in the rearing period within a 3-month period, beginning 4 months after the start of
Table III-1 Analysed composition of the experimental feeds (per kg DM; average of monthly analyses)

<table>
<thead>
<tr>
<th>Feed</th>
<th>Bermuda grass hay</th>
<th>Wheat bran</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>s.d.</td>
</tr>
<tr>
<td>Dry matter (g/kg as fed)</td>
<td>905</td>
<td>19</td>
</tr>
<tr>
<td>Organic matter (g)</td>
<td>899</td>
<td>14</td>
</tr>
<tr>
<td>Crude protein (g)</td>
<td>45</td>
<td>6</td>
</tr>
<tr>
<td>NDF (g)</td>
<td>752</td>
<td>31</td>
</tr>
<tr>
<td>ADF (g)</td>
<td>436</td>
<td>25</td>
</tr>
<tr>
<td>ADL (g)</td>
<td>68</td>
<td>4</td>
</tr>
<tr>
<td>Gross energy (MJ)</td>
<td>17.4</td>
<td>0.5</td>
</tr>
<tr>
<td>P (g)</td>
<td>2.0</td>
<td>0.4</td>
</tr>
<tr>
<td>Ca (g)</td>
<td>3.9</td>
<td>0.8</td>
</tr>
<tr>
<td>Mg (g)</td>
<td>1.8</td>
<td>0.6</td>
</tr>
<tr>
<td>Cu (mg)</td>
<td>10</td>
<td>3</td>
</tr>
<tr>
<td>Zn (mg)</td>
<td>22</td>
<td>4</td>
</tr>
</tbody>
</table>

the experiment, and secondly within the first reproduction cycle (between months 2 and 4 of lactation). The work was performed on 5 days per week either by pulling sledges (weighing 0.15 of BW on a 50 m path) or by walking on a 0.5 km long path at a rate of 5h/day. As members of each feeding level x genotype sub-group were allocated in a balanced manner to the two types of exercise, working treatment was not specifically regarded in data evaluation.

### III.2.2 Housing and experimental procedures

Cows were tethered in individual pens with concrete floor (1.2 x 2.2 m) in a well-ventilated barn at ILRI Debre Zeit Research Station, Ethiopia, located at 1900 m above sea level. Once daily after milking at 6 a.m. wheat bran followed by hay was provided in concrete troughs which were separated by concrete walls of 1 m height. Daily feed offered and refused was recorded during all stages of the experiment, and every second week approximately 500 g samples of feed were taken and thoroughly mixed. Samples of feed refusals of individual animals were collected. Both feed and refusals were pooled to monthly samples. Animals had free access to fresh tap water offered in buckets. Body condition scores were estimated every second week as outlined by Nicholson and Butterworth (1986) along with BW measurements by one trained person.
and a trained co-examiner (out of five persons in total) at each date. A nine-grade scale ranging from extremely meagre to extremely obese was applied. Milk yield was recorded twice daily from the second day after parturition, and proportional milk samples were taken every second week from one evening and the subsequent morning milk, treated with potassium dichromate (Laboratory supplies, Poole, UK) and stored at +4°C until analysis. Because of the need of a stimulus for milk release, purebred Boran calves were allowed to suckle for the first 60 days. In that case, milk yield was determined by a combination of the weigh-suckle-weigh method and actual milking. In detail, calves suckled before milking until milk let-down was observed. Then calves were tied near the head of the cow throughout milking and finally calves were allowed to suckle the remaining milk (c.f. Combellas et al., 2003). Cows were milked in both lactations either, similar to farmers practice in Ethiopia, until the exploitation of milk was less than 0.5 kg/day or until 3 months in advance of the calculated calving date. In the third month of the first lactation, total faecal amount was registered via complete collection immediately upon excretion during 24 h per day over a period of 6 days. The favourable consistency of the faeces (high DM content) and the shape of the concrete floor allowed collection with only negligible losses. Proportionate (0.1) faeces samples were obtained after thorough mixing and subsequently frozen at -20°C until analysis. The experiment was performed under the guidelines of, and retrospectively approved by, the Institutional Animal Care and Use Committee (IACUC, ILRI, Nairobi, Kenya). Animals were under permanent observation by an in-house veterinarian.

III.2.3 Laboratory analyses and calculations

Feed and faeces were analysed for contents of DM and organic matter (OM) according to AOAC (1990). In milk samples, contents of fat were determined by the Gerber method with BS 696 (British Standard Institute, 1989) and protein by formaldehyde titration (6.38 × N) as suggested by Scott (1986). Total solids contents were analysed according to BS 1741 (British Standard Institute, 1990). Energy-corrected milk (kg/day) was calculated, using the NRC (2001) formula, as milk (kg/day) × [0.3887 × fat (g/100 g) + 0.2289 × protein (g/100 g) + 0.8033]/3.14. Dry matter contents of grass hay and wheat bran on average amounted to 917 and 894 g/kg, respectively. A posteriori, digestible
energy (DE) of wheat bran was assumed to be constant (13.51 MJ/kg, NRC 2001) and the
conversion into ME was supposed to be \(0.81 \times \text{DE}\) (AFRC, 1993). Additionally, from
digestibility studies on the same animals with the same feed composition but not in
this experimental period, DE of the feed was calculated to actually account for 12.35
MJ/kg, with feed and faeces being analysed by ballistic bomb calorimetry (CBB-330-
20L, Gallenkamp, Loughborough, U.K.). This meant that actual ME contents were
probably 11.0 and 9.6 MJ/kg for wheat bran and grass hay (AFRC, 1993) which is higher
than the corresponding contents of 10.1 and 7.5 MJ ME/kg that had been used for feed
allocation.

According to AFRC (1993), \(\text{ME}_m (\text{MJ/day})\) were calculated as \([F+A]/k_m\) where \(F = \) fasting
energy metabolism (MJ/day) calculated as \(0.53 \times [\text{BW (kg)/1.08}]^{0.67}\); \(A = \) activity
allowance (MJ/day) for standing lactating dairy cows calculated as \(0.0095 \times \text{BW (kg)}\); \(k_m\)
= efficiency of energy utilisation for maintenance calculated as \(0.35 \times q_m + 0.503\); \(q_m\) =
metabolisability of gross energy (ME/GE; here set to 0.612). The ME required for milk
production \((\text{ME}_i \text{MJ/day})\) was computed as \([\text{milk yield (kg/day)} \times \text{EV}_i]/k_p\) where \(\text{EV}_i\)
(MJ/kg) = 0.0376 \times milk fat (g/kg) + 0.0209 \times milk protein (g/kg) + 0.948 and \(k_i = 0.35 \times q_m + 0.420\) (AFRC, 1993). Metabolisable energy content of BW change \((\text{ME}_g)\) was
assumed to amount to 19 MJ/kg (AFRC, 1993). Total ME allowances for retention \((\text{ME}_r)\)
comprised \(\text{ME}_i\) and \(\pm \text{ME}_g\).

### III.2.4 Statistical analysis

Some animals had serious reproductive problems, mainly as a result of treatment
feeding. Therefore, seven cows had to be excluded from the statistical evaluation of the
first reproduction cycle and additional four from the second reproduction cycle (details
concerning experimental group numbers see table headings). The latter four cows were
also excluded from all calculations referring to calving interval in reproduction cycle 1.
Results of the measurements performed in the heifer rearing period are described
elsewhere (Jenet et al., 2002). The data of the first and the second reproduction cycle
were compared with two between-subject factor analysis, one within-subject factor
analysis and the statement sum of square type 4 (SS4) by the general linear model (model 1) of SAS (2001):

\[ Y_{ijkl} = \mu + \alpha_i + \beta_j + \alpha \beta_{ij} + \gamma_{kij} + \delta_i + \alpha \delta_{ij} + \beta \delta_{ij} + \gamma \delta_{ijk} + e_{ijkl} \]  \hspace{1cm} (1)

where \( Y_{ijkl} \) = individual observation, \( \mu \) = overall mean, \( \alpha_i \) = effect of feed level, \( \beta_j \) = effect of genotype, \( \gamma_{kij} \) = variation in between cows, \( \delta_i \) = effect of reproduction cycle and \( e_{ijkl} \) = residual error. In the case of significant feeding level \( \times \) genotype interactions \((P < 0.05)\), the means of both reproduction cycles were separately compared by the least square means option of the general linear model of SAS (2001), and multiple comparisons of the means were carried out using the PDIF option and the Bonferroni method applying model 2:

\[ Y_{ijk} = \alpha_i + \beta_j + \alpha \beta_{ij} + e_{ijk} \]  \hspace{1cm} (2)

The tables give arithmetic means and standard errors (s.e.). Additionally, for all cows, data from the first 6 weeks of both reproduction cycles were subjected to regression analysis to predict MEr based on the effects of genotype, ME intake (MEi) and the interaction of genotype \( \times \) MEi. In this regression analysis the intercept represents MEi during fasting, the genotype effect represents the difference between genotypes in the amount of MEi required at zero retention (or the differences in their MEi at any given level of MEi in the absence of a significant MEi \( \times \) genotype interaction). The regression coefficient associated with MEi is an estimate of the efficiency of conversion of MEi to MEr and the MEi \( \times \) genotype interaction represents the difference in feed efficiency between the two genotypes. Not significant \((P > 0.05)\) terms were excluded in subsequent analysis. The regression analysis was carried out by the general linear model of SAS (2001) using the SOLUTION statement and the sum of square type 4 (SS4).

### III.3 Results

The realised daily intake of dry matter (DM) and organic matter (OM), as calculated over the entire reproduction cycles 1 and 2, differed \((P < 0.001)\) between feeding levels as intended (Table III-2). Intake was lower \((P < 0.05)\) in Boran than in crossbreds with, on average, 5.32 (±0.116) and 5.87 (±0.130) kg OM/day, respectively. This and the non-linear increase in feed intake from low to high feeding level (+0.19 and +0.56 of OM intake with medium and high level over both reproduction cycles and genotypes) resulted
from corresponding differences in body weight (BW) since feed allocation was oriented towards actual BW. Organic matter intake did not significantly differ between reproduction cycles. With increasing feeding level, digestibility of OM, as determined in reproduction cycle 1, declined \( (P < 0.05) \) by 0.069 in the crossbreds, while the Boran cows showed no corresponding response. Overall, OM digestibility was higher \( (P < 0.05) \) by 0.047 in the Boran than in the crossbred cows.

Feeding level clearly affected BW and, less so, BW changes during lactations and dry periods which illustrates that the major differences in BW developed before the first parturition (Table III-2). On average of both complete reproduction cycles, BW was not significantly affected by genotype and was low with low and medium feeding level and higher \( (P < 0.001) \) with the highest feeding level. Figure III-1 illustrates the evolution of BW over the respective first 40 weeks of the two reproduction cycles. In all groups there was an initial decline in BW which was followed by a slow increase after approximately 15 to 20 weeks from calving (earlier in reproduction cycle 2). In reproduction cycle 1, there were obvious group differences in BW concerning both genotypes and feeding levels. Genotype differences had disappeared in reproduction cycle 2, where even the large gap, which had been observed between Boran and crossbreds of the high feeding level in BW in cycle 1, was no longer present. Furthermore, in reproduction cycle 2, the BW of the remaining four treatment groups was almost indistinguishable. This resulted from the corresponding differences in the extents of BW losses during lactation and BW gains in the dry period (Table III-2) and also from taking into account the large differences in the lengths of lactation and dry periods (c.f. Table III-3).

