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# **Strategies for the prevention and reduction of the main health problems in the beef cattle rearing**

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# **CHAPTER 1**

## **Foreword**



# 1 Foreword

In every productive process, commercial, industrial or zootechnical, there is a “critical point” that is able to influence the good result of the productive system in terms of economic profitability.

The good results of the investment are strictly influenced by the production cost that should be balanced between a satisfactory profit and a good quality of the final product in order to comply thoroughly the requests of the customers. In this way the demand and the supply are well balanced and the entire productive sector could stay alive.

Therefore the “quality” of a product is a primary feature and the goal to pursue during the entire productive process.

This concept applies usually in every productive sector, but is maximally important for all the business that involves in some way the human foodstuff sector.

The “quality goal” is really important because every single phase could definitively compromise the qualitative characteristics of the final product, and usually it happens as gravely as the productive process is quick.

This observation shouldn't be limited to the industrial sector, but should be applied to the zootechnical field due to the really intensive productive level that is typical for farms of every size.

In light of the above shows clearly how, in the beef, the process of farming is seen as having a role of fundamental importance for the influence it exerts on the economic results, but particularly because it is during this phase that it's built and defined the quality and features of the final product.

To define the phases of the farming process that could have an important influence on the profitability of the investment and those that could obstruct to achieve the best result in terms of beef quality, is of the utmost importance to emphasize and enquire every critical point that influences the productive phases.

## 1.1 Adaptation Phase

For animals, as occur also in the humans, the adaptation to new environment and social conditions gives rise to stress condition; this situation could drive the subject to severe physiologic and psychological reactions and compromise the health.

The relationship between stress and immunity response of the organism and so the susceptibility to various diseases was demonstrated in the 1936 by Selye.

This is the reason because the adaptation phase for the imported beef cattle is the most important critical point.

The problems related with the adaptation at the new environment of the cattle take place not only to imported cattle that travels for many hours, but also to cattle moved from neighboring farms and even when the cattle has been moved inside the same farm. Evidently the severity of the problems is also related with the length of the travel, so in the last two cases isn't comparable with the first case. We highlight the morbidity of the pulmonary disease that occurs when the cattle are moved from their barn inside the farm to remove the manure from the paddocks (especially in the winter because the temperature decrease quickly), or when the management of the farm requires to mix groups of healthy animals without distinction of breed or sex.

The dimension of the problems doesn't extent only by the gravity of the stress due to the environmental factor that occurs on the cattle, like the varied temperature and humidity conditions, social competition, bacterial environmental load and so on and so forth. The transport and the management of the cattle before it produce a stress condition that plays a crucial role to bring about the typical problems of this farming phase. (Young, 1996, Sgoifo Rossi et al., 1996)

Besides the external factors, whom significance strictly depends by characteristics of its (duration of the transport or hydric and food deprivation, management of the cattle at the arrival, like treatments, and so on and so forth), also the subjective characteristics, like weight of the animal, sex, breed (Sgoifo Rossi et al., 1997) have a crucial influence on the capability of adaptation to new environments and then to determine the incidence of problems (Santoro, 1996; Grandin, 1997).

The most careful farmers apply specific procedures to the newly arrived herds because they recognize the relationship between those aspects and the problems (Young, 1996; Sgoifo Rossi et al., 1996).

Although is really hard to evaluate how every aspect contribute to compound those problems because the health compromission derives from a multifactor effect where only one of those couldn't change the physical, psychological and metabolic balance and so compromise the health status (Santoro, 1996).

The improvement of the health and nutritional management of the newly arrived herds, haven't cut the occurrence of the problems during the adaptation.

The market conditions need the herds to have fast growth and short periods of breeding, since the first phases of farming, and so brought the herds in a condition of high susceptibility to illness.

During the adapting phase of the imported beef cattle, the three primary problems are the pulmonary disease, the parasites infestations, and the locomotor system disease. In fact only the pulmonary disease causes about 60-80% of the disease morbidity in the adapting cattle (Church and Radostis, 1981; Young , 1996) and the 40-80% of the mortality or convenience culling (Young, 1996; Horton, 1984).

The parasites infestations conversely, could cause a heavy impairment of the performance of the cattle and predispose the animals to other infectious disease and in some cases this situation could drive the subject to death (Laval and Remy, 1994; Muneer et al, 1988).

The incidence of the articular disease is raising in the last years and represents the 40% of culling of the animals, because the recovery after this illness is difficult and long (Tondello et al, 1996; Sgoifo Rossi et al, 1997).

During the adaptation phase, depending on the various factors listed above, the problems that occur in a group of animals could vary from 15% to 80%, with a mortality rate that could be even about 20% (Sgoifo Rossi et al, 1997).

In the well managed farms the morbidity of the different diseases is subjected to a wide variability, but the mortality rate is between 0.2 and 3%. In this regard, it should be specified that for mortality rate we mean all the subjects that dies in the farm, but we exclude the subjects culled for convenience.

If this rate is upper than the values listed before, it means that there are some weakness during the entire process; then the farmer should evaluate all the management, with more attention to the nutritional and sanitary management.

If we examine the zootechnical parameters of a newly arrived group of animals, is really interesting to evaluate the number of animals that requires a specific management in suitable pens (like sick-bay) in consequence of the severity of the problems. On the average the incidence of these situations is between 0.5 and 3%, but there are some reports of very high values (about 20%).

The complexity of this situation makes this phase the most important critical point of the entire breeding process, so we could clearly understand how many problems could cause a lack in the management or an unsuitable adaptation

program. This situation in turn causes a decrease in the productive performances, an increase of the pharmacological costs, of the technical and veterinary assistance, of the convenience culling, of the death of subjects, and finally of the length of the breeding process and then of the financial liabilities.

## **1.2 Fattening Phase**

Fattening phase is very important in order to reach the “quality goal”, that because even in this phase, every error could compromise the sanitary status of animals and the qualitative characteristics of the final product.

Fattening status intervene on two aspects of meat quality fundamental for consumers, that are health-nutritional and organoleptic characteristic.

With an appropriate nutrition it's possible to satisfy consumers and to avoid slaughtering mishap due to animals too fat or too thin.

The choice of nutritive level, as well as diet feeds, will be function of farmer objective and cattle age, sex and genetics.

At the present time farmer target must be to obtain high quality meat, and so nutritional level will be the most of the time high, to stimulate the greater growth than possible.

Energy and protein concentration of diets should be based on the different development phase and on breed precocity.

Diets for growing cattle aging 8-10 months (300-320 kg), should be formulated considering that these animals are in a phase in which they are growing fast, and they need adequate protein amount.

Differently cattle aging 15-16 months are in a phase completely different, and nutritional management should be aimed to maximize, since the very early days in farm, the correct ripeness at slaughtering.

In function of these aspects and the variable precocity of cattle, high in female and in milk breed and very low in breed characterized by muscular hypertrophy, we should decide if to adopt high energy concentration since the early phase of breeding or to begin with high energy from the half of production phase, or again to use high energy only during the final phase, fattening period; to each of these choice should correspond the correct protein concentration in the diets.

For example, breed like Piemontese, Blonde d'Aquitaine, Garronaise or every cattle with a marked muscular hypertrophy needs high energy diets from the early phase of breeding, because the main problem with these bovine is that it's

very hard to have animal with the correct fattening at the right age of slaughtering that should be 17-28 months of age.

The risk, frequently, is to slaughter or too lean cattle if they are sold when weight and age are thought to be ideal, or too old and heavy cattle if they are sold when maturity and fattening are ideal for that kind of beef.

Such mistake can be avoided breeding young cattle fed, even from 6-8 months of age, high energy (0.96 UFC/kg D.M.) and protein (16 % CP based on D.M.) diets, because feeding high energy involves higher protein synthesis, that need to be adequately sustained (Hocquette et al., 1998).

Nutritional management should be different, reflecting breed characteristic, for female or for high precocity cattle.

The increase in energy concentration should be gradual during productive cycle, to prevent a too early fattening, and protein concentration should be medium-high (16-17% CP based on D.M.) to stimulate myogenesis, guarantying sufficient ADG (Average Daily Gain) and little risk of early adipogenesis.

### *1.2.1 Feeding and feeding management critical points*

The arising of metabolic disease is above all due to risk factors that can modify the whole gastro enteric environment.

It is in balance, and it's regulated by endogen and hexogen factors, that are related with animal, feeding, management and environment.

But this balance can be altered by various causes that can generate disease and also death.

Control and limiting of ruminal and enteric micro flora misbalance predisposing factors include various nutritional and management aspects.

Nutritional factors are for sure of primary importance in development of digestive metabolic disease in beef cattle.

Ruminal and enteric environment, and precisely their pH, is regulated by an adequate balance between feed choice and feeding and farm management.

Any variation of this fragile balance leads to alterations of digestive functions, of feed transit quickness and can induce the passage in the enteric tract of excessive undegraded nutrient, like starch or protein.

In the end, considering that italian typical beef cattle diet are characterized by high energy concentrations that predispose to digestive metabolic disease, the correct management of feed level and feed administration in the fattening period is very important.

Furthermore, high amount of concentrate, and so high amount of NSC (Non Structural Carbohydrate) induce increased feed speed in gastroenteric tract, and cause ruminal environment alterations and favourable intestinal conditions to enterotoxaemia development.

### **1.3 Factors that compromises the beef cattle rearing**

#### *1.3.1 Stress*

Stress is a biological response to a stimulus that could perturb the homeostasis of the subject. Broom (1986) argues that this situation comes true every time a subject is put through adverse or dangerous situations. The response is behavioral, psychological and physiological.

Several authors describe the welfare condition like a status of total health, both psychological and physical in which the animal is harmonized with the environment (Huges and Dun, 1986); another definition is the conditions of the animal searching to adapt to the environment (Broom, 1986).

The entity of the reactions depends not only by the seriousness of stressing factors, but also by some subjective characteristics, like breed, sex, weight, age and temperament of animals (Sgoifo Rossi et al, 1996), and by some other factors like previous sensitization with similar situations (Grandin, 1997; Santoro, 1996).

After stress and panic situation, the organism reacts with a secretion of epinephrine and norepinephrine. If the situation that generated this modification in the hormonal secretion persists, there is a modification of the hypothalamus-hypophysis-adrenal gland line with the consequent secretion of glucocorticoids. It is demonstrated that the immune response is influenced by these hormonal mechanisms. In event of acute stress the animals have a “fight and escape” reaction with secretion of catecholamine an activation of the sympathetic nervous system. There is an increase of the heart rate, of the systolic pressure and of the perfusion of several districts. Besides all this systemic reactions, triggered by the sympathetic nervous system and by the secretion of adrenaline and noradrenaline, there also an involvement of the immune defense system (Santoro, 1996).

When an animal is subjected to stress there is a hypotalamic secretion of the corticotrophin releasing factor (CRF). This peptide promotes the releasing of the adrenocorticotrophic hormone (ACTH) by the hypophysis that has his target in



the adrenal gland cells. These cells release then the glucocorticoid hormones. It has been demonstrated that the transport of cattle induce an increased level of these hormones in the blood, with top level after 24h. The level remains at this point for about a day and then decrease in seven days.

The immune depression caused by glucocorticoids and catecholamines probably has the role to modulate the immune response, with the inhibition of the proliferation of T-lymphocytes activated by antigens in order to prevent the eventuality of autoimmune reactions. In fact it has been demonstrated that the presence of antigens determine an increase in the concentration of these hormones in the blood; furthermore there is evidence of a receptors induction on the surface of lymphocytes and white cells during an infection. The immune system is modulated by the cortisol through the inhibition of 1 and 2 interleukin and by the hyposensitization of its receptors (Blecha, 1986). The interleukin mechanism could modulate the secretion of the CRF and then the ACTH and glucocorticoids. Also the growth hormone (GH), thyroxine and prolactin could play a role in the modulation of the immune response after a stress condition; In fact, in vitro, all these three hormones have the effect to increase several cellular functions of the immune system.

The immunodepression is due to a reduction of the blastogenetic activity of the T-lymphocytes in spite of leucocytosis that come true, probably effect of the mobilization of the marginal pool caused by the sympathetic response. These changes in the hormone balance could last for a long time after the end of the stressing situation (even ten days) (Axelrod et al., 1984; Blecha et al., 1984; Crookshank et al., 1979; Kelley, 1998; Locatelli et al., 1985; Murata, 1989; Ponti et al., 1989).

The impairment of the health of the animals and of the growth performance during the adaptation phase is more severe as the management skill and the attention to the welfare is lower. (Camp et al., 1981; Church and Radostis, 1981; Hutchenson, 1990; Moonsie-Shageer and Mowat, 1993; Sgoifo Rossi et al., 1996). In this regard has been evidenced that with an adequate management the stressing event could be reduced drastically, improving both the health of the subject, and the productive performance (Grandin, 1997).

### *1.3.2 Transport*

Transport is a stressing event that could influence the next phase of imported cattle adaptation. In fact during the transport by train or by truck, animals bear

different disadvantaged situations as water and food deprivation, fear of the sudden movement during the transport, noise, adverse climatic conditions, restricted spaces and low comfort, the contact with other animals, and more other. The length of the transport exert an influence on the severity of the stress, because a long journey means that the adverse conditions interact with the animals for a longer time and so the cattle imported from remote farms have worse immune defense than those transported from nearby and the morbidity of the respiratory disease is related with the distance (Church et al., 1981); this situation is frequent in the Italian intensive farming because the animals are often imported from abroad. In addition there is an influence of season, in fact it has been demonstrated that there is a relationship between the climatic condition, the weight loss and the pulmonary disease morbidity (Camp et al., 1981).

Another aspect that should be considered is the discomfort in which the animals are involved before and after the transport. During the phases preceding the travel the cattle is often grouped together in gathering cowshed, where the stressing factors are important: the handling, the shuffling of different groups of animals mutually unknown, the crowding in narrow spaces, the variations in the rationing, the fastening and the water deprivation. In these cowshed there is a continue transition of animals and often the animals are crowded and so there is a high load of microorganism in the environment.

There are specific laws that regulate the transport of the animals because the public opinion is sensitive for the animal welfare. There are contrasting opinions about the convenience of the stop during long transports, because there are studies that highlight a higher morbidity in animals that stopped than others that, on the same journey with similar traveling time, didn't stopped (Grandin, 1997). In fact during the stops the animals are unloaded from the trucks and recovered in cowshed and so are subjected to supplementary stressing handlings.

### *1.3.3 Weight loss*

The weight loss that occurs in cattle after the transport could vary from 2 to 12% (Table 1) and should be attributed to all the handling phase with the situations before the transport and the operation that are typical after the arrival in the farming. The weight loss is related with some external conditions, like the duration of the transport, weather conditions, the entity and the type of the stressing factors that affect the animals; some studies highlighted that there is a

higher weight loss in animals transported than in animals only fastened for same time.

The nutritional management of the animals before the transport is a factor that could influence the weight loss, in fact saline rehydrating solutions administered to animals reduce the weight loss. It has been demonstrated that the administration in the drinking water of an electrolytic solution constituted of 0,02-0,04% of sodium chloride, 0,02-0,08% of potassium bicarbonate, 0,01-0,04% of magnesium sulphate, 0,005-0,009% of some amino acids (lysine, alanine, phenylalanine, threonine, tryptophane, methionine, isoleucine, leucine, hystidine, valine in equal amounts) and phosphoric acid to reach pH 7, in the phases before the transport reduces the weight loss and make easier the adaptation phase of the imported cattle (Schaefer et al., 1992; Cole et al., 1996).

Table 1. Weight loss (%) in beef cattle of average weight 213 Kg during the entire handling phase and only during the transport (1600 Km).

<b>Heads, n.</b>	<b>Transport weight loss, %</b>	<b>Total weight loss, %</b>	<b>Difference, %</b>
275	7.7	11.1	3.4
152	6.8	11.4	4.6
185	8.5	12.1	3.6
159	9.9	12.3	2.4
194	8.5	12.7	3.7

The weight loss that occur during the handling phase is due to reduction of the urinary bladder and gastrointestinal tract content, but also to some tissutal catabolic events and to dehydration, event of utmost seriousness for the balance of the organism, second only to the oxygen deprivation (Hutchenson and Cole, 1986). A severe weight loss during market, transport and handling phase is related with increase of morbidity and worse growth indexes (Camp et al., 1983; Griffin, 1983).

