# Trypanotolerance and Phenotypic Characteristics of Four Ethiopian Cattle Breeds

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Cover: Bull from the Mizan-Teferi area in Ethiopia (photo: J. Stein)

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#### Abstract

Trypanosomosis is the single most important livestock disease in sub-Saharan Africa. Infected animals suffer from weight loss, anemia and miscarriages. Trypanotolerant breeds, however, can survive, produce and reproduce even under disease pressure. In Ethiopia some breeds live in infected areas and could possess trypanotolerance. The aim of this study was to compare the four Ethiopian cattle breeds Abigar, Gurage, Horro and Sheko in aspects related to trypanotolerance. These breeds are all kept in different tsetse infested parts of South-Western Ethiopia.

To learn about constraints for livestock production and about cattle diseases in particular, a survey was carried out among livestock keepers in the areas where the breeds are normally kept. Trypanosomosis was considered the most important disease in all areas. Sheko and Abigar had the best milk production according to the livestock keepers. The reproductive performance was the least favorable for Gurage. Blood samples were collected during the peak trypanosomosis challenge period of the year, to determine parasitaemia and Packed red Cell Volume (PCV) as indicators of trypanosomosis. Sheko cattle were least infected by trypanosomes (6%), compared to the other breeds (17-23%). Horro and Sheko had the best PCV. The number of trypanocidal treatments varied greatly, where Sheko had the fewest and Gurage the most (1 vs. 24 treatments per animal and year). The four breeds were also compared at a field experimental station in a tsetse infested area (Ghibe valley), where a total of 375 animals were kept. Abigar and Gurage showed poor reproductive characteristics compared to Horro and Sheko. The survival rates were best in Horro and Sheko. Sheko and Abigar had the highest PCV. Sheko had the fewest average number of trypanocidal treatments and the lowest infection rate, 9% compared to 21-26% for the other breeds.

Sheko cattle stand out as the most trypanotolerant animals; they rarely get infected by trypanosomes, and have good PCV, production and reproduction. A broader use of the Sheko breed in tsetse infested areas could improve animal health and household welfare. Immediate actions are needed to avoid extinction of this valuable breed.

Keywords: cattle, Ethiopia, trypanosomosis, trypanotolerance, phenotypic characteristics, Abigar, Gurage, Horro, Sheko

Author's address: Jennie Stein, SLU, Department of Animal Breeding and Genetics, P.O. Box 7023, 751 24 Uppsala, Sweden *E-mail:* Jennie.Stein@slu.se Pappa, jag önskar du vore här.

Maja, du är ljuset i mitt liv!

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# List of Publications

This thesis is based on the work contained in the following papers, referred to by Roman numerals in the text:

- I Stein, J., Ayalew, W., Rege, J.E.O., Mulatu, W., Malmfors, B., Dessie, T., Philipsson, J. (2009). Livestock keeper perception of four indigenous cattle breeds in tsetse infested areas of Ethiopia. *Tropical Animal Health and Production* 41(7), 1335-1346.
- II Stein, J., Ayalew, W., Rege, J.E.O., Mulatu, W., Lemecha, H., Tadesse, Y., Tekle, T., Philipsson, J. (2011). Trypanosomosis and phenotypic features of four indigenous cattle breeds in an Ethiopian field survey. *Veterinary Parasitology*. In Press. doi: 10.1016/j.vetpar.2010.12.025
- III Stein, J., Mulatu, W., Ayalew, W., Lemecha, H., Malmfors, B., Dessie, T., Rege, J.E.O., Philipsson, J. Production, reproduction and trypanotolerance in four Ethiopian cattle breeds kept on station in a tsetse infested area. Manuscript.

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# Abbreviations

AAT	Animal African Trypanosomosis
AFC	Age at First Calving
AI	Artificial Insemination
BCS	Body Condition Score
BCT	Buffy-coat technique
EIAR	Ethiopian Institute of Agricultural Research
HAT	Human African Trypanosomosis
HCT	Haematocrit Centrifugation Technique
ILRI	International Livestock Research Institute
NAHRC	National Animal Health Research Centre
PCV	Packed Red Cell Volume
SSA	Sub-Saharan Africa

# Introduction

Trypanosomosis is the single most important livestock disease in Sub-Saharan Africa (SSA) and is present in 37 countries in that region. More than one third of the land (~9 million km<sup>2</sup>) is infested by tsetse flies (Rogers & Robinson, 2004; Reid et al., 2000) and approximately 60 million heads of cattle are under disease threat for a direct costs of approximately 1.34 billion USD every year (Kristjanson et al., 1999). It has been estimated that Africa looses 3 million heads of cattle each year due to trypanosomosis (FAO, 1992). The direct losses caused by trypanosomosis in livestock include lowered production and reproduction, higher morbidity and mortality as well as direct costs for treatments and counter actions such as tsetse control. There are also indirect losses, that are perhaps more difficult to quantify, namely reduced or no animal populations in tsetse infested areas, as well as reduced crop production due to insufficient animal draft power. Consequently, based on all factors above, trypanosomosis causes poor food safety and a worse economy both for individual livestock keepers and for the countries or regions affected. In areas with trypanosomosis, the off-take of meat and milk is reduced by at least 50 % (Swallow, 1999).

Ethiopia is one of the countries suffering from trypanosomosis. The disease covers approximately 15 % of the total arable land, mainly in the west and southwest of the country. It is estimated that some 10 to 14 million heads of cattle in Ethiopia, and an equivalent number of small ruminants together with a significant number of equines and camels, are exposed to the risk of trypanosomosis. Additionally, trypanosomosis in the form of sleeping sickness is a large problem for humans.

As many control methods of the disease are failing, there is a need for alternative and sustainable solutions (Itty *et al.*, 1995; Peregrine *et al.*, 1994). One possible and potentially cost effective solution is to use the trypanotolerant breeds that can be found in different parts of Africa.

Trypanotolerant cattle are able to survive and reproduce even under trypanosomosis challenge (d'Ieteren *et al.*, 1998). A better use of such cattle breeds would greatly improve cattle production in many areas of SSA, through for instance higher productivity, higher survival rates and decreased cost of treatment. The most well known and investigated trypanotolerant breed is the taurine N'Dama from West Africa (Murray & Trail, 1984; Murray *et al.*, 1982). It is known for being able to live, produce and reproduce better than zebu cattle under trypanosomosis challenge.

The overall aim of this thesis is to clarify the variation in trypanotolerance among the four indigenous cattle breeds Abigar, Gurage, Horro and Sheko, in tsetse infested areas of Ethiopia. This is done by field studies in the home areas of the breeds as well as by a comparative study of the four breeds at a field experimental station in the tsetse infested Ghibe valley.

# General background

# Trypanosomes

*Trypanosoma* spp. are unicellular flagellate protozoa. All trypanosomes are heteroxenous which means they require more than one host to complete a life cycle. There are numerous different trypanosomes and the effect of an infection varies greatly based on the trypanosome specie. There are some species that hardly seem to be virulent at all, as for instance *T. theileri*, where effects only can be seen if the host is already sick. Other species are highly virulent and cause severe diseases in their mammalian hosts.

Trypanosomes cause a large number of diseases in different species of vertebrate hosts. Some trypanosomes can infect several vertebrate hosts whereas others are host-specific.

In humans there are two diseases that are caused by trypanosomes; Human African Trypanosomosis (HAT), also called sleeping sickness, which is caused by *T. brucei gambiense* or *T. brucei rhodesiense*, and Chagas disease in America which is caused by *T. cruzi*. It has been estimated that approximately 50-60 million people are at risk of contracting HAT (WHO, 1998; Kuzoe, 1991), and that there were at least 300,000 infected people in 1995.

Other examples of diseases caused by trypanosomes are Dourine in horses caused by *T. equiperdum*, and Surra in horses and camels caused by *T. evansi*.

Cattle are infected by the virulent *T. brucei brucei*, *T. congolense* and *T. vivax* as well as the avirulent *T. theileri*. Often *T. vivax* is found to be responsible for the first infections in cattle, whereas *T. congolense* often is considered to be the most virulent of the trypanosomes in cattle (d'Ieteren *et al.*, 1998).

The complete life cycle of the trypanosomes contains several phases both in the intermediate invertebrate host (the tsetse fly) and in the mammalian host (figure 1).



*Figure 1.* The main phases in the life cycle of the trypanosome, both in the intermediate host (tsetse fly) and in the mammalian host.

Recent molecular techniques lead to the discovery of new trypanosome species continuously (Adams *et al.*, 2010). At least 10 new variants of trypanosomes have been found, however only one has been classified as a new specie (McNamara *et al.*, 1994). Little has been done to investigate these new variants and it is not known how virulent they are, which hosts they have or how they respond to trypanocidal treatments (Adams *et al.*, 2010). The new variants of trypanosomes are often impossible to separate from well known species by solely looking at the morphology of the parasite; they can therefore pass undetected (Adams *et al.*, 2010).