Accordingly, in the lactation period the average BW losses of the crossbred cows were lower \( (P < 0.01) \) than in the purebred Boran, and they were also lower \( (P < 0.05) \) in reproduction cycle 2 than in cycle 1. There were also differences \( (P < 0.05) \) between reproduction cycles 1 and 2 in BW gain in the dry period (lower in reproduction cycle 2). Crossbreds, due to a short dry period and particularly when exposed to the high feeding level, showed higher daily BW gains in both dry periods (significant against most other groups; significant feeding level \( \times \) genotype interactions).
Table III-2 Effect of feeding level, genotype and reproduction cycle (C) on feed intake and digestibility as well as body weight and condition

<table>
<thead>
<tr>
<th>Genotype (G)</th>
<th>Boran</th>
<th>Boran x Holstein Friesian</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feeding level (F)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Number of animals (c %)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Low 5/5</td>
<td>Medium 7/7</td>
<td>High 8/8</td>
</tr>
<tr>
<td>Dry matter intake (kg/day)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>4.73</td>
<td>5.66</td>
<td>7.08</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>4.86</td>
<td>5.80</td>
<td>7.48</td>
</tr>
<tr>
<td>Organic matter intake (kg/day)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>4.36</td>
<td>5.23</td>
<td>6.52</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>4.44</td>
<td>5.29</td>
<td>6.85</td>
</tr>
<tr>
<td>Organic matter digestibility†</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>0.623</td>
<td>0.647</td>
<td>0.639</td>
</tr>
<tr>
<td>Body weight (kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>318</td>
<td>322</td>
<td>374</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>330</td>
<td>328</td>
<td>382</td>
</tr>
<tr>
<td>Body weight change (g/day)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lactating period</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>-191</td>
<td>-248</td>
<td>-53</td>
</tr>
<tr>
<td>Dry period</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>319</td>
<td>447</td>
<td>339</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>360</td>
<td>306</td>
<td>250</td>
</tr>
<tr>
<td>Body condition score (points)‡</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lactating period§</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>4.63</td>
<td>4.67</td>
<td>6.57</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>4.60</td>
<td>4.78</td>
<td>7.06</td>
</tr>
<tr>
<td>Dry period‖</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>4.98</td>
<td>4.86</td>
<td>6.89</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>4.84</td>
<td>5.24</td>
<td>7.96</td>
</tr>
</tbody>
</table>

abc Differing superscripts denote significant differences among means within rows (P < 0.05) separate for each lactation as determined by variance analysis model; +, P level < 0.1; † Measured in week 17 of reproduction cycle 1 and not determined in reproduction cycle 2. ‡ Body condition score ranks animals from extremely meagre (1 point) to extremely obese condition (9 points); § Significant GxC interaction (P < 0.05); ‖ FxGxC interaction (P < 0.1).
Figure III-1 Effect of feeding level (low, --; medium, ----; high, ——) and genotype (Boran, ○; crossbreds, ●) on the evolution of body weight in reproduction cycles 1 (a) and 2 (b).

Body condition scores (BCS) were significantly affected by feeding level and genotype in the lactation and in the dry periods, and feeding level × genotype interactions were significant (Table III-2).
In detail, purebred Boran cows generally showed a higher \( P < 0.001 \) BCS than crossbred cows, and BCS was higher with the high feeding level (more pronounced in the Boran) than with low and medium level. In reproduction cycle 2, there was a trend towards higher BCS in the Boran cows receiving the medium and the high feeding level and in the crossbreds offered the low level of feed, whereas all other groups showed slightly lower BCS values compared to those found in reproduction cycle 1 (significant three-way interaction among feeding level, genotype and reproduction cycle).

The calving interval was not significantly affected by feeding level and genotype, and was shorter \( P < 0.1 \) in reproduction cycle 2 than in cycle 1 by 108 days on average (Table III-3). This was accompanied by a shorter \( P < 0.05 \) length of lactation 2. The latter was almost exclusively due to the Boran cows (interaction of genotype and reproduction cycle: \( P < 0.05 \)), with their lactation 2 being shorter by 79 days on average, in contrast to a difference of only 8 days in the crossbreds. Feeding level did not significantly affect lactation length.

Whole lactational milk yield was 3.1 times higher \( P < 0.001 \) in the crossbred cows than in the purebred Boran (uncorrected data), and the difference in energy-corrected milk was only slightly lower (2.7 times of Boran). Also milk fat and protein yields were 2.6-fold and 3.1-fold higher \( P < 0.001 \) in the crossbreds compared to the Boran, respectively. Expressed on a daily basis, energy-corrected milk yield over the whole lactation period was not only different between genotypes, but was also affected \( P < 0.01 \) by feeding level. Additionally, there were feeding level \times \) genotype interactions \( P < 0.01 \) and daily yields were higher \( P < 0.01 \) in lactation 2 than in lactation 1. Plane of nutrition was only effective in the crossbreds and with the high feeding level (in lactation 2 significant only against the group with low feeding level). There was a non-significant curvilinear trend in the Boran, with the numerically highest milk yield being found with medium feeding level. In daily milk yield over the first 13 weeks similar kinds of effects occurred (Table III-4), but differences among treatment groups were more pronounced than for the whole lactation.
Table III-3 Effect of feeding level, genotype and reproduction cycle (C) on calving interval and whole lactational performance

<table>
<thead>
<tr>
<th>Genotype (G)</th>
<th>Boran</th>
<th>Boran×Holstein Friesian</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feeding level (F)</td>
<td>Low</td>
<td>Medium</td>
<td>High</td>
</tr>
<tr>
<td>Number of animals (c. 1/2)</td>
<td>5/5</td>
<td>8/7</td>
<td>5/3</td>
</tr>
<tr>
<td>Calving interval (d)†</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>525</td>
<td>475</td>
<td>601</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>382</td>
<td>395</td>
<td>445</td>
</tr>
<tr>
<td>Lactation length (d)‡</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>265</td>
<td>256</td>
<td>221</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>156</td>
<td>185</td>
<td>164</td>
</tr>
<tr>
<td>Milk (kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>485</td>
<td>647</td>
<td>586</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>369</td>
<td>656</td>
<td>553</td>
</tr>
<tr>
<td>Energy-corrected milk (kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>605</td>
<td>800</td>
<td>658</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>456</td>
<td>775</td>
<td>668</td>
</tr>
<tr>
<td>Fat yield (kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>28.4</td>
<td>37.6</td>
<td>29.5</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>21.2</td>
<td>33.7</td>
<td>30.1</td>
</tr>
<tr>
<td>Protein yield (kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>16.5</td>
<td>21.5</td>
<td>18.3</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>12.7</td>
<td>24.2</td>
<td>19.6</td>
</tr>
<tr>
<td>Energy-corrected milk (kg/day)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>2.80b</td>
<td>3.27a</td>
<td>2.94b</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>3.23b</td>
<td>4.28a</td>
<td>3.23b</td>
</tr>
<tr>
<td>Feed conversion rate (kg OM intake/kg milk)§</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>5.49</td>
<td>4.96</td>
<td>6.33</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>4.33</td>
<td>4.79</td>
<td>11.97</td>
</tr>
</tbody>
</table>

a,b,c Differing superscripts denote significant differences among means within rows (P < 0.05) separate for each lactation as determined by variance analysis model 2; † P level < 0.1; ‡ Number of animals for this trait are 5/4, 8/7, 5/3 for Boran receiving low, medium and high feeding level, respectively, and 7/7, 7/6, 8/8 for the respective Boran groups; ‡ Significant GxC interaction (P < 0.05); § Significant FxGxC interaction (P < 0.05).
Thirteen-week averages of daily milk yield of the crossbreds receiving the high feeding level were 1.36 and 1.62 times higher ($P < 0.05$) than that of the crossbreds with medium and low feeding level, respectively, and 2.73 times higher than the average of all Boran groups. Milk yield curves of the crossbreds showed a steady decline with time from parturition in both lactations whereas in the Boran cattle at least a flat initial part of the lactation curve was observed (Figure III-2). The pronounced drop in milk yield of the Boran cows between week 8 and week 10 of lactation reflects the time of weaning when even two out of three cows of the group with high feeding level in reproduction cycle 2 terminated milk production. Therefore, their lactation curve was not prolonged in Figure III-2 from that time-point on. Curves of both lactations were similar, with the exception that certain differences in the crossbreds fed either low or medium feeding level occurring in lactation 1 had disappeared in lactation 2.

Milk composition (contents of fat, total solids and protein), as measured on average of the first 13 weeks of lactation, was different between genotypes ($P < 0.001$) and reproduction cycles ($P < 0.001$), whereas feeding level had no significant effect (Table III-4). On average, the respective contents of fat, total solids and protein were 1.30-fold, 1.15-fold and 1.20-fold higher in the Boran compared to the crossbred cows.

Feed conversion rate, described here as the amount of OM required to produce one kilogram of milk and calculated over the complete reproduction cycle (Table III-3) and over the first 13 weeks of lactation (Table III-4), was affected by genotype ($P < 0.001$) and differed slightly between reproduction cycles 1 and 2 during the first 13 weeks of lactation. Using either time period, three-way interactions of feeding level x genotype x reproduction cycle ($P < 0.1$ and $P < 0.05$, respectively) were noted. Feed conversion ratio over the two reproduction cycles was 4.9 (±0.93) kg OM/kg milk in Boran with low feeding level, but increased up to 7.7 (±2.01) kg/kg with higher feeding level, while in the crossbreds the ratio was maintained with increasing feed level and ranged at 1.9 (±0.19) kg/kg on average. The corresponding results for the first 13 weeks of lactation were 1.6 (±0.12), 2.7 (±0.48) and 0.9 (±0.02) kg OM/kg milk (significantly higher in the Boran fed the high level than in the other groups).
Figure III-2 Effect of feeding level (low, ---; medium, ----; high, —) and genotype (Boran, ○; crossbreds, ●) on the evolution of milk yield in reproduction cycles 1 (a) and 2 (b).
The regression analysis of the ME spent for retention on ME intake \((\text{ME}_r)\) over the respective first 6 weeks of lactations 1 and 2 showed a significant linear equation in the crossbreds (Figure III-3) which explained 0.29 of the variation. According to the extrapolation to \(y = 0\), the estimate for maintenance requirements \((\text{ME}_m, \text{MJ/day})\) was 0.492 MJ/BW\(^{0.75}\) for the crossbred cows. The regression equation was not significant for Boran cows. Regressions analysis over both genotypes yielded the following equation:

\[
\text{ME}_r = 20.3 (\pm 19.7) + [-59.3 (\pm 24.0), \text{crossbreds}; 0, \text{Boran}] - 7.5 (\pm 24.1) \times \text{ME}/\text{BW}^{0.75} + [86.9 (\pm 28.8) \times \text{ME}/\text{BW}^{0.75}, \text{crossbreds}; 0 \times \text{ME}/\text{BW}^{0.75}, \text{Boran}], (R^2 = 43.0; P < 0.001).
\]

The coefficient associated with the genotypes indicates that the amount of \(\text{ME}/\text{BW}^{0.75}\) required for zero retention was different between genotypes, and the significant \(\text{ME}/\text{BW}^{0.75} \times \text{genotype}\) interaction suggests that the efficiency of ME use for maintenance purpose was also different between genotypes.

III.4 Discussion

III.4.1 Effect of feeding level and genotype on milk yield and feed conversion efficiency

The crossbreds, but not the purebred Boran, significantly responded in milk yield to additional feed. Crossbreds receiving the low level of feeding were able to produce about 4 kg of milk per day over two lactations at reasonably long lactations and short dry periods. This may be at least partially explained when assuming that the energy contents of the feeds initially were underestimated (c.f. materials and methods section). Accordingly, the calculations based on actual digestibility measurements suggested that the low feeding level was equivalent to 1.1 to 1.2 times of \(\text{ME}_m\) rather than 1.0 times of \(\text{ME}_m\). The lack of response to feeding level of purebred Boran cows to additional feed in milk yield suggests that even the lowest feeding level, which was supposed to be deficient in relation to maintenance and production requirements together, was sufficient to achieve the maximum milk yield; this at cost of body reserves.
Table III-4. Effect of feeding level, genotype and reproduction cycle (C) on milk yield and composition as well as feed conversion efficiency during the first 13 weeks of lactation

<table>
<thead>
<tr>
<th>Genotype (G)</th>
<th>Boran</th>
<th>Boran×Holstein Friesian</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Low Medium High</td>
<td>Low Medium High</td>
<td></td>
</tr>
<tr>
<td>Number of animals (c. 1/2)</td>
<td>5/5 8/7 5/3</td>
<td>7/7 8/7 8/8</td>
<td>s.e. FL G FxG C</td>
</tr>
<tr>
<td>Milk (kg/day)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>3.18&lt;sup&gt;d&lt;/sup&gt; 3.56&lt;sup&gt;d&lt;/sup&gt; 3.18&lt;sup&gt;d&lt;/sup&gt;</td>
<td>5.35&lt;sup&gt;b&lt;/sup&gt; 6.66&lt;sup&gt;ab&lt;/sup&gt; 8.42&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.355***</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>2.95&lt;sup&gt;c&lt;/sup&gt; 3.56&lt;sup&gt;bc&lt;/sup&gt; 2.88&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.40&lt;sup&gt;b&lt;/sup&gt; 6.25&lt;sup&gt;a&lt;/sup&gt; 8.90&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.400***</td>
</tr>
<tr>
<td>Fat (g/kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>56.4 61.9 57.3</td>
<td>42.2 45.2 47.0</td>
<td>1.77***</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>53.2 41.9 47.3</td>
<td>33.0 39.4 42.2</td>
<td>0.19</td>
</tr>
<tr>
<td>Total solids (g/kg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>138 145 136</td>
<td>122 122 125</td>
<td>2.1</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>141 129 130</td>
<td>112 116 122</td>
<td>2.4</td>
</tr>
<tr>
<td>Protein (g/kg)&lt;sup&gt;†&lt;/sup&gt;</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>32.0 33.3 32.9</td>
<td>29.0 27.9 29.1</td>
<td>0.05***</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>36.3 37.4 38.8</td>
<td>27.5 31.7 31.4</td>
<td>0.20 ***</td>
</tr>
<tr>
<td>Feed conversion rate (kg OM intake/kg milk)&lt;sup&gt;†‡§&lt;/sup&gt;</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reproduction cycle 1</td>
<td>1.65&lt;sup&gt;b&lt;/sup&gt; 1.72&lt;sup&gt;b&lt;/sup&gt; 2.53&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.97&lt;sup&gt;ab&lt;/sup&gt; 0.96&lt;sup&gt;a&lt;/sup&gt; 0.94&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.116***</td>
</tr>
<tr>
<td>Reproduction cycle 2</td>
<td>1.58&lt;sup&gt;a&lt;/sup&gt; 1.67&lt;sup&gt;a&lt;/sup&gt; 3.67&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.83&lt;sup&gt;a&lt;/sup&gt; 0.88&lt;sup&gt;a&lt;/sup&gt; 0.85&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.136 ***</td>
</tr>
</tbody>
</table>

*<sup>abc</sup> Differing superscripts denote significant differences among means within rows (P < 0.05) separate for each lactation as determined by variance analysis model 2.
†, P level < 0.1.
‡ GxC interaction (P < 0.1).
§ FxC interaction (P < 0.1).
§§ FxGxC interaction (P < 0.05).
Figure III-3 Regression of retained metabolisable energy, i.e. milk, ±body weight change \(y, \text{MJ/day}\) on intake of metabolisable energy \(x, \text{MJ/kg BW}^{0.75/\text{day}}\) during the first 6 weeks of lactation. Boran (○), no line; \(y = -20.3 (±12.3) - 7.5 (±15.0) x; R^2 = 0; \) not significant; Crossbreds (●), \(y = -39.1 (±16.5) + 79.4 (±18.8) x; R^2 = 0.29; P < 0.001 \) (line; extrapolated to \(y = 0\)).