#### *1.3.4 Digestive disorders*

Nutritional conditions before the arrival in the livestock and before the adaptation have a role of utmost importance in the success of the adaptation phase, because it influences the health status of the beef cattle.

Several authors have widely described how the nutritional state is important as a factor of conditioning the immune response and how specifically the lack of energetic support and particular nutrients, like amino acids, vitamins and

micronutrients could cause an immunodepression state and the reduction of the growth of the animals (Hutcheson and Cole, 1986; Muneer et al., 1988; Hutcheson, 1990; Moonsie-Shageer and Mowat, 1993; Dubeski et al., 1996; Fluharty et al., 1996; Cole, 1996). In addition, it must be considered that during the stress and the inflammatory status there is an increased renal excretion of some micronutrients, and so the deficiency of these elements is exacerbated (Moonsie-Shageer and Mowat, 1993). This situation added to the several therapeutic and prophylactic treatments supplied to the animals, make easier to compromise the hepatic functionality. Usually after the arrival in the destination farm, the animals are in a nutritional stress condition, due to the food and water deprivation typical of the transport phases and to the unsuitable diets fed in the starting farms or in the markets places, often with the only purpose to reduce the feeding cost. The malnutrition, the fast, and the water privation are exacerbated by stressing events like transport, competition, handling of the newly arrived beef and determine an immediate weight loss and a reduction of the appetite (Phillips et al., 1985; Hutchenson and Cole, 1986).

The energy deficiency plays a role in the worsening of the appetite because the negative energetic balance causes a mobilization of the body fat, specifically the long chain fatty acids, with further appetite depression. The accentuation of the catabolism mobilizes also the muscular proteins because the amino acids are used by the liver for the gluconeogenesis.

After the arrival in the farm the animals, which often came from the pasture, have to adapt to different nutritional conditions. A study compared the daily intake of animals fed at the crib versus other fed at the pasture. The authors describes that the first seven days after the arrival in farm, the animals used to eat at the crib have an intake 33% higher than the animals used to fed at the pasture. In the first two weeks of adaptation the intake is severely reduced and sometimes in the first five days the animals eat nothing (Hutchenson and Cole, 1986; Chang and Mowat, 1992).

Table 2. Daily feed intake (% of normal intake) in imported beef cattle with the respiratory disease and without it. (Hutchenson, 1990. Modified).

Adapting phase	Feed intake, % of normal intake	
	Healthy	With disease
1th – 7 th day	51 %	29.7 %
1th – 28 th day	89 %	60.7 %
1th – 56 th day	100 %	88.4 %

The feed intake is influenced obviously by external and subjective factors, like is evidenced in table 2 and 3, in which are reported the feed intake of cattle subjected to respiratory disease, water deprivation and fasting for different times. As evidenced in table 2 the beef affected with respiratory disease have an intake 50% lower than the healthy and this lower intake lasts for 28-56 days after the arrival as well the animals are cured. In table 3 is evidenced how water deprivation and fasting have an impact on the feed intake, that is halved when the restriction ends (day 0) in subjects exposed for 48 hours compared with animals not deprived and in beef subjected to fastening for 48 hours versus 72 hours. Is interesting to observe that 4-7 days after the end of the restriction the intake is not completely restored.

Table 3. Effect of 48 and 72 hours of food and water restriction on the DM intake (kg/d) in a group of crossbreed beef (average weigh Kg 237 Kg) at the moment of the restriction, 4 and 7 days after it. (Fluharty et al, 1996)

Days after restriction	Duration of fastening and water restriction, h		
	0	48	72
0	4.413	2.295	1.148
4°	5.586	4.266	3.796
7°	5.826	4.766	4.325

The low intake is caused by the reduced digestive efficiency that is characteristic in newly arrived animals and depends by the compromised rumen functionality due to the fasting and water restriction during the transport (Fluharty et al., 1996).

It has been observed that the fasting reduces the rumen volume (Fluharty et al., 1996), the frequency and duration of rumen contractions (Galyean et al., 1981), the rumen turnover (Fluharty et al., 1996) and finally there is a modification in the rumen microflora (Galyean et al., 1981; Hutchenson and Cole, 1981; Fluharty et al. 1996). Some authors noticed a direct action of the stress on the rumen activity, highlighting how some catecholamines have an inhibiting action on the rumen and reticulum motility (Graham et al., 1982; Cole et al., 1986), and increase the intestinal one. In this way there are the optimal conditions for the proliferation of anaerobic microorganism (like clostridium) and the production of toxins that compromise the health status and reduce the appetite.

After the fasting it is reduced the rumen content and so there are few substrates to ferment. The rumen bacteria start to replicate very quickly after the restore of

the optimal condition, but the protozoa aren't able to do the same. It needs about a week to restore an optimal condition (Cole and Hutchenson, 1985; Cole, 1995; Fluharty, 1996). The transport could have an influence on the fermentation capability of the rumen that could be about 25% lower than normal conditions (Cole and Hutchenson, 1981) and this situation could last about three or more days after the arrival in the farm (Cole and Hutchenson, 1985).

The impairment of the growth performances and the health of the animals due to the transport and first adapting phases are caused both by the water and food restriction and also by several factors that influence the homeostasis of the animal. Furthermore it is important to highlight that the nutritional and water restriction limit the stress response of the cattle worsening its negative effects (Hutchenson and Cole, 1986).

### *1.3.5 Management*

Another factor of the most importance is the nutritional, hygienic and the handling management of the animals inside the farm. A bad management could drive to several problems with a so important economic prejudice to make the investment uneconomic. These problems harm also the industrial farms managed with specific and strict protocols. It's evident that the animals have to be received and stalled in the best way, in disinfected sheds, isolated from the other adaptation structures of the farm, with the purpose to separate the newly arrived animals from pathogens yet present in the farm and contrariwise that the newly arrived cattle could introduce in the farm some pathogens. It's also important that the microclimate inside the sheds should be controlled and void of harmful gas and dust in order to reduce the irritation that could impair the local defense of the respiratory ways against virus and bacteria (Barbari et al, 1995).

Other important factors that affect the morbidity and the mortality are the numerosity of the groups, the shuffling of the animals and the space available for every single head. The crowding raises the infecting charge of the environment and the contacts between animals with mix up of pathogens. The availability of space, conversely, could improve the weight gain (Martin et al., 1982; Martin et al., 1986). The handling usually applied to the newly arrived animals, like vaccinations, parasitocidal treatments, marking with ear tag, dehorning and metaphylaxis, should carried out with the maximum respect for the animals

welfare and reducing the stress and the natural defensiveness or the fear against the humans, with the goal not to compromise the productivity and the immune defense (Pasillè et al., 1996).

#### *1.3.5.1 Feeding management*

Another important factor that has influence on the typical disease of receiving period and on fast adaptation is feeding management with the aim to reestablish as fast as possible the ruminal normality conditions (Dell'Orto e Sgoifo Rossi, 2000; Dell'Orto et al., 2002; Dell'Orto et al., 2005b). It's clear that a delayed reestablishment of normality nutritional conditions, and the whole physio-metabolic homeostasis, induce a sub clinical and prolonged suffering that leads to greater susceptibility and incidence of health problems and a reduction of growth performance.

Transport, handling before and after the trip, fasting, water restriction and stress induce a severe compromising and limitation of digestive activity and functionality, that can last for several days and that needs a fast and affective rebalance of all the nutrients.

This objective isn't obtainable with diets containing mainly straws or exclusively hays. New receiving beef cattle needs, as well as mechanic function fiber, also and mainly of high degradability fiber, of specific energy and protein sources, of vitamin, micro and macro elements, proportioned not only to requirements but also to reduced feed intake, due to a lack of appetite, and for these reasons it's explained the need of a correct use of glucose precursors, probiotics, social balancing substances and ruminal fermentations modulators (Dell'Orto et al., 2002; Dell'Orto et al., 2005a)

Feeding adequate energy since the first phase after arrival is of fundamental importance to limit mobilization of body reserve induced by transit and adaptation stress that produce a lower appetite, and sometimes a lower or absent feed intake.

For this reason is very useful to include in the diet glico plastic substances, like propylene glycol, that bypassing rumen for more than 60 %, and by originating in the rumen propionate, give to cattle rapidly use energy.

The use of rumen protected fatty acids, too, is an effective strategy, because they allow to give a big amount of energy also under conditions of lower feed intake (Vandoni et al., 2006).

As well these substances reduce probability of acidosis that, in the opposite direction, can take place when energy is given only with the use of cereals.

Talking about protide, besides the importance of a correct balance between soluble, ruminal degradable and undegradable protein, particularly attention must be given to the presence in the diet of those amino acids that can influence immune response.

Also the use of probiotic like yeast, that enhance ruminal protein synthesis and feed intake, produce an increase of available proteic amount.

In fact yeast enhances appetite, promotes fiber degradation and fibrolitic bacteria and lactic acid user bacteria activity, and reduces N-NH<sub>3</sub> and methane concentration in the rumen, with positive effects on ruminal ecosystem balance.

Protein don't accomplish only the task to satisfy synthesis needs of the body, but interact with it in several other manners, like attending directly on metabolism, enhancing or reducing feed intake and conditioning immunity.

Several studies highlight decrease of immune response in subjects fed hypoproteic diets or diets poor in some specific amino acids.

Mineral rebalance is essential, and particular importance must be given to K, Na, P, Ca, Mg, Cu, Zn and Fe, that are useful to re-establish the correct electrolytic and acid-base balance of the organism.

Same attention must be given to vitamins, also to those vitamins that usually are synthesized in adequate amount to satisfy requirements, like group B vitamins and vitamin C.

In fact feed and water restriction, or some disease, can induce a lack because, in such conditions, requirements can increase from 2 to 15 times, it depends on vitamin and amount and length of stress (Zinn, 1987), and there are marked alterations of haematic concentrations of pantothenic acid, ascorbic acid, B6 and B12 vitamins.

Also vitamin E, with Se, deserves specific attention. Several researches show an increase of immune response in different species animals fed with specifically integrated diets. The stimulus of immune response involves several mechanisms. Vitamin E holds a protective role for leucocytes and macrophages during phagocytosis process, protecting these cells by toxic action of some substances product during elimination of pathogens.

Vitamin E reduces productions of glucocorticoids that have immunosuppressive action. Vitamin E could stimulate immune response by altering arachidonic acid metabolism, and so prostaglandin, thromboxans and leucotriens synthesis; they are usually produced in great amount during stress status, and they have negative effects on immunity cells activity (Williams et al., 1993).



Besides vitamin E promotes haematic neutrophyl, peritoneal and pulmonary macrophages and T lymphocytes activity, enhances phagocytosis activity and antibody production, limits lack effects of essential nutrients like zinc, reduces toxic effect of several heavy metals and mycotoxins and stimulate vitamin C synthesis.

### *1.3.6 Respiratory disease*

Bovine respiratory syndrome is the most frequent health problem during the adaptation phase of the imported cattle, responsible of 60-80% of the morbidity (Church and Radostis, 1981; Young, 1996) and of 40-80% of the death causes (Young, 1996; Horton, 1984); it represents the first expense because is responsible of the increase of the costs of pharmacological treatments, veterinary or technical manpower and make slower the adaptation process. Furthermore there are also negative effects on the quality of the meat (Gardner et al., 1999).

#### *1.3.6.1 Immunity and defense of the respiratory tract*

There are several defense systems in the respiratory tract, classified in mechanical, biochemical, cellular, secretory antibodies and interferon.

*Mechanical.* Cough, epithelium ciliate and mucosal integrity are the most important mechanical defense. Mucosal necrosis, adverse environmental conditions, like gas, dust, hot and cold could cause a total inhibition of mechanical defense and the deposition of mucus due to the stop of the ciliary activity is an optimal pabulum for the bacteria and could obstruct the aerial tract. During respiration the swirl of the air make the small particles of dust (<5 micron) to hit the mucosa and the mucosal film catches it. Then the progression of the cilia brings the dust particles in the superior tracts and then they're expectorated or swallowed.

*Biochemical factors.* Lysozyme, transferrin takes action against bacteria. Surfactant balances the ventilation in the alveolus, supporting a regular gas exchange. Without this action the lung is no longer elastic with a bad ventilation.

*Cellular factors.* Harmful substances, viral infections and severe vitamin deficiency could compromise the action of macrophage and non specific phagocytic cells, worsening the reactivity to infections.

*Interferon.* It is a specie specific, virus non specific, glycoprotein that is secreted by viral infected cells with the objective to protect the adjacent cells.

*Secretory antibodies.* They are secreted by respiratory mucosa after the contact with the antigen.

### *1.3.6.2 Etiology*

Several microorganisms are responsible of the respiratory disease. The viral infection is the “key factor” that allow the bacterial secondary infection. The more frequent described viruses are bovine herpes virus type 1, para influenza virus 3, bovine viral diarrhea virus, adenovirus, rhinovirus, reovirus, enterovirus, coronavirus. On the gravely modified mucosa take place the bacteria, like *pasteurella* spp, *haemophilus somnus*, *mycoplasma* spp, *chlamidya* spp, *corynebacterium pyogenes*, *salmonella* spp. In our farms the *mycoplasma bovis* is responsible of 25-30% of the pulmonary disease (Gevaert, 2006).

The respiratory syndrome is a typical conditioned pathology, related to factor that have a negative interaction with the animals. In the intensive farms the animals are subjected to several and frequently unknown of these factors and so it is really difficult to identify and eliminate it.

Furthermore at the typical respiratory syndrome there is the correlation of the compromise in other districts. For example the herpesvirus causes alteration also in the genital and eyes, with an opacification of the cornea. Also the other etiological agents could have not only respiratory localizations.

### *1.3.7 Parasitic diseases*

One of the more important causes that compromise the growth performances and the optimal utilization of the nutrients is the parasite infestation. The parasites directly worsen the performances of the animals and indirectly they predispose the animals to the action of the infective antigens driving, in the worst situations, to the death of the subject (Laval and Remy, 1994; Muneer et al., 1988).

In the imported cattle the transport and the adaptation are factors that increase the reproduction of the parasites and the awakening of the asleep larvae in the animals fed at the pasture in the origin country.

The animals are usually affected by different species of parasites; among nematoda, like *ostertagia ostertagi* and *trichostrongilus axei* parasitize the abomasum, *nematodirus filicollis* and *helvetianus*, *strongiloides* and *neoascaris* parasitize are in the small intestine and they are responsible of enteritis and bad digestion and malabsorption with strongyle that parasitize the caecum and colon. In the respiratory tract it is possible to find *Dictyocaulus viviparus*, responsible of wormy bronchitis and pneumonia. Among trematoda the more common are *fasciola hepatica*, *dicrocoelium dentriticum* and *paramphistomum*. The cestoda

family, in particular *Echinococcus granulosus*, arrives in the farms with the animals imported from the pastures. Another important parasite is *Moniezia benedeni*, localized in the small intestine. The insects have importance not for the wounds that they cause during their activity, but as vectors of other parasites and infections.

#### *1.3.7.1 Coccidiosis*

A special consideration would be reserved to the coccidiosis because the prevalence of these parasites is very high in the imported cattle. Usually the coccidia don't cause clinical evidence of the disease, but the chronic infestation of these protozoa limits the growth of the animals. The typical infestations are caused by eimeriidae, like *Eimeria* and *Isospora* with more frequency, and more rarely also *Cystoisospora* and *Cryptosporidium*. The principal symptoms of the acute disease are rare, but the chronic type of the disease is really frequent in the intensive breeding and it generally slows down the growth of the animals and so also the daily food conversion index is reduced significantly (Fox, 1983; Fox, 1987; Laval and Remy, 1994). The incidence of the subclinical disease on the zootechnic parameters makes this pathology to be considered the fifth disease of the beef cattle, with recovery costs higher than the clinical form (Fitzgerald and Mansfield, 1984).

The environment plays a crucial role in the effects that the parasites could have on the animals. Stalls characterized by bad aeration, high ammonia concentration, carbon dioxide, manure, and environmental moisture could increase the risk of infestation and the dangerousness of the parasites (Fox, 1983).

Every condition that exerts on the animal an immune depressing action could influence the disease and its diffusion in the farm. So the transport, the sudden changes of temperature, the nutritional unbalancing or the modification in the composition of the diet plays an important role with the parasite load in the environment in the occurrence of the disease.

There is no vaccine against the coccidia, but luckily there are several pharmacological principles against the coccidiosis, that could stop the reproduction of the parasites waiting that the animal could react against it. An important consideration is that it's evident the effect of the parasites only when they have yet damaged the intestinal wall, so it's really important to use those drugs as preventive drug rather curative. It should not be excluded also the occurrence of secondary infections in the intestinal tract.

Between the substances with higher efficacy, the decoquinate is the one that has been wider used also because it is constant in different environmental conditions and is really desirable for the cattle (Laval and Remy, 1994). The effect of this drug is exerted on the DNA of the *Eimeria* spp particularly on the sporozoite (Hodgson, 1968) and during all the agamic phase, on the schizonts, (Fitzgerald and Mansfield, 1984). All these combined actions could reduce drastically the excretion of the oocysts and the occurrence of the clinical disease.

Another drug class that could be used in the fight against the coccidian is the sulphonamide, but the efficacy of the administration depends by the timeliness of the intervention (Grafner et al, 1965). Another advantage is the antibacterial effect of the sulpha drugs, that is helpful for the secondary infections. (Burger, 1983). In any case there are some studies that highlight the low efficacy of these drugs in the mammalians (Snoep and Potters, 2004).

During the last years the most used drug was the sodium monensin, used widely in the beef cattle breeding since the 1990 as auxinic and anti coccidia, but now its use is no more allowed by the EC rule (1831/2003) since the 1st January 2006 and there is nothing that could replace it.

So is necessary to use alternative substances. Like the yeasts, the malate or the phyto derived drugs (Sgoifo Rossi et al., 2006). The use of those drugs promote the balancing of the digestive process and reduce the occurrence of some diseases and of the coccidian infestations. Furthermore these drugs aren't subjected to the need of veterinary prescription, haven't interruption time and could be used as prophylactic drugs.

### 1.3.8 Lameness

Another problem that could compromise the adaptation phase are the pathologies of the locomotor system, that occur with a certain frequency.

These pathology could concern all tissues of the limb, but in every case they generate severe conditions because the *quoad functionem* prognosis is often gloomy. In every situation, the recovery is really slow and the subject stay for a long time lying down and then they don' eat enough; so it's important to segregate the animals in suitable cowshed in which there is no social competition.

The pathology of the locomotor system could affect the articulation, the soft tissues or the corn.

#### *1.3.8.1 Joint diseases*

In the adapting imported cattle the disease have always traumatic or septic inflammatory causes. Extremely rare are the secondary diseases and the immunological reactions, while the neoplastic origin it must be excluded because the animals have a lifetime too short to develop this kind of disease.

#### *1.3.8.2 Soft tissues diseases*

The lesions that interest the soft tissues of the bovine finger are frequently septic inflammatory processes localized to the heel and to the interdigital region.

Interdigital dermatitis and phlegmon is related to unbalanced diets, like acidogenic diets rich in carbohydrates or to deficiency of micronutrients, like zinc. Also the environment plays an important role in the development of these diseases: the excessive humidity of the bedding make easier the development of the phlegmon. The more frequent etiology are bacteria like fusiformis necrophorus, bacillus nodosus streptococcus spp and polimicrobial gram negative flora.

#### *1.3.8.3 Corneous tissue diseases*

There are several types of corneous lesions and they are caused by nutritional and traumatic factors. Pododermatitis circumscripta and white line disease of the tip are frequently caused by lesions of the keratogenous tissue due to clinical or subclinical laminitis. Also the circulatory alterations that occurs when the animals stand for long time on hard surfaces like concrete or grated floors and the adverse environment conditions could support the occurrence of these disease. The disease caused by traumatic factors are the heel white line disease, the sole lesions and, rarely, the traumatic desungulation .

#### *1.3.9 Ruminal acidosis*

Acute and chronic acidosis, conditions that follow ingestion of excessive amounts of readily fermented carbohydrate, are prominent production problems for ruminants fed diets rich in concentrate. Often occurring during adaptation to concentrate rich diets in feedyards, chronic acidosis may continue during the feeding period. With acute acidosis, ruminal acidity and osmolarity increase markedly as acids and glucose accumulate; these can damage the ruminal and intestinal wall, decrease blood pH, and cause dehydration that proves fatal. Laminitis, polioencephalomalacia, and liver abscesses often accompany acidosis. Even after animals recover from a bout of acidosis, nutrient absorption may be

retarded. With chronic acidosis, feed intake typically is reduced but variable, and performance is depressed, probably due to hypertonicity of digesta. Feeding higher amounts of dietary roughage, processing grains less thoroughly, and limiting the quantity of feed should reduce the incidence of acidosis, but these practices often depress performance and economic efficiency (Owens et al., 1998).

By definition, acidosis is a decrease in the alkali in body fluids relative to the acid content (Stedman, 1982). Because pH of body fluids is buffered by bicarbonate, the pH of body fluids may or may not be depressed during acidosis, depending on the degree to which bicarbonate compensation is possible. Central nervous system function can be disturbed by low bicarbonate concentrations even if blood pH is not depressed. Although clinical diagnosis of acidosis requires blood pH to fall below 7.35, other clinical signs such as ruminal pH, anorexia, variable feed intake, diarrhea, and lethargy are the routine diagnostic indications of acidosis of feedlot cattle.

#### *1.3.9.1 Acidosis in cattle*

Anaerobic microbes in the rumen and cecum ferment carbohydrates to VFA and lactate. Ruminal production of more than 55 mol of VFA daily has been measured in steers fed feedlot diets (Sharp et al., 1982). Herbivores absorb these organic acids from the rumen and/or cecum for metabolism by tissues. When carbohydrate supply is increased abruptly the supply of total acid and the prevalence of lactate in the mixture increase. Normally, lactate is present in the digestive tract at only low concentrations, but when carbohydrate supply is increased, lactate can accumulate; ruminal concentrations occasionally reach 100 mM. Acidosis of ruminants often is separated into two forms, acute and chronic or subclinical. Animals exhibit acute acidosis as an overt illness following consumption of readily fermented carbohydrates in amounts sufficient to reduce ingesta pH. With chronic acidosis, feed intake and performance are reduced, but animals may not appear sick.

Meal frequency may be as important as total feed intake as a cause of acidosis. For example, cattle with implants typically have greater feed intakes.

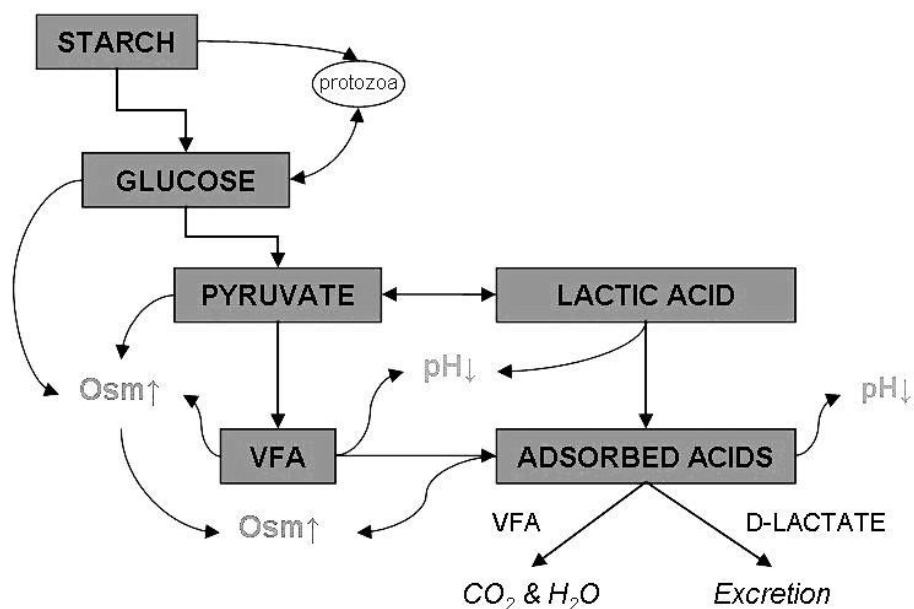
The roles of ruminal protozoa in acidosis are not clear. By engulfing starch particles and storing glucose as polysaccharide, protozoa delay starch fermentation by bacteria, help to retard acid production, and stabilize ruminal fermentation (Slyter, 1976; Nagaraja et al., 1990). In view of the large amounts of starch consumed by ruminants, the quantitative significance of starch

consumption by protozoa seems questionable. However, the population of ruminal bacteria normally decreases when protozoa are present; this decrease also could delay fermentation. Protozoa numbers in the rumen typically decline when high-concentrate diets are fed, probably because long dietary fiber provides a fibrous mat in the rumen to which protozoa attach and remain long enough to replicate. Free fatty acids and detergents reduce protozoa numbers, as well, and a low pH may cause defaunation. However, in addition to stabilizing normal fermentation, protozoa presence in the rumen can be deleterious. Because they have much higher amylase activity per unit of protein than bacteria (Mendoza and Britton, 1991), protozoa, when rupturing due to changes in acid or osmolarity associated with acidosis, release large amounts of amylase that in turn accelerates glucose production from starch and increases the likelihood of acidosis.

### 1.3.9.2 Etiopathogenesis

The relevant steps involved with acid production in and output from the rumen are illustrated in Figure 1.

Figure 1. Key reactions in acidosis of ruminants (Owens et al., 1998 – mod.).



Bacteria in the rumen often are classified as “lactate producers” or “lactate users.” Balance between these two groups determines whether lactate accumulates. End products of bacterial strains may change depending on substrate availability and culture conditions (Russell and Hino, 1985). Most

lactate-using microbes are sensitive to low pH, whereas most lactate producers are not. Under anaerobic conditions, pyruvate is converted to lactate to regenerate the NAD used in glycolysis. Under “normal” conditions, lactate does not accumulate in the rumen at concentrations above 5 mM. In contrast, ruminal concentrations exceeding 40 mM are indicative of severe acidosis. Ruminal and silage microbes produce two forms of lactate, the D+ and L form. The L form, identical to that produced from glucose by exercising muscle, can be readily metabolized by liver and heart tissue. In contrast, D+ lactate, typically 30 to 38% of the total lactate found in the rumen, is not produced by mammalian tissues. In addition to D-lactate and VFA being involved with acidosis, other microbial products including ethanol, methanol, histamine, tyramine, and endotoxins often are detectable during acidosis and can exert systemic effects (Koers et al., 1976; Slyter, 1976).

Conversion of pyruvate to VFA involves multiple steps and generates approximately half the ATP for microbial growth in the rumen; the other half is derived from conversion of glucose to pyruvate. Normally, VFA do not accumulate at sufficient concentrations in the rumen to reduce pH drastically. However, when the rate of acid production exceeds the rate of acid absorption, due either to rapid production, inhibited absorption, or reduced dilution, VFA accumulate to higher concentrations. In some studies, ruminal pH falls below 5.0 even without lactate being present. This has led to the suggestion that total acid load, not lactate alone, is responsible for acidosis (Britton and Stock, 1987), particularly with chronic acidosis.

Ruminal pH represents the consortium of relative concentrations of bases, acids, and buffers. The primary ruminal base is ammonia.

The two primary buffers under neutral pH conditions are bicarbonate and phosphate. In addition, VFA and lactate act as buffers when pH falls below 5, as discussed by Counotte et al. (1979). When pH decreases to 5.0 during acidosis, ionization of acids increases slightly, but the added lactate is primarily responsible for the increased hydrogen ion concentration. Lactate depresses pH more drastically than similar amounts of other ruminal acids because its pK (the pH point of maximum buffering) is considerably lower (3.8 vs 4.8). With low pH, osmotic pressure is increased by greater ionization of acids and presence of free glucose. Compared with normal concentrations, the change during acidosis is much greater in osmolarity than in the hydrogen ion concentration.

Absorption from the rumen normally prevents acid accumulation;



Lactate and VFA are absorbed passively through the rumen and intestinal epithelium. Rate of absorption is greater when concentrations are high, pH is low, and osmolality is normal (Tabaru et al., 1990). The lower the pH, the higher the percentage of each organic acid in the non-dissociated (acid) form and the greater the absorption rate. During absorption, butyrate is partly metabolized as an energy source for the rumen wall and glucose is partly converted to D-lactate. Lactate also is produced in and absorbed from the intestines (Godfrey et al., 1992), so total lactate load for the liver may greatly exceed the lactate absorption from the rumen.

However, high osmolarity of ruminal contents reduces the rate of acid absorption (Tabaru et al., 1990). Furthermore, acidity enhances activity of lactate dehydrogenase, increasing conversion of pyruvate to lactate and complicating recovery from acidosis. Combined with the fact that a low pH enhances activity of pyruvate hydrogenase and favours pyruvate conversion to lactate (Russell and Hino, 1985), a severe drop in ruminal pH is difficult to reverse.

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## **CHAPTER 2**

### **Objectives**



## 2 Objectives

The objective of the present doctoral study was to evaluate the incidence and the severity of the health problems in the Italian beef cattle rearing focusing on the two of the most important pathology that characterized this sector: the Bovine Respiratory Disease and the parasitosis. An important part of the study was dedicated to evaluate the effectiveness of drugs administration to reduce the incidence and severity of BRD and to understand how specific plant extracts could reduce the parasitic infestation.

In specific, the aim of the first study was to perform a wide data collection to evaluate the incidence and the severity of the health problems in the adaptation phase of the beef cattle imported in Italy. Furthermore, has been reported all the primary factors that have an influence on the adaptation phase and the data of the health conditions of a significant number of beef bred in a farm characterized by highly specialized management, with the purpose of highlight that there are a lot of different factors that have an influence on the adaptation phase of the beef cattle, despite all the efforts to prevent and cure these problems.

Furthermore, the 4 subtrials of the second study were conducted to investigate the efficacy of gamithromycin in the prevention and treatment of bovine respiratory disease (BRD) in newly received beef cattle.

In the end, the third study was managed to verify the effects of some different commercial plant extracts on growth performance and coccidia infestation in 235 newly received Charolaise beef cattle, imported from France, during the adaptation period. This trial was divided in four subtrials to test four different plant extracts.



## **CHAPTER 3**

# **Incidence and severity of the health problems in the adaptation phase of newly received beef cattle**





### **3 Incidence and severity of the health problems in the adaptation phase of newly received beef cattle**

#### **3.1 Abstract**

The typical Italian beef cattle rearing of fattening imported young animals makes the adaptation phase the first important critical point of the entire breeding process, so it's easy to understand how many problems could cause a lack in the management or an unsuitable adaptation program. This situation in turn causes a decrease in the productive performances, an increase of the pharmacological costs, of the technical and veterinary assistance, of the convenience culling, of the death of subjects, and finally of the length of the breeding process and then of the financial liabilities.

In this study it has been enquired the incidence and the importance of the more important sanitary problems of the adaptation phase of the imported beef cattle related with some animal's parameters. The enquired parameters were body weight, weight loss, incidence of pulmonary disease, incidence of locomotion disease due to traumatic and nutritional causes, incidence of animals moved in sick-bay pens due to pulmonary or locomotion diseases and finally mortality and the cause of it. To analyze the parameters listed above, the weight of the animals was divided in four classes (<300 Kg, 300-380 Kg, 381-450 Kg and > 451 Kg), the weight loss was divided in four classes (<2%, 2-3%, 3-7%, and >7%), the breeds were Charolaise, Limousine and crossbreed, and finally it has been considered the two sexes.

The data collected showed that pulmonary disease is inversely proportional to the body weight while the locomotion system diseases are directly proportional to that. Subjects with a low weight loss (<2%) had a high morbidity of pulmonary disease. Females have a morbidity of the pulmonary disease higher than the male subject, primarily because the female have a body weight lower than male and there is evidence that the body weight is directly related with the weight loss and the incidence of the problems. Conversely the males are more affected by locomotion disease, probably because those subjects are more competitive than the female. Limousine breed is more affected by pulmonary disease than the others, but it's also to be considered that those animals arrive

from France with a body weight lower than the other imported breeds. Charolaise breed manifested high rates of locomotion disease, both traumatic and nutritional, due to the higher body weight and the higher ruminal capacity that induce to speed up the adaptation program.

## **3.2 Introduction**

The arrival of young beef cattle in the farm is the beginning of a critical phase of the breeding cycle that needs to be carefully overseen to obtain high performance and low incidence of pathology and nutritional disease.

In fact it's known that often the main health problems are concentrated during the early days in the new farm, and that the consequence, also due to relapses, is a reduction in growth performance.

Several surveys carried out in Italian farm highlight that, despite the criticality of the receiving period can differ for numerous variable, like weight and sex, season, management before, during and after the transport and the weight loss, the influence on farm economy is always considerable (Dell'Orto and Sgoifo Rossi, 1998).

Despite such evidence is well documented in literature and deeply-rooted in the experience of every breeder, frequently the large number of possible strategies available to face this critical moment are not adequately esteemed and correctly applied.

## **3.3 Aim of the study**

The aim of this study was to perform a wide data collection to evaluate the incidence and the severity of the health problems in the adaptation phase of the beef cattle imported in Italy. Furthermore, has been reported all the primary factors that have an influence on the adaptation phase and the data of the health conditions of a significant number of beef breded in a farm characterized by highly specialized management, with the purpose of highlight that there are a lot of different factors that have an influence on the adaptation phase of the beef cattle, despite all the efforts to prevent and cure these problems.

## **3.4 Materials and Methods**

The data had been collected for an entire year from a group of 18.000 heads of cattle imported from France in a large farm specialized in the importing of beef

cattle. Was evaluated the incidence and the gravity of the more important diseases of the imported beef cattle related to some parameters, like the weight of the animals, the breed, the sex and the transport weight loss.

The collected data were:

- mean weight of the animals at their arrive in the farm;
- weight loss due to the transport;
- incidence of respiratory disease;
- incidence of locomotor system diseases, caused by trauma or dietary trigger;
- incidence of beef moved in sick-bay stall;
- mortality and causes.

The parameters has been evaluated distributing the animals in classes based on the arrival weight: <300 Kg, 300-380 Kg, 380-450 Kg, >450 Kg; the breeds were crossbreed, charolaise and limousine; there were animals of both sex; and the weight loss was distributed in four classes: <2%, 2-3%, 3-7%, and >7%.

The animals have been stabled in permanent bedding and the number of the animals in every stall varied depending the extent. The animals were received in adapting stalls that were far off from the fattening and from the sick bay stalls. The diets (Table 1) have been distributed with the ad libitum unifeed system, with long hay available for the first ten days of adapting phase. The protein an energy concentration has been increased gradually during the first 15 days.

There was a veterinary surgeon in the farm that managed the sanitary aspects of the cattle. During the first days has been administrated a prophylaxis based on a quadrivalent vaccine against the BHV-1, BRSV, BVDV and PI3; a parasiticial prophylaxis based on ivermectina and clorsulon was also administered within the first week. All the handling of the animals has been managed in specific corral with the purpose to reduce the stress induced by the contact with the humans.

The health of the animals has been controlled daily by the veterinary and when there was suspicious of infection of one beef, the technical staff administered an antibacterial therapy to it inside the stall without restraining the subject. If the treatment failed the animal was moved to the sick bay stall to be managed individually.

Table 1. Characteristics of the arrival diet.

<b>Feed</b>	<b>kg/head/d</b>
Corn silage	6.00
Beet pulp (dry)	1.70
Corn meal	1.40
Soybean meal (50% CP)	1.01
Micronutrients and vitamins	0.14
Straw	0.70
Dry matter, kg	6.01
UFC, kg dm	0.86
CP, % kg dm.	13.58
Fiber, % kg dm	17.63
NSC, % kg dm.	44.14
NDF, % kg dm	36.84
Ca, % kg dm	0.76
P, % kg dm	0.32

### **3.5 Results and Discussions**

#### *3.5.1 Diseases and sex of the animals*

##### *3.5.1.1 Respiratory disease*

The morbidity of this pathology is lightly higher in the females than in the males, as shown in table 2. A factor that explains clearly this result is that the females arrived at a lower body weight than the male beef. As described in the first chapters, several authors described the existing relationship between the bodyweight and the incidence of the respiratory disease in the adaptation of the imported beef cattle (Dell'Orto and Sgoifo Rossi, 1998; Sgoifo Rossi et al, 2001) Another factor that differentiate the male from the female is the higher nervousness of the females. This situation could influence the reactivity to the stressing events and so the immune response and feed intake.

Table 2. Influence of sex on the health of cattle

<b>Sex</b>	<b>Male</b>	<b>Female</b>
Number of subjects	9261	8435
Mean weight, Kg	434 ± 52	285 ± 50
Weight loss, % of Body Weight	4.1 ± 1.06	4.9 ± 1.09
Incidence of respiratory disease, %	10.21	12.06
LSD–nut, %	2.24	0.38
LSD–tra, %	1.73	0.04
“problem beef”, %	6.37	4.42
Respiratory disease, %	53.22	91.96
LSD–nut, % *	25.64	1.07
LSD–tra, % *	16.94	6.97
Dead rate, %	0.745	0.12
Meteorism/enterotoxin, %	23.19	-
Peritonitis, %	7.25	-
Urolithiasis, %	8.69	-
Respiratory disease, %	57.98	100.00
Other, %	2.89	-

\*LSD–nut: Locomotor system disease due to nutritional factors

\*LSD–tra: Locomotor system disease due to traumatic events

### 3.5.1.2 *Locomotor system disease*

It has been evidenced that there is a higher incidence in the males than in the females, giving proof of a higher risk of this kind of disease due to the higher competitiveness typical of the males and due to the early and fast diet concentration that characterizes these subjects. Anyway, the traumatic disease could be generated also by quick direction change or sudden escape from novelty, situations that are really common and more disordered in the groups of female; this is a good reason not to approach the animals in a rough way, mainly the group of young females.

### 3.5.1.3 *“Problem beef”*

The occurrence of the need to move the animals from their stall to the sick bay stalls due to severe respiratory or locomotor system compromise had been influenced by the sex of the animals. According with previous survey there were a higher occurrence of “problem beef” in the male due to the locomotor system disease.

It must be considered that if the cattle have a good management there are more animals that are moved in the sick bay stall due to the more scrupulous respect of the animal welfare. It is difficult to make a correct interpretation of this data, because if there are many animals moved to the sick bay stalls it could be an indicator of the correct management, and so the animals are moved early from their stalls, or conversely it could be an indicator of poor management, because is really high the incidence of disease or there are incorrect health practices on the cattle. Again, low incidence of “problem beef” could be an indicator of poor animal welfare, because there aren’t the conditions to move the animals to the sick bay stall even if severely sick.

The mortality has been really low and almost caused by respiratory disease both in males and in females. In males there were some evidence of meteorism and of toxæmic disease, strengthening this occurrence that the males are subject to early and rapid diet adjustments.

### *3.5.2 Diseases and breed of the animals*

#### *3.5.2.1 Respiratory disease*

In Europe there are only few scientific surveys of relationship between breed and reactivity against the respiratory disease and so is impossible to formulate a hypothesis on the different susceptibility to the diseases related to the breed. In United States the research activity on this argument have demonstrated that there is a relationship between breed and susceptibility to diseases. Engle et al (1999), demonstrated that infecting with BHV-1 some Angus and Simmenthal animals, the first had a higher immune response than the others. In Italy many worker, both veterinary and not, have the perception that the subjects with high muscular development are really susceptible to respiratory disease, more than other breeds, and that the charolaise breed have a higher incidence of diseases of the joints. These considerations have a confirmation in some American studies that highlight a lower respiratory potential, a higher susceptibility to the stress, particularly the thermal one and a lower resistance to the physical effort with a quick overworking of these animals (Shackelford et al, 1994).

In this survey the limousine cattle had the higher morbidity, then the charolaise and last the crossbreed cattle (Table 3). It’s interesting to notice that the limousine cattle have an arrival weight lower than the others and so it could be a cause of the higher morbidity.

Table 3. Influence of sex on the health of cattle.

<b>Breed</b>	<b>Charolaise</b>	<b>Limousine</b>	<b>Crossbreed</b>
Number of subjects	4323	10646	2727
Mean weight, Kg	443 ± 66	328 ± 82	372 ± 70
Weight loss, % of Body Weight	3.9 ± 0.74	4.69 ± 1.24	4.63±1.02
Incidence of respiratory disease, %	9.25	12.47	8.6
LSD–nut, %	2.61	1.13	0.81
LSD–tra, %	1.22	0.98	0.22
“problem beef”, %	6.38	5.48	3.81
Respiratory disease, %	49.27	75.51	67.32
LSD–nut, %	19.56	7.54	16.34
LSD–tra, %	31.17	16.95	16.34
Dead rate, %	0.640	0.667	0.367
Meteorism/enterotoxin, %	36.42	9.86	21.43
Peritonitis, %	10.00	1.40	17.85
Urolithiasis, %	3.58	2.82	-
Respiratory disease, %	50.00	73.24	60.72
Other, %	-	12.68	-

\*LSD–nut: Locomotor system disease due to nutritional factors

\*LSD–tra: Locomotor system disease due to traumatic events

### 3.5.2.2 *Locomotor system disease*

The breed is a factor strictly related to the incidence of the locomotor system disease, especially if it's considered in relationship with the weight of the animals. The high incidence in the charolaise could be related to the weight and to the size of the animals, factors that predispose to severe traumatic events during the social fighting inside the stalls. Another factor could be the provenience of the animals in consideration of the flat pastures that characterizes the regions of France from whom the charolaise come. In fact in these circumstances the joints don't need to adapt to the intense solicitations or to sudden movements typical of the mountain pastures, and so the joint system isn't strong as in other breeds coming from uplands.

In any case the incidence of this pathologic condition should be high only in the first phases of the adaptation, because the social fighting are characteristics of the first moment of this phase. If the incidence of joint disease due to traumatism lasts also in the subsequent phases of the breeding, it's a signal that evidence poor management or inadequate cowsheds.

The charolaise cattle had also a higher incidence of foot diseases due to nutritional causes, probably because those subjects have a high rumen capacity and so those subjects have a high intake, with consequent high risk of rumen acidosis. It's important to considerate that all the nutritional foot disease could depend also by the dramatic change in the diet composition that could occur in the moment of the arrival in the destination farm and so it's of utmost importance to implement an adequate nutritional management.

#### *3.5.2.3 "Problem beef"*

It has been noticed a higher percentage of charolaise subjects moved in sick bay stalls due to locomotor system diseases both traumatic and nutritional. The limousine cattle had the highest percentage of isolation in sick bay stalls due to severe respiratory disease; probably in this case there was an important influence of the low weight of the subjects in this genetic type of animals.

The mortality was really limited in all three breeds, with a higher incidence of mortality for respiratory disease in the limousine and for meteorism/toxins disease and peritonitis in charolaise and crossbreed.

### *3.5.3 Diseases and weight of the animals*

#### *3.5.3.1 Respiratory disease*

A scrupulous consideration of the weight of the animals at the arrival should be done to evaluate the animal welfare in the stalls, because the respiratory disease resulted strictly related to the weight of the subjects. As shown in table 4, the higher morbidity of the respiratory disease was related to the weight class <300 Kg; this evidence is probably related to a lower reaction capability against the stressing conditions and against the adverse situations, to the difficult of adaptations to the new environmental, social, nutritional and handling situations of the farm (Dell'Orto and Sgoifo Rossi, 1998).

#### *3.5.3.2 Locomotor system disease*

The collected data show that the incidence of the locomotor system disease is related to the weight class >380 Kg, with the explanation of a higher social competitiveness during the adapting phase and a faster and high protein and energetic concentration of the diet during all the adapting phase.



### 3.5.3.3 “Problem beef”

The incidence of severe disease that drive to move the animals to sick bay stalls are related to the higher incidence of respiratory disease in the lighter subjects and of locomotor system disease in the heavier animals.

The respiratory disease is the most important cause of death apart of the weight of the animals. The lower mortality rate is in the <300 kg class for the meteorism/enterotoxins diseases probably due to the gradual nutritional concentration of the animals that are known to be more sensible to this factor. The >450 Kg class have also a low level of mortality due to the high capability of these animals to adapt to environmental and nutritional severe conditions.

Table 4. Weight and health problems.

<b>Weight, kg</b>	<b>&lt; 300</b>	<b>300 – 380</b>	<b>380 – 450</b>	<b>&gt; 450</b>
Number of subjects	5226	4360	4594	3516
Mean weight, Kg	254 ± 31	338 ± 24	416 ± 20	486 ± 31
Weight loss, % of Body Weight	4.9 ± 1.13	4.67 ± 1.15	4.27 ± 1.13	3.49 ± 0.91
Incidence of respiratory disease, %	13.7	11.7	9.9	8.0
LSD–nut, %	0.325	0.826	2.35	2.303
LSD–tra, %	0.12	0.55	1.74	1.53
“problem beef”, %	5.28	4.75	5.92	5.88
Respiratory disease, %	93.48	81.64	51.10	43.47
LSD–nut, %	4.35	14.49	33.82	33.34
LSD–tra, %	2.17	3.87	15.08	23.19
Dead rate, %	0.746	0.481	0.457	0.796
Meteorism/enter, %	5.13	23.81	33.33	7.14
Peritonitis, %	5.13	4.76	-	14.28
Urolithiasis, %	5.13	4.76	-	-
Respiratory disease, %	74.35	52.38	57.14	78.57
Other, %	10.26	14.29	9.53	-

\*LSD–nut: Locomotor system disease due to nutritional factors

\*LSD–tra: Locomotor system disease due to traumatic events

### *3.5.4 Diseases and weight loss*

#### *3.5.4.1 Respiratory disease*

The few articles that it's possible to find in bibliography about the weight loss highlight that it is a high influence between the weight loss and the health of the animals. The relationship between these two factors depends by the consideration that the weight loss during the transport depends not only by the reduction of the rumen and bladder content, but also by catabolic effect in the tissues and by the dehydration, that are able to impair the health of the animal.

In this study there was a result contrasting with the bibliography (Sgoifo Rossi et al, 2001); it has been evidenced that at the lower weight loss class, it is related the higher morbidity (Table 5). This result could be explained considering the nutritional management before the transport. In fact, if the weight loss is a valid indicator of the stress that have acted on the animals with a good nutritional and hydric status before the transport, it isn't so for animals that are subjected to water and feed deprivation before the transport. The weight loss could be considered as indicator of the conditions of the animals only if there is the guarantee of an optimal nutritional management before the transport. A low weight loss after a long transport indicate that before the transport the animals were subjected to a water and food deprivation and so the transport act as stressing event in conditions of animals yet stressed with an increase in the morbidity.

#### *3.5.4.2 Locomotor system disease*

Also for this type of disease it seems that the animals with lower weight loss have a higher incidence of pathology, and probably this situation could be related to high stress conditions caused by water restriction and fast before the transport. This justify the pathology of the locomotor system due to nutritional causes, but for the traumatic diseases there is probably an effect of the higher body weight of the animals with lower weight loss.

#### *3.5.4.3 "Problem beef"*

Mortality and incidence of "problem beef" has resulted higher in the lower weight loss category, as a confirmation of the results described before. Also for this parameter the respiratory disease is the more frequent cause for the mortality and for moving the animals from their stalls to the sick bay stalls.

Table 5. Weight loss and health problems

<b>Weight loss</b>	<b>&lt; 2 %</b>	<b>2 – 3 %</b>	<b>3 –7 %</b>	<b>&gt; 7 %</b>
Number of subjects	515	907	15965	309
Mean weight, Kg	400 ± 80	376 ± 77	363 ± 91	302 ± 76
Weight loss, % of Body Weight	1.46 ± 0.32	2.67 ± 0.32	4.62 ± 0.84	7.88 ± 1.18
Incidence of respiratory disease, %	31.84	14.55	10.16	13.92
LSD–nut, %	2.32	0.99	1.39	0.97
LSD–tra, %	7.77	0.55	0.73	0.65
“problem beef”, %	11.07	4.74	5.27	6.80
Resp. disease, %	68.42	69.77	67.81	76.19
LSD–nut, %	21.05	18.60	21.50	14.29
LSD–tra, %	10.53	11.60	10.69	9.52
Dead rate, %	1.750	0.990	0.56	0.320
Meteorism/enter, %	-	22.22	15.56	-
Peritonitis, %	-	11.12	6.67	-
Urolithiasis, %	-	-	3.33	-
Resp. disease, %	88.89	55.54	66.66	100.00
Other, %	11.11	11.12	7.78	-

\*LSD–nut: Locomotor system disease due to nutritional factors

\*LSD–tra: Locomotor system disease due to traumatic events

### 3.6 Conclusions

According with the bibliography, from the analysis of the collected data, emerge that there are several influences on the sanitary status by the factors sex, breed, weight and weight loss due to transport.

Female have a morbidity of the pulmonary disease higher than the male subject, primarily because the female have a body weight lower than male and there are evidence that the body weight is directly related with the weight loss and the incidence of the problems. Conversely the males are more affected by locomotion disease, probably because those subjects are more competitive than the female. The subjects moved in the sick-bay pens are mostly males affected by locomotion diseases. The mortality resulted really low both in females and males, for various causes, but for the males are more represented the meteorism or the toxiemic disease.

Even in the international bibliography the authors report that there is an effect of the breed on the susceptibility of the animals to the pulmonary disease (in fact the animals with strong muscular hypertrophy have a lower immune defense), there are no such data from Italy and Europe.

From the data analysis it was evident that the limousine breed is more affected by pulmonary disease than the others, but it's also to be considered that those animals arrives from France with a body weight lower than the other imported breeds. The breed, with other simultaneous factors, like the body weight and the mode of stabling, resulted an important factor for the development of the locomotion disease. The charolaise breed manifested high rates of locomotion disease, both traumatic and nutritional, due to the higher body weight and the higher ruminal capacity that induce to speed up the adaptation program.

According with the above, there were a high percentage of charolaise animals moved in the sick-bay pens for the locomotion disease. The mortality rate resulted higher in limousine for pulmonary disease, and higher in charolaise and crossbreed for meteorism and toxiemic disease.

The pulmonary disease is inversely proportional to the body weight, in fact the subject that are lower than 300 Kg have a high incidence due to several factors, among the others there is the low reaction capacity against stress, new situations, and modified environmental, social, nutritional and managerial conditions. On the locomotor system the effect is directly proportional to body weight, because animals that weight more than 380 Kg are more competitive and because the adaptation program is quicker than in lighter subjects.

As seen above, the lighter subjects are moved in sick-bay pens due to the pulmonary disease, and the subject with high body weight are moved for locomotion system diseases. The mortality rate is related primarily to the pulmonary disease independently by the body weight, and then to the meteorism and toxiemic diseases more frequently in the mean body weight classes because the adaptation phase is more prudent for low body weight subjects and the adaptation capability is better for heavy body weight subjects.

The limited bibliography available on this argument highlight that the weight loss could heavily influence the sanitary conditions of the animals. In this study there is an evidence in contrast with the bibliography because the subjects with a low weight loss (<2%) had a high morbidity of pulmonary disease. Because the weight loss is related with the nutritional management before the transport, the animals subjected to bad management, like water and food deprivation before

the transport, are already dehydrated and so the weight loss is lower, but the conditions of the subjects are the same of those animals subjected to these stress during the transport and so with high weight loss. This aspect emerged also for the diseases of the locomotion system, due probably to the same conditions. Furthermore, the low weight loss classes are typical for high body weight animals and so these subjects are characterized by a strong competitiveness that justifies an increased rate of articular diseases.

According with above, the mortality rate is higher for the weight loss class <2% and the causes of the mortality resulted primarily for the pulmonary disease.

From the results of this study it has been evidenced that the parameters listed above could be considered to evaluate the sanitary conditions of the cattle. Furthermore only the data of the weight loss effect were in contrast with the little information available in bibliography and so this parameter could be used to evaluate the sanitary conditions of the subjects if the management of the cattle before the transport is well known by the operators.

The typical diseases of the adaptation and the resultant sequels could have a negative effect also on the subsequent phases of the breeding, with reduction of the zootechnical parameters and severe economic loss for the breeder. In fact the occurrence of this situation could slow down the growth process and so also the quality of the meat is compromised because the animals reach elder age with consequence on the color and tenderness of the meat. The consumer consider hygiene and salubrity of the meat as factors of first choice for the buying, but in the last years also the color of the meat at the bought and the tenderness eating it are factors that could orient the choice of the people.

This survey was inspired by the need to reduce the use of the drugs in the breeding of beef cattle, to improve the animal welfare and the quality of the foodstuffs. Also the difficulties that the breeders encounter during the adaptation phase of the imported beef cattle, the actual hard market conditions and the perspective of the future of the beef cattle breeding in Italy play an important role in stimulate the research to find specific strategies to reduce the incidence of the costs related to this critical point of the beef chain.

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## **CHAPTER 4**

# **Effects of arrival medication with gamithromycin on Bovine Respiratory Disease in Feedlot cattle in Italy**

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## **4 Effects of arrival medication with gamithromycin on Bovine Respiratory Disease in feedlot cattle in Italy**

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### **4.1 Abstract**

A series of trials were conducted in feedlots Italy to investigate the efficacy of gamithromycin in the prevention and treatment of bovine respiratory disease (BRD) in newly arrived cattle. Three studies were conducted on its preventive efficacy when compared to either an untreated control, a long-acting oxytetracycline formulation or tulathromycin. The therapeutic responses to tulathromycin and gamithromycin were compared in the therapeutic study. Preventive treatment with gamithromycin significantly reduced the morbidity due to BRD by 86%, 86% and 35% compared to the untreated control group, the oxytetracycline group and the tulathromycin group respectively. In the therapeutic trial, the number of animals that required re-treatment during the 14 days following the initial medication was significantly reduced in the gamithromycin group, compared to the positive control group. These results suggest that the dual therapeutic and preventive action of gamithromycin provides a valuable addition to the veterinarians' armamentarium for the medical management of BRD.

### **4.2 Introduction**

Bovine Respiratory Disease (BRD) is a common, complex condition of young cattle. It is common firstly because, compared to other domestic animals, cattle have a relatively small lung volume and less efficient pulmonary function and are therefore vulnerable to perturbations of the respiratory tract rendering them more susceptible to infections (Kainer and Will, 1981; Veit and Farrell, 1978), secondly because many of the pathogens that are associated with disease are

themselves common and frequently occur as commensals in healthy animals (Angen et al., 2009; Aution et al., 2007) and thirdly because many of the risk factors that are associated with BRD, such as mixing (Ribble et al., 1995; Step et al., 2008) and transportation (Chirase et al., 2004; Ishizaki et al 2005) of animals, are integral to commercial cattle production (Mintert, 2003).

The complexity of BRD is a consequence of the variety of risk factors that can be involved (Cavirani et al., 2007; Hodgson et al., 2005), the diversity of viral and bacterial agents that can be present (Cavirani et al., 2007; Hodgson et al., 2005), the nature of the inflammatory response in the lungs (Ackermann and Brogden, 2000) and associated pathology (Dowling et al., 2000; Reeve-Johnson, 2001) and the differences in innate and immune responses amongst individual cattle, which may, in part, be genetically based (O'Neill et al., 2006; Snowden et al., 2006).

BRD remains the most important single cause of mortality and morbidity within the cattle feedlot industry, regardless of geographical location (Ribble et al., 1995, Smith, 1998; Thompson and Schltheiss, 2006; Sgoifo Rossi et al., 2009) and correspondingly is responsible for losses and costs that undermine the profitability of such enterprises (Snowden et al., 2006; Scheneider et al., 2009). Various measures to mitigate the risk and impact of BRD, including vaccination against viral and/or bacterial pathogens (Schunicht et al., 2003; Mosier et al., 1998), pre-conditioning of cattle before transport to the feedlot (Ribble et al., 2008; Schwartzopf-Genswein et al., 2007) and sympathetic management on arrival (Duff and Galyean, 2007), have been studied and implemented, but none are wholly effective.

Because of the central importance of bacteria - *Mannheimia haemolytica*, *Pasteurella multocida*, *Histophilus somni* and *Mycoplasma* spp. - in BRD and because there are currently no antiviral drugs registered for cattle, antibiotics form the cornerstone for the treatment and control of BRD.

Approaches to the use of antibiotics in BRD are normally classified as either therapeutic or preventive, which can be defined as:

- Therapeutic: treatment of individual cattle that are suffering from clinical BRD.
- Preventive: simultaneous treatment of cohorts of cattle in order to help prevent them from acquiring dangerous loads of pathogenic bacteria. Preventive can be further sub-divided into:
  - Prophylactic: treatment of whole groups of apparently healthy cattle, determined to be at high risk of BRD.
  - Metaphylactic: when the number of cases of BRD within a group reaches a threshold, the remainder of the in-contact animals are treated simultaneously in order to restrict the spread and impact of BRD (Lees and Shojaee Aliabadi, 2002).

Irrespective of these approaches, the objectives common to all are to reduce bacterial populations in the lungs in order that clinical and pathological changes can be reversed or prevented and to reduce the overall bacterial pathogen load within the group in order to reduce transmission within and between cohorts.

Gamithromycin is a novel 7a-azalide that has recently been developed for the treatment and prevention of bovine respiratory disease. The compound belongs to the 15-membered semi-synthetic macrolide antibiotics of the azalide sub-class with uniquely positioned alkylated nitrogen at 7a-position of the lactone ring. As a class, the azalides are characterized by having low serum concentrations, high tissue concentrations and extended tissue elimination half-life (Amsden, 2001). They also preferentially accumulate in host defense cells, predominantly polymorphonuclear leukocytes and macrophages, which can enhance the exposure of some bacterial pathogens to the antibiotic (Jain and Danziger, 2004). Gamithromycin has been developed as single subcutaneous administration in cattle to provide clinical efficacy against respiratory diseases while minimizing stress from animal handling and maximizing compliance with treatment regimens. Following subcutaneous injection at 6 mg/kg, absorption is rapid and average plasma concentrations reach a maximum within twenty-four hours of administration (Huang et al., 2009). Gamithromycin is extensively and rapidly distributed in lung tissue where concentrations reach 18,500 ng/g 24 hours after injection. Concentrations of gamithromycin in lung are 247 to 410 times higher than in plasma over the period from 1 to 15 days post-injection. The high volume of distribution ( $V_{ss}$ ) of 24.9 L/kg after intravenous administration is

reflective of this finding and the low level of binding to plasma proteins (26%) indicates that the availability of gamithromycin in tissues should be high (Huang et al., 2009).

In studies involving field strains isolated from cattle in various European countries, gamithromycin was shown to have minimum inhibitory concentration (MIC<sub>90</sub>) values of 0.5, 1, and 1 µg/mL against *M. haemolytica*, *P. multocida*, and *H. somni*, respectively, and corresponding minimum bactericidal concentrations (MBC<sub>90</sub>) values of 1, 2, and 2 µg/mL 32,33. Field studies have shown that the pharmacokinetics and pharmacodynamics are reflective of clinical responses in that a single subcutaneous dose of gamithromycin at 6 mg/kg body weight provides rapid therapeutic efficacy in BRD cases and persistent activity to control existing and to prevent new infections for an extended period (EMEA, 2008).

The studies reported here were conducted under commercial feedlot conditions in Italy within a development program to extend the European field data that have already been generated for the registration of gamithromycin (Zactran®).

### **4.3 Aim of the study**

These trials were conducted to investigate the efficacy of gamithromycin in the prevention and treatment of bovine respiratory disease (BRD) in newly received beef cattle.

### **4.4 Materials and Methods**

An outline of the four studies that contribute to this paper is provided in Table 1. All the procedures were conducted according to the guidelines of the Council Directive 86/609/EEC of 24 November 1986 on the protection of animals used for experimental and other scientific purposes (European Communities, 1986) and to “The welfare for cattle kept for beef production” of the Scientific Committee on Animal Health and Animal Welfare, 2001.

#### *4.4.1 Allocation*

For the three prevention studies, the normal procedures were as follows. The animals arrived from France at the Italian feedlots in trucks containing approximately 60 cattle each: the cattle from each consignment were off-loaded and randomly divided into two batches to ensure that the subsequent treatment

groups were evenly matched for origin. Pens, which typically had space for around 60 animals, were then filled with batches of cattle from the trucks to ensure an even distribution within each pen.

After arrival and allocation to pens, the cattle were then processed through a handling breed on Friday and Saturday according to the normal procedures for each feedlot, which included weighing as well as medication. The induction treatments comprised a range of commonly used vaccines and parasiticides (Table 1). After processing, animals were treated alternatively in order of presentation with either gamithromycin or the control antibiotic except for subtrial 1 in which the control group was unmedicated, but was not treated with a placebo. In the therapeutic study (subtrial 4), the clinically affected animals were separated from the main group of cattle and then treated as above.

#### *4.4.2 Treatment groups*

The three prevention studies were in effect a progression from the first, in which the control group received no antibiotic treatment, to the second, in which a positive control group was treated with a conventional long-lasting oxytetracycline product at 300 mg/kg, to the third, in which a relatively new product with prolonged activity was used – tulathromycin at a dose of 2.5 mg/kg. The therapeutic study was opportunistic insofar as a group of young Limousine heifers within a larger batch of animals suffered from clinical BRD very soon after arrival; they were separated off from the main group, divided into two batches and treated according to allocation with either gamithromycin or tulathromycin, in both cases plus ketoprofen at 3 mg/kg.

In Subtrial 1, in an effort to limit contact between treated and untreated animals, the treatment groups were kept in separate pens; in subtrials 2 and 4, the animals in each treatment group were mixed within pens and in subtrial 3 the cattle were penned separately by treatment groups.

In subtrial 1 naso-pharyngeal swabs were taken from a random selection of 16 animals prior to treatment at the start of the study and from 29 control animals, clearly affected by BRD, on day 7 and 14.

Table 1. Background details of the four subtrials.

Subtrial, type & Farm		Animals			Induction treatment*	Treatment groups	Duration (clinical)	Evaluations
<i>Prevention</i>	<i>Number</i>	<i>Sex</i>	<i>Breed</i>	<i>Starting weight</i>				
1. Verona	250	Male	Charolaise	~350 kg	<b>Vaccination</b> IBR, PI3, Pasteurella <b>Parasiticide</b> Ivermectin + clorsulon	125 animals gamithromycin 125 animals untreated control	14 days	Microbiology Morbidity Problem cases Growth
2. Alessandria	546	Male & female	Charolaise Limousine Ch x Lim	~345 kg	<b>Vaccination</b> IBR, PI3, RSV, BVD <b>Parasiticide</b> Ivermectin + clorsulon	235 animals gamithromycin 235 animals oxytetracycline	14 days	Morbidity Problem cases
3. Alessandria	1328	Male & female	Charolaise Limousine Ch x Lim	~325 kg	<b>Vaccination</b> IBR, PI3, RSV, BVD <b>Parasiticide</b> Ivermectin + clorsulon	568 animals gamithromycin 568 animals tulathromycin	14 days	Mortality Morbidity Problem cases Growth
<b><i>Therapy</i></b>								
4. Alessandria	24	Females	Limousine	~258 kg	<b>Vaccination</b> IBR, PI3, RSV, BVD <b>Parasiticide</b> Ivermectin + clorsulon	13 animals gamithromycin + ketoprofen 11 animals tulathromycin + ketoprofen	14 days	Animals requiring 2 <sup>nd</sup> treatment Problem cases

\*IBR=Infectious Bovine Rhinotracheitis; PI3=Parainfluenza 3 virus; RSV=Respiratory Syncytial Virus; BVD=Bovine Virus Diarrhoea virus.



#### 4.4.3 Management

In each study, subtrial cattle were subject to the same management in terms of feeding, watering, handling and housing as was normally carried out at each site. In subtrial 1 the cattle were penned in outside yards with shelter, in the other studies, the cattle were kept in open-sided sheds.

#### 4.4.4 Observations

Following allocation and treatment, the subtrial animals were examined daily by the on-site veterinarian, who was blinded as to the identity of the treatment groups. Any animals that were seen to be affected by BRD during the 14-day observation period were examined clinically and treated individually, at the discretion of the veterinarian, with antibiotics and non-steroidal anti-inflammatory agents (NSAIDs). The number of BRD-affected animals was used to calculate morbidity rates and in addition, re-treatments were recorded. If individuals were removed from the main pens to hospital pens, they were recorded as 'problem animals'. Live weight was measured at the start of the studies for the purposes of ensuring accurate dosing of any treatments and again on Day 30 in Subtrials 1 and 3 to calculate short-term growth rates.

### 4.5 Statistical Analysis

In subtrial 1 body weight and average daily gain (ADG) were individually recorded and statistically analyzed using a General Linear Model procedure (SAS institute 2004). The following model was fitted:  $Y_{im} = \mu + T_i + TP_{ik} + e_{iklm}$  where  $Y_{iklm}$  is the dependent variable,  $\mu$  is the overall mean,  $T_i$  is the fixed effect of the treatment,  $TP_{ik}$  is the fixed effect of treatment  $\times$  pen interaction and  $e_{iklm}$  is the random residual error. Due to lack of significance ( $P > 0.05$ ) of treatment  $\times$  pen interaction, this effect was not considered.

In subtrial 3 body weight and average daily gain (ADG) were individually registered and statistically analyzed using a General Linear Model procedures (SAS institute 2004). The following model was fitted:  $Y_{iklm} = \mu + T_i + TP_{ik} + bP_{ijlk} + e_{iklm}$  where  $Y_{iklm}$  is the dependent variable,  $\mu$  is the overall mean,  $T_i$  is the fixed effect of the treatment,  $TP_{ik}$  is the fixed effect of treatment  $\times$  pen interaction,  $b$  is the linear regression coefficient of the starting weight of the animals ( $P_{ijlk}$ ) on the dependent variable ( $Y_{iklm}$ ) and  $e_{iklm}$  is the random residual error.

In all the subtrials the association among Incidence of problematic animals, relapsing animals and mortality was evaluated by means the  $\chi^2$  test for a 2x2 contingency table using the FREQ procedure of SAS (SAS institute 2004).

## 4.6 Results

The main clinical results and their statistical significance are summarized in Table 2.

Table 2. Summary of clinical results in subtrials 1-4

<b>Trial number</b>	<b>Parameter</b>	<b>Treatment group</b>		<b>Significance (P)</b>
<b>1. Verona</b>		Negative Control	Gamithromycin	
	% Morbidity	34.4	4.8	<0.0001
	% Problem animals	1.6	0.8	NS
<b>2. Alessandria</b>		Oxytetracycline	Gamithromycin	
	% Morbidity	14.5	1.7	<0.0001
	% Problem animals	5.1	1.7	<0.05
<b>3. Alessandria</b>		Tulathromycin	Gamithromycin	
	% Morbidity	14.6	9.3	0.006
	% Problem animals	1.8	0.9	NS
	% Mortality	0.7	0.4	NS
<b>4. Alessandria</b>		Tulathromycin	Gamithromycin	
	% Animals re-treated	81.8	30.8	0.004
	% Problem animals	27.7	0	0.04

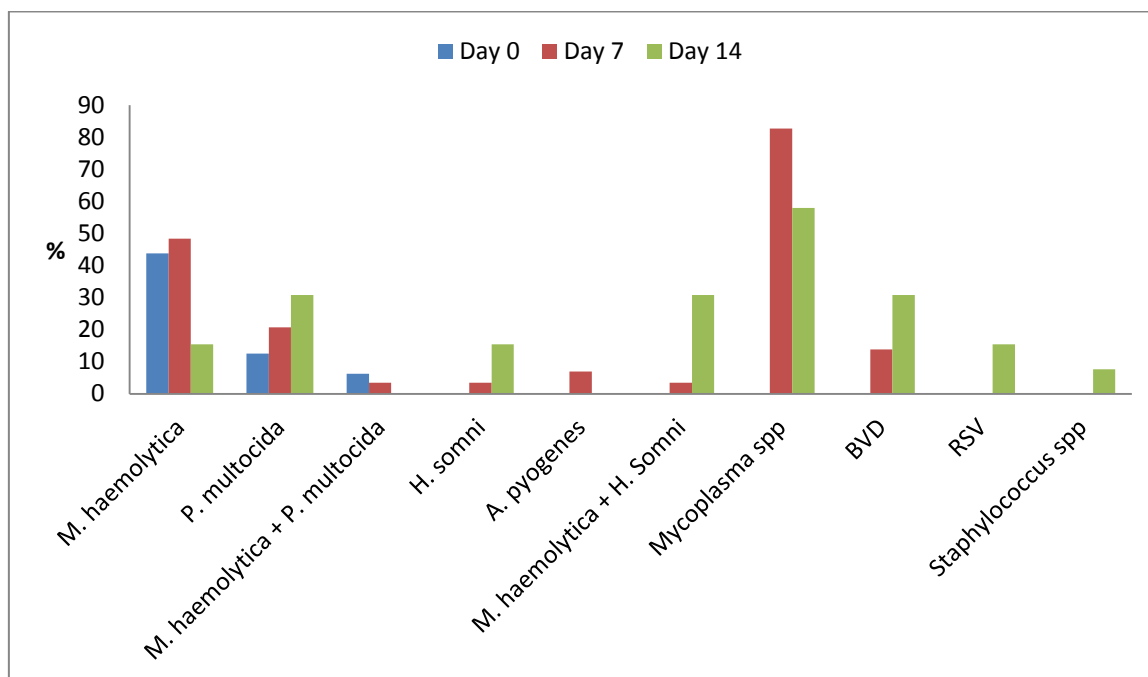
### 4.6.1 Subtrial 1

#### 4.6.1.1 Microbiology

The identity and proportion of organisms isolated from the nasopharyngeal swabs are shown in Figure 1. Swabs that were taken prior to medication yielded bacterial pathogens only – *M. haemolytica* and *P. multocida*. However subsequent samplings of control animals with BRD 7 and 14 days later revealed an evolving and more diverse biota with the addition of several other pathogens, including *H. somni*, *Arcanobacterium pyogenes*, *Staphylococcus* spp,

## Mycoplasma spp. Respiratory Syncytial Virus (RSV) and Bovine Virus Diarrhoea (BVD) virus.

Figure 1. Subtrial 1. Pathogens isolated from naso-pharyngeal swabs Days 0 (pre-treatment), 7 & 14 (untreated controls with clinical BRD).



*M. haemolytica* = *Mannheimia haemolytica*

*P. multocida* = *Pasteurella multocida*

*H. somni* = *Histophilus somni*

*A. pyogenes* = *Arcanobacterium pyogenes*

BVD = *Bovine Virus Diarrhoea*

RSV = *Respiratory Syncytial Virus*

### 4.6.1.2 Clinical observations

Morbidity in the untreated control group (34%) was significantly ( $P < 0.0001$ ) different from that in the gamithromycin group (5%), but the percentage of problem animals in both groups was low (1.6% and 0.8% for the control and treated groups respectively) and not significantly different. The incidence pattern for BRD over the 14-day observation period is shown in Figure 2.

### 4.6.1.3 Growth

There was a significant ( $P = 0.0001$ ) difference between the daily growth rate of the control group, 1.08 kg, and the treated group, 1.83 kg, over the first 30 days of the study.

Figure 2. Subtrial 1. Pattern of BRD morbidity Days 0-14.

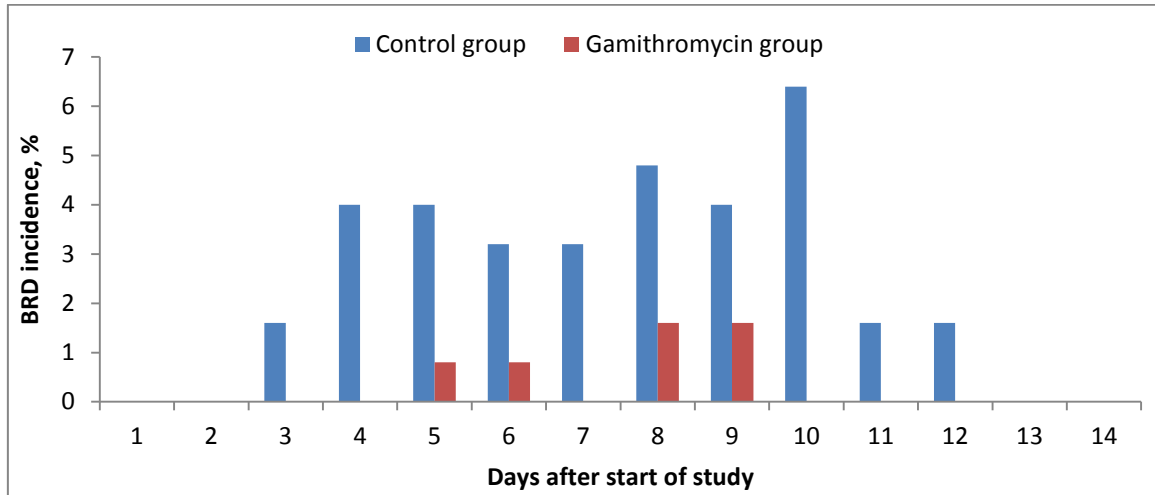


Figure 3. Subtrial 2. Pattern of BRD morbidity Days 0-14

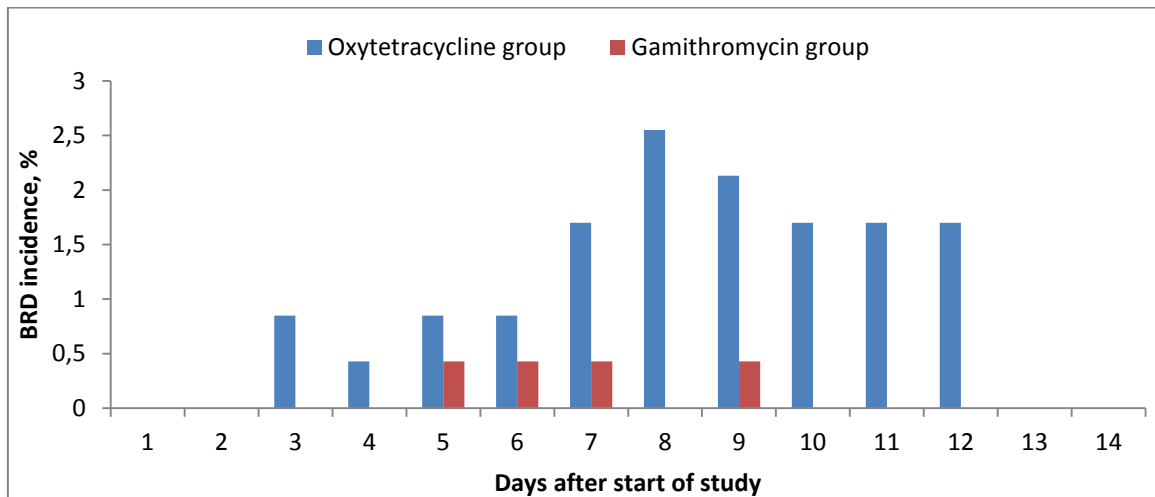
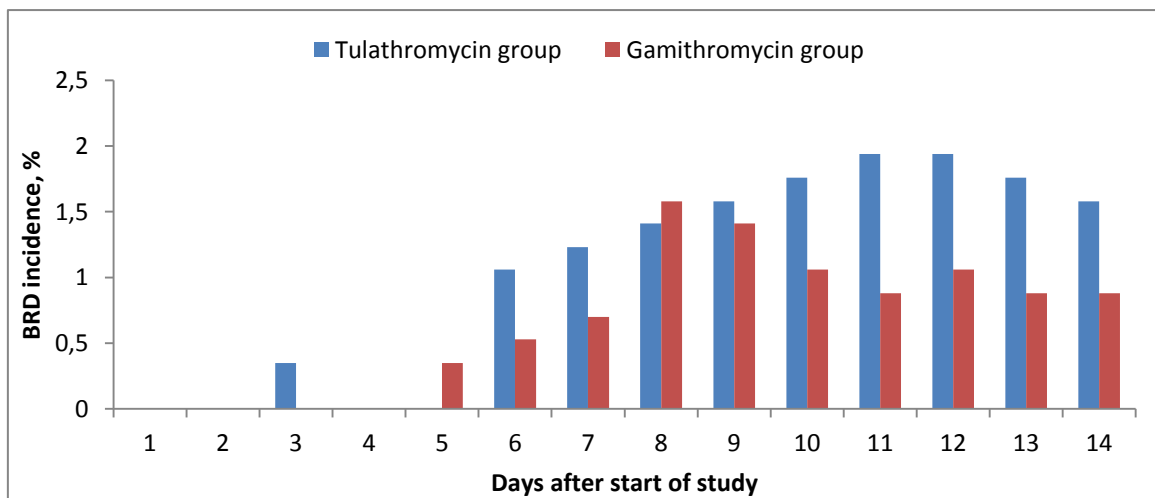


Figure 4. Subtrial 3. Pattern of BRD morbidity Days 0-14



## 4.6.2 *Subtrial 2*

### 4.6.2.1 *Clinical observations*

Morbidity in the oxytetracycline group of animals (15%) was significantly ( $P < 0.0001$ ) different from that in the gamithromycin group (2%), and there was also a significant ( $P < 0.05$ ) difference in the percentage of problem animals (4.8% and 1.6% for the oxytetracycline and gamithromycin groups respectively). The incidence pattern for BRD over the 14-day observation period is shown in Figure 3.

## 4.6.3 *Subtrial 3*

### 4.6.3.1 *Clinical observations*

Morbidity in the tulathromycin group (14%) was significantly ( $P < 0.05$ ) different from that in the gamithromycin group (9%). The percentage of problem animals and mortality in both groups was low (1.6% and 0.9%; 0.6% and 0.4% for the tulathromycin and gamithromycin groups respectively) and not significantly different. The incidence pattern for BRD over the 14-day observation period is shown in Figure 4.

### 4.6.3.2 *Growth*

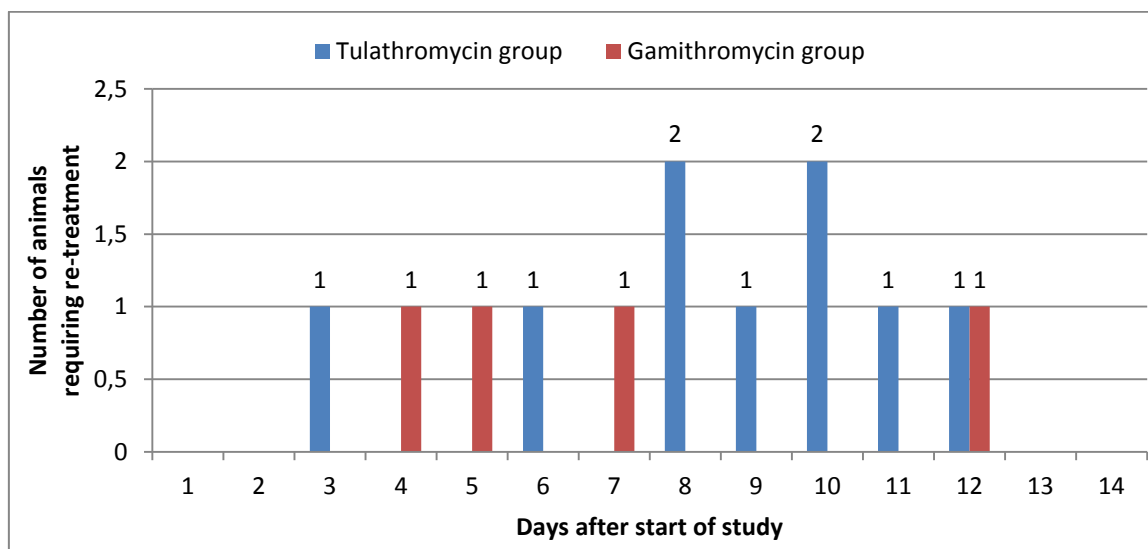
There was no significant difference between the daily growth rate over the first 30 days of the study of the two groups (1.03 kg and 1.10 kg for the tulathromycin and gamithromycin groups respectively).

## 4.6.4 *Subtrial 4*

### 4.6.4.1 *Clinical observations*

Following the initial therapeutic treatment on Day 0, the proportion of heifers that required a 2nd treatment over the subsequent 14 days differed significantly ( $P = 0.004$ ) between the tulathromycin group (82%) and the gamithromycin group (31%). The percentage of problem animals also differed significantly ( $P = 0.04$ ) between the groups (28% and 0% for the tulathromycin and gamithromycin groups respectively). The incidence pattern of the animals requiring a 2nd treatment for BRD over the 14-day observation period is shown in Figure 5.

Figure 5. Subtrial 4. Pattern of re-treatment for BRD Days 0-14.



## 4.7 Discussions

The complexity of BRD, particularly in terms of the range of pathogens that may be present and the severity of lung pathology, means that the outcomes of antibiotic treatment can be correspondingly variable. Examples from the scientific literature indicates a range of responses from <50% to >90% success in both therapeutic and prevention studies (Galyean et al., 1995; Catry et al., 2008; Wellman and O'Connor, 2007), irrespective of the antibiotic that was used, although within studies, there may be differences in efficacy between products. The performance of gamithromycin in the current studies falls within this range, but direct comparisons are not possible because of differences in protocol etc. For logistical reasons, it was not possible to continue the intensive clinical observations for more than 14 days after treatment in this series of studies and this could be seen as a short-coming when compared to studies that were conducted for longer. Nevertheless in subtrials 1 and 2, the morbidity had declined to zero by the end of the 14 days; in contrast, in subtrial 3, the outbreak of BRD continued up to Day 14. Nevertheless, the efficacy rates reported over a longer period, e.g. 60 days are generally reflective of results at 14 days, so the success rate over a 14-day period can be taken as indicative of longer term results (Godinho et al., 2005).

The microbiological evaluations in subtrial 1 showed that on arrival the majority of sampled cattle harboured *M. haemolytica* and/or *P. multocida* the

predominant organisms involved in the classical ‘transit’ or ‘shipping’ fever of cattle (DeRosa, 2000). Subsequently other bacteria were isolated and, most notably *Mycoplasma* spp became the most common pathogen. The other observation of note is the isolation of BVD virus on days 7 and 14, which strongly indicates the presence of individual cattle amongst the arrivals that were persistently infected with the virus. As BVD is known to result in general and local immunosuppression in BRD (Confer et al., 2005), its presence is likely to exacerbate the impact of the disease. The continued preventive efficacy of gamithromycin throughout this 14 day period during which the mix of pathogens changed supports its versatility as a BRD treatment.

#### **4.8 Conclusions**

It is important that antibiotics are used in a rational and controlled way in order to optimize their therapeutic effectiveness and to avoid unnecessary use and over-dependence (Anon, 2008; Barrett, 2000). Because feedlot cattle are frequently considered to be at high risk of BRD, mass preventive treatments before transport or on arrival are commonly used and are effective (Wellman and O’Connor, 2007; Duff, 2000). In addition, some studies have shown that selective treatments on arrival, based on body temperature, can provide equivalent levels of control to mass treatments (Galyean et al., 1995; Martin et al., 2007). The introduction of gamithromycin, with its pharmacokinetic and clinical profile of rapid therapeutic activity and prolonged preventive efficacy, offers additional opportunities for managing BRD on commercial farms.

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## **CHAPTER 5**

**Effects of administration of  
different plant extract on growth  
performance and *Eimeria* spp.  
infestation level of beef cattle**





## 5 Effects of administration of different plant extract on growth performance and *Eimeria* spp. infestation level of beef cattle

### 5.1 Abstract

Cattle usually do not show clinical signs of coccidiosis unless stressed by weaning, weather, shipping or other diseases. In any case the disease can compromise the animal homeostasis and nutritional up-take worsening the weight gain. Several natural substances have capability to improve physiological and health bred animal status and some of them have anti parasite properties. The trial was managed to verify the effects of some different commercial plant extracts on growth performance and coccidia infestation in 235 newly received Charolaise beef cattle, imported from France, during the adaptation period. This trial was divided in four subtrials to test four different plant extracts or mix of them (Subtrial 1: *Calendula officinalis*, *Castanea sativa*, *Plantago major*, *Silybum marianum*, *Trigonella foenum-graecum*; Subtrial 2: *Castanea sativa*, *Vitis vinifera*, *Citrus* spp, *yucca shidigera*; Subtrial 2 and 3: *Origanum vulgare*).

Plant extracts mix administered in the first subtrial shower to increase growth performance, reducing damages inducted by *Eimeria* coccidia acting as anticoccidial. In fact both number of animals infested and number of oocysts in feces were strongly decreased. The plant extract mix used in the second subtrial showed to decrease infested animals, but the same animals emitted with the feces a greater number of oocysts, denoting higher infestation and probably greater damages. Growing performances didn't show any difference.

The administration of *Origanum vulgare* didn't show any difference, denoting no effects on coccidia.

### 5.2 Introduction

#### 5.2.1 Coccidiosis

Coccidiosis is caused by a small, single celled parasite, called protozoa, that lives inside the cells of an infected animal's intestinal tract. These protozoa belong to subphylum Sporozoa, class Coccidia, family Eimeridae, and the most

represented belong to genus *Eimeria*, *Isospora*, *Cystoisospora* or *Cryptosporidium*. The most involved in bovine coccidiosis is *Eimeria*.

Coccidiosis is a serious disease in bovine, goats, sheep, pigs, poultry and rabbits. (Fayer, 1989; Foreyt et al., 1986; Gregory et al., 1987; Coudert, 1989).

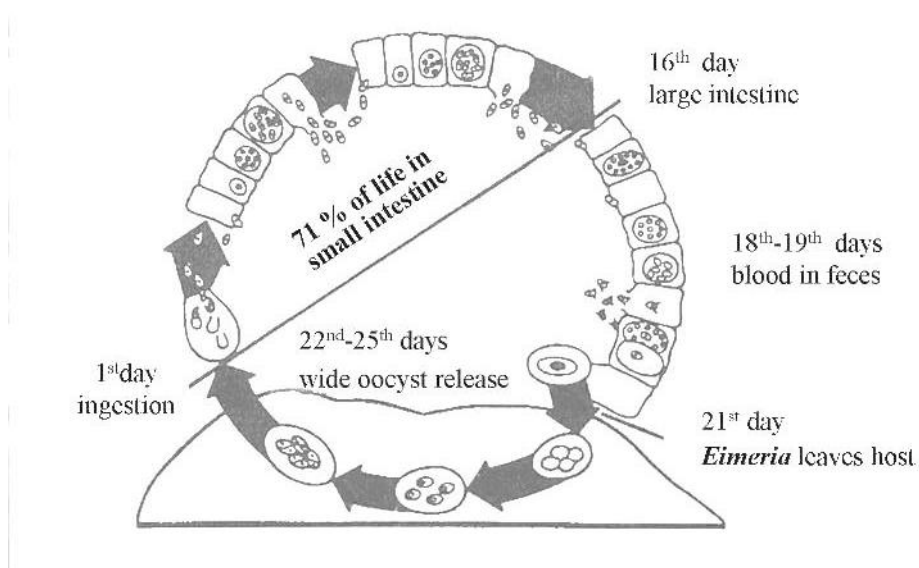
Infection with no clinical signs of disease is common. In clinical cases the disease may appear as a diarrhea and anemia due to the loss of red blood cells. In chronic cases a decrease in growth rates and an increase feed conversion rate (Fox, 1983; Fox, 1987; Laval and Remy, 1994). *Coccidia* cause a greater economic loss among domestic animals in temperate climates than any other protozoa.

Transmission of coccidiosis is fecal-oral and occurs when an animal consumes manure from an infected animal, usually due to manure contamination of the feed or water. They are commonly found in the intestinal cells, though they also attack the liver and other internal organs. The organism passes in the faeces as a freeliving form or oocysts. Once passed in the faeces, the oocyst develops into an infective form in the environment, is protected from adverse environmental conditions by a double cyst wall, until digested by a host animal. Development into an infective form can occur as soon as 5 to 10 days if it is in a moist environment with cool temperatures. *Coccidia* oocysts have survived as long as 2 years under favorable environmental conditions (Maas J.).

### *5.2.2 Coccidia life cycle*

The life cycle of *coccidia* is complex with both sexual and asexual stages in the intestines of cattle (figure 1) Cattle ingest the infective oocyst liberating an infective form called a sporozoite. This form penetrates the cells of the intestine, and goes through a cycle of rapid growth and reproduction known as the asexual phase. One infective oocyst can produce up to 900 asexual forms, each invading a cell in the intestine. The asexual phase is repeated several times during a 21 day cycle. Eventually the asexual form becomes a precursor of a sex cell that results in an oocyst that is passed in the faeces. Thus *coccidia* harm the host by destroying the cells and tissues in the lower intestines, cecum, and the colon. The loss of intestinal lining may lead to blood and fluid loss and may alter food absorption. Secondary bacterial invasion of the intestine may follow. *Coccidia* are extremely prolific, one ingested oocysts has is capable of producing 23,040,000 oocysts destroying an equal number of intestinal cells (Maas J.).

Figure 1. Coccidia life cycle



### 5.2.3 Bovine coccidiosis

Cattle usually do not show clinical signs of the disease unless stressed by weaning, weather, shipping or other diseases. Clinically apparent coccidiosis in cattle is deceptive. Clinical signs, if present are often not demonstrated until 3 weeks after initial infection. Oocyst cycle in other animals in the pen or feedlot, and significantly, most of the damage to the intestinal tract has already occurred. If the infection is mild, the most characteristic sign is foul smelling, dark, and watery faeces. Usually no blood is seen in these less severe infections. The animal may have a mild fever, but in most cases its temperature is normal or possibly below normal. Cattle become anorexic, depressed, dehydrated for loss of electrolytes and albumins and lose weight (Ferguson, 1996). Severely affected animals may develop a diarrhoea that is thin and bloody. Some cattle will pass formed faeces that contain streaks or clots of blood and shreds of mucus. The diarrhoea will usually last 3 to 4 days, but may continue for a week or more. The area around the anus and tail is often stained with blood and straining is common. The animals lose their appetite, become depressed and dehydrated, and lose weight. Cattle can also suffer a central nervous disorder from coccidiosis. Affected animals show muscular tremors, convulsions, and bending of the neck and head. Infected calves may die within 24 hours after the onset of dysentery and nervous signs, or they may live for several days, but usually are unable to rise. Nervous coccidiosis is difficult to treat and requires intensive veterinary care if the animal is to survive.

A clinical diagnosis is usually made when the characteristic coccidial symptoms, dysentery, straining, mild systemic involvement, and dehydration are present. Coccidian oocysts can be diagnosed by routine microscopic fecal examinations. The presents of fecal oocysts lags behind the onset of clinical signs.

#### *5.2.4 Control of coccidiosis*

Control of coccidiosis in cattle has been based on good management of premise sanitation, treatment of clinical cases as they appear, and especially the use of preventive medications. An infective dose of coccidia is necessary to produce signs of disease. Keeping the level of contamination down by sanitation and preventing contamination of feed and water we will have reduced the economic impact of the disease on the farm. The oocyst wall provides protection against chemical disinfectants. Coccidia are ubiquitous and unlikely to be destroyed in nature. Symptomatic treatment is necessary to stop the bacterial infections that accompany clinical coccidiosis. Treatment of clinical animals is not very rewarding, since clinical signs result from the final stage of parasite cycling in the host. The most acceptable method of control is prevention achieved by timely medication. The approved drugs for prevention of coccidiosis in cattle were Rumensin (Monensis sodium), and Decoquate, Decoquate is a chemotherapeutic feed additive that is effectively used as a preventative treatment in confined cattle. It can also be used as treatment to reduce the effects of an acute outbreak. The clinically affected animals should be treated with sulfa drugs, and then the coexistent cattle should receive Decoquate to prevent further cycling of the oocysts. The medication should be fed for 28 to 56 days or longer. Rumensin (monensin sodium) is growth-promoting feed additive that is also effective at preventing coccidiosis (Sgoifo Rossi et al., 2006).

#### *5.2.5 Plant extracts and phytotherapy*

Several natural substances have capability to improve physiological and health bred animal status and, for the reasons expressed before, there is a growing interest in some of these substances, particularly plant extract.

They are a group of substances that can be used as every other feeds.

In fact the European Union in the Council Directive 96/51/CE and Regulation 1804/99, about organic animal productions, indicated that plants and plant extracts are to simply included in the category of feed of vegetal origin usable in animal feeding, and are not regulated as therapeutic substances.

The term “phytotherapy” means prevention and treatment of pathologies with medicinal plant and plant extracts.

In the last years phytotherapy was especially revalued, following the discover of the effects of new plants and for some research that confirmed and clarified most of the pharmacologic activities, traditionally acquired in empiric way.

Phytotherapy isn't an alternative therapy, but it's part of orthodox pharmacology.

In fact plant extracts are not to be proposed like an unique alternative of synthetics medicines, because they can profitableness be combined with them, or synthetics medicines can complete a prevalently natural therapy.

Plant extracts, like synthetics medicines, act on men and animals organism for their chemical components.

In fact plants contain secondary metabolites that usually don't have nutritional properties, but showed to have several biological activities.

These metabolites can have more than one effects, and can operate on more than one metabolism at the same time.

Plant extracts components have synergic activities, which enhance the effects of just one substance, and the active compound generally have higher bioavailability than the original compound.

Generally these products don't have toxicity, and if they could be toxic, for example salicylic glycosides, they are less detrimental than synthetic molecule.

Plant extract showed to have several different activities, all of them to be considered really important in cattle breeding.

Different studies showed as different plant extract have antimicrobial effects, due to their metabolites (Smith-Palmer et al., 1998; Craig, 1999; Yam et al., 1997).

Also mixes of different plant extracts showed bactericidal or bacteriostatic activity on different bacteria strains, in in vivo or in vitro studies, and also exhibited beneficial effects on viral infections, for example against HIV virus (Murphy-Cowan, 1999).

Another aspect that makes these compound interesting in cattle breeding is the aspecific anti diarrhoea action that some plant extracts showed, in different animal species (Kyriakis et al., 1998; Tsinas et al., 1998a,b; Wu et al., 1998).

Besides, and even more interesting, the anti diarrhoea effects can be due to the anti parasite effect that some plant extracts showed (Morar et al., 1983; Omeke and Ezema, 2005).

### 5.3 Aim of the study

The trial was managed to verify the effects of some different commercial plant extracts on growth performance and coccidia infestation in 235 newly received Charolaise beef cattle, imported from France, during the adaptation period. This trial was divided in four subtrials to test four different plant extracts.

### 5.4 Materials and Methods

The trial was divided in four subtrials. In each subtrial we tested the efficacy of four different commercial plant extract on coccidiosis. In the following sections are reported data about trial length, animals, diets and treatment for each subtrial. Evaluated parameters are reported at the end of these sections.

#### 5.4.1 Trial duration

The subtrial 1 lasted 19 days since the arrival of the cattle in the farm from France. The subtrial 2 lasted 28 days, the subtrial 3 lasted 21 days and the subtrial 4 lasted 34 days.

#### 5.4.2 Animals and treatment

##### 5.4.2.1 Subtrial 1

The subjects of the trial were 94 Charolaise beef cattle, imported from France, averaging 373 ( $\pm$  26) kg, that were divided in two groups homogeneous for weight, age and conformation:

- Treated group (T): 47 beefs fed basal TMR diet and administered 90 g/head/day of plant extract mix, which was included in the concentrate.
- Control group (C): 47 beefs fed basal TMR diet.

The plant extract mix contained *Calendula officinalis*, *Castanea sativa*, *Plantago major*, *Silybum marianum*, *Trigonella foenum-graecum*.

##### 5.4.2.2 Subtrial 2

The subjects of the trial were 48 Charolaise beef cattle, imported from France, averaging 398 ( $\pm$  29) kg, that were divided in two groups homogeneous for weight, age and conformation:

- Treated group (T): 24 beefs fed basal TMR diet and administered 50 g/head/day of plant extract mix, which was included in the concentrate.
- Control group (C): 24 beefs fed basal TMR diet.

The plant extract mix contained *Castanea sativa*, *Vitis vinifera*, *Citrus* spp, *Yucca schidigera*.

#### 5.4.2.3 Subtrial 3

The subjects of the trial were 43 Charolaise beef cattle, imported from France, averaging 471 ( $\pm$  29) kg, that were divided in two groups homogeneous for weight, age and conformation:

- Treated group (T): 23 beefs fed basal TMR diet and administered 30 g/head/day of plant extract, that was included in the concentrate.
- Control group (C): 20 beefs fed basal TMR diet

The plant extract contained *Origanum vulgare*.

#### 5.4.2.4 Subtrial 4

The subjects of the trial were 50 Charolaise beef cattle, imported from France, averaging 453 ( $\pm$  34) kg, that were divided in two groups homogeneous for weight, age and conformation:

- Treated group (T): 25 beefs fed basal TMR diet and administered 40 g/head/day of plant extract, that was included in the concentrate.
- Control group (C): 25 beefs fed basal TMR diet

The plant extract contained *Origanum vulgare*.

### 5.4.3 Diets

During the experimental period animals were fed the following TMR diets reported in table 1, which were administered ad libitum.

### 5.4.4 Evaluated parameters

#### 5.4.4.1 Weight and Average Daily Gain (ADG)

Individual weight was registered at the discharge of cattle in the farm, day 0 of the subtrials, and at days of the end of each subtrial.

With these data Average Daily Gain for each animal was calculated.

Table 1. Diets (% as fed).

	Subtrial 1	Subtrial 2	Subtrial 3	Subtrial 4
Corn silage	34	48	39	47
Corn	24	21	-	22
Corn cracked	-	-	22	-
Soybean meal 44%	6	10	12	7
Beet pulp	20	8	17	6
Middlings	6	6	4	12
Straw	-	5	4	-
Distiller	8	-	-	4
MinVit	2	2	2	2
D.M, %	64.23	60,98	62.4	61.2
UFC/kg	0.90	0.91	0.92	0.91
C P, %	15.54	14,62	15.01	14.8
E.E, %	3.28	4,24	4.01	4.17
C F, %	14.37	13,84	14.2	14.01
NDF, %	34.34	28,58	32.5	30.5
NSC, %	40.68	39.3	39.8	39.4
Ca, %	0.81	0,72	0.77	0.74
P, %	0.42	0,46	0.44	0.46

#### 5.4.4.2 *Coccidiosis evaluation*

Parasite surveying was effected with copromicroscopic examinations.

The first day of all the subtrials and on the final day feces samples were collected, per each animal, directly from rectal ampulla.

Feces sample analysis was done using flotation method; 3 grams of each sample was put in a mortar with 42 ml of saturated solution of NaCl in water, and accurately diluted. The solution was filtered in a sieve and placed in McMaster slide.

It was observed with an optical microscope with 10X magnification, and parasite oocysts were counted, and with the proper formula, transformed in number/g of feces.

#### 5.4.4.3 *Health status*

During the experimental period pathology and therapeutic actions were registered.



## 5.5 Statistical analysis

Statistical analyses were conducted using the GLM procedures of SAS (SAS Inst. Inc). Initial weights, infested animals and number of oocysts were used as covariate.

## 5.6 Results and Discussions

### 5.6.1 Subtrial 1

#### 5.6.1.1 Average Daily Gain (ADG)

Data about ADG between the two groups, the Treated group receiving a mix of plant extracts and the Control group, are showed in table 2.

Table 2. ADG during the treatment period (kg).

	<b>ADG 0-19</b>
Treated	1.65
Control	1.36
<i>P</i>	<i>0.0347</i>

It appears that treatment with plant extracts increased ADG during the adaptation period.

#### 5.6.1.2 Coccidiosis evaluation

Data about percentage of beef cattle infested with coccidia, and about the number of oocysts that beef released with the feces are reported in tables 3 and 4.

Table 3. Percentage of infested bovine

	<b>day 0</b>	<b>day 19</b>	<b><i>P</i></b>
Treated	100	15	<i>0.0087</i>
Control	89	85	<i>0.1174</i>
<i>P</i>	<i>0.2434</i>	<i>0.0087</i>	

Table 4. Number of oocysts released with feces (n/g of feces).

	<b>day 0</b>	<b>day 19</b>	<b><i>P</i></b>
Treated	307	33	<i>0.0332</i>
Control	284	225	<i>0.1986</i>
<i>P</i>	<i>0.1185</i>	<i>0.0475</i>	

Analysing these data it's possible to highlight that both number of animal infested and the number of oocysts disseminated with feces by these animals, are statistically lower in treated group than in control cattle.

Fox (1983), Fox (1987) and Laval and Remy (1994) showed in their study how coccidiosis, also chronic, can reduce ADG and FCR; the anticoccidic effect of the plant extract used in this trial, due to a probable protection of intestinal mucosa, could explain also higher ADG registered in treated animal.

### 5.6.1.3 Health status

During experimental period no pathology were detected in the two groups.

## 5.6.2 Subtrial 2

### 5.6.2.1 Average Daily Gain (ADG)

In table 5 are reported data about ADG between the two groups.

Table 5. ADG during the treatment period (kg).

	<b>ADG 0-28</b>
Treated	1.32
Control	1.31

During the first 20 days of breeding animals registered the same gain of weight. No differences were also noticed in DMI. Also live weight didn't show any difference.

### 5.6.2.2 Coccidiosis evaluation

Coccidia infested bovine percentage and oocysts released gave interesting results (tables 6 and 7).

Table 6. Percentage of infested bovine

	<b>day 0</b>	<b>day 28</b>	<b>P</b>
Treated	50	18	0.0382
Control	50	34	0.4388

Table 7. Number of oocysts released with feces (n/g of feces).

	<b>day 0</b>	<b>day 28</b>	<b>P</b>
Treated	350	100	0.0929
Control	235	25	0.0793
<i>P</i>	0.3543	0.0833	

It's amazing to highlight that number of animal infested was statistically lower in treated animals, but only compared to the same group on day 0, not compared with control group animal.

Number of oocysts in treated group decreased significantly, but was statistically higher than number of oocysts released by control group on day 28.

So plant extract mix used in this trial seems to reduce number of infested animals, but these bovine produce more oocysts than the control.

### 5.6.2.3 Health status

During experimental period no pathology were detected in the two groups.

### 5.6.3 Subtrial 3

#### 5.6.3.1 Average Daily Gain (ADG)

In table 8 are reported data about ADG between the two groups.

Table 8. ADG during the treatment period (kg).

	<b>ADG 0-21</b>
Treated	1.14
Control	1.04

During the experimental period administration of *Origanum* showed to increase ADG in treated group but only numerically, not statistically.

#### 5.6.3.2 Coccidiosis evaluation

In tables 9 and 10, are reported data about infestation percentage and numbers of oocysts in feces.

Table 9. Percentage of infested bovine.

	<b>day 0</b>	<b>day 21</b>
Treated	65	39
Control	84	75

Table 10. Number of oocysts released with feces (n/g of feces).

	<b>day 0</b>	<b>day 21</b>
Treated	86	141
Control	185	170

There were no statistically significant differences between treated and control group, both in infested bovine percentage and in number of oocysts in feces.

There was a greater decrease of infected bovine in treated group, but this difference was only numerical. Besides percentage of infected animal in treated group at day 21 was close to 40 %, which is unacceptable. As reported in the most representative literature (Ferguson, 1996; Fox, 1983; Fox, 1987; Laval and Remy, 1994) also chronic coccidiosis cause depression of animals, weight loss and decreased growth performance, and such a number of infected animal is too dangerous.

Difference in number of oocysts showed not only to be not significant, but even an increase of oocysts in treated group from day 0 to day 21.

#### 5.6.3.3 Health status

During the experimental period three animals needed to be treated for clinical coccidiosis, two of them were treated group animal; this situation confirm that the treatment used in this trial were not effective as anticoccidial.

#### 5.6.4 Subtrial 4

##### 5.6.4.1 Average Daily Gain (ADG)

ADG between the two groups didn't show statistical difference, despite the numerical difference between the two groups; in fact animals treated with *Origanum* growth an average of 160 g more than control animals (table 11)

Table 11. ADG during the treatment period (kg).

	<b>ADG 0-34</b>
Treated	1.77
Control	1.51

##### 5.6.4.2 Coccidiosis evaluation

In tables 12 and 13 are reported infestation percentage and numbers of oocysts in feces.

Table 12. Percentage of infested bovine.

	<b>day 0</b>	<b>day 34</b>
Treated	52	24
Control	47	63

Table 13. Number of oocysts released with feces, n/g of feces

	<b>day 0</b>	<b>day 34</b>
Treated	258	253
Control	125	84

Analyzing data about number of infested animals it's possible to highlight that in this trial, like subtrial 2, infested animals treated with *Origanum* decreased of about 50%, but this decrease was not significant.

Number of oocysts remained unvaried during the experimental period, confirming ineffectiveness of this plant extract in coccidial oocysts control.

#### 5.6.4.3 *Health status*

During experimental period no pathology were detected in the two groups.

## 5.7 Conclusions

These four subtrial confirmed that in Italian beef cattle rearing there is a worrying incidence of animal infested with coccidia.

This infestation, in the past unconsciously controlled with Monensin, it's particularly hard to be defeated, but there are some plant extracts that can be useful.

In fact plant extracts mix administered in the first subtrial shower to increase growth performance, reducing damages inducted by *Eimeria* coccidia acting as anticoccidial. In fact both number of animals infested and number of oocysts in feces were strongly decreased.

The plant extract mix used in the second subtrial showed to decrease infested animals, but the same animals emitted with the feces a greater number of oocysts, denoting higher infestation and probably greater damages. Growing performances didn't show any difference.

Plant extracts used for the third and fourth subtrial didn't show any difference, denoting no effects on coccidia.

Any substance used as anticoccidic should have as target the almost complete elimination of these parasites, and not just a numerical reduction. For example reduce infested animals to 50 % isn't enough, because these animals, as reported by Fox (1983), Fox (1987) and Laval and Remy (1994), will have a decrease in growth rates and an increase feed conversion rate, with a loss of farm profit and a reduced animal welfare.

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# **CHAPTER 6**

## **General Discussion**



## 6 General discussion

The present doctoral study clearly showed that BRD is one of the most important diseases in the Italian beef cattle rearing. Because of its consequences on sanitary costs and animal growth performance, farmers and veterinarians have to consider some characteristics of imported animals like weight at arrival, sex and breed, and not only the length and quality of transport or other stressful events before that. Those individual characteristics can play a significant role in the BRD etiopathogenesis causing increment in morbidity, mortality and in severity of clinical signs. Furthermore, those aspects shall be carefully considered with the aim to adopt specific nutritional strategies and correct sanitary protocols in new arrival beef cattle and to prevent and/or to treat the sick animals.

In “high risk situations”, the administration of long acting slow release antibiotics is an effective strategy to reduce the BRD incidence with proportional effects to the characteristics of drug used. The same considerations could be applied even to the use of antibiotic for the treatment of BRD.

In addition, the study highlighted how even the presence of coccidia could play an important role on animal health balance increasing the susceptibility to BRD.

In the matter of this, particular attention should be paid to the effectiveness of coccidiosis preventive treatment especially if it is performed with natural extract. Indeed, as reported in the third trial of this study, not every plant extract or mix are actually able to contain or prevent the coccidiosis in ruminants.

During the investigation regarding the incidence and type of health problem has been also revealed the crucial role of proper nutritional management in order to reduce the incidence of diseases such as lameness, meteorism, enterotoxaemia, peritonitis or urolithiasis. Furthermore, the study showed that must not be forget the importance of the correct animal's groups creation, the characteristics of the rearing facilities and human-animal interaction in order to prevent arthropathy.



# CHAPTER 7

## Summary





## 7 Summary

Italian beef cattle's rearing is mainly based on fattening imported young animal from abroad. For animals, as occur also in the humans, the adaptation to new environment and social conditions gives rise to stress condition; this situation could drive the subject to severe physiologic and psychological reactions and compromise the health. To meet the consumer needs, at the present time farmer target is to obtain high quality meat as quick as possible in order to reduce the rearing cost and to improve the animal welfare. Therefore, nutritional level and sanitary condition must be the best to stimulate the greater growth than possible. Considering that, the italian typical beef cattle rearing needs to be based on diets characterized by high energy concentrations and on sanitary programs. The correct management of vaccination, antibiotic treatment and nutrition are important both in the fattening period and in the adaptation phase.

The objective of the present doctoral study was to evaluate the incidence and the severity of the health problems in the italian beef cattle rearing focusing the attention on Bovine Respiratory Disease and parasitosis. With this aim, an important part of the study was dedicated to evaluate the effectiveness of drugs administration to reduce the incidence and severity of BRD and to understand how specific plant extracts could reduce the parasitic infestation.

As mentioned, the adaptation phase is the first important critical point of the entire breeding process and any problem that occurs during this phase can compromise the entire rearing period. BRD, nutritional diseases and parasitosis cause a decrease in productive performances, an increase in pharmacological costs, technical and veterinary assistance, convenience culling, mortality, and consequently the length of breeding process and financial liabilities.

In the first study it has been enquired the incidence and the importance of the more important sanitary problems of the adaptation phase in imported beef cattle related with some animal's parameters. The enquired parameters were body weight, weight loss, incidence of pulmonary disease, incidence of locomotion disease due to traumatic and nutritional causes, incidence of animals moved in sick-bay pens due to pulmonary or locomotion diseases and finally mortality and the cause of it. To analyze the parameters listed above, the weight of the animals was divided in four classes (< 300 Kg, 300-380 Kg, 381-450 Kg and > 451 Kg), the weight loss was divided in four classes (< 2%, 2-3%, 3-7%,

and > 7%), the breeds was charolaise, limousine and crossbreed, and finally it has been considered the two sex.

The data collected showed that pulmonary disease is inversely proportional to the body weight while the locomotion system diseases are directly proportional to that. Subjects with a low weight loss (< 2%) had a high morbidity of pulmonary disease. Female have a morbidity of the pulmonary disease higher than the male subject, primarily because the female have a body weight lower than male and there are evidence that the body weight is directly related with the weight loss and the incidence of the problems. Conversely the males are more affected by locomotion disease, probably because those subject are more competitive than the female. Limousine breed is more affected by pulmonary disease than the others, but it's also to be considered that those animals arrive from France with a body weight lower than the other imported breeds. Charolaise breed manifested high rates of locomotion disease, both traumatic and nutritional, due to the higher body weight and the higher ruminal capacity that induce to speed up the adaptation program.

After the evaluation of the incidence of the main disease and the factors related, a series of subtrials were conducted in feedlots Italy to investigate the efficacy of a long acting and slow release antibiotic (gamithromycin) in the prevention and treatment of bovine respiratory disease (BRD) in newly arrived cattle. Three studies were conducted on its preventive efficacy when compared to either an untreated control, a long-acting oxytetracycline formulation or tulathromycin. The therapeutic responses to tulathromycin and gamithromycin were also compared in the therapeutic study. Preventive treatment with gamithromycin significantly reduced the morbidity due to BRD by 86%, 86% and 35% compared to the untreated control group, the oxytetracycline group and the tulathromycin group respectively. In the therapeutic trial, the number of animals that required re-treatment during the 14 days following the initial medication was significantly reduced in the gamithromycin group, compared to the positive control group. These results suggest that the dual therapeutic and preventive action of gamithromycin provides a valuable addition to the veterinarians' armamentarium for the medical management of BRD.

Other than BRD, parasitic infestation is considered one of the main problems related to an increase in morbidity and bad growing performance. Cattle usually do not show clinical signs of coccidiosis unless stressed by weaning, weather, shipping or other diseases. In any case the disease can

compromise the animal homeostasis and nutritional up-take worsening the weight gain. Several natural substances have capability to improve physiological and health bred animal status and some of them have anti parasite properties. The trial was managed to verify the effects of some different commercial plant extracts on growth performance and coccidia infestation in 235 newly received Charolaise beef cattle, imported from France, during the adaptation period. This trial was divided in four subtrials to test four different plant extracts or mix of them (Subtrial 1: *Calendula officinalis*, *Castanea sativa*, *Plantago major*, *Silybum marianum*, *Trigonella foenum-graecum*; Subtrial 2: *Castanea sativa*, *Vitis vinifera*, *Citrus* spp, *yucca shidigera*; Subtrial 3 and 4: *Origanum vulgare*).

Plant extract mix administered in the first subtrial showed to increase growth performance, reducing damages inducted by *Eimeria* coccidia acting as anticoccidial. In fact both number of animals infested and number of oocysts in feces were strongly decreased. The plant extract mix used in the second subtrial showed to decrease infested animals, but the same animals emitted with the feces a greater number of oocysts, denoting higher infestation and probably greater damages. Growing performances didn't show any difference. The administration of *Origanum vulgare* didn't show any difference, denoting no effects on coccidia.

In conclusion, the present doctoral study showed that, for a correct management of the adaptation phase, but even of the entire rearing period, it should necessary to consider some of newly received cattle's characteristics like weight at the arrival, sex and breed because are often connected with an increase of morbidity.

Furthermore, the study brings out the fact that it's also very important to choose the most appropriate protocol of vaccination and antibiotic treatment to reduce the incidence of infectious diseases and also to adopt specific nutritional strategies to promote a quickly reestablishment of the normal ruminal conditions after transport and to prevent coccidiosis.



# CHAPTER 8

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## 8 Acknowledgements

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