Content

1. Introduction

Metabolic profile test is a tool for the diagnosis of metabolic disorders in dairy cattle before any clinical manifestation of those metabolic disorders can be detected. The first metabolic profile test was done In England in the early seventies. The Compton metabolic profile test was done (Payne and other, 1970) on dairy cattle in farm in England close to the Compton research instituted for agriculture. The goal of the test was to see if by using wide range metabolic analytic tools on blood samples from groups of cows in different stages of lactation, an overall picture of the metabolic condition of the cows can be drawn.

The introduction of intensive dairy farming techniques in and around the world brought to the surface a new type of diseases not common before the metabolic diseases. In the intensive farm a cow may produce 10000 liter in lactation or more. The dairy farm today is much more similar to a factory then to a traditional farm. In the modern production view a raw material is put into the system where it is processed and transformed into a finished product. Metabolic diseases (production diseases) are usually a manifestation of lack or improper raw material input into the system. The luck of raw material in the system cause the system to break down, the cows will start using their own body to compensate the lack of raw material. In the beginning of this process there will be no clear damages this is the sub clinical stage of the metabolic disorders. When clinical signs start to appear, the cattle metabolic disorder is already severe and can't be compensated by the cattle own body. For example a case of hypomagnesmia will usually appear in an acute neurological signs without any warning (Andrews 1992)

In Israel the average lactation milk yield corrected to 300 day in kg is over 11500 litters (Israel dairy board, 2011). Metabolic disorders are one of the most common causes of loss in the dairy practice. The metabolic profile test is a common tool for the diagnosis of subclinical metabolic disorder that is mainly connected to the nutrition of the dairy cows. Israels' cow are less prune to metabolic disorder connected to pasture feeding like hypomagnesmia as they never eat on pasture (Andrews, 1992). The Israeli cow is sensitive to negative energy balance and to ketonemia due to the huge milk yield hypocalcaemia is also common among Israel's cows.

The aims of the metabolic profile test:

Cow level interpretation:

The metabolic profile test can be used by the farmer with the help of the veterinarian to assess the condition of a specific cow in order to see if she is suffering from any metabolic diseases (Duffield 2009).

Herd level interpretation:

The farmer with the help of a veterinarian can use the metabolic profile test to detect any problems in feeding and management of the farm (Duffield 2009).

.

2. Review of literature

2.1. Metabolic profile test

Metabolic profiling is used extensively in the human medicine practice. In the early 60th the first try to establish a reference values for cattle took place. The first to establish a complete metabolic test was Peyne in the Compton agricultural institute in the United Kingdom (Peyne and other, 1970). The original Compton metabolic test checked only blood samples from three groups of cows. The groups consisted of seven cows; each of the groups represented a different stage of lactation. The three groups as follow: dry cows, middle yielding cows, and high yielding cows. The metabolites checked were: glucose, urea, inorganic phosphorus, calcium, magnesium, sodium, potassium, albumin, globulin, hemoglobin, copper.

Between 1999 and 2004 a large metabolic profile survey took place in the UK to try and see if any pattern of metabolic abnormalities can be found in theirs dairy cattle farms. The large scale analysis took blood samples from 35,506 lactating cows, in this survey it was found that at list three quarter of the cows sampled postpartum (8- 10 days after parturition) had at list one parameter in the abnormal range. As well there was no group of cows that showed no abnormalities with the cows in mid lactation had the best result with 57% with a one abnormal parameter (Macrae, 2006). A major finding was that cows in dry period 10 days prior to lactation was just as prune to metabolic abnormalities as cows in lactation. This finding indicates that it is likely that subclinical metabolic diseases are a consequence of nutrition and management of the cows and less dependent on the huge milk yield of the cows (Macrae, 2006). Most metabolic disorders can lead from one to another with a chain like reaction if the primary subclinical disorder is not treated it may lead to others. Preparturent negative energy balance with subclinical ketonemia is believed to be one of the leading factors in the lower DMI in postparturent cows that in turns leads to the manifestation of LDA (Epperson, 2005). Preparturent hypocalcaemia is believed to be the main cause of dystocia in cows this later leads to immune suppression that leads to metritis, mastitis or other infectious diseases (Andrews, 1992). This connection between subclinical metabolic disorders and

clinical disease is most prevalent in the transition cow up to 75% of clinical disease of cows take place in the three weeks prior to calving and three weeks postpartum. This can show the connection between the metabolic load on the cows and the clinical manifestation of diseases (Kevin Lager, 2012).

2.2. Most important metabolic disorders

Production diseases known as well as metabolic diseases are characterized by unbalance in the metabolic preference of dairy cows. Those diseases are a consequence of imbalance between the input and output of the cows. They can be categorized by their clinical pictures and by the main metabolite involved in the disorder.

Milk fewer

Milk fewer is the clinical picture of hypocalcaemia. The homeostasis of calcium in the body is a complicated system involving absorption excretion and ability to store and recruit calcium from the body reserves. The calcium blood levels are regulated by two hormones: calcitonin and parathyroid hormone (PTH). The main calcium stores of the body are the bones (Andrews, 1992). Subclinical hypocalcaemia is a wide spread metabolic disorder present in most dairy herds. The clinical form of the disease milk fewer is most frequent just prior to pregnancy or in the following weeks after pregnancy (Andrews, 1992). The calcium levels required for the milk production during peak lactation is extremely high and even calcium supplementation into the feed postpartum maybe not be sufficient to cover the requirements of milk production (1-1.5 g/l of milk) (Andrews, 1992). For this reason in order to achieve the amounts of calcium required for milk production the cow utilizes the body stores. Hypocalcaemia develops due to the fact that the cow is unable to recruit enough calcium from the bones for maintaining the calcium concentration in the blood. The dropping calcium concentration in the blood leads to different clinical pictures depending on the severity of the hypocalcaemia. The normal blood concentration of calcium is 2.2-2.6 mmol/l (Andrews, 1992). Clinical signs of milk fewer usually appear at values of 0.75-1.5 mmol/l (Andrews, 1992). Subclinical hypocalcaemia is connected to many diseases. Dystocia is one of the most common complications of subclinical hypocalcaemia (book). Many predisposing factors are connected to the development of clinical hypocalcaemia:

1. The age of the cow is a relevant parameter as older cows have a much higher prevalent of clinical hypocalcaemia than cows in first lactation which practically never have the disease. The ability of the cow to recruit calcium from the body stores diminishes with the age (Macare, 2006).

2. Estrogens inhibit the mobilization of calcium stores, this can lead to higher frequency of milk fewer just postpartum and during estrus (Andrews, 1992).

3. During the transition period the food intake is frequently depressed, this leads to a decreased intake of feed and to lower calcium availability for the animal (Andrews, 1992).

4. A common failure of management in the form of calcium overfeeding during the dry period is often leading to a depressed function of the parathyroid gland and causes it to underfunction in the postparturient period. In the past it was a common practice to increase calcium concentration in the feed prior to calving in order to increase the cow`s calcium stores. This led to a high incidence of milk fewer due to depressed function of the parathyroid hormone (Andrews, 1992).

5. Low magnesium levels in the feed can lead to depressed calcium ingestion, due to the fact, that there is a connection between the uptake of calcium and magnesium. Hypomagnesaemia depresses the body's ability to recruit calcium from the bones, as well. Hypomagnesaemia is one of the predisposing conditions of hypocalcaemia (Andrews, 1992).

The incidence of clinical hypocalcaemia has been estimated to affect 6% of cows in the postparturient period. The high incidence of the disease makes it one of the most frequent clinical conditions that affect dairy cattle. Following an episode of milk fewer, the chance of having further complications disorders increases. The most frequent disorders are ketosis and dystocia.

While the incidence of clinical hypocalcaemia is in the reasonable range, the prevalence of subclinical hypocalcaemia is much higher in average can be even as high as 25% to 57% of all cattle in the dairy farm (Epperson, 2005). The effect of subclinical hypocalcaemia is no well-established and few researches have been dealing with the effect that subclinical hypocalcaemia have on the production of dairy cows. One of the conclusions regarding hypocalcaemia points on a connection between preparturient hypocalcaemia and postparturient ketosis.

Milk fewer requires the involvement of veterinary treatment which can increase the economic impact of the clinical disease (Andrews, 1992). Detecting the subclinical hypocalcaemia in an earlier stage can save economical resources from farmers.

Fat cow syndrome

Fat cow syndrome is a collection of metabolic disorders that usually appear in the postparturient stage of lactation. The main reason for the syndrome is the over-conditioning of preparturient cows (Andrews, 1992). In the body condition scoring system the cows should reach a maximum score of 3 to 3.5 during the dry period. If the cow is over-conditioned usually around BCS of 4 it will be more prone to the fatty cow syndrome (Epperson, 2005).

A hypothesis exist that a complex mechanism involving the fat tissue of over-conditioned cows has a negative control on the DMI of cows in the postparturient period. The mechanism is supposed to function in two levels:

1) Larger fat cells in over-conditioned cows are more sensitive to the negative energy balance in the transition period. By that they can release high level of NEFA into the blood circulation. The liver will transform the NEFA into keton bodies due to the high levels of NEFA in the blood and the lack of ability of other tissues to utilize them. The high blood concentration of NEFA and keton bodies in return will lower the appetite of the effected cow (Epperson, 2005).

2) Over-conditioned cows might show insulin-resistance. This fact can predispose the over conditioned cows to a poor response to the increase in energy content of the feed (Andrews, 1992).

Ketosis

Clinical ketosis

Ketosis is the most important metabolic disorder in dairy cattle (Oetzel, 2007) The disease is usually associated with high milk production but can be present also in case of anorexia or starvation. Ketosis is characterized by high concentrations of keton bodies in the blood. Keton bodies include acetone, aceto-acetate beta-hydroxy-butyrate (BHBA) (Andrews, 1992). Ketosis often occurs when for any reason the dry matter intake of the cow decreases. Typically just after calving dairy cattle shows decreased appetite which can result in up to 30% decrease in feed intake. During the lactation period of the cow a stress from any source

can initiate decreased appetite. The most common is incorrect feeding, or spoiled feed, the other one is infectious diseases. The decrease or insufficient feed intake results in recruitment of body stores. The cow will utilize its body stores in order to try and maintain milk production. Energy stores in the form of NEFA will be released from fat tissue and circulated into the liver. In the liver they will be stored in the form of triglycerides and then they will be sent to other body tissue for energy production. Most tissues can utilize fats by incorporating them into the citric acid cycle in the form of acetyl-CoA. Over-utilizing of fat tissue will cause depletion of oxalate from the other body tissues and will prevent proper utilization of acetyl-coA. This will result in recruitment of amino acid and glucose for the synthesis of oxalate. If the body recruits too high amount of NEFA and cannot utilize it due to the mentioned reason the liver will transform the NEFA into keton bodies and mainly into beta-hydroxi-butirates as well as acetone which will give the typical smell to the ketosis effected cow's breath.

Keton bodies are potent sources of metabolic energy for the body. Most body tissues can utilize them as energy source except red blood cells and the CNS tissues. As keton bodies cannot be stored by the body they will be accumulated in all body-fluids. Keton bodies will be secreted from the body into the breath the urine and the milk. Due to this fact the body is basically draining itself of its main energy giving metabolite. High concentration of keton bodies and NEFA in the blood has been shown to decrease appetite in the affected animals even more than what they had been in the first place.

The negative energy balance in the transition period resulting from the insufficient DMI was shown to be the main stress behind clinical ketosis in cows in the transition period. The first signs of negative energy balance can already be detected in one or two weeks prior to calving. Although some efforts to increase concentrated feed intake in this time have been taking place its assumed that the size of the embryo limits the amount of feed a cow can consume in a day. For this reason the rumen is too small during this time of pregnancy and also during the postparturient period. Some efforts to try an increase in the volume of the rumen by increasing the amount of water content in the feed in order to increase the total volume that the cow will consume in a day is a common practice. This method is an effort to try and stretch the rumen and facilitate it for higher feed intake. As well the habituation of the ruminal flora is a critical step in the transition period for the improvement of the postparturient feed intake. These steps have been shown to be able to relieve to a certain extent the negative energy balance and to decrease the incidence of clinical ketosis.

It has been shown that clinical ketosis is a cause and also an outcome of different metabolic and metabolic-related disorders. For example a cow with a retrained placenta has a higher chance of suffering from clinical ketosis. The same cow with clinical ketosis later will be in much higher risk of development of abomasal displacement.

Subclinical ketosis

Subclinical ketosis, it's by far the most prevalent metabolic disease in dairy cattle. In certain surveys the incidence of subclinical ketosis was higher than 50% of sampled animals. Those surveys were conducted on seemingly healthy animals. The definition of subclinical ketosis can be modified by the threshold of the sampling. In most sources the threshold for subclinical ketosis is between 1000-1400 micromole/liter of beta-hydroxy-butyrate. By changing between the two extreme values the effect attributed to subclinical ketosis can be magnified or diminished. Although the amount of cows with blood concentration of 1000 micromole of BHB is much higher in dairy practice, the effects attributed to it are much less severe. When using a threshold of 1400 micromole/l BHB the number of effected cows is much lower but the effect of the subclinical ketosis is much higher. For example a research by (Duffield, 1997) found that the chance of a cow with subclinical ketosis of 1000 micromole/ liter to suffer from a displaced abomasum is higher than the no sick cow, while the chance of a cow with blood concentration of 1400 micromole/l BHB is three times higher to have a clinical ketosis or displaced abomasum. This can show us that by using a different threshold for the definition of subclinical ketosis the effects and prevalence of the condition can be dramatically changed.

Subclinical ketosis usually effect cows in their transition period but in most farms can be detected in cows in any stage of lactation as well in the dry period. The effect of sub clinical ketosis was summarise in a paper by Duffield (2009)

- Subclinical ketosis (BHBA > 1200 – 1400 μmol/L) in early lactation is associated with
  - 3 to 8 times increased risk of LDA (; Geishauser et al, 2000b; LeBlanc et al 2005; Duffield et al, 2009)
  - Decreased probability of pregnancy at first AI (Walsh et al, 2007)
  - Decreased milk production (Duffield et al, 2009)
  - Increased duration and severity of mastitis (Suriyasathaporn, 2000)

The wide effect of the disease on the cows and the very high prevalence of it in dairy cattle are putting it in a good position to be the most economically significant sub clinical or clinical disease in the dairy practice (Duffield et al, 2009). It was calculated that each cow with subclinical ketosis will cost the farmer up to 78 us dollars. With the high prevalence of the disease and the fact that as a sub clinical disease it is unnoticed and cannot be treated without a herd level metabolic profile testing (Duffield et al, 2009).

Fatty liver syndrome

Fatty liver syndrome is the clinical manifestation of high concentration of NEFA in the blood (Andrews, 1992). It is common in over conditioned cows during the transition period. The negative energy balance around parturition leads to recruitment of energy sources of the cow's body mainly fat storage. In the case of over conditioned cows the high level of fat tissue mobilization can exceed the ability of the cow's body to utilize the free fatty acids. The free fatty acids accumulate in the liver as triglyceride droplets, this leads to liver damage that can irreversibly damage the normal function of the liver (Andrews, 1992). Decreased albumin levels and increased AST in the blood is good indicator for this disease (Andrews, 1992). As well the ability of the GI tract to absorb food is diminished. The liver damage as well may cause decreased ability of the cow to detoxify the content of the blood. The fatty liver syndrome increases dramatically the incidence of peri- and post parturient diseases like retained placenta, milk fewer, mastitis and ketosis (Andrews, 1992).

Displaced abomasum

Due to depressed eating and high keton and NEFA in the blood anorexia develops in the postparturient cow (Andrews, 1992). The decreased amount of food intake leads to ruminal hypomotility that in turn leads to gas production in the abomasum. The liquid and gas content in the abomasum prevents the peristaltic motion from removing the gas from the abomasum (Andrews, 1992). The inflating of the abomasum causes it to dislocate usually to the left side of the abdominal cavity. This usually subacute or chronic disease can cause significant economic losses due to depressed feed intake and milk production (Andrews, 1992). Usually

only surgical intervention can solve the displacement. Cows with subclinical ketosis in the transition period and high NEFA concentration were shown to be much more predisposed to the disease (Epperssom).

Ruminal acidosis

Acute ruminal acidosis

A complication of feeding to much easily fermentation concentrates in the feed (Burmin 1990). The production of volatile fatty acids lowers the PH of the rumen hindering proper function of microbes and protozoa in the rumen. If the ruminal acidosis last for days it can hurt. Parakeratosis will develop in the rumens wall leading to increased permeability of metabolites and toxins as well bacteria can cross from the rumen into the blood circulation. Travelling in the portal circulation the bacteria will be filtered by the liver which may lead to further complication liver abscessation (Burmin 1990).

Hypomagnesaemia

Hypomagnesaemia known as well grass grass-tetany or grass staggers is a common clinical disease of magnesium deficient cows. The disease is common in dairy cattle which are on pasture. It can occur in dairy cattle in all ages but it is most common during peak lactation (Andrews, 1992). The combination of three factors causes the disease to develop into an acute form. Magnesium cannot be stored in the body tissues and is only regulated in the uptake and excretion of access magnesium. Many types of grasses have low magnesium content. If the cattle is kept only on pasture and no magnesium supplementation takes place, there is a limited possibility for the cow to forage sufficient levels of magnesium. Milk contains low amount of magnesium 0.12g/L (Andrews, 1992). But the amount of magnesium required for the milk production during peak lactation can exceeds the intake from pasture. Subclinical and clinical hypomagnesaemia is frequently leads to development of hypocalcaemia in the transition period of dairy cows. The clinical picture of hypomagnesaemia is nervousness decreased milk production and in the severe cases rigid paralysis in sternal or lateral recumbence (Andrews, 1992).

2.3. Minerals and metabolites

Every metabolic profile test is composed of a selected group of metabolites and minerals. The laboratory check for each parameter in the test is an additional cost to the performer. For this reason most metabolic profiles are focused on the most important metabolites and minerals. The criteria the metabolites and mineral is decided by a set of parameter.

1. The possibility to detect and measure them in an accurate way (Andrews, 1992)

2. The connection between the parameter and clinical disease (lager, 2012)

3. Easy and cost effective way to collect transport and analyses in the lab (Andrews, 1992)

Using this parameter we can collect a selected parameter for the best metabolic profile test.

Energy related metabolites are a group of metabolites that can indicate how balanced is the energy status of the cow (Macrae, 2006). This group includes different metabolites and usually is they are coupled with the body condition score of the cow. Non esterified fatty acids (NEFA) keton bodies (beta-hydroxy-butyrate, aceto-acetate and acetone) and glucose are to most common energy related metabolites checked in the metabolic profile test. Each of them gives a small part of the overall energy balance picture.

Energy related metabolites

Glucose

Glucose is the main metabolic fuel of the organism. The most efficient and common way to get energy is by complete oxidation of glucose. Ruminant don't absorb large amounts of glucose foe the GI tract (Lager 2012). The most common way for ruminants is to synthesis glucose in the liver by means of gluconeogenesis. Volatile fatty acids the main product of the microbial fermentation in the rumen is absorbed in to the blood stream by the rumen wall. From all the VFA absorbed the most important for the gluconeogenesis in propionic acid. When reaching the liver the propionic acids will be coupled and synthesized in to glucose by means of the gluconeogenesis (Lager 2012). Then the glucose will be stored in the liver as glucogen or secreted in to the blood for the use of other tissues the rumen.

Glucose levels are strictly regulated by homeostatic mechanisms. This fact shows us that great variations in glucose levels are rare. On the other hand glucose level tends to change relative to the nutrition of the cow and to the time of sampling (Macrae, 2006).

In a large scale metabolic profile survey done in England in 1999 to 2005 it was found that the most common metabolic disorder is too low glucose blood levels. It was found that: "The most common abnormality was low plasma glucose, which was below the optimum range in 49.7 per cent of cows in early lactation, 22.6 per cent of cows in mid-lactation and 27.2 per cent of dry cows within 10 days of their predicted calving date." (Macrae, 2006)

NEFA

NEFA is one of the best anilities for the diagnosis of NEB. High levels of NEFA can come only for large scale mobilization of fat tissues. The fat cell secrete big amount of energy stored in them in the form of triglyceride (lager 2012). The triglyceride will break into NEFA and will be transported by blood to the liver where they will be built into VLDL and secreted to the blood again. The cattle liver has a limited capacity to produce VLDL (Andrews 1992). In the case of mobilizing a large scale of fat there will be limited amount of VLDL and the NEFA will be stored in the liver as triglycerides (Andrews 1992). the liver will use NEFA as an energy source by oxidation in the citric acid cycle. If there is a deficiency in glucose and oxalate the liver will produce keton bodies from the NEFA. Although in the short run keton bodies can serve as energy source for muscle, This will lead in the long run to first of all to sub clinical ketosis and later to ketosis.

Fat cow syndrome and fatty liver syndrome are both the outcome of this metabolic load on the liver. Ketosis is a later complication of this metabolic pathway.

NEFA has been associated as a risk factor for many diseases. In a study by Duffield (Duffield, 2009) it was summarized that level of NEFA exciding a level of 0.5mmol/L will increase the rate of different disorders:

1. The risk of LDA is 2 to 4 times higher (LeBlanc 2010)
2. Retained placenta is 1.8 times more common (LeBlanc 2010)
3. A cow has twice the chance to be culled in the first 60 days of milking (LeBlanc 2010)
4. Milk loss of 1.2kg/day in the first 120 days of milking (LeBlanc 2010)

NEFA levels should be monitored prior to calving as it was found by many researchers that NEB starts to have its effect on the cow already before calving. In a survey in England it was found that 10% of cows had high level of NEFA more than 10 day before calving and 13% in the 10 day before calving. Cows that start to show signs of NEB before calving are much more prune to clinical disease latter in lactation.

Table taken from: Duffield (2009)

Interpretation of Serum Metabolic Parameters around the Transition Period

Table 1. Cow Health and Production Parameters associated with Herd Level Precalving NEFA status Low Risk Herd Proportion of Precalving High NEFA (≥ 0.5 mmol/L) cows < 25% N=26		High Risk Herd Proportion of Precalving High NEFA (≥ 0.5 mmol/L) cows ≥ 25% N=29
Displaced Abomasum Incidence (DA)	3.6%	4.7%
Proportion of Herds with DA incidence > 3.3%	38%	65%
Milk Production for 1st 120 days (kg/d)	39.5	39.3
Proportion of Herds with milk production > 39.6	58%	27%
1st DHI test Protein	3.07	2.97
Proportion with 1st Test Milk Protein > 3.00	60%	37.5%

Beta-hydroxybutarete (BHB)

Beta-hydroxybutarete (BHB) is the most commonly used keton body with its highest concentration in blood test. The main reason of using it is due to its longer half-life that allows to analyses it later in a lab. BHB can be found in blood in the highest concentration from all keton bodies including acetone and aceto-acetate. Ketone bodies are the product of the incomplete oxidation (Stephen LeBlanc, 2010). The appearance of ketone bodies in the blood is the signal for ketosis the border between clinical and sub clinical ketosis is usually higher or lower than 1400 micromole per liter blood. As well clinical ketosis must show

abnormal changes in vital signs on the cow. BHB level can increase in many cases not only in the postparturient period. Many diseases can cause ketosis the decrease in feed intake due to fewer or impaired GI function can induce a secondary ketosis.

Detecting ketone bodies in field condition usually take place with urine stick. This common practice does not rely on BHB but on acetone and aceto-acetate. For this reason it's less indicative for the presence of ketosis. BHB is found in urine and milk in limited levels and for this reasons it is recommended to use blood chemistry for the measuring of BHB in the blood. If BHB level are higher than 1400 BHB micromole/L the incidence of some diseases is much higher. These cows have a likelihood of three times more to develop displaced abomasum. In higher concentrations of 2000 BHB micromole/L the cow is already likely to suffer from decreased milk production (Oetzel. 2007). Cows with clinical ketosis usually have much higher BHB blood levels. Clinical signs start when the concentrate reaches 3000 BHB micromole/L or more. Some cows even have such high level of BHB in their blood without any clinical manifestation (Oetzel. 2007)

Detection of keton bodies in the field is usually done with the help of urine or milk tests. Different commercial kits and tools exist for the diagnosis of ketosis. Those different tool show different effectively.

Table 2. Use of Cowside Ketone Tests in Screening Programs for Identifying Subclinical Ketosis (Duufield. 2009)

Test	20% Prevalence			40% Prevalence			60% Prevalence		
	PV +ve	PV -ve	Apparent Prevalence	PV +ve	PV -ve	Apparent Prevalence	PV +ve	PV -ve	Apparent Prevalence
Keto-Test® using 100 µmol/L	62%	93%	23%	81%	83%	35%	91%	68%	48%
Ketocheck™ (milk)	90%	86%	8%	96%	70%	16%	98%	51%	23%
Urine Acetest Tablet	38%	100%	53%	62%	100%	65%	78%	100%	76%
Urine Ketostix	83%	94%	19%	93%	87%	34%	97%	74%	48%

PV +ve: Predictive Value of a positive test result. PV –ve: Predictive Value of a negative test result.

Protein related metabolites

Nutrition wise the second most important parameter to energy is protein metabolism. Two standard metabolites are used in the protein level analysis. One is urea the other is albumin. This two have a very different life spend and metabolic pathways and give indication for different disorders.

Two type of protein is digested by cattle. The ruminal undegradable protein (RUP) and the ruminal degradable protein (RDP) can be differentiated. RDP is broken in the rumen by proteolytic bacteria. The product of this brake down is amino acids and ammonia. Those will later be used by another type of bacteria to build up their own protein. This generated bacterial protein will be digested later in the abomasum. PRUP is a type of protein the dose not denaturant in the rumen environment and for this reason can't be attacked by the ruminal flora. These proteins will be broken down in the abomasum (Blowey, book).

Urea

Urea is a nitrogen based molecule it is a very good indicator for the short term protein metabolism of cattle. Urea is conjugated in the liver from ammonia. There two main metabolic pathways for the production of ammonia. Rumen microbes produce ammonia in the metabolic brake down of RDP. Some of this ammonia is taken up by the ruminal wall and transferred to the liver by the blood. The second pathway is by deamination of amino acids in the liver.

Urea is the best metabolite for the estimation of the protein status in the rumen and in the food ration as its levels are directly connected to the ability of microbes to brake protein. The standard value for urea concentration in the blood is 1.7 mmol/L (Macrae. 2006) . there can be some reasons why the urea concentration in the blood is to low or high.

Low urea levels are usually a outcome of low intake of protein. This can be for two reasons. The feed ration of the cattle is too low in protein so microbes in the rumen are starving. The second reason is that the cow is not eating the entire feed and is missing the required feed intake for are metabolic demand.

High levels of urea are typical to a deficiency in fermentable metabolisable energy (FME). Or to an overload of protein or other nitrogenous feeds. This disorder is common in cattle herds which are feed with too much protein rich feed or ammonia supplementation was done in an unprofessional fashion (Andrews1992). RDP feed to the cow is being broken by microbes but

the energy demanding step of protein build up by microbes cannot take place as there is a energy shortage in the rumen.

In a metabolic profile survey in England It was found that most cows with protein associated metabolic disorders were in the transition period. 20% of cows in the 10 days of estimated calving time had lower urea levels then the standard of 1.7 mmol/L. while 21.9% of all cows had urea level exuding 3.3 mmol/L which indicate ammonia overload of the rumen (Macare, 2006).

Albumin

Albumin is a blood protein its synthesis take place in the liver. Albumin levels are a good indictor metabolite for the long term protein supplies. Liver disease cause albumin level drop due to decreased ability of the liver to produce it. Normal blood levels of albumin are higher than 30g/L.

Albumin exists in a state of equilibrium with protein and amino acid balance. Low level of albumin can indicate suboptimal protein feeding. Decreased albumin levels have been reported to be a common finding in case of liver disease, kidney disease, inflammatory conditions, and malnutrition. Serum albumin levels have been shown to decrease as the severity of fatty liver increases (Lager. 2012)

Minerals

Calcium

Calcium is an important mineral its functions are related to many metabolic processes in the body. For dairy cattle calcium play a major rule in the production of milk. The main problems attributed for calcium deprivation is low blood level of calcium ions in the blood. This can result in clinical disease milk fewer. Other disease connected to low calcium levels are dystocia and retained placenta (Mulligan. 2006)

Calcium is stored mainly in the bones. It is the most abundant mineral in the cow's body. The main reason for low calcium levels in the blood as less to do with the total amount of calcium in the body but more to do with the ability to recruit stores when needed and to increase absorption from feed to its highest efficiency. Skeletal problems are very rare in modern dairy cattle practice (Suttle 2010)

In order to prevent hypocalcaemia it is suggested to build a counter hypocalcaemia strategies. The first part is to increase control of BCS during the pregnancy. It is important to keep a low calcium diet before parturition. And to supplement magnesium in the postparturient period this step was found to increase the absorption of calcium from the GI track.

Magnesium

Hypomagnesmia is the main disorder of this mineral, magnesium can't be stored in to body and its concentration is regulated by absorption and mainly by secretion in the urine. In the case of magnesium deficiency magnesium levels in the urine are practically zero. If an over load of magnesium rich feed is digested by the cow the magnesium concentration in the urine can become very high (Suttle 2010).

Clinical signs can appear relatively quickly due to the lack of magnesium stores in the body. Clinical hypomagnesaemia can develop rapidly and will result in nervous system signs. Because most cows encountering this metabolic disorder are kept on pasture the disease was given the name "grass tetany". Some types of grass are poor in magnesium which predisposes to the development of the disease. Subclinical hypomagnesemia may cause decreased milk yield as well lower milk fat was reported as a possible outcome. Magnesium supplementation showed to increase fat content in milk in hypomagnesemic cows (Suttle 2010).

A correlation was found between high levels of magnesium in the feed of cattle and decreased magnesium absorption (Suttle 2010). In order to prevent hypomagnesaemia the best and simplest method is to increase magnesium intake with the feed. This can be done by direct adding of magnesium to the feed or by fertilizing the fields with a magnesium containing fertilizer (Suttle 2010).

Phosphorus

Phosphorus is the second most abundant mineral in the animal body. 80 percent of the phosphorus can be found in the bones the other 20% can be found in all body tissues. It is a major part in all the metabolic activities in the body. It's one of the building blocks of DNA and plays an important role in the metabolism of energy in the body in the form of ATP. Disorders related to phosphorus deficiency are usually connected to the type of keeping of the herd and to the quality of the postures on which it is kept. The first time phosphorus deficiencies was reported was a century ago in south Africa in a region with very low phosphorus content in the ground.

Phosphorus deficiency can lead to many different clinical and subclinical disorders. Loss of appetite is a common manifestation of deficiency. In dairy cows one of the most important out comes of a deficiency is decreased milk yield in the time of pick lactation this can reach even to 33% loss of the estimated milk yield (Suttle 2010,)

Sodium

Sodium is the major kation in the blood of all animals. It is responsible for maintaining the osmotic pressure in the intravascular space. From history we can learn that supplying livestock with sufficient salts in their feed is a way to insure good husbandry. The most common salt deprivation symptom is salt hunger seen by the urge of the animal to try and eat abnormal objects like: wood floors and walls.

If a dairy cow suffers from sodium deficiency it may show mild signs till it reaches a braking point and a drop in production and even death might happen. Sodium is a cardinal part of the acid base balance and any deficiency in it can lead to acid base balance abnormalities (Suttle 2010).

Potassium

The third most common mineral in the animal body potassium is the most abundant in the intracellular space. Potassium plays a key role in the electric nature of the ion balance between the intracellular and the extracellular spaces. The most common outcome of potassium deprivation is loss of appetite (Suttle 2010).

In case of deficiency in potassium normal blood level can remain stable but the intracellular potassium levels drops. This make the diagnosis of hypokalemia more complicated than other minerals (Suttle 2010). In lactating cow the daily milk yield is diminished. Abnormal fair and skin signs may develop. The most severe symptom is anorexia usually it develops four week after the deprivation of potassium started. Normal potassium levels play a major role in keeping normal acid base balance in the body. If potassium levels in the blood drop the acid base balance will shift to be more acidic. Acidosis will cause general sickness and loss of production (Suttle 2010).

Optimal potassium levels give cattle a better chance of combating heat stress. During heat stress respiratory alkalosis develops and can be countered with potassium-carbonate but not with sodium-carbonate (Suttle 2010).

2.4. Subjective methods

Body condition scouring

Body condition scoring (BCS) is a subjective method for the assessment of the amount of body reserves the cow have in form of body fat and muscle (W. Kellogg). By looking on the animal a professional trained in assessing the BCS of the cow will give a score of 1 to 5. The main point of focus is the rump region and the hip bones. A score of 1 indicate extremely thin cow while 5 indicate an abuse cow. The ideal BCS is 3 to 3.5 this showed that the cow is in good condition and have a little of extra reserves of energy (W. Kellogg).

Picture from: Chapter 12: Body Condition Scores Michel A. Wattiaux, The Babcock Institute

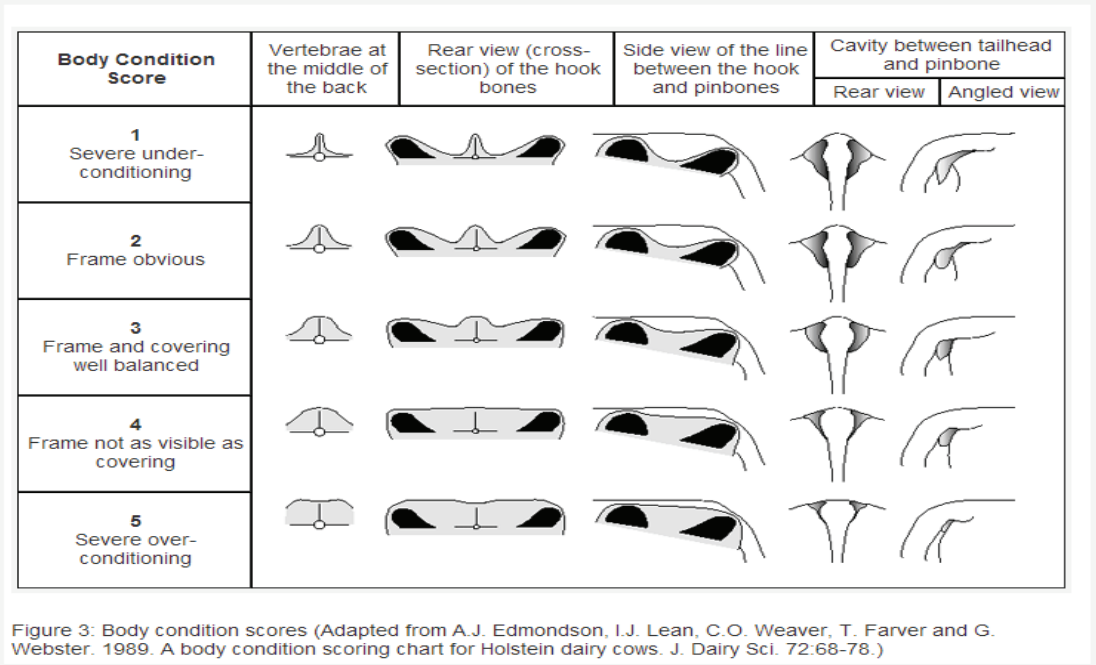


Figure 3: Body condition scores (Adapted from A.J. Edmondson, I.J. Lean, C.O. Weaver, T. Farver and G. Webster. 1989. A body condition scoring chart for Holstein dairy cows. J. Dairy Sci. 72:68-78.)

In different stages in the cows life BCS take place. Usually those times are:

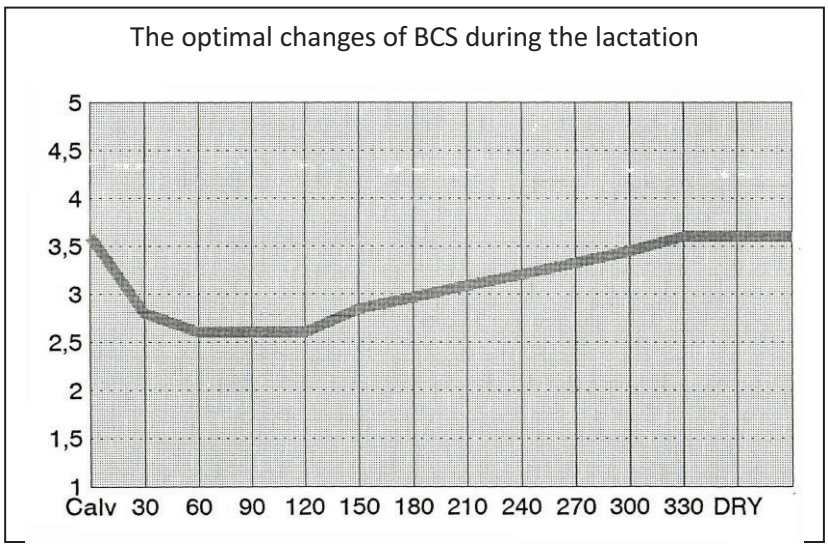
1. Drying off
2. Calving
3. First service

Each one of those stages of the cow's life cycle has its own optimal BCS.

Recommended body condition scores at various stages of lactation are:

Table 3: Michel A. Wattiaux, Chapter 12: Body Condition Scores, The Babcock Institute

Calving	3.0 to 3.5
Breeding	2.5
Late lactation	3.0 to 3.5
Dry period	3.0 to 3.5



Using the BCS give the management of the farm a tool to assess the loss and gain of body weight. This tool is used to optimize the fat reserves of the cows and to provide a way to insure that the cow will not get to its lactation period with too make body fat or too low body fat. Cow which is to thin or too fat is known to suffer for severe metabolic disorders or for low milk production due to low energy reserves (W. Kellogg).

2.5. Abbreviation keys

AST = Aspartate Amino Transferase

ATP = Adenosine Tri Phosphate

BCS = Body Condition Scoring

BHB = Beta-Hydroxybutarete

DMI = Dry Mater Intake

DNA = Deoxyribo Nucleic Acid

FME = Fermentable Metabolic Energy

GI = Gastro Intestinal

LDA = Left Displaced Abomasum

NEB = Negative Energy Balance

NEFA = Non Esterified Fatty Acid

PTH = Para Thyroid Hormone

UK = United Kingdem

3. Own examination

My metabolic profile testing was done on a dairy farm on the Western border of Hungary close to the Austrian border. The samples were collected by students of the veterinary faculty on their farm animal ten week's practical during their 11 semester with help from the staff of the department of animal hygiene and the farm vet and workers. The sampling was on the June 26th 2012.

3.1. Aims and goals

The aim of the study was to detect the presence and prevalence of subclinical metabolic disorder in healthy cattle. By using metabolic profile test created by the department of animal hygiene to fit the needs of Hungary I tried to create a picture of all metabolic errors in the herd. The metabolic profile test was design to detect the metabolic errors by the milking status of the cows. This design gave the farm a good view of the nutritional and management conditions of the farm divided by status of lactation. This gave the farm management a tool to analyze and fix specific errors before they show as clinical disease.

3.2. Placement of examination

The farm is placed in west Hungary close to Austria. The farm has approximately 650 head of cows and the same amount of replacement heifers. The milk yield per lactation corrected to 300 days is about 9500 Kg. the high yielding cows usually closer to their calving are milked three times a day. The cows with the lower milk production are milked only twice a day.

The cows are kept in loose keeping system. The high yielding cows are kept in sheds with cubicles. There are approximately one and a half cubicles for every cow in the shed. The low yield cows the dry cows and the sick cows are kept on deep straw. The cows are groups according to their milk yield and time from calving. Groups 1 and 2 are the freshly calved cows. Groups 3 and 4 are the high yielding cows usually from day 30 to day 120 day after calving. The other groups are the low milk producing and the dry cows. The cows will go from group to the next group by order till they will reach the dry cows group six weeks before calving. Just before calving they will move to the Calvary where they will stay until two days after a calving.

The heifers are kept till the age of one week in the Calvary in separated boxes. Then they are moved to outside hutches till the age of two month. Only the heifers will be moved to be

housed in groups in sheds. When positively diagnosed as pregnant they will move to the pregnant heifers group.

3.3. Design of the examination

The examination was designed to include the all herd. Six groups where formed. Each group will represent a different stage in the life of the dairy cattle. For each group five cows were separated by the farm administrator. The groups are presented in table 3.

Stage in life	Short description of the group status
1. close up	Cows in the last weeks before their expected calving date
2. fresh cows	Cows who just calved and are still kept in the maternity shed
3. early lactation primiparous	Cows that calved in the last few weeks and it is their first lactation
4. early lactation multiparous	Cows that calved in the last few weeks and this is not their first lactation
5. pick lactation primiparous	Cows in their highest milk yield period in their first lactation
6. pick lactation multiparous	Cows in their highest milk yield period which it's not their first lactation

Samples were collected from the cows after the morning milking and before the first feeding of the day. Only clinically healthy cows were sampled. Blood and urine samples were collected from each cow. The blood samples were taken from the milk vein with a needle and a glass tube with a rubber cork. The urine samples were taken from the bladder with the help of a clean metal catheter and collected into a glass tube with a rubber cork.

Three samples were taken from each cow two blood samples and a urine sample. One blood tube contained EDTA and the other one contained heparin. The samples were stored in a insulted container with cooling bottles. Each sample was given a number from 1 to 30. Each cow code was registered next to the sample number into an information sheet. Later the

production and nutritional information was added to the information sheet. The BCS was assessed by the veterinarian of the farm.

The following table summarizes the metabolites and mineral analyzed from the blood and urine samples.

Table. 4 metabolites and minerals analysed from samples

blood samples 1	**blood samples 2**	**urine samples**
Hemoglobin (Hb)	Carotene (Vit. A, precursor)	pH
Glucose	Calcium (Ca)	Net Acid Base Excretion (NABE)
Beta-Hydroxy-Butyrate (BHB)	Inorganic Phosphorus (Ing. P)	Urea
	Magnesium (Mg)	Calcium (Ca)
Free Fatty Acids (FFA)	Cupper (Cu)	Phosphorus (P)
Aspartate Aminotransferase (AST)	Zinc (Zn)	Magnesium (Mg)
	Glutathione Peroxidase (GSH-Px)	Sodium (Na)
		Potassium (K)

All blood and urine samples were analyzed in the laboratory at the Department of Animal Hygiene, Herd-Health and Veterinary Ethology, Szent István University, Budapest.

All the parameters were measured by Biosystems A25 Chemistry Analyzer Diamond Diagnostics – USA, 333 Fiske Street Holliston, MA 01746

4. Results

4.1. Introduction to the results

Haematological status of the cow was assessed by the haemoglobin levels. Energy status was determined by the glucose, BHB and FFA concentration in the blood analysis. Protein metabolism was controlled by looking on the urea levels in urine and in the blood samples. The function of the vital organs like liver and kidney was also checked. Indictors of the liver function were AST and FFA levels. Kidney parameters are pH, Na, K and Urea from blood and urine analysis. The general status of the cow was represented by the BCS and the minerals balance.

Table 1. Ják-Felsőnyírvár 25. 06. 2012

Results of the laboratory examination of blood samples

Group of cows	Ear tag number	DIM	BCS	HB mmol/l	Glükóz mmol/l	BHB mmol/l	FFA mmol/l	AST U/l	Urea mmol/l
Reference values			2,5-3,5	5,0-5,6	2,3-3,9	<0,8	<0,800	<80	3,3-5,0
Close-up	6801		3.5	4.6	3.0	0.3	0.400	63	4.3
	8881		3.5	5.7	2.8	0.5	0.580	62	3.7
	8318		3.5	5.1	2.4	0.6	0.320	64	4.0
	8818		4.0	7.1	2.9	0.4	0.650	79	2.7
	7536		4.0	6.4	2.9	0.6	0.370	66	4.8
Average			**3.7**	**5.8**	**2.8**	**0.5**	**0.464**	**67**	**3.9**
TSD			**0.3**	**1.0**	**0.2**	**0.1**	**0.143**	**7**	**0.8**
Fresh cows	8905	0	3.5	7.8	2.0	0.6	0.440	81	3.7
	7372	0	3.5	8.9	1.6	0.6	0.540	88	4.5
	6514	4	2.0	5.7	1.8	0.7	0.250	124	5.4
	8617	1	4.0	8.0	2.8	0.7	1.020	94	3.7
	8908	3	3.5	6.0	1.6	0.7	0.590	78	3.9
Average			**3.3**	**7.3**	**2.0**	**0.7**	**0.568**	**93**	**4.2**
TSD			**0.8**	**1.4**	**0.5**	**0.1**	**0.284**	**18**	**0.7**
Early lact. first parity	8914	23	2.5	5.4	0.5	0.7	0.760	72	5.4
	8927	20	2.5	6.2	1.7	0.7	0.360	90	4.3
	8892	14	3.0	5.5	0.4	1.1	0.800	73	3.1
	8746	21	3.5	4.6	2.0	0.8	1.080	89	3.4
	78763	50	2.5	5.3	2.0	1.0	0.480	71	3.8
Average			**2.8**	**5.4**	**1.3**	**0.8**	**0.696**	**79**	**4.0**
TSD			**0.4**	**0.6**	**0.8**	**0.2**	**0.284**	**10**	**0.9**
Early lact. multi-parous	8182	35	**1.5**	5.8	0.4	0.6	0.070	80	4.2
	5320	61	2.5	5.3	0.9	0.4	0.330	105	4.1
	7740	46	3.0	5.9	2.6	0.6	0.750	66	5.7
	8347	20	2.0	4.8	0.4	1.6	0.590	78	5.0
	7653	36	2.5	5.9	0.9	0.3	0.450	86	4.6
Average			**2.3**	**5.5**	**1.0**	**0.7**	**0.438**	**83**	**4.7**
TSD			**0.6**	**0.5**	**0.9**	**0.5**	**0.259**	**14**	**0.6**
Peak lact. first parity	8755	89	**1.5**	5.4	2.1	0.5	0.370	80	4.3
	8698	143	2.5	5.6	1.9	0.4	0.160	113	4.5
	8656	112	**1.5**	5.0	1.4	0.6	0.060	121	6.9
	8832	136	3.5	6.3	1.6	0.4	0.210	70	3.8
	8576	181	2.5	5.3	1.1	0.7	0.060	93	4.9
Average		**132**	**2.3**	**5.5**	**1.6**	**0.5**	**0.172**	**95**	**4.9**
TSD		**35**	**0.8**	**0.5**	**0.4**	**0.1**	**0.128**	**22**	**1.2**
Peak lact. multi-parous	7988	154	**1.5**	5.6	2.8	0.4	0.400	114	5.3
	6749	82	**1.0**	5.3	2.1	0.3	0.260	124	5.4
	7862	95	2.0	5.4	1.1	0.3	0.330	102	4.8
	7281	108	**1.5**	5.1	1.8	0.6	0.250	84	4.2
	7000	149	**1.5**	4.6	1.6	0.6	0.090	106	5.6
Average		**118**	**1.5**	**5.2**	**1.9**	**0.4**	**0.266**	**106**	**5.1**
TSD		**32**	**0.4**	**0.4**	**0.6**	**0.2**	**0.115**	**15**	**0.6**

Table 2. Ják-Felsőnyírvár 25. 06. 2012

Results of the laboratory examination of blood samples

Group of cows	Ear tag number	Carotene µmol/l	Ca mmol/l	inorg. P mmol/l	Mg mmol/l	Cu µmol/l	Zn µmol/l	Gsh-Px U/g Hb
Reference values		>5,6	2,1-3,0	1,6-2,3	0,8-1,2	10,0-18,9	10,0-30,6	20-30
Cole-up	6801	3.5	2.5	1.9	1.0	16.0	11.7	26.0
	8881	3.3	2.5	1.7	1.0	18.0	8.1	24.0
	8318	2.2	2.5	2.0	1.0	17.1	9.2	24.0
	8818	1.6	2.7	2.3	1.0	18.4	13.6	29.0
	7536	3.3	2.5	1.8	1.1	18.3	9.9	22.0
Average		**2.8**	**2.5**	**1.9**	**1.0**	**17.6**	**10.5**	**25.0**
TSD		**0.8**	**0.1**	**0.2**	**0.0**	**1.0**	**2.2**	**2.6**
Fresh cows	8905	4.8	2.4	1.9	1.1	20.7	7.3	
	7372	3.1	2.0	1.7	1.1	17.8	7.3	
	6514	2.4	2.2	1.9	0.9	17.6	8.5	
	8617	2.3	2.2	1.3	1.0	22.7	3.7	
	8908	1.2	2.2	1.8	0.9	19.1	6.5	
Average		**2.8**	**2.2**	**1.7**	**1.0**	**19.6**	**6.7**	
TSD		**1.3**	**0.1**	**0.2**	**0.1**	**2.1**	**1.8**	
Early lact. first parity	8914	1.2	2.4	2.1	1.1	17.4	10.1	
	8927	1.1	2.2	2.4	1.0	19.0	11.0	
	8892	1.2	2.3	2.2	1.0	20.4	10.0	
	8746	1.2	2.2	2.0	0.9	17.5	15.7	
	78763	1.5	2.4	1.8	1.0	16.5	13.7	
Average		**1.2**	**2.3**	**2.1**	**1.0**	**18.2**	**12.1**	
TSD		**0.2**	**0.1**	**0.2**	**0.0**	**1.5**	**2.5**	
Early lact. multi-parous	8182	3.3	2.2	1.9	0.9	20.5	5.7	
	5320	4.3	2.3	2.5	0.8	21.3	6.0	
	7740	4.0	2.6	1.9	1.2	17.2	10.5	
	8347	1.0	2.4	2.7	1.2	16.7	10.7	
	7653	2.5	2.2	2.5	1.2	18.7	10.6	
Average		**3.0**	**2.4**	**2.3**	**1.0**	**18.9**	**8.7**	
TSD		**1.3**	**0.2**	**0.4**	**0.2**	**2.0**	**2.6**	
Peak lact. first parity	8755	2.5	2.6	1.9	1.1	16.5	11.6	24.0
	8698	3.2	2.4	2.6	1.2	19.4	12.7	23.0
	8656	2.8	2.2	2.8	1.2	18.3	13.9	23.0
	8832	3.1	2.5	2.3	0.9	19.0	12.4	24.0
	8576	3.3	2.5	2.1	1.2	18.2	15.3	26.0
Average		**3.0**	**2.4**	**2.3**	**1.1**	**18.3**	**13.2**	**24.0**
TSD		**0.3**	**0.2**	**0.4**	**0.1**	**1.1**	**1.4**	**1.2**
Peak lact. multi-parous	7988	6.8	2.5	2.2	1.1	17.7	10.8	24.0
	6749	5.1	2.5	1.8	1.5	18.4	6.8	23.0
	7862	4.1	2.5	2.1	1.2	18.5	12.1	23.0
	7281	6.0	2.3	1.9	0.9	18.9	11.5	26.0
	7000	7.6	2.4	2.2	1.1	20.7	8.4	26.0
Average		**5.9**	**2.4**	**2.0**	**1.2**	**18.8**	**9.9**	**24.4**
TSD		**1.4**	**0.1**	**0.2**	**0.2**	**1.1**	**2.3**	**1.5**

3. táblázat Ják-Felsőnyírvár 25. 06. 2012

Results of the laboratory examination of urine samples

Group of cows	Ear tag number	pH	NABE mmol/l	Urea mmol/l	Ca mmol/l	P mmol/l	Mg mmol/l	Na mmol/l	K mmol/l
Reference values		7,8-8,4	>100	130-300	0,1-1,5	0,3-5,2	6,2-16,5	20-80	140-320
Close-up	6801	8.3	144	209	3.4	0.9	19.0	54	239
	8881	8.3	132	303	9.2	1.0	22.0	6	253
	8318	8.4	157	276	9.3	1.2	19.0	12	272
	8818	8.3	145	113	0.3	2.6	11.0	75	170
	7536	8.5	207	285	1.1	1.2	14.0	23	288
Average		**8.4**	**157**	**237**	**4.7**	**1.4**	**17.0**	**34**	**244**
TSD		**0.1**	**29**	**78**	**4.3**	**0.7**	**4.4**	**29**	**46**
Fresh cows	8905	8.4	56	223	0.3	1.2	13.0	17	170
	7372	8.4	114	134	0.1	1.0	8.0	5	183
	6514	**6.5**	**-6**	353	0.5	8.9	15.0	22	145
	8617	8.0	67	459	0.5	1.9	20.0	14	233
	8908	8.1	67	382	0.9	1.5	21.0	24	212
Average		**7.9**	**60**	**310**	**0.4**	**2.9**	**15.4**	**16**	**189**
TSD		**0.8**	**43**	**130**	**0.3**	**3.4**	**5.3**	**8**	**35**
Early lact. first parity	8914	8.3	130	268	1.9	1.2	15.0	137	108
	8927	8.3	109	148	0.4	1.7	13.0	145	85
	8892	8.3	133	143	0.7	0.9	9.0	118	84
	8746	8.6	95	118	0.4	0.8	11.0	86	81
	78763	8.3	186	128	0.4	0.9	9.0	136	116
Average		**8.3**	**131**	**161**	**0.8**	**1.1**	**11.4**	**124**	**95**
TSD		**0.1**	**35**	**61**	**0.7**	**0.4**	**2.6**	**24**	**16**
Early lact. multi-parous	8182	8.1	71	378	1.1	1.2	27.0	108	117
	5320	8.4	222	290	0.7	2.2	28.0	140	186
	7740	8.4	190	178	5.8	1.1	17.0	104	140
	8347	8.2	69	108	2.5	0.7	8.0	89	65
	7653	7.8	81	246	0.4	11.8	15.0	128	100
Average		**8.2**	**127**	**240**	**2.1**	**3.4**	**19.0**	**114**	**122**
TSD		**0.3**	**74**	**103**	**2.2**	**4.7**	**8.5**	**20**	**45**
Peak lact. first parity	8755	8.4	169	190	0.5	1.1	10.0	137	133
	8698	8.1	74	89	0.3	1.0	5.0	64	60
	8656	8.1	110	199	0.4	12.0	13.0	143	98
	8832	8.4	248	208	4.5	1.4	16.0	132	172
	8576	8.4	108	64	0.1	0.9	6.0	87	76
Average		**8.3**	**142**	**150**	**1.1**	**3.3**	**10.0**	**113**	**108**
TSD		**0.2**	**68**	**68**	**1.9**	**4.9**	**4.6**	**35**	**45**
Peak lact. multi-parous	7988	8.2	98	102	0.7	1.3	9.0	67	88
	6749	8.2	135	128	0.3	1.0	12.0	115	105
	7862	8.4	213	197	0.5	1.2	17.0	136	115
	7281	8.3	232	195	3.1	1.3	19.0	118	174
	7000	8.2	145	249	0.9	1.3	28.0	96	150
Average		**8.3**	**165**	**174**	**1.1**	**1.2**	**17.0**	**106**	**126**
TSD		**0.1**	**56**	**59**	**1.9**	**0.1**	**7.3**	**26**	**35**

5. Evaluation

Body condition scoring

Body condition scoring was the first parameter to be diagnosed on every animal that was sampled. A strong variation between groups can be easily recognised in the results. The groups which are not intensively milked yet show relatively high BCS while the groups in peak lactation show extremely depressed BCS.

In the close up and the fresh cows group high body condition average was found. The average of the BCS on the close-up group was 3.7. Out of the five cows samples three had 3.5 and two had 4.0. This over condition in this group which is predisposed to metabolic disorders can be considered as a major error. In the fresh cow group the average of the BCS was 3.3. One cow out of the five samples was in BCS 4.

The farm should try a restructure the nutrition of dry cows in order to optimize the BCS of the cows before calving. The high BCS found in the two relevant groups is an opened gate for metabolic disorders like ketosis and fatty liver syndrome. Those disorders can increase the prevalence of many other management and infectious disorders, as mentioned in the literature.

The BCS measured in the two groups in the peak lactation was extremely low. In the first parity peak lactation group the BCS average was 2.3, and in the multiparous peak lactation it was 1.5. This very low body condition score can lead to major decrease in reproductive performance in their cows which are already supposed to be in their reproductive period. Low body condition can also decrease the immune function of the effected animals and by that increase the rate of infectious diseases.

In the multiparous peak lactation group five out of five cows were in depressed body condition score. The farm should consider to increase the energy content of the daily ration of feed given to the cows.

Haemoglobin

This parameter is the main indicator of the homeostatic condition. Most cows had the haemoglobin concentration levels in the normal. The close up and fresh cows group showed a

strong diversion from the rest of the herd. Those two groups showed higher levels of haemoglobin concentrations resulting in haemoconcentration. In the close up group it was not as clear as in the fresh cows. In the close up group two cows out of five showed haemoconcentration and the average result was 5.8, higher that the maximum normal level. In the fresh cow group only one out of the five cows sample had normal haemoglobin concentration and the average sampled was 5.3, much higher than the maximum levels required.

These results can be explained by the relative shortage of drinking water. If 20% of cows in a group show haemoconcentration the most typical causative is relative drinking-water shortage. This is usually typical after milking or feeding. The fresh cow group is probably susceptible to this problem due to the sudden increase in milk production.

The farm should consider reforming the water supply for those two groups.

Glucose

90% of the cows sampled showed extremely depressed glucose blood levels. The exception was in the close up cows in which no cow showed low glucose level. This parameter is highly influenced by the outside temperature. The blood sampling took place in late morning in the month of June which can explain the low level of glucose which was measured. During hot weather glycolysis increases, this can explain the extremely low levels measured that day.

Beta-hydroxy-butyrate

Slight hyperketonaemia was found in the lactating cows with sporadic prevalence. Out of all the lactating cows sampled only one had high enough keton body concentration to be considered as suffering from subclinical hyperketonaemia. This prevalence is highly acceptable and indicates a favourable condition for the prevention of hyperketonaemia.

Free fatty acids

Low incidence with sporadic prevalence of high FFA count was detected during the metabolic profile test. Out of twenty five sampled lactating cows only two were found to suffer from high free fatty acid concentration. In general all the groups' average was in the optimum level.

Aspartate Aminotransferase

High levels of AST were found in at least 50% of all sampled cows. Two groups showed strong difference from one another and from the total average. In the close up group all cows sampled were in the required concentration of AST. In contrast, in the multiparous peak lactation group all cows sampled recorded higher levels than the maximum acceptable level. All the other groups had either two or three sampled cows with high level of AST.

During peak lactation liver cell activity is much higher than in any other time in the cow's life cycle. This results in frequent cell damage that increases the AST concentration in the blood. Due to this the higher AST levels recorded in the peak lactation group can be considered as acceptable.

Strong correlation was found between two factors that induces liver cell damage with high level of AST in the blood. High concentration of copper in the blood can induce liver cell damage. As well high level of free fatty acids is a common cause of liver cell damage. Out of the ten cows with high AST levels which are not in the multiparous peak lactation two had free fatty acid count and four had high copper concentration in the blood.

Urea

Urea levels in the blood were found to be in the acceptable range very few of the sampled cows showed higher or lower urea levels than the optimum. The only exception was the multiparous peak lactation group in which three out of five cows had slightly elevated urea levels in the blood. This can indicate either an overfeeding of protein or a shortage in rumen

fermentable energy in the ration. Only one cow was found to have a strong deviation from the required levels. The group average is quite ideal.

Carotene

Out of all the cows sampled the only cows that had high enough blood concentration of carotene considered as optimal were in the multiparous peak lactation group. All other cows showed either low level or very low level of blood carotene concentration; especially negative was the first parity, early lactation with a group average of 20% of the required levels.

Major importance can be attributed to the low levels of carotene measured in the first parity peak lactation group. Carotene is extremely important for the proper reproductive performance of dairy cattle especially in the stage of corpus lutea build-up. Low levels of carotene can lead to increased insemination rate for every pregnancy. These two facts can lead us to the conclusion that this group will show poor reproductive performance.

Calcium

Calcium level in was normal in the great majority of cows sampled. This indicates a good supplement of calcium in the feed in the lactation period. Early lactation groups all showed good blood levels ((2.1 – 3.0 mmol/L). this last parameter is a proof of the good management of feeding low calcium level feed in the dry period as recommended.

Inorganic phosphorus

Most of the cows sampled showed normal levels of inorganic phosphorous. Some cows show high level of phosphorous in the urine. High level of phosphorous in urine can be an indication to acid load or even to an imminent acidosis. In some cows that suffer from high phosphorous level in the urine there is either acidaemia or very low acid base balance.

Magnesium

All cows sampled have normal levels of magnesium in their blood. Some of the cows show higher levels of magnesium in the urine which can indicate overconsumption of magnesium. Excretion of magnesium in the urine is connected to the amount of magnesium intake in the feed.

Copper

One third of the sampled cows show slightly elevated copper concentration in their blood. The distribution is approximately equal all over cows in lactation. Dry cows show normal levels of copper concentration. High level of copper can be a cause to liver cell damage and increased blood concentration of AST. A strong correlation exists between those two values in the sampled cows. No correlation between haemostatic variation (haemoconcentration) and the copper concentration was found.

Zink

All of the cows in the freshly-calved group show a subclinical shortage of zinc. As well the multiparous groups show a 40% incidence of zinc shortage. This shortage in zinc might increase the risk of infectious diseases due to immune suppression especially in the mammary glands. Hoff problems and reproduction failures are connected to zinc shortage.

Glutathione-peroxidase

All the cows sampled had normal levels of GSH-PX.

Urine pH

Out of all the cows sampled only one cow showed abnormally low pH values, indicating a specific problem of acid load in that animal. Subclinical metabolic acidosis can be determined in this cow. The most common cause of this problem is subclinical or subacute ruminal acidosis due to feed which is too rich in concentrates.

Net Acid Base Excretion

Values below a 100 mmol/litre are indication of an acid load in case of values of 0 or negative clinical or subclinical acidosis is likely. The freshly calved group shows the lowest NABE value in average. This can be due to the more concentrate-rich feeding ration they are given.

Sodium

Sodium levels in urine samples are highly connected to the group from which the samples were collected from. Almost all the samples coming from close up and freshly-calved group show very low sodium concentration. 90% of the samples coming from cows in lactation show very high concentration of sodium in the urine. This can be explained by the fact that during the dry period and just after calving the farmer wants to prevent other oedema by decreasing sodium intake. On the other hand low sodium levels in the feed can decrease the appetite of the cow. In the lactation groups the high concentration of sodium in urine can be explained due to the overfeeding of buffer (sodium bicarbonate). This is given by the farmer in order to prevent rumen acidosis caused by concentrate-overfeeding. Overfeeding of sodium bicarbonate might cause rumen alkalosis.

Potassium

75% of lactating cows show low urine levels of potassium. This is usually caused by low level of potassium in the forage given to the animal. The outcome of potassium shortage can negatively impact the reproductive performance of the cows. As well, a normal level of potassium helps the cows to combat heat stress. Good balance of minerals and primarily potassium will support the animal's appetite and immune system, by that improving the general well-being of the animal.

6. Summary

The following statements should be considered as the cardinal issues of the evaluation.

- BCS was too high in the close up and freshly calved groups while it was very low in the rest of the groups
- Haemoconcentration in the close up and freshly calved groups
- Sporadic high blood concentration of AST
- Carotene deficiency in all sampled cows in exception of the peak lactation multiparous group.
- Sporadic high cupper concentration in the blood
- Zink shortage in the freshly caved group
- Sodium level in the urine of close up and freshly calved groups is very low
- Sodium level in the urine of all the lactating groups is very high
- High prevalence up to 75% of animals showed hypokalemia

7. Conclusion

The metabolic profile test preformed on the farm was able to surface some subclinical disorders. This outcome of the test is a positive one as it will allow the farm management to correct major mistakes before they will develop into a clinical disease. From the evaluation of the results we can see a clear picture of the farm management and nutritional condition.

Energy status

The energy status of the cows is not perfect. The main problem is with the suboptimal BCS of all animals in the farm. most of the cows before lactation are in a relatively high BCS then the recommended. The majority of the cows in lactation groups have very low BCS which might put them in risk of many disorders mainly infectious and traumatic disease. Surprisingly the prevalence of subclinical metabolic diseases which is connected with energy status is relatively low. A very small group of cows show values that indicate the presence of the most common metabolic disorders like ketosis, fat cow syndrome, fatty liver syndrome and milk fewer.

Protein metabolism

Urea is the main indictor of the protein metabolism. Most of the cows in all of the different groups show acceptable levels of urea in blood and in urine samples. The balance between the energy and the protein is good and should be kept as well the total protein supply is good.

Minerals and vitamins

Some macrominerals are normal and some are in clear herd level disorder. Calcium magnesium and phosphorus are all in the recommended blood level. Sodium blood concentration is too low in nonlactating cow and too high in lactating one. Low levels of sodium in the blood can decrease appetite and so predispose to many metabolic disorders related to the transition period. High sodium blood levels are an indicator of overfeeding with sodium-bicarbonate. This can lead to rumen alkalosis which will lead to farther complications. Potassium levels are very low in all the lactating cows and this can lead to many complication including problems with appetite and immune function. Recommended level of potassium can help the cows overcome the heat stress during the summer days.

From all the microminerals copper and zinc are the one that showed herd level abnormalities. High cupper levels were found in most groups in high prevalence this can increase liver damage and risk the cow's metabolic performance. Low zinc levels can chose increase risk of hoof problems and infection in the mammary gland due to immune-suppression. Zinc is a major component of normal reproductive function.

Carotene is the only vitamin precursor in the metabolic profile test. All cows except the one in the pick lactation multiparous have extremely low carotene blood concentration. Low carotene levels can cause decreased reproductive function. Increased insemination rate for every pregnancy is also a typical outcome.

8. Discussion

The impotence of metabolic disorders in the modern dairy industry is beyond doubt. The increasing load on cows to deliver greater milk yield every lactation period is the main cause to the increase in metabolic failures. Metabolic profile testing is an ideal tool for the diagnosis of subclinical metabolic disorders.

With subclinical disorders it's of the highest importance to diagnose them in early stage before. When subclinical disorders become clinical the economic losses are already great. Regular metabolic testing of herd can be a major tool for combating metabolic disorders.

9. References

1. A.H. ANDREWS, R.W.BLWEY, H. BOYD, R.G.EDDY, 1992, BOVINE MEDICINE DISEASES AND HUSBANDRY OF CATTLE. P 575 - 606
2. A. I. Macrae, D. A. Whitaker, E. Burrough, A. Dowell, J. M. Kelly, 2006, Use of metabolic profiles for the assessmentof dietary adequacy in UK dairy herds, *Veterinary Record* (2006) 159, 655-661
3. A. J. EDMONSON, I. J. LEAN, L. D. WEAVER, T. FARVER, and G. WEBSTER, A Body Condition Scoring Chart for Holstein Dairy Cows, Journal of Dairy Science Vol. 72, No. I, 1989
4. Brydl, E., Könyves, L., Jurkovich, V., Mrs. Tegzes, L. and Tirián, A., 2005, SUBCLINICAL METABOLIC DISORDERS IN PERIPARTAL DAIRY COWS IN HUNGARY IN 2005**,** ISAH-2007 Tartu, Estonia, p 423 – 427
5. Finbar Mulligan, Luke O'Grady, Desmond Rice and Michael Doherty, 2006, Production diseases of the transition cow: Milk fever and subclinical hypocalcaemia, Volume 59 (12) December, 2006, Irish Veterinary Journal
6. G. H wentink, VPMG. Rutten, Th. Wensing, NUTRITION, METABOLIC DISEASES AND IMMUNITY IN DAIRY COWS, Proceedings of the World Buiatrics congress, Edinburgh 8 – 12 july 1996,
7. Garrett R. Oetzel, DVM, MS, Herd-Level Ketosis – Diagnosis and Risk Factors, Preconference Seminar 7C: Dairy Herd Problem Investigation Strategies: Transition Cow Troubleshooting AMERICAN ASSOCIATION OF BOVINE PRACTITIONERS 40th Annual Conference, September 19, 2007 – Vancouver, BC, Canada
8. Garrett R. Oetzel, 2007 -2008, Production Medicine Fresh Cow and Calves Rotation, Fall Semester2007 and Spring Semester 2008, p 1 – 19
9. J.M. Payne, S.M. Dew, R. Manston, M. Faulks, The use of a metabolic profile test in dairy Herds, Vet Rec. 1970 Aug 8;87(6), 150-8
10. J. M. Payne, The Future of Presymptomatic Diagnosis, 182 Proc. roy. Soc. Med. Volume 65 February 1972
11. Kevin Lager, Ellen Jordan, 2012, The Metabolic Profile for the Modern Transition Dairy Cow, Mid-South Ruminant Nutrition Conference
12. Michel A. Wattiaux, Chapter 12: Body Condition Scores, The Babcock Institute, http://babcock.wisc.edu/node/170
13. Neville Sutlle, 2010, *Mineral Nutrition of Livestock*, 4th EDITION, p. 54 – 200

14. Stephen LeBlanc, Health in the Transition Period and Reproductive Performance, 2010, WCDS Advances in Dairy Technology (2010) Volume 22: 97-110
15. Todd F. Duffield, Minimizing Subclinical Metabolic Diseases in Dairy Cows , WCDS Advances in Dairy Technology (2006) Volume 18:43-55
16. T.F. Duffield, S.J. LeBlanc, 2009, Interpretation of Serum Metabolic Parameters Around the Transition Period,
17. The Dairy Industry in Israel ,2011, Israel dairy board, http://www.icba-israel.com/Dary_Ind_2011_english.pdf
18. Wayne Kellogg, Body Condition Scoring With Dairy Cattle, University of Arkansas, United States Department of Agriculture, and County Governments Cooperating, http://www.uaex.edu/other_Areas/Publications/PDF/FSA-4008.pdf
19. William B. Epperson, 2005, Risk Factors for Metabolic Disease, Tri-State Dairy Nutrition Conference

10. Acknowledgements

First and for most I would like to thank Professor Brydl Endre for is support and expertise he was the guiding hand being all of my actions. I would like to thank the stuff of the animal hygiene department with the help with collecting the samples and for the laboratory work. I would like to thank my Colleagues on the farm and to the veterinarian on the farm Dr. varga Tamas for their help and support during the collection of the samples.