The rapid decline in milk production of this genotype after 8 weeks of lactation was not dependent on nutrition but caused by the dependency of milk ejection on the stimulus of suckling which was terminated thereafter. Boran cows receiving increasing amounts of feed showed increasingly poor feed efficiencies due to the absence of response in milk yield, and even Boran with low feeding level were inferior to the crossbreds in feed efficiency. Concerning milk yield, this even did not account for the cows culled because of reproductive problems. The accumulated milk production of the experimental
groups of initially eight animals each over the entire period of the two reproduction cycles was 5305, 11,825 and 5294 kg milk for Boran cows receiving the low, medium and high feeding level, respectively, while milk amounts were 23,450, 25,286 and 34,368 kg in the corresponding crossbred cow groups. However, culling infertile cows as under farm conditions did not impair overall feed conversion efficiency further since no extra feed was consumed by these cows. Taking into account the huge genotype differences in the proportion of the inter-calving cycle as dry or lactation would cause Boran cows to make very inefficient use of energy for BW gain while not being under pressure to repartition energy to milk production. Additionally, comparisons including the cows culled for infertility reasons neglect the presumably superior resistance of the Boran against other than pure nutrient deficiency stressors and the negative impact of weaning after 60 d on milk production of the Boran genotype. Boran is a multipurpose type of cattle (milk, meat and draught yield) for smallholders in East Africa, and represents an important asset (Cossins and Upton, 1987). Over centuries African Zebu cattle was selected to cope with drought and extreme feed fluctuations, and furthermore has developed a certain resistance against tropical diseases (Tawah et al., 1997). In large parts of East Africa milk products are a major staple food (Cossins and Upton, 1987), being a significant source of smallholder household income (Gonfa et al., 2001). In Ethiopia, for instance, fermented dairy products with high fat and protein contents substantially contribute to human protein supply (Lindtjorn et al., 1993). Butter is a high price trade good used for oil and cosmetics in rural areas (Gonfa et al., 2001). It is likely that pastoralists selected Boran cows not predominantly for milk yield, because high fat and protein contents were more valuable. However, in spite of the higher concentrations of these milk constituents, Boran cows were less efficient in converting feed into milk fat and protein.

III.4.2 Effect of feeding level and genotype on body condition

The pattern of BW changes within the reproduction cycles was similar in all feeding levels and genotypes, showing the expected decline in the first weeks of each lactation and a plateau or an increase in later lactation and dry period. Additionally, initial BW differences between genotypes receiving the same feeding level were reduced from
reproduction cycle 1 to 2. Although showing an apparently low daily BW gain, the extremely long dry period allowed the Boran cattle with the high feeding level to approach the BW of the crossbreds receiving the same feeding level. Both the massive BW gain and the high BCS illustrate that the high feeding level was clearly excessive for Boran leading to increasingly obese animals. Similar BW at the same feeding level, despite genetically different target BW, meant that in the two genotypes nutrients were utilised differently for body fat stores and milk production, with the consequence that, at the same BW, BCS were clearly higher in the Boran cattle. Crossbreds fed on low and medium level were in poor condition, like the unsupplemented cows in the study of Gemeda et al. (1995). It seems likely that any disease would have been fatal in these permanently undernourished crossbred groups. However, it remains unexplained why there were no apparent changes in BW and BCS, and only a very moderate difference in milk yield, when crossbreds received the medium instead of the low feeding level. One explanation for this phenomenon could be the extremely long exposure to the low feeding level allowing adaptive mechanisms such as reduced muscular activities, digestive secretions, nutrient transports and substrate cycling for a reduced energy expenditure to develop.

III.4.3 Effect of feeding level and genotype on calving interval and lactation length

In contrast to the Boran cows subjected to medium feeding level, where nutrient and energy supply was sufficient to avoid deficiency-caused anoestrus (Zerbini et al., 1996), the high plane was clearly excessive, with the consequence of a disturbed reproduction in five out of the eight Boran cows. There is evidence for excessive body fat stores causing conception problems. Progesterone, stored in body fat tissue and released along with body fat during mobilisation at the start of the lactation, could suppress heat and prevent ovulation (Schopper and Claus, 1989). The average calving interval found in the Boran agrees with the 442 and 421 days reported by Haile-Mariam and Kassa-Mersha (1994) as well as Alberro (1983), but shorter calving intervals of Boran × Friesian cows (Alberro, 1983) were not observed. Length, particularly of lactation 2, was considerably shorter in Boran cows than in crossbreds partly caused by the different response to weaning. The correlation with calving interval was only 0.48 ($P < 0.05$) in
Boran but 0.72 ($P < 0.001$) in the crossbreds. Concerning calving interval, crossbreds again responded in a different way to feeding level than Boran. Zerbini et al. (1996) showed that calving interval of Boran crossbred cows was affected by a low feeding level through a prolonged anoestrus period. Also in our study, both, low and medium feeding level showed a non-significant trend to prolonged calving intervals in crossbreds in both reproduction cycles, suggesting anoestrus due to energy deficiency. Zerbini et al. (1996) determined that cows resumed with oestrous when half of their proportional 0.21 BW loss had been regained. Lactation length, although not significantly affected by feeding level, showed similar trends in crossbreds as calving interval, thus explaining part of the differences between feeding levels in that trait.

### III.4.4 Genotype differences in energy requirements

There is a limited possibility to estimate maintenance ME requirements ($\text{ME}_m$) of the cows from the present data. The estimate for the crossbred cows of 0.492 MJ ME/BW$^{0.75}$ (using the \textit{a posteriori} determined higher ME contents of the feedstuffs) was close to the values of 0.543 and 0.540 MJ ME/BW$^{0.75}$ calculated by using MAFF (1987) and AFRC (1993) assumptions, respectively, both after excluding safety margins. The estimate was also within the range of other results reported for crossbred cows comprising values from 0.384 MJ ME/BW$^{0.75}$ for Brahman × Angus cows (Solis et al., 1988) to 0.519 MJ ME/BW$^{0.75}$ for Nelore × Holstein steers (Martin and Garcia, 1995). The $\text{ME}_m$ estimation as performed in our study is valid only under the assumption that $\text{ME}_m$ was fairly constant at the different feeding levels and physiological states. This may not be entirely the case since cows may have adapted their energy metabolism to feeding environment. Thus the levels of intake before first calving could have established their $\text{ME}_m$ requirements after calving when the animals presumably tried to readjust to this situation by altering energy partitioning.

The present data did not allow the determination of $\text{ME}_m$ of Boran cows as these cows showed no response to feeding level. This would indicate that $\text{ME}_m$ was lower than assumed by MAFF (1987) and as found in the crossbreds. However, $\text{ME}_m$ values reported for purebred \textit{Bos indicus} cattle were in a similar range as for crossbreds with 0.481 and
0.447 MJ ME/BW\(^{0.75}\) for Brahman cattle (Ferrell and Jenkins, 1998) and Nelore bulls (Tedeshi et al., 2002), respectively (data transformed under the assumption that NE\(_m\)/k\(_m\) = ME\(_m\) with 0.72 for k\(_m\); AFRC, 1993). The value of 0.375 MJ ME/BW\(^{0.75}\) reported by Ferrell and Jenkins (1998) for Boran is exceptionally low and suggests that this Bos indicus breed may actually have a lower ME\(_m\). Many authors claim differences in ME\(_m\) between Bos indicus and Bos taurus cattle (e.g. Hotovi et al., 1991; Reid et al., 1991). Some references recommend to apply ME\(_m\) assumptions for Bos indicus breeds, including Africander, Barzona, Brahman and Sahiwal, which are proportionately lower by 0.1 than for Bos taurus breeds (Solis et al., 1988; NRC, 2000), others (Ledger, 1977) noted a factor of 0.05. Hotovi et al. (1991) did not detect differences, either in efficiency of utilisation of ME for maintenance (k\(_m\)) or in fasting heat production between Bos indicus \(\times\) Bos taurus (Barzona \(\times\) Hereford) and Bos taurus (Angus \(\times\) Hereford) heifers and steers. However, heritability estimates for fasting heat production, ME\(_m\) and k\(_m\) were high in their study with 0.75, 0.52 and 0.34 indicating that fasting heat production has a strong genetic component. Possible explanations for a lower heat production per unit of BW\(^{0.75}\) of the Boran cows compared to the crossbreds could be (i) a more favourable shift in the ratio of metabolically active to inactive organs occurring with under-nutrition (Burrin et al., 1990) as a result of endocrine changes (Chilliard et al., 1998), and (ii) a lower heat increment from production (body tissue accretion, milk production, work etc.) as can be assumed from results of Reid et al. (1991). Accordingly, Ferrell and Jenkins (1998) found Boran crossbred steers to have lower ME\(_m\) under feed restriction as opposed to supplying a high concentrate diet; however, in the same study the opposite was observed for Brahman crossbreds. The second explanation would actually represent a production-related increase in efficiency rather than genetic differences in ME\(_m\), and may particularly reflect the well known lower heat energy loss for metabolic synthesis of fat instead of protein. Boran had a fat : protein ratio in milk of about 2.0 : 1 compared to 1.7 : 1 in the crossbreds, and the body of the Boran probably also contained more fat relative to protein than that of the crossbreds receiving the same feeding level. The assumption that metabolic energy turnover per unit of metabolic BW might be lower in obese compared to lean animals was ruled out by Ortigues et al. (1993). Results obtained in the same animals, but before first parturition, showed that the energy
intake for zero weight gain did not differ between genotypes, except when heifers were pregnant, i.e. when an additional form of yield contributed to heat production (Jenet et al., 2002). A final, but indirect, explanation for lower energy needs of Boran to be covered for maintenance was their generally higher OM digestibility. Additionally, the crossbreds, but not the Boran, responded with a higher digestibility in a situation of low and medium feed supply compared to the high plane of nutrition. Grimaud et al. (1998) found no significant difference in OM digestibility between Bos indicus and Bos taurus cattle in tropical environments both at supplies according to ME_m and 0.5 ME_m. It seems that restricted feed intake may increase digestibility, mainly by a lower passage rate, whereas drastic under-nutrition to levels far below ME_m may (Grimaud et al., 1998) or may not (Michalet-Doreau and Doreau, 2001) impair digestibility.

### III.5 Conclusions

The present study showed an entirely different pattern of response to different feeding levels of indigenous breeds and crossbreds. This was probably the result of a combination of genetic limitations in milk yield and differences in energy requirements. Accordingly, in the view of tropical smallholders extra feed resources appear to be wasted when supplied to indigenous breeds even though this could improve body condition and, indirectly and mostly unnoticed, longevity of the cows. Sumberg (2002) underlined the need to distinguish between implications for different market-oriented production systems and for remote areas with poor natural resources, where productivity is often not the priority issue. Strategies to cope best with feed fluctuations in small-holder systems therefore have to acknowledge the availability of both feed and water. Opportunities to effectively use these resources are given in peri-urban areas, where an increase in milk production is feasible with improved feeds for crossbred cows. Indigenous breeds, better adapted to low water and feed supply, would be the choice for remote tropical regions with unfavourable conditions where cattle often fulfill various purposes and the body condition of the cows mostly is more important than the level of milk yield.
Acknowledgements
We are grateful to Ato Bekele Andarge, Worku Getachew, Dr. Olga Karenfil, and Alemeshet Zewdie for assistance in handling of the animals and in the laboratory. We also appreciate the assistance of Ato Amare Atale and Ato Aklilu Bogale for managing the data base and data analysis. This study was partially supported by the Swiss Agency for Development and Cooperation (SDC) through the Swiss Centre for International Agriculture (ZIL), ETH Zurich.

References


Evidence for different nutrient partitioning in Boran (Bos indicus) and crossbred cows when re-allocated from low to high or high to low feeding level

based on:
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Animal Science (submitted June 2004)

Abstract
We tested the hypothesis that Bos indicus cows and crossbreds of Bos indicus and Bos taurus respond differently to long-term changes of feeding level in nutrient partitioning to milk and body fat stores. A total of 27 Boran and Boran Holstein Friesian crossbred dairy cows had been subjected either to a low or a high feeding level (equivalent to 1.0 and 1.4 times of maintenance energy requirements) from their first oestrus as heifers until birth of their third calf. Half of the cows of each genotype were fed these levels further on, while the other half was switched to the respective other feeding level for the third reproduction cycle. Besides milk and body weight records, body composition was determined with the deuterium oxide dilution technique and several metabolic variables were analyzed in blood and fat tissue (hump, tailhead). If at all, Boran cows responded to feeding level almost exclusively by corresponding changes in body weight but not in milk yield. Crossbreds kept continuously on the low feeding level had a lower milk yield than those continuously fed the high level, but lost similar amounts of body weight. In crossbreds, changes in feeding level from high to low were accompanied by a mobilization of body reserves, whereas changes from low to high were followed mostly by an increase in milk yield. Certain other genotype differences in metabolic responses were obvious from differences in body composition (fat and ash content) and from the metabolic profile either reflected in blood (particularly insulin-like growth factor I) or in adipose tissue (lipoprotein lipase). Other traits including insulin, thyroid hormones and RNA expression levels of leptin, tumour necrosis factor-alpha and glucose transporter-4 showed less pronounced genotype effects. Generally, the effects of feeding history affected the traits investigated at least as much as the actual feeding level. Reproductive performance was different between genotypes, with shorter lactations associated with earlier occurrences of the first oestrus in the Boran cows. Calving interval and number of services required for successful conception showed weak trends toward reduced values when the high feeding level followed the low level. In
conclusion, the present experiment suggests a clearly different metabolic response pattern of purebred Boran and crossbreds in a way that Boran seem to react to long-term food fluctuations mainly by mobilizing and restoring body fat reserves whereas crossbreds tend to spend most of additional energy for milk production.

**Keywords:** metabolism, adaptation, under-nutrition, food fluctuation, Zebu, tropics

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### IV.1 Introduction

Ruminants in tropical countries are commonly offered seasonally available forages or post-harvest products. Food supply often shows long-term fluctuations depending on the intensity, duration and occurrence of the rainy season. Because food is rarely stored, rural farmers are particularly affected by seasonal food fluctuations and food scarcity caused by extended droughts, whereas smallholders in peri-urban areas often purchase food or industrial by-products. The increasing demand for animal products in less developed countries (Delgado *et al.*, 1999) is, therefore, often covered from peri-urban and urban production systems, i.e. from systems where health care as well as continuous food and water availability are secure and where improved breeds guarantee a high productivity (McLeod *et al.*, 2002), or from international trade. The rural sector, however, lacks adequate strategies and is in danger of being excluded from the dynamic growth. To improve this situation, it is essential to understand the options for and the constraints of tropical livestock systems, especially for the rapidly growing dairy sector in developing countries. There are two major options as far as genotype is concerned, i.e. the use of local and adapted *Bos indicus* breeds or of cattle upgraded by crossbreeding with *Bos taurus*. Genotype differences in the long-term response of these breeds to an unchanged feeding level were described in a previous study (Jenet *et al.*, 2004b), in which cows of the East African Boran breed were found to produce milk almost independently from feeding level, covering a range from deficient to excessive energy supply, whereas crossbreds transformed additional energy into milk and did not increase body fat stores to the same extent as the purebred Boran cows. There was also a complex interaction between lactation length, milk yield, body fat loss and re-storage, as well as reproductive performance throughout the reproduction cycles, which added to the genotype differences in performance and reproduction at a given feeding level. These results suggest that also the response to
long-term changes in feeding level may differ between these two genotypes. Other evidence, mainly derived from shorter-term changes in feeding level or supplementation, supports this assumption. Differences in metabolic utilization and partitioning of energy seem to be small between contrasting *Bos taurus* dairy breeds such as Holstein, Simmental and Jersey (Münger, 1996), but response was found to differ between *Bos taurus* and *taurus × indicus* (Hotovi et al., 1991). More specifically, purebred *Bos indicus* cows, when severely undernourished after calving, were observed to initially replenish body reserves at the cost of lactation (Mukasa-Mugerwa et al., 1997) while the opposite was observed in *Bos taurus* cows (Röhrmoser and Kirchgessner, 1982). Accordingly, *Bos indicus* cows obviously show no or only a very moderate increase in milk yield in response to improved feeding (Osuji et al., 1995), whereas *Bos indicus × Bos taurus* appear to respond more like purebred *Bos taurus*. However, experiments comparing responses of these genotypes in one study (e.g., Kurtu et al. 1999) are scarce so far. This, and the lack of studies on genotype differences in the regulation of nutrient partitioning to cope with nutrient deficiency, illustrate that the ideal genotype in order to optimise animal productivity and health in situations of long-term food scarcity and fluctuation under otherwise constant conditions still has to be identified.

In this study the hypothesis was tested that, due to different breeding goals, local *Bos indicus* and *Bos taurus* as well as their crosses with *Bos indicus* now show a different metabolic response to long-term food fluctuations. A unique experimental population of cows of Boran and crossbred genotype, either undernourished or well-fed for their whole productive life-time, i.e. from the onset of reproductive activity in the heifer stage (Jenet et al., 2004a) to the end of reproduction cycle 2 (Jenet et al., 2004b), was available. In the present study, reproduction cycle 3 of the cows was investigated where half of the cows were allocated to a different feeding level and the others served as controls. This allowed determining carry-over effects of feeding levels unchanged for long periods and the type of response to the new feeding level. Various indicative blood and adipose tissue traits were followed in order to determine whether or not different metabolic pathways were utilized in the two genotypes.
IV.2 Materials and methods

IV.2.1 Experimental design

A total of 16 heifers of Zebu origin (Boran) and 16 crossbreds (Holstein × Boran, F1 and F2) were selected at body weights (BW) of 302 (±35.7) and 297 (±36.3) kg. Despite the clearly different ages of 4.3 (±0.98) and 2.2 (±0.29) years in Boran and crossbreds, respectively, it is likely that, because of the known genotype differences in maturing (Mukasa-Mugerwa, 1989), their physiological age was approximately similar at that time (discussed in more detail in Jenet et al., 2004a). The animals were fed on a high or a low feeding level (H, or H, where H stands for the feeding history and the indices H and L for high and low feeding level) until the end of lactation 2 (for details see Jenet et al., 2004b). These feeding levels were calculated to cover 1.0 and 1.4 × maintenance energy requirements, respectively (ME<sub>m</sub>; Agricultural and Food Research Council (AFRC), 1993). At the beginning of lactation 3, half of the H<sub>H</sub> cows were switched to the low feeding level (F<sub>L</sub>, where F is the actual feeding level in lactation 3), and half of the H<sub>L</sub> cows to the high feeding level (F<sub>H</sub>). The remaining H<sub>H</sub> and H<sub>L</sub> cows continued on their original feeding level. Sub-grouping was done randomly considering a balanced proportion of F<sub>1</sub> and R<sub>1</sub> crossbred cows. This yielded the following four groups for each breed: H<sub>H</sub>F<sub>L</sub>, H<sub>H</sub>F<sub>H</sub>, H<sub>L</sub>F<sub>H</sub>, and H<sub>L</sub>F<sub>L</sub>. Overall, this resulted in a 2 × 2 × 2-factorial arrangement with genotype, feeding history and actual feeding level as factors in lactation 3. Only 27 of the original 32 cows entered lactation 3, as some of the H<sub>L</sub> cows of both breeds failed to conceive. Unfortunately, the Boran H<sub>L</sub>F<sub>H</sub> data set was eventually reduced to one cow. At the start of lactation 3, H<sub>H</sub> Boran and crossbred cows were 8.6 (±0.57) and 7.2 (±0.70) years old, whereas H<sub>L</sub> Boran and crossbred cows had ages of 8.1 (±1.25) and 5.7 (±0.30) years, respectively. The experiment was performed under the guidelines of, and approved by, the Institutional Animal Care and Use Committee (IACUC, ILRI, Nairobi, Kenya). Animals were under permanent observation by an in-house veterinarian.

IV.2.2 Housing, feeding and mating

Cows were housed individually in a well-ventilated barn in tie-stall pens (1.2 × 2.2 m) equipped with concrete troughs (1.1 × 0.9 × 1.0 m) and concrete floor at the Research
Station of the International Livestock Research Institute in Debre Zeit (Ethiopia), located at 1850 m a.s.l. Ambient temperatures varied during days and nights with 12 h averages of 31°C (±3.5) and 15°C (±1.2), respectively. Relative humidity averaged at 40% during 7 months and at 61% during 5 months, reflecting the variation in monthly precipitation of 22 and 128 mm, respectively. Diets always consisted of grass hay, mainly *Cynodon dactylon* (proportionally 0.65 of total dietary dry matter (DM)), and wheat bran (0.35). Wheat bran was provided once per day after milking followed by the hay. The nutrient and mineral contents of the grass hay (903 (±17.5) g DM/kg) and the wheat bran (886 (±14.0) g DM/kg) on average of 24 monthly determinations were (g/kg DM): organic matter (OM), 900 (±13.0) and 955 (±5.3); crude protein, 46 (±5.9) and 161 (±16.9); neutral detergent fibre, 752 (±30.9) and 446 (±48.0); acid detergent fibre, 435 (±28.1) and 115 (±13.8); acid detergent lignin, 68 (±4.7) and 26 (±1.2); ether extract, 6.5 (±1.74) and 27.5 (±9.44); Ca, 3.89 (±0.771) and 2.52 (±1.584); P, 1.99 (±0.461) and 7.57 (±1.590); Mg, 1.70 (±0.407) and 3.19 (±1.348); Na, 0.131 (±0.201) and 0.054 (±0.026); K, 17.2 (±2.77) and 14.3 (±3.38); Zn, 0.024 (±0.0043) and 0.055 (±0.0222); Cu, 0.011 (±0.0027) and 0.010 (±0.0012). Daily energy requirements were calculated according to Ministry of Agriculture Fisheries and Food (MAFF, 1987) assuming metabolizable energy (ME) contents of 7.5 and 10.1 MJ for the grass hay and the wheat bran, respectively. Animals were provided with bucket water ad libitum and with 50 g/day of bone meal (in g/kg: DM, 938; OM, 314; ash, 686; Ca, 0.127 g; P, 0.103 g). Sodium chloride was provided in amounts of 8 and 12 g/day with the low and the high feeding level, respectively.

On the first occurrence of oestrus in lactation 3, Boran cows were naturally inseminated by four station herd Boran bulls, whereas artificial insemination was practiced for crossbred cows with Holstein Friesian semen of five Dutch bulls. For that purpose, oestrus activity was daily monitored by a veterinarian, by barn attendants, and with the help of a teaser bull freely walking in the barn. Crossbreds were inseminated on detected oestrus. Sixty days after insemination, pregnancy diagnosis was conducted. Further details on animals, site and housing are described in Jenet *et al.* (2004b).
IV.2.3 Determination of food intake and performance

Food amounts offered and refused were recorded daily throughout lactation. Samples of food and refusals were analyzed for contents using standard methods (Association of Official Agricultural Chemists, 1990; Van Soest and Robertson, 1985; cf. Jenet et al., 2004b). The P content of food was determined on an autoanalyzer (Sampler CS40/80, Chemlab Instruments, Southfields, Laindon SS15 6TQ Essex, UK) at a wave length of 660 nm, while the other minerals were determined by atomic absorption spectrometry (300 AAS, Perkin Elmer Analyst, Boston, MA, USA). Body weight and body condition scores (BCS) were recorded at the same time by trained staff every second week. The scale used for BCS assessment ranged from extremely meagre (1) to extremely obese (9) as outlined by Nicholson and Butterworth (1986). Milk yield was recorded twice daily by a combination of hand milking and the weigh-suckle-weigh method, beginning 2 to 4 days after parturition in order to ensure that calves completely received colostrums. To initiate milk led-down calves were allowed to suckle followed immediately by hand milking. During milking calves were tied in front of the cow, and were returned to suckle after milking. Calves were allowed to suckle approximately 2 kg milk. Proportionate milk samples from the hand milked part of the yield, were taken every second week from morning and afternoon milk, preserved with potassium dichromate (Laboratory Supplies, Poole, UK) and stored at +4°C until analysis. Calves were weaned on day 60 of lactation. This practice was implemented because Boran cows required the stimulus for milk release. Adopting farmers' practices, cows were milked either until handmilked yield was <0.5 kg/d or the calculated calving date was not longer than 3 month in advance. Contents of milk fat and protein were determined by the Gerber method with BS 696 (British Standard Institution, 1989) and by formaldehyde titration (6.38 N) as suggested by Scott (1986), respectively. Energy-corrected milk (kg/day) was calculated, using the National Research Council (NRC, 2001) formula, as:

\[ \text{Energy-corrected milk (kg/day)} = \frac{0.3887 \times \text{fat (g/100 g)} + 0.2289 \times \text{protein (g/100 g)} + 0.8033}{3.14} \]

IV.2.4 Organisation of an intensive sampling period

A 10-day collection period was conducted between days 60 and 70 after calving. The period consisted of 7 days of water intake measurements in addition to food intake. On
day 8 of the collection period, blood samples were taken, followed on day 9 by subcutaneous adipose tissue sampling. On day 10 deuterium injection and subsequent blood collection was accomplished.

IV.2.5 Blood collection and determinations in blood plasma and serum
Blood samples were collected from the jugular vein at 13.00 h, i.e. 5 to 6 h after the last allocation of food, in sterile (10 ml) and heparinized (20 ml) vacutainer tubes. Blood samples for serum were centrifuged after 1 h of clotting. The heparinized samples were immediately centrifuged (IEC, Needham Heights, MA, USA) at 1500 × g for 20 min, and the plasma was stored at -20°C for later analysis of glucose, triglycerides, protein, urea (Roche, Rotkreuz, Switzerland), non-esterified fatty acids (NEFA; Wako, Neuss, Germany), β-hydroxy-butyrate (BHB), and albumin (Sigma, St. Louis, MO, USA) according to previously described methods, using a Cobas Mira autoanalyser (Roche, Rotkreuz, CH). In addition, serum levels of insulin (CIS Schering, Gif sur Yvette Cedex, France), triiodothyronine (T3) and thyroxine (T4) (DPC, Los Angeles, CA, USA) were analyzed by radioimmuno assays using a γ-counter (Packard Instrument, Meriden, CT, USA). Plasma levels of insulin-like growth factor-1 (IGF-1) were determined by an immuno enzymometric assay, using sample diluents according to the recommendation of the manufacturer (Octeia, IDS, Boldon, UK), and a micro titre plate reader (Dynatech MR 5000, Dynatech Laboratories, Chantilly VA 22021, USA).

IV.2.6 Collection of and determinations in adipose tissue
Adipose tissue samples were taken at 07.00 h before feeding. The skin was shaved in advance, disinfected by iodine soap and ethanol, and a local anaesthetic (7 ml of 2% Lidocain) was applied. A pocket cut incision of 2 to 3 cm in length was made with a scalpel between the tailhead and the tuber ischii (in 8 cm distance from the tailhead), followed by an incision frontal of the hump or, in the crossbreds, the same dorsal location between neck and withers to remove 3 × 50-70 mg subcutaneous adipose tissue from each site with the scalpel. Incisions were sutured with #2 cat gut suture material and treated with a topical antibiotic and an antifungal agent. The samples were stored in 400 µl RNAlater (Ambion, Austin, TX, USA) in cryo tubes and stored at -
70°C until analysis. RNAlater of the embedded adipose tissue biopsies was then replaced by lysis buffer (Qiagen, Hilden, Germany) and homogenized with a Polytron (Kinematica, Littau, Switzerland) operated for 5 min at about 25,000 rpm. Total RNA was extracted using RNeasy Mini spin columns (Qiagen, Hilden, Germany). The cDNA was synthesized with the AMV reverse transcriptase system (Promega, Madison, WI, USA; Invitrogen Life Technologies, Carlsbad, CA, USA) using 30 μg total RNA and following the manufacturer’s recommendations. Real time PCR was employed to quantify the expression of adipose tissue total RNA transcripts of bovine leptin, tumour necrosis factor-α (TNFα), glucose transporter-4 (GLUT-4), lipoprotein lipase (LPL), and GAPDH as a referencing housekeeping gene. Quantitative PCR was performed on an ABI Prism 7700 Sequence Detection System (Applied Biosystems, Foster City, California, USA) using Taq-Man PCR reagents (Applied Biosystems) in a 25-μl reaction mixture (12.5 μl master mix, 2.5 μl reverse and forward primer, 2.5 μl probe (TAMRA-quenchers and FAM-reporters), 2.5 μl DEPC, and 2.5 μl of the 1:5 diluted cDNA synthesis reaction product). Polymerase chain reaction (PCR) was performed over 40 cycles at 95°C for 15 s and 60°C each for 1 min after initial incubations at 50°C for 2 min and 95°C for 10 min. The PCR product specificity and purity was evaluated by generating a dissociation curve following the manufacturer’s (Applied Biosystems) recommendations. The RNA quantification of the target and treatment differences was accomplished by comparing the amount of target with the standard GAPDH gene expression. Relative gene induction values were calculated following the same manufacturer’s recommendations.

IV.2.7 Estimation of body composition

Animals were fasted for 12 h before deuterium oxide application at 07.00 h. Thereafter fasting continued for another 9 h. Before deuterium application, animals were weighed (±1 kg) and a baseline blood sample was collected from the jugular vein in heparinized vacutainers. Then an amount of 99.8 atom % deuterium oxide (ARMAR AG, Döttingen, Switzerland), equivalent to 0.3 g/kg fasted BW was weighed in a syringe by using a micro-balance (±0.01 g). Deuterium was slowly administered directly into the jugular vein and the syringe was rinsed with refilled blood. Blood samples were taken at 5, 7,
and 9 h post infusion, collected in heparinized tubes and centrifuged at 1500 × g. The plasma was stored immediately in cryo vials at -70°C to minimize evaporation. Deuterium concentrations in plasma were analyzed using an isotope mass spectroscopy (Delta-S, Finnigan, Bremen, Germany) after conversion into its elementary form in a chromium reactor at 850°C (H-Device, Finnigan) and comparison with Vienna Standard Mean Ocean Water. Data from samples obtained 5, 7 and 9 h after deuterium administration were combined. Deuterium space (kg) was calculated according to Speakman (1998) as 1000 × [tracer (ml) × concentration] / (D/H₀ % - D/Hᵣ %), where D/H is the deuterium/hydrogen ratio corrected by the, water standard t in the sample mean and to the deuterium concentration before injection. The gastrointestinal tract (GIT) of the cows was assumed to amount to 0.20 of BW, and empty BW was described as BW - GIT (NRC, 2001). The following equations (Brown et al., 1989) for the estimation of body composition were applied:

Empty body water (kg) = 0.4717 × BW (kg) + 0.1536 × D₂O-space (kg) - 25.046;  (1)

Empty body fat (kg) = 0.3790 × BW (kg) - 0.2955 × D₂O-space (kg) - 42.163;  (2)

Empty body protein (kg) = 0.1624 × BW (kg) + 0.0165 × D₂O-space (kg) - 11.488;  (3)

Empty body ash (kg) = 0.0363 × BW (kg) + 0.0231 × D₂O-space (kg) - 5.755.  (4)

Data were either related to empty BW or to total body DM.

IV.2.8 Calculations and statistical analysis

The MEₘ (MJ/day) was calculated according to AFRC (1993) as extensively described in Jenet et al. (2004a and b). The metabolizability of energy (qm) required for this formula was set to 0.612. The energy content of BW change (MEᵣ) was assumed to amount to 19 MJ/kg (AFRC, 1993). Metabolizable energy balance was calculated as ME intake (MEᵢ) - MEᵣ. Total ME allowances for retention comprised ME included in milk and ±MEₑ. For analysis of variance, the general linear model (GLM) and the statement sum of square type 4 (SS₄) of Statistical Analysis Systems Institute (2001) was employed to account for unbalanced treatments based on the following model.

$$ Y_{ijkl} = \mu + \alpha_i + \beta_j + \gamma_k + \alpha\beta_{ij} + \alpha\gamma_{ik} + \beta\gamma_{jk} + \alpha\beta\gamma_{ijk} + e_{ijkl} $$  (5)

where $Y_{ijkl}$ = individual observation, $\mu$ = overall mean, $\alpha_i$ = effect of genotype (G), $\beta_j$ = effect of actual feeding level (F), $\gamma_k$ = effect of previous feeding level (H), and $e_{ijkl}$ =
residual error. In order to conduct multiple comparisons of the means, the PDIF option and the Bonferoni adjustment were used. Treatment means in tables are displayed as least square means. For the determination of the effects on water intake as well as energy balance and retention, regression analysis was employed using GLM procedure, sum of square type 4 and the solution statement. Genotype was set as class variable. Water intake was regressed against milk yield in combination with genotype and their interaction, while energy retention and energy balance were regressed against ME, genotype and their interaction. Non-significant terms were excluded in subsequent analyses. Confidence intervals at zero retention ($X_0$) were calculated by inverse regression (Draper and Smith, 1966).

IV.3 Results

The differences between low and high actual feeding level ($F_L$ and $F_H$) in lactation 3 in amounts of DM offered and consumed were smaller in absolute terms in Boran (1.3 kg) than in crossbred cows (2.7 kg) (genotype (G) x feeding level (F) interaction, $P < 0.05$; Table IV-1).

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Actual feeding level</th>
<th>Boran</th>
<th>Boran x Holstein</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Low</td>
<td>High</td>
<td></td>
</tr>
<tr>
<td>Dry matter intake (kg/day)</td>
<td>Previously low (n = 3)</td>
<td>4.74</td>
<td>6.22†</td>
<td>4.34</td>
</tr>
<tr>
<td></td>
<td>Previously high (n = 4)</td>
<td>5.16</td>
<td>6.37</td>
<td>5.32</td>
</tr>
<tr>
<td>Organic matter intake (kg/d)</td>
<td>Previously low</td>
<td>4.44</td>
<td>5.93</td>
<td>4.54</td>
</tr>
<tr>
<td></td>
<td>Previously high</td>
<td>4.74</td>
<td>5.80</td>
<td>4.87</td>
</tr>
<tr>
<td>Metabolizable energy intake‡</td>
<td>Previously low</td>
<td>47.4</td>
<td>62.2</td>
<td>43.4</td>
</tr>
<tr>
<td></td>
<td>Previously high</td>
<td>51.6</td>
<td>63.7</td>
<td>53.2</td>
</tr>
<tr>
<td></td>
<td>[kJ/kg BW$^{0.75}$]</td>
<td>657</td>
<td>853</td>
<td>574</td>
</tr>
<tr>
<td></td>
<td>Previously low</td>
<td>637</td>
<td>843</td>
<td>642</td>
</tr>
<tr>
<td>Drinking water intake (kg/day)$§$</td>
<td>Previously low</td>
<td>19.5</td>
<td>23.2</td>
<td>28.8</td>
</tr>
<tr>
<td></td>
<td>Previously high</td>
<td>23.4</td>
<td>21.3</td>
<td>34.5</td>
</tr>
</tbody>
</table>

† Values in brackets because of n = 1. ‡ ME calculated according to AFRC (1993).
§ Determined in the 10th week of lactation. || Approaching significance ($P < 0.1$).
This was mostly due to the persistently smaller BW in the Boran cows (Table IV-2), which was the basis for food allocation. Accordingly, there was no significant genotype difference in ME intake per unit of metabolic BW (BW0.75; Table IV-1).

**Table IV-2 Effect of genotype (G), actual feeding level (F) and previous feeding level (H) on body weight, condition and composition (LSmeans)**

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Boran</th>
<th>Boran x Holstein</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Low</td>
<td>High</td>
<td>s.e.</td>
</tr>
<tr>
<td>Body weight (kg)†</td>
<td></td>
<td></td>
<td>8.7</td>
</tr>
<tr>
<td>Previously low</td>
<td>300</td>
<td>[305]</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>351</td>
<td>320</td>
<td>n.s.</td>
</tr>
<tr>
<td>Change (g/day)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Within the first 12 weeks</td>
<td>-486</td>
<td>[300]</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>-482</td>
<td>-325</td>
<td>n.s.</td>
</tr>
<tr>
<td>Within the lactating period</td>
<td>-225</td>
<td>[-31]</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>-318</td>
<td>-134</td>
<td>n.s.</td>
</tr>
<tr>
<td>Body condition score‡</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Points</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>4.06</td>
<td>[4.86]</td>
<td>0.190</td>
</tr>
<tr>
<td>Previously high</td>
<td>4.98</td>
<td>5.16</td>
<td>** n.s.</td>
</tr>
<tr>
<td>Change (points/month)</td>
<td></td>
<td></td>
<td>0.0561</td>
</tr>
<tr>
<td>Previously low</td>
<td>-0.072</td>
<td>[-0.017]</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>-0.163</td>
<td>-0.150</td>
<td>n.s.</td>
</tr>
<tr>
<td>Body composition§</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>per kg body DM</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fat (%)</td>
<td></td>
<td></td>
<td>2.26</td>
</tr>
<tr>
<td>Previously low</td>
<td>14.4</td>
<td>[17.4]</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>25.6</td>
<td>22.6</td>
<td>n.s.</td>
</tr>
<tr>
<td>Protein (%)</td>
<td></td>
<td></td>
<td>1.73</td>
</tr>
<tr>
<td>Previously low</td>
<td>68.1</td>
<td>[65.6]</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>59.0</td>
<td>61.6</td>
<td>n.s.</td>
</tr>
<tr>
<td>Ash (%)</td>
<td></td>
<td></td>
<td>0.54</td>
</tr>
<tr>
<td>Previously low</td>
<td>17.4</td>
<td>[17.1]</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>15.3</td>
<td>15.7</td>
<td>n.s.</td>
</tr>
<tr>
<td>per kg empty BW</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fat (%)</td>
<td></td>
<td></td>
<td>0.718</td>
</tr>
<tr>
<td>Previously low</td>
<td>5.62</td>
<td>[5.38]</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>8.53</td>
<td>7.78</td>
<td>n.s.</td>
</tr>
<tr>
<td>Energy (MJ)</td>
<td></td>
<td></td>
<td>0.284</td>
</tr>
<tr>
<td>Previously high</td>
<td>7.77</td>
<td>7.41</td>
<td>n.s.</td>
</tr>
</tbody>
</table>

† Average of the first 12 weeks of lactation.
‡ Within the lactation period; body condition score ranks animals from extremely meagre (1 point) to extremely obese condition (9 points).
§ Determined in week 10 of lactation using the deuteriated water dilution technique.
|| Approaching significance (P < 0.1).
There was a large genotype difference in water intake, which could not be explained by the variation in feeding level when applying a regression analysis. Instead, the variation was mainly explained by genotype and milk yield ($R^2 = 0.74; P < 0.001$). Proportionally, both genotypes required approximately the same ($P > 0.05$) amount of water to produce one kg of milk (1.2 ($\pm 0.39$) kg/kg). At zero milk yield, however, crossbred cows on average consumed 5.8 ($\pm 1.8$) kg more ($P < 0.001$) water than Boran cows, which required 18.7 ($\pm 1.4$).

At the start of lactation 3, there were certain residual differences in BW from previous feeding levels. Yet, these differences persisted only for about the first 12 weeks of the lactation in the groups $F_L$ and $F_H$ (Table IV-2). $F_H$ cows prevented ($P < 0.1$) a severe BW loss (35% less than $F_L$) during the first 12 weeks of the lactation, a trend which was not significant any more for the rest of the lactation. Nutritional history ($H$) had no significant effect on BW and BW changes. Figure IV-1 shows the evolution of BW depending on actual feeding levels ($F_I$ and $F_H$), separately for the genotypes and the two previous feeding levels ($H_L$ and $H_H$) (Fig. IV-1). $H_IF_H$ cows of both genotypes lost more BW than $H_iF_h$ cows (Fig. 1a and b); however, a change in actual feeding level to high did not cause changes in BW of $H_i$ cows (Fig. IV-1c and d). The actual feeding level ($F$) caused a similar trend in BCS as in BW changes, but the differences for BCS did not reach statistical significance. Boran cows generally had a higher BCS than crossbreds. This is supported by the numerically higher body fat contents of the $H_h$ Boran cows compared to $H_I$ groups, whereas such a relationship was not obvious in the crossbreds. The body of the Boran cows had a lower ($P < 0.1$) ash content than the crossbreds, which was mainly due to the numerically higher fat contents. All other changes in body composition were not systematic with respect to genotype, actual and previous feeding level. The calving interval was slightly ($P < 0.15$) shorter in the $F_h$ cows and did not differ between genotypes (Table IV-3). In contrast, lactation was clearly shorter ($P < 0.01$) in Boran cows compared to crossbred cows, and was not influenced by $H$ and $F$. 
Figure IV-1 Milk production (bold lines) and body weight changes (fine lines) during the first 20 weeks after calving of Boran and crossbred cows either fed previously the high feeding level, with, or without changes (Fig. IV-1a,b; —, always high; --, change from high to low) or when previously exposed to low feeding level (Fig. IV-1c,d; ..., always low; ---, change from low to high).

Boran cows showed their first oestrus on average 89 days after calving, which was approximately half of the time found in the crossbreds. F₁ crossbred cows seemed to have an anoestrus period prolonged by 45 days on average, but this effect was not significant. In both genotypes, H₄F₁ cows showed a trend ($P < 0.1$) to conceive earlier than H₁F₁ cows, as can be seen from the number of services needed for a successful pregnancy.

Crossbred cows were more productive ($P < 0.01$) than purebred Boran cows in terms of yields of milk (uncorrected, energy-corrected), milk fat and milk protein at any feeding level. Yet, their milk had a lower ($P < 0.01$) fat content (Table IV-4).
Table IV-3 Effect of genotype (G), actual feeding level (F) and previous feeding level (H) on reproductive performance

<table>
<thead>
<tr>
<th>Actual feeding level</th>
<th>Genotype</th>
<th>Boran</th>
<th>Boran x Holstein</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calving interval (days)</td>
<td>Previously low</td>
<td>439</td>
<td>528</td>
<td>19.2</td>
</tr>
<tr>
<td></td>
<td>Previously high</td>
<td>423</td>
<td>493</td>
<td>n.s.</td>
</tr>
<tr>
<td>Lactation length (days)</td>
<td>Previously low</td>
<td>151</td>
<td>262</td>
<td>30.1</td>
</tr>
<tr>
<td></td>
<td>Previously high</td>
<td>141</td>
<td>384</td>
<td>**</td>
</tr>
<tr>
<td>First oestrus after calving (days)</td>
<td>Previously low</td>
<td>73</td>
<td>181</td>
<td>14.2</td>
</tr>
<tr>
<td></td>
<td>Previously high</td>
<td>87</td>
<td>195</td>
<td>n.s.</td>
</tr>
<tr>
<td>Services until pregnancy</td>
<td>Previously low</td>
<td>3.0</td>
<td>3.0</td>
<td>0.27</td>
</tr>
<tr>
<td></td>
<td>Previously high</td>
<td>2.3</td>
<td>1.2</td>
<td>n.s.</td>
</tr>
</tbody>
</table>

Genotype differences were particularly pronounced when seen over the whole lactation and reproduction cycle resulting in a four-fold higher milk production of the crossbreds compared to the Boran cows. Relative to the genotype differences, effects of F on milk yield were small. Average milk yield (kg/day) over the complete lactation and reproduction cycle tended to be influenced by H (P < 0.1), but differently in the two genotypes. Whole lactational yield was low (P < 0.1) in H_L compared to H_H cows (0.75 and 0.50 of the yield with H_H in Boran and crossbred cows, respectively). Figure IV-1 shows the evolution of milk yield during the first 20 weeks of lactation 3 in addition to BW changes. Boran cows did not noticeably respond either to H or F, but expressed a drop in yield at weaning around week 10 (Fig IV-1a and c). Also, H_H crossbred cows showed a similar milk yield in lactation 3 when fed the low (F_L) or the high (F_H) level (Fig. IV-1 b), whereas in H_L crossbred cows the milk yield was higher in F_H than in F_L groups (Fig IV-1d). Food and water use efficiency for milk production significantly differed between genotypes (Table IV-4). Crossbred cows required less dietary organic matter (-83% and -162% during the first 12 weeks of lactation and during the complete reproduction cycle) and less water (-34%) than Boran cows per unit of energy-corrected milk (ECM). For the indigenous genotype the food conversion ratio was more
favourable with F₁ than with F₉ due to the lack of a clear response in milk yield to actual food supply.

Table IV-4 Effect of genotype (G), actual feeding level (F) and previous feeding level (H) on milk yield and food and water use efficiency

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Actual feeding level</th>
<th>Boran</th>
<th>Boran x Holstein</th>
<th>s.e.</th>
<th>G</th>
<th>F</th>
<th>H</th>
<th>GxF</th>
<th>FxH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Whole lactation (kg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uncorrected milk</td>
<td>Previously low</td>
<td>311</td>
<td>[334]</td>
<td>191.1</td>
<td>**</td>
<td>n.s.</td>
<td>§</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>341</td>
<td>576</td>
<td>1695</td>
<td>2161</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Energy-corrected milk (ECM)</td>
<td></td>
<td>346</td>
<td>[422]</td>
<td>169.3</td>
<td>**</td>
<td>n.s.</td>
<td>§</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously low</td>
<td>1078</td>
<td>937</td>
<td>910</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously high</td>
<td>403</td>
<td>585</td>
<td>1470</td>
<td>2060</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uncorrected milk (kg/day)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>First 12 weeks</td>
<td>Previously low</td>
<td>2.22</td>
<td>[1.72]</td>
<td>0.509</td>
<td>***</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>2.53</td>
<td>2.59</td>
<td>5.55</td>
<td>5.38</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Whole lactation (kg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>0.81</td>
<td>-</td>
<td>3.09</td>
<td>1.39</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously high</td>
<td>0.98</td>
<td>0.79</td>
<td>3.04</td>
<td>3.20</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Milk fat (whole lactation)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Content (g/kg)</td>
<td>Previously low</td>
<td>52.9</td>
<td>[60.3]</td>
<td>0.22</td>
<td>**</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>52.6</td>
<td>55.0</td>
<td>37.4</td>
<td>41.7</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yield (kg)</td>
<td>Previously low</td>
<td>16.5</td>
<td>[20.1]</td>
<td>8.10</td>
<td>**</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>17.9</td>
<td>13.9</td>
<td>63.1</td>
<td>86.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Milk protein (whole lactation)*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Content (g/kg)</td>
<td>Previously low</td>
<td>25.3</td>
<td>[33.5]</td>
<td>0.18</td>
<td>n.s.</td>
<td>n.s.</td>
<td>§</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>35.0</td>
<td>20.3</td>
<td>19.9</td>
<td>27.3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yield (kg)</td>
<td>Previously low</td>
<td>7.8</td>
<td>[11.2]</td>
<td>5.17</td>
<td>***</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>12.0</td>
<td>5.1</td>
<td>32.7</td>
<td>55.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Food and water use efficiency (kg/kg ECM)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dietary organic matter</td>
<td>First 12 weeks of lactation</td>
<td></td>
<td></td>
<td>0.126</td>
<td>***</td>
<td>**</td>
<td>§</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously low</td>
<td>1.39</td>
<td>[1.87]</td>
<td>0.74</td>
<td>1.52</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously high</td>
<td>1.38</td>
<td>2.38</td>
<td>0.81</td>
<td>1.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Within calving interval†</td>
<td>Previously low</td>
<td>6.06</td>
<td>-</td>
<td>0.510</td>
<td>***</td>
<td>**</td>
<td>n.s.</td>
<td>n.s.</td>
<td>n.s.</td>
</tr>
<tr>
<td>Previously high</td>
<td>5.08</td>
<td>6.75</td>
<td>1.86</td>
<td>2.30</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Water (week to of lactation)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>6.09</td>
<td>7.29</td>
<td>4.77</td>
<td>6.35</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously high</td>
<td>6.97</td>
<td>7.78</td>
<td>5.72</td>
<td>4.64</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

† OM intake of the reproduction cycle per total energy corrected milk production.
‡ G × F × H, P level < 0.1. § Approaching significance (P < 0.1).
Figure IV-2 Estimated energy balance (top) and retention (bottom) of Boran (○) and crossbred (●) cows during the first six weeks of lactation 3 in relation to calculated intake of metabolisable energy.
### Table IV-5 Effect of genotype (G), actual feeding level (F) and previous feeding level (H) on blood plasma metabolites (week 10 of lactation)

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Actual feeding level</th>
<th>Boran</th>
<th>Boran x Holstein</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Low</td>
<td>High</td>
<td>Low</td>
<td>High</td>
</tr>
<tr>
<td>Glucose (mmol/l)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>2.57</td>
<td>[2.76]</td>
<td>3.32</td>
<td>4.64</td>
</tr>
<tr>
<td>Previously high</td>
<td>3.37</td>
<td>3.44</td>
<td>3.16</td>
<td>2.89</td>
</tr>
<tr>
<td>Triglycerides (μmol/l)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>170</td>
<td>[180]</td>
<td>213</td>
<td>270</td>
</tr>
<tr>
<td>Previously high</td>
<td>177</td>
<td>233</td>
<td>147</td>
<td>192</td>
</tr>
<tr>
<td>NEFA (μmol/l)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>131.3</td>
<td>[114.0]</td>
<td>65.7</td>
<td>49.0</td>
</tr>
<tr>
<td>Previously high</td>
<td>248.0</td>
<td>426.3</td>
<td>128.8</td>
<td>307.8</td>
</tr>
<tr>
<td>β-hydroxy butyrate (μmol/l)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>1134</td>
<td>[770]</td>
<td>676</td>
<td>1199</td>
</tr>
<tr>
<td>Previously high</td>
<td>759</td>
<td>563</td>
<td>822</td>
<td>718</td>
</tr>
<tr>
<td>Protein (g/l)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>62.9</td>
<td>[69.7]</td>
<td>69.4</td>
<td>66.5</td>
</tr>
<tr>
<td>Previously high</td>
<td>79.8</td>
<td>75.9</td>
<td>70.7</td>
<td>69.8</td>
</tr>
<tr>
<td>Albumin (g/l)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>31.7</td>
<td>[30.6]</td>
<td>33.1</td>
<td>35.3</td>
</tr>
<tr>
<td>Previously high</td>
<td>35.0</td>
<td>35.0</td>
<td>32.5</td>
<td>33.4</td>
</tr>
<tr>
<td>Urea (mmol/l)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Previously low</td>
<td>4.48</td>
<td>[5.42]</td>
<td>4.55</td>
<td>5.01</td>
</tr>
<tr>
<td>Previously high</td>
<td>3.86</td>
<td>4.48</td>
<td>4.85</td>
<td>4.88</td>
</tr>
</tbody>
</table>

† Approaching significance ($P < 0.1$).

This effect was less pronounced in the crossbreds. Carry-over effects of the previous feeding level were not pronounced. Figure IV-2 relates either the estimated energy balance (non-significant regression) or the energy retention (MJ/day) in milk and body tissue to the calculated ME intake ($ME_i$, MJ/day) for the first six weeks of lactation. The pertinent regression equation calculated over all data was as follows:

\[
\text{Energy retention} = 11.6 (±11.90) + 0.069 (±0.216) \times ME_i - [\text{crossbreds} = 32.5 (±14.45); \text{Boran} = 0] + [\text{crossbreds} = 0.631 (±0.854) \times ME_i; \text{Boran} = 0]; R^2 = 0.44; P < 0.001. \quad (6)
\]

Accordingly, the two genotypes retained energy with different efficiency ($G, P < 0.05; ME_i, P < 0.001; G \times ME_i, P < 0.05$) with $0.70$ and $0.07$ MJ/MJ ME intake in crossbreds and Boran cows, respectively. The calculated $ME_m$ of the crossbreds was $29.8 (±16.91)$ MJ ME/day (confidence interval (at $X_0$) $0.8$ to $58.8$ ME MJ/day).
Table IV-6 Effect of genotype (G), actual feeding level (F) and previous feeding level (H) on hormones in blood and RNA expressions of metabolic traits in adipose tissue (week 10 of lactation)

<table>
<thead>
<tr>
<th>Genotype Actual feeding level</th>
<th>Boran Low</th>
<th>Boran High</th>
<th>Boran x Holstein Low</th>
<th>Boran x Holstein High</th>
<th>s.e.</th>
<th>G</th>
<th>F</th>
<th>H</th>
<th>GxF</th>
<th>FxH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Concentrations in blood Insulin (µg/l)‡</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.0624</td>
<td>n.s.</td>
<td>*</td>
<td>n.s.</td>
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† Tissue level expressions are arbitrary (n x GAPDH) compared to the assumed constant expression of the housekeeping gene glyceraldehyde-3-phosphate dehydrogenase (GAPDH).
‡ Determined in blood serum.
§ Determined in blood plasma.
|| Approaching significance (P < 0.1).
In the Boran cows ME$_m$ could not be determined due to the lack of response in energy retention to additional ME supply.

There were only few significant effects of F and G on plasma metabolite levels (Table IV-5). Plasma triglyceride (TG) concentration was high ($P < 0.1$) in all F$_H$ cows, independent of H, whereas H played a certain role for plasma NEFA levels with high ($P < 0.15$) values being found in H$_H$ cows in contrast to low values in H$_L$ cows. Similarly, plasma protein was elevated ($P < 0.05$) in H$_H$ cows. Blood serum levels of insulin tended ($P < 0.1$) to be high in H$_H$ cows and were higher ($P < 0.05$) in F$_H$ than in F$_L$ cows. This resulted in an F $\times$ H interaction ($P < 0.05$). The plasma level of IGF-1, however, was higher ($P < 0.05$) in Boran cows than in crossbreds. There were no significant effects of either F or G on plasma thyroid hormone levels.

The RNA expression levels of the ob-gene product leptin in adipose tissue of the tailhead showed a trend to be high in H$_L$ cows and to be lower in H$_H$ cows (H, $P < 0.1$). Moreover, leptin levels of H$_L$ cows were elevated in the F$_H$ treatments, when compared to the F$_L$ treatments; by contrast, levels in H$_H$F$_H$ were lower compared to the H$_H$F$_L$ treatments ($F \times H, P < 0.001$). RNA expression levels of the cytokine TNF-$\alpha$ ($G \times F, P < 0.1$) and enzyme lipoprotein lipase (LPL; $G \times F, P < 0.05$) in subcutaneous hump adipocytes were elevated in F$_L$ crossbred cows compared to F$_H$ treatments, conversely to the reaction of the Boran cows, respectively. Generally, LPL expression was higher in Boran cows compared to crossbreds, which was significant in tailhead adipocytes.

The large variation of GLUT-4, a facilitative glucose transporter, was not systematically explained by the factors investigated, although the expression of GLUT-4 in tailhead showed a similar pattern as the BW change over the lactation.

IV.4 Discussion

IV.4.1 Long-term effects of unchanged feeding levels (H$_L$F$_L$ and H$_H$F$_H$)

Half of the cows of the present study were fed at the same low (H$_L$F$_L$) or high (H$_H$F$_H$) feeding level from the beginning of the heifer stage until the end of the third reproduction cycle (see also Jenet et al., 2004a and b). The surprising trends found in
milk yield in lactations 1 and 2 (Jenet et al., 2004b), i.e. the previously observed response of milk yield to feeding level in crossbreds but not in Boran, continued in lactation 3. The level of $\text{ME}_m$ of 0.38 MJ ME/kg BW$^{0.75}$ (31 MJ ME/day) found in the crossbreds was lower than the 0.49 MJ ME/kg BW$^{0.75}$ found in the previous lactations (Jenet et al., 2004b). Due to the lack of response to feeding level we could not estimate $\text{ME}_m$ in the Boran cows. This was also reflected in the efficiency of food utilization for milk production, which was higher in the crossbreds than in Boran, an effect which was also true for water use efficiency. Per capita and per unit of BW, as well as at zero milk production, however, purebred Boran had an advantage in that respect. Boran cows have probably developed efficient coping strategies against water loss under stress (Finch, 1986), an important aspect for the breed choice as scarcity of water is often more serious than that of food. Body fat contents, estimated by deuteriated water labelling technique, mostly showed the expected trends. However, the high variation, probably caused by both individual differences and methodological uncertainties related to the GIT water pool, prevented the expression of statistically significant differences. Cows, particularly those kept under tropical regimes and confronted with food scarcity, are reported to vary their metabolic rate in order to be able to adjust to harsh conditions (Western and Finch, 1986) and to improve efficiency of the utilization of the food offered (Grimaud et al., 1998).

We also measured various blood and adipose tissue traits to better characterize the mechanisms of metabolic adaptation. The effects of G and F on the selected traits remained small, though. Retrospectively, the most likely reason for these small effects was that we collected the samples around the tenth week of lactation, when cows already had passed peak lactation and shifted from the catabolic to the anabolic state. Plasma glucose concentrations in Boran cows reflected differences in nutrient supply, whereas $F_H$ crossbred cows adapted to high milk potential had low plasma glucose concentrations. In $F_H$ cows TG plasma concentrations were 20-30% higher ($P < 0.10$) than in $F_L$ treatments indicating a high de novo synthesis, this parallel to elevated NEFA concentrations. The difference was not related to a different clearance activity of FA and subsequent uptake into adipose tissue by LPL, because LPL activity was not correspondingly expressed between treatments. Plasma NEFA concentrations in $H_1 F_L$
cows were low, in contrast to high levels in $H_n F_n$ cows. Probably lipolysis in $H_n F_n$ cows was limited since these animals lacked adequate body fat reserves, in contrast to $H_n F_n$ cows which still were able to mobilize fatty acids from stored TG for milk production. Low NEFA levels may indicate decreased energy expenditure. Chilliard et al. (2000) reported high plasma NEFA levels in long-term under-nourished ewes which after 3 mo declined close to a level of normal fed groups confirming the ability of ewes to decrease their energy expenditure during long term under-nutrition.

Levels of $T_3$ and $T_4$ were non-significantly lower in crossbred cows compared to Boran and, within Boran, in $F_1$ compared to $F_n$. This can be explained by the cows' different energy status, as an increase in thyroid hormones with positive energy balance stimulates energy turnover (Reist et al., 2002). The observed decrease of plasma insulin in $F_1$ cows is consistent with similar findings in under-nourished cattle (Chilliard et al., 1998). An elevated level of insulin stimulates glycogen formation, increases tissue uptake of glucose, inhibits lipolysis and gluconeogenesis, and stimulates lipid accretion. Our results revealed a genotype difference in plasma IGF-1 (28% lower in crossbreds) but no clear dependence on $F$, which is different from the findings of Jolly et al. (1996). Thus, IGF-1 was low in cows of high milk production. According to Boone et al. (2000) IGF-1, whose concentration reflects the growth hormone, increases adipose conversion of pre-adipose cell lines. Consequently, plasma concentrations of IGF-1 are typically reduced in starvation (McGuire et al. 1998). Consistent with the low plasma IGF-1 in starvation, the re-alimented, i.e. the $H_n F_n$, crossbred cows, in our study had high plasma levels of IGF-1 and partitioned less energy into milk, in contrast to other crossbred groups.

Leptin is considered to signal adiposity and/or to reflect dynamic changes in metabolite flow in and out of the adipocyte. Leptin expression in adipose tissue was weakly correlated to body fat content ($r = 0.28$, $P < 0.1$), but was not influenced by $F$, suggesting that the differences in the homeostatic state of the adipocytes of $F_n$ and $F_1$ cows were not sufficiently large to induce changes in leptin expression. Chilliard et al. (2001) reported that leptin is decreased in under-nutrition. The decrease in leptin in turn increases cortisol, which contributes to metabolic adaptations to under-nutrition (e.g.
enhanced protein mobilization and gluconeogenesis, stimulated feeding, inhibited reproduction). Leptin expression in adipose tissue of the tail head was increased with re-feeding (H1F1 cows) but also under recent food restriction (H1,F1 cows) (F × H, P < 0.01). This puzzling finding may be explained by the body fat reserves in cows of the latter treatment groups. Interestingly, the response of leptin expression to changes in fat deposits was less pronounced in Boran cows than in crossbred cows. It is therefore tempting to speculate that the weak leptin response in Boran cows allows for maximum storage of energy during the vegetative season, hence, providing an evolutionary advantage for survival during harsh conditions.

In addition to leptin, we measured the adipose tissue mRNA expression of TNFα as another regulator of adipocyte size. TNFα is known to increase lipolysis by decreasing fatty acid uptake and inhibiting lipoprotein lipase and lipogenesis (Kern, 1997, Boone et al., 2000). F1 crossbreds tended to have a higher expression of TNFα in adipose tissue from the hump (G × F, P < 0.10; and non-significantly in tail-head) compared to F2 crossbreds, whereas Boran cows showed a partly different reaction, with TNFα expression being higher in F1 compared to F2 cows. TNFα opposes LPL as well as GLUT-4 and induces insulin resistance. On the other hand it stimulates hormone sensitive lipase (Kern, 1997). The high expression of TNFα in under-nourished crossbred cows may reflect an acute inhibition of lipogenesis and stimulation of lipolysis (Kern, 1997), whereas the enhanced TNFα action in F1 Boran cows may have occurred to prevent the accretion of adipose tissue by a reduction of lipogenesis and a stimulation of adipose tissue mobilization even under conditions of high plasma insulin levels (Kern, 1997). Breed differences and effects of F in adipose tissue expressions of the insulin-responsive GLUT-4 were not observed in the present study, and overall GLUT-4 expression was low. Its expression in adipose tissue reflects energy partitioning into adipose tissue and plays a key role in nutrient oxidation rates. GLUT-4 is the main peripheral site of insulin action (Zhao et al., 1996). One explanation for low GLUT-4 expression may be that in mid lactation glucose supply to adipose tissue is limited. Lactation resulted in a 20-60% decrease in GLUT-4 in muscles and this was associated with a 50% decrease in whole body glucose utilization, suggesting that in lactation
glucose disposal in insulin sensitive tissues is limited, possibly at the expense of the mammary gland (Balage et al., 1997). Chronic under-nutrition decreased GLUT-4 contents in muscle cells and was associated with low insulin plasma levels (Agote et al., 2001). In obese Boran, however, the high levels of TNFα may have reduced GLUT-4 expression as well (c.f. Kern, 1997). LPL hydrolyses TG in the capillary endothelium and controls the partitioning of TG between adipose tissue fat stores and muscle. Thus, at maintenance feeding level about 60% of NEFA originate from LPL-mediated TG hydrolysis (Hocquette et al., 1998). The adipose tissue expression of LPL transcripts were more than 2-fold higher in Boran cows than in crossbreds and seemed to be elevated in hump tissue. F₄₅ Boran cows responded with an elevated expression of LPL in hump tissue, in contrast to the generally low LPL expression in crossbred cows. The generally higher LPL expression in Boran cows indicates a higher ability to accumulate energy in adipose tissue in contrast to crossbred cows. In hump tissue LPL followed a similar expression pattern as TNFα, contrary to the commonly inverse relationship between LPL and TNFα in adipose tissue (Kern, 1997). This may be related to the fact that the Zebu hump, though containing large fat deposits, is morphologically a combination of two muscles, the ligamentum nuchae that supports the head and its antagonist rhomboideus cervicis, both probably relics of pre-domestic era (Heath, 1979). The LPL in muscle and adipose tissue are inversely regulated, and the LPL ratio adipose tissue/muscle is high during hyperinsulinemia, favouring the uptake of lipids into adipose tissue (Fielding and Frayn, 1998). The LPL expression is further affected by level of nutrition, insulin and TNFα (Kern, 1997).

IV.4.2 Carry-over effects of previous feeding levels in performance

The changes in the feeding level revealed clear carry-over effects of the previous feeding level (H), which differed between genotypes (Fig. IV-1). Comparing the changes in BW and milk yield over the first 20 weeks of lactation 3, Boran cows seem to respond exclusively by changes in BW (not obvious in Fig. IV-1c, but there the group H₄₁F₄₅ consists of only one animal). Unless even the low feeding level was sufficient to achieve the genetically determined maximum in milk yield, which is unlikely (Jenet et al., 2004b), this has to be a peculiarity of the Boran cows. The response to F was much more
differentiated in the crossbreds and differed between unchanged and changed feeding levels. \(H_1F_1\) crossbreds produced less milk in lactation 3 \((P < 0.1)\) but lost a similar amount of BW compared to \(H_1F_H\). In contrast, \(H_1F_1\) crossbreds lost much BW, but maintained milk production, whereas \(H_1F_H\) crossbreds responded with an increase in milk yield. Overall, the presence of clear carry-over effects, particularly in the crossbreds, underlines the importance of a history of not too poor nutrition for replacement crossbred heifers in areas with fluctuating food supply. Carry-over effects in milk yield and BW changes in relation to under-nutrition and re-alimentation cycles are mostly known from experiments with Bos taurus cattle kept in temperate regions. These cows, which tend to compensate by both reductions in milk yield and mobilization of body reserves when exposed to energy deficiency, show higher milk yields during re-feeding but are not able to recover completely (Röhrmoser and Kirchgessner, 1982; Coulon and Ollier, 1996). Yet, different responses than those to short-term under-nutrition could be expected from whole-lactational periods of under-nutrition followed by re-alimentation. Severe under-nutrition was found to impair milk production in a subsequent lactation, if not lifetime performance (Broster et al., 1989; Bell et al., 2000). Wiktorsson (1979) found a positive linear relationship between milk yield and high feeding level, whereas cows on low feeding level had larger BW losses, but larger gains when re-fed than those previously receiving high feeding levels. Interestingly, this resembles the pattern of responses found in the crossbred cows in the present investigation and differs completely from the response pattern of the Boran cows. Similar findings as for Boran cows, however, were also reported by Kurtu et al. (1999) comparing the effect of supplementary feeding before and after parturition in Bos indicus (Arsi) and crossbreds. The \(H_1F_H\) group of Boran, though under-represented in number, retained more energy for BW than other Boran treatments. Mukasa-Mugerwa et al. (1997) stated similarly that Bos indicus cows first restore body reserves after a period of under-nutrition and therefore do not noticeably respond in milk production. Yet, the lack of a clear production response to increases in the food supply after an extended preceding period of under-nutrition was also noted in several shorter-term studies (Osuji et al., 1995; Kurtu et al. 1999), while few studies with indigenous cows reported a response in yield to supplementary feeding (Wilson et al., 1987; Islam et al., 2000). We also found an interaction related to fertility of F and H in services needed for
conception. Treatments F_L following H_H (both genotypes) and F_H after H_L (crossbreds) improved fertility, presumably because either excessive fat stores or poor body condition were removed.

Ruminants, especially those adapted to tropical environments, must be able to re-adjust metabolism to large changes in nutrient supply. Long-term endocrine or metabolic adaptations in underfed animals result in the orderly mobilization of endogenous body reserves, sparing glucose and amino acids, as well as in a decrease of metabolic rate and energy expenditure. Some obvious, but often not significant, influences from the previous feeding level (e.g. reduced NEFA, protein, albumin and elevated urea in H_L treatments) suggest that low dietary protein tended to increase the recycling activity in cows adapted to low nutrition. The tendency of enhanced accumulation of BHB in the blood of H_L cows reflects an adaptation to energy deficiency and may be due to a limited gluconeogenesis but an enhanced tissue mobilization. Lower BHB plasma levels in crossbred cows pointing towards a higher uptake of BHB by the mammary gland for milk production as noted by Whitaker et al. (1999). The serum thyroid hormone (T3 and T4) and plasma IGF-1 concentrations in H_H Boran cows were high compared to the HF and the crossbred treatment groups, which is indicative for a high oxidative metabolism, cell growth and high heat production. Given the high BCS, BW and estimation for ME_m, these results support our previous conclusion that H_H Boran cows had an elevated heat production. Continuously higher serum insulin levels were found in cows of H_H treatments compared to H_L, though low actual feeding treatments F_L affected the blood level of insulin at the same time (F × H, P < 0.05).

A carry-over effect (F × H; P < 0.01) was also found for leptin in tailhead adipose tissue, indicating that cows adapted to the low feeding level responded with an increase in leptin to re-alimentation. This may be a mechanism to block insulin and cortisol, thus supporting gluconeogenesis (Chilliard et al., 2001) and supports the notion that changes in leptin also reflect changes in the inflow and outflow of energy into adipocytes rather than merely changes in adipocyte size. Other traits such as LPL
activity, which was described to sharply increase in re-alimentation (Bonnet et al., 1998), did not follow this pattern in the present experiment. However, GAPDH may not be the ideal housekeeping gene as a reference in experiments dealing with energetic under-nutrition during lactation (Rhoads et al., 2003).

IV.5 Conclusions
The present data suggest that Boran, selected involuntarily for survival in remote areas with frequent food scarcity and marked food fluctuations, have developed a different metabolic response pattern changes in to food supply than Bos taurus breeds. Boran cows appear to respond to long-term food fluctuations mainly by mobilizing and restoring body fat reserves. This is likely to be associated with a massively prolonged calving interval, thus reducing the necessity to produce milk for the offspring at cost of body reserves. Crossbred cows (here represented with 50 to 75% Bos taurus blood), by contrast, are inclined to spend most of the energy, which is not explicitly needed to maintain BW, for the production of milk for the calf at a level as high as possible. This genotype obviously follows the homeorhetic strategy of predominantly guaranteeing the survival of the calf, by increasing energy expenditure at cost of body condition and body reserves. That might be explained by the fact that extent and duration of under-nutrition was and is typically lower in temperate regions. Although being apparently favourable for the livestock keeper in the short-term by generating more income from milk production even in times of food scarcity, metabolic disorders and diseases under the common poor housing conditions in the tropics may more than de-compensate for this advantage. A number of metabolic responses to long-term food fluctuation were found. Leptin and TNF-α probably were involved as key regulators for energy stores. Insulin, IGF-1, and thyroid hormones signalled carry-over effects from the previous feeding level. Given the limited number of observations in the subgroups available for the present study, the specific metabolic differences of the genotypes in response to long-term food fluctuation have to be confirmed in further studies.
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V General Conclusions

V.1 Differences in energy utilisation in *Bos indicus* and crossbred cows at high or low feeding levels

Results revealed that crossbred heifers and cows respond to higher feed supply under any physiological state. Under practical conditions, however, stress related to diseases and feed fluctuation could be detrimental to poorly fed or undernourished crossbred animals. Nevertheless, these animals performed well under the controlled conditions of the present study. Despite the fact that Boran heifers, with a genetically lower growth potential, also responded well when switched from a low to a medium feeding level, the overall results suggest that extra feed resources are more efficiently utilised by the crossbred cows.

The estimation of energy requirements was lower for crossbreds and conversion efficiency was not statistically different between genotypes. It was hypothesised that Boran cows with high intake were at their genetic limit and not able to efficiently retain energy. By analysing genotypes separately, efficiency of conversion and also energy requirements tended to be far lower in Boran associated with a high variability. These results suggest the need to differentiate between cows adapted to a specific feeding level when analysing energy requirements. Boran cows in this study, although fed at maintenance level, seemed to exhibit far lower levels of intake. The energy intake associated to zero energy retention for Boran that are adapted to low feed intake may be different to that in Boran cattle adapted to high feeding levels. The efficiency of utilisation of ME for growth was similar in both genotypes and during the different stages investigated, although generally speaking found to be unfavourably low, accounting for only 0.26 to 0.31 (ME retained MJ per kg W<sup>0.75</sup>/ME intake MJ per kg W<sup>0.75</sup>), due to the poor quality of the diet. This underlines a high potential for improving metabolic and overall energy utilisation, at least in crossbred heifers, when using diets of higher digestibility. Further, the large differences found in ME requirements between physiological states indicate the need for review of the current energy standards in the tropics.
Differences in energy utilisation were found to be potentially related to the roles of insulin-like growth factor-1 (IGF-1), lipoprotein lipase (LPL) and tumor-necrosis factor-α (TNFα). The higher TNFα expression in previously undernourished crossbreds may indicate an endocrine shift towards higher energy deposition during a subsequent phase of diet level restoration. In contrast, IGF-1 which regulates fat synthesis at the level of cell differentiation, appeared to be naturally by 28% higher in the Bos indicus. Similarly, the higher LPL expressions in Boran cows indicated a higher genetically based ability to accumulate adipose tissue compared to crossbred cows.

There is a need to further investigate the role of hormones and other traits in blood and tissue in relation to energy partitioning between Bos indicus and their crossbreds. In contrast to feeding levels, unpublished results indicate even stronger relationships between fat stores, as well as milk production, and the expression of metabolites and hormones in blood and tissue.

V.2 Carry-over effects of previous feeding levels in Bos indicus and crossbred cows

Boran, selected over centuries for survival in remote areas with frequent feed scarcity and serious feed fluctuations, have developed a different metabolic response pattern to feed fluctuation than Bos taurus breeds. Boran cows responded to long-term feed fluctuations mainly by mobilising and restoring body fat reserves. This seemed to be associated with a prolonged calving interval, thus reducing the necessity to produce milk for the offspring at the expense of body reserves. Other carry-over effects of malnourishment on reproduction were not significant in Boran cows. Crossbred cows are inclined to spend most of the energy that is not explicitly needed to maintain body weight for milk production for the calf at its highest possible level. This genotype seemed to follow a homeorhetic strategy of predominantly guaranteeing the survival of the calf not by increasing or reducing calving interval, but rather by expenditure of body reserves and body condition. This might be explained by the fact that the extent and duration of under-nutrition was and is typically lower/shorter in temperate regions than in tropical ones. Although being apparently favourable for the farmer in the short-
term, by generating more income from milk production even in times of feed scarcity, metabolic disorders and diseases under the common poor housing conditions in the tropics may more than outweigh this advantage of the crossbreds. Thus, previously undernourished crossbred cows did not recover their milk potential when diet levels were restored in a subsequent lactation and, conversely, well-fed crossbred cows did not reduce milk production during periods of under-nutrition proportionally to the amount of feed restriction.

V.3 Implications of the results for feeding strategies

The experiments showed different response patterns to different feeding levels of Boran and crossbreds. This is probably the combined result of genetic limitations in milk yield and differences in energy requirement. Accordingly, in the view of tropical smallholders, extra feed resources appear to be wasted when supplied to indigenous breeds even though this could improve body condition and, indirectly and mostly unnoticed, the longevity of the cows.

There is a need to distinguish between implications for different market-oriented production systems and for remote areas with poor natural resources, where output productivity is often not the priority issue. Opportunities to effectively use these resources are in peri-urban areas, where an increase in milk production is feasible given improved feeds for crossbred cows. By contrast, indigenous breeds, better adapted to low water and feed supply, may be the better option for remote tropical regions with unfavourable conditions. Breed choice and production goals, therefore, need to carefully account for feed fluctuations, in the development of strategies for smallholder livestock farmers.
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