# **Tsetse flies**

Tsetse flies are large biting flies that inhabit 37 countries in SSA and they inhabit an estimated  $8.7 \text{ million } \text{km}^2$  (Rogers & Robinson, 2004). A



characteristic of the fly is that one wing rests on top of the other when they are resting. All tsetse flies belong to the genus *Glossina* and they are normally also placed in their own family Glossinidae. There are currently 31 recognized species and subspecies of tsetse flies, and they are normally divided into three different subgroups; the savannah (subgenus *Morsitans*), forest (subgenus *Fusca*) and riverine type (subgenus *Palpalis*) (Solano *et al.*, 2010). As for the trypanosomes, different tsetse species are located in different areas of Africa.

The tsetse flies act as the intermediate vector for the trypanosome parasite and the parasite needs the fly to complete its life cycle. The fly ingests the parasite when taking a blood meal from an infected animal. After the parasite has gone through evolvement steps in the fly (figure 1), a new host will be infected with the parasite when the fly takes another blood meal. There is also a risk for mechanical transmission of trypanosomes from an infected animal to an uninfected one. In these cases a biting fly (tsetse fly or for instance a horse fly) bites an infected animal and within a short period of time, bites an uninfected animal. This is more common if the fly gets interrupted during the first blood meal, and soon after takes another meal.

# Trypanosomosis

Animal African Trypanosomosis (AAT), often also called Nagana, is, as the tsetse fly, spread over 37 countries in SSA. The disease in cattle is caused by *Trypanosoma congolense*, *Trypanosoma brucei* and *Trypanosoma vivax*. Consequences of the disease can be that affected areas are underutilized, either because they are inhabited by poorly producing animals, or because no animals can be kept there at all.

### Symptoms, effects and diagnosis

There are many different signs of trypanosomosis and also many different ways to diagnose it. There are also large differences regarding what ways of diagnosing that can be used in the field by individual livestock keepers and what is possible to do with extensive laboratory resources. Clinical signs of trypanosomosis are generally not regarded as being specific for the disease. Therefore, diagnoses that are made by the livestock keepers are not as precise and reliable as laboratory tests but on the other hand, under usual circumstances they are often the only option.

To clinically diagnose trypanosomosis there is no other option but to physically examine the animal directly. Clinical signs of acute trypanosomosis include anemia, weight loss, abortion of fetuses, roughness

of the hair, enlargement of peripheral lymph nodes, fever and reduced milk production. If the disease stays untreated the animal may also die (Eisler *et al.*, 2004).

Anemia is one of the most important effects of trypanosomosis, and it is highly linked to overall losses in production and reproduction (d'Ieteren *et al.*, 1998; Murray *et al.*, 1991; Trail *et al.*, 1991). The breakdown of red blood cells begins at infection, and progresses over the following weeks. The degree of anemia can be objectively quantified by measuring the haematocrit i.e. the Packed red Cell Volume (PCV), given as a percentage of the total blood volume (Figure 2).

Parasitological diagnosis includes several different techniques, such as wet blood film (thick or thin), buffy-coat technique (BCT), antibody and antigen detection (Ab and Ag) and polymerase chain reaction (PCR) (Eisler *et al.*, 2004). With only a little more equipment needed compared to the clinical diagnosis, thick or thin blood films have been used widely for diagnosis over a long time. A drop of blood can easily be examined in a microscope, to reveal parasitaemia. The blood can either be examined directly for parasitaemia alone, or fixated and Giemsa-stained in a laboratory where the trypanosome specie also can be determined. These methods with blood films are not sensitive enough to detect low levels of parasitaemia (Eisler *et al.*, 2004).

A more effective and reliable method of detecting parasitaemia is to concentrate the parasites before microscope detection. This can be done by centrifugation of the capillary tubes containing the blood samples, the so called haematocrit centrifugation technique (HCT) (Woo, 1970). After centrifugation the buffy-coat/plasma junction is located between the plasma and the red blood cells and contains white blood cells as well as the parasites (figure 2). In HCT capillary tubes were investigated in a microscope without any further steps. However, with BCT the capillary tubes are cut at the point of the buffy-coat to be able to extract and investigate it in a microscope (Murray *et al.*, 1977). The advantage of this technique is a higher concentration of parasites, which then are easier to detect. Another advantage of this technique is that it gives the possibility to measure PCV at the same time. The BCT was the method of choice for the parasitaemia evaluation of the studies in this thesis.



Figure 2. Schematic image of a blood filled capillary tube after centrifugation.

#### Control methods

There are a number of possible strategies and control methods for fighting trypanosomosis, both for targeting the parasite and the vector. So far, no single method has been successful on its own, and a range of methods are mostly practiced in the same area.

#### Tsetse control

Control methods can be aimed at reducing the number of tsetse flies as these serve as the intermediate vector. If the fly can be controlled, so can trypanosomosis. Some of the methods tried and used are fly traps, the sterile insect technique, insecticide spraying and pour-on of insecticide on livestock.

Fly traps often consist of urine baited conical traps or cloth targets, where, in both cases, the tsetse fly is attracted to the odor of cattle urine. The baits (trap or target) are generally impregnated with insecticides which kills the flies.

The use of insecticides has been used in a number of ways including ground and aerial spraying (by the use of aircrafts) and animal dipping or pour-on (Allsopp, 2001). When using animal dipping or pour-on, the insecticides are applied to the livestock. A drawback when using insecticides is the effect the compounds have on the environment and on mammals getting in contact with it. However, through research new compounds are developed that have less effect on the environment and less toxicity for mammals.

The sterile insect technique decreases the number of flies in an area by the release of large number of sterile male flies. The sterile males compete with the viral males and when a sterile male mates with a female fly, she will become sterile as well (Feldmann, 2004). Over generations the population will gradually decrease. An advantage of the sterile insect technique is the absence of environmental effects, apart from the effect on the tsetse population. However, lack of sustainability in sterile insect technique programs and reinvasion of flies from adjacent areas, have caused several trials to fail.

Trying to eradicate the tsetse fly has proven to be difficult and there are reports showing that in areas where the tsetse was eradicated a long time ago, they are once again present (Oluwafemi *et al.*, 2007). Reinvasion is inevitable unless complete populations of tsetse flies are eradicated or that the cleared areas stay protected (Hursey & Allsopp, 1982).

#### Trypanocidal treatments

There are currently three types of antibiotics used as trypanocidal treatments; isometamidium chloride, homidium (bromide and chloride) and diminazene aceturate (Holmes *et al.*, 2004). Isometamidium is used almost solely as a prophylactic drug and treatments can give protection against trypanosomosis for up to six months. Homidium can be used both for curative and preventive purposes but the most common is the curative. The third compound, diminazene, has only therapeutic effects. Approximately 35 million trypanocidal doses are administered in SSA every year, to a cost of about 30 million USD.

Over the years there has been a shift in who administers trypanocidal treatments. Previously local animal health representatives and veterinarians performed all trypanocidal treatments, but now the individual livestock keepers play a much larger role in the treatment of their cattle, which has made it possible for more animals to receive treatment. However, when livestock keepers take over the responsibility for trypanocidal treatments, it also includes some new risks. Livestock keepers tend to overuse trypanocidal treatments as they treat animals even if no correct diagnosis is performed. It has also been shown that livestock keepers have a strong tendency towards using curative treatments instead of the sometimes more appropriate preventive treatments (Van den Bossche *et al.*, 2000). If trypanocidal compounds are misused, over-dosed or under-dosed, there may be many different implications, for instance unnecessary high costs, drug resistance,

environmental impact and drug remnants in meat or milk that is used for human consumption.

There is an emerging problem with drug resistance among the trypanosomes (Holmes *et al.*, 2004; Afewerk *et al.*, 2000; Mulugeta *et al.*, 1997; Codjia *et al.*, 1993; Rowlands *et al.*, 1993). Drug resistance has been found to be higher in areas with a more intensive treatment practice.

# Vaccine

There has been extensive research trying to produce a vaccine against the trypanosomes, but so far it has not been successful due to a fast antigen change on the surface of the trypanosome (Black *et al.*, 2001; Teale, 1993). It has been estimated that the trypanosomes have more than one thousand different variations of antigens and only one at a time is expressed on their surface (Dempfle, 2009).

# Trypanotolerance

One definition of trypanotolerance is "the relative capacity of an animal to control the development of the parasites and to limit their pathological effects" (d'Ieteren *et al.*, 1998). It is also possible to define this trait as the characteristic of an animal that enables it to remain productive under tsetse-trypanosomosis challenge (Courtin *et al.*, 2008; Murray *et al.*, 2004). In other words, trypanotolerant animals can be defined as animals that do not get infected as often, and when infected, they survive, produce and reproduce better than susceptible animals. These definitions are all very wide and can possibly contain factors ranging from coat color of the animals that does, or does not, attract the tsetse fly, to acquired antibodies in the immune system targeting the trypanosomes.

It is commonly believed that trypanotolerance in cattle is a result of natural selection where cattle and trypanosomes have co-existed for a long time. Differences between breeds and breed groups are explained by the way cattle arrived to the African continent (Epstein, 1971). The first cattle to arrive were of *Bos taurus* type, whereas *Bos indicus* (zebu) cattle came later. Taurine cattle types in Africa are often characterized by a small body size and a relatively low productivity compared to most zebus. However, they have over the years acquired hardiness to harsh climates and different disease resistances among which trypanotolerance is the most notable one. Trypanotolerance can therefore most easily be found in taurine cattle breeds, whereas indicus breeds more often are trypano-susceptible (Murray *et al.*, 2004; Rege, 1999). The most well known and well documented

trypanotolerant breed is the N'Dama in West Africa (Paling & Dwinger, 1993; Murray *et al.*, 1982; Trail *et al.*, 1979). The idea that N'Dama, due to a small size, also had a low production has been widely spread (Mugambi, 2009), and livestock keepers has been reported to deliberately select more indicus than trypanotolerant taurine breeds partly based on milk production (Jabbar & Diedhiou, 2003).

The ability to control anemia is an important characteristic of trypanotolerant cattle and it is highly linked to the overall production and reproduction (d'Ieteren *et al.*, 1998; Murray *et al.*, 1991; Trail *et al.*, 1991). After the initial breakdown of red blood cells, leading to a lowered PCV, trypanotolerant cattle recover, whereas susceptible animals continue to have a lower PCV. The control of anemia in N'Dama was found not to be influenced by the acquired immune response (Naessens *et al.*, 2001).

Trypanotolerant animals, such as the N'Dama, are also known for a better response to trypanocidal treatments, and hence need less treatments (Rowlands & Teale, 1994).

However, even though trypanotolerant animals are able to survive and reproduce in tsetse and trypanosomosis infested areas, they may still be affected by the level of the challenge (ILCA, 1979). Thus, production and reproduction of tolerant animals could be increased by strategic control methods during high challenge periods (Murray *et al.*, 2004).

There are studies that show that not only *Bos taurus* breeds display levels of trypanotolerance. In a comparison between the three Bos Indicus breeds Maasai Zebu, Orma Boran and Galana Boran significant differences were found in their ability to prevail trypanosomosis challenge (Mwangi *et al.*, 1998; Njogu *et al.*, 1985). The Maasai Zebu and Orma Boran were less susceptible judged by lower trypanosomosis prevalence, higher PCV, need for fewer trypanocidal treatments, higher survival and higher growth rate then the Galana Boran cattle. Previously it was believed that only taurine breeds were trypanotolerant and the finding that also zebu cattle can possess levels of trypanotolerance is promising. By also being able to include zebu cattle in the quest for trypanotolerance, the chance of finding suitable breeds for all the different areas in SSA increases.

Trypanotolerance has also been investigated and found in other species of livestock such as small ruminants, pigs, horses and donkeys (Murray *et al.*, 2004) For instance, the Djallonke sheep in West Africa and the Red Maasai sheep (Murray *et al.*, 1982) in East Africa have shown trypanotolerant characteristics.

# Ethiopia

In 2010 Ethiopia had a human population of almost 87 million people, out of which nearly 72 million live in rural areas (FAO, 2011a). Almost 40 % of the population lives in poverty (WorldBank, 2011). The cattle population in Ethiopia is one of the largest in SSA with almost 51 million live cattle in 2010, after an increase by 12 million since 2005 (FAO, 2011a; FAO, 2011b).

In Ethiopia, approximately 15 % of all arable land is under tsetse and trypanosomosis challenge. Extensive research as well as trypanosomosis control programs has been carried out in different parts of the country and in the Ghibe valley in particular (Tadesse & Tsegaye, 2010; Kebede & Animut, 2009; Codjia *et al.*, 1993; Leak *et al.*, 1993; Rowlands *et al.*, 1993). However, despite many years of research trypanosomosis is still a major problem in many areas of Ethiopia.

#### The Ghibe valley

The Ghibe valley, located 230 km southwest of Addis Abeba, is part of the Great Rift valley and it is an approximately 30 km long gorge following the Ghibe River. Up until the mid 1980-ies the Ghibe valley was noninhabited due to the large problems with tsetse flies in the area. After a large human re-settlement scheme decided upon by the Ethiopian government and the start of an extensive tsetse eradication program, new settlers started to arrive. After a fast settlement of people and livestock there were over 10,000 farming families and over 25,000 heads of cattle in the area (Kemp, 2006; Ayalew & Mulatu, 2004; Reid et al., 2000). However, trypanosomosis was still a constraint for keeping cattle in the area, and there have since been a number of research projects and tsetse eradication projects in the valley. The International Livestock Research Institute (ILRI) and the National Animal Health Research Center (NAHRC) of the Ethiopian Institute of Agricultural Research (EIAR) have also guided livestock keepers into forming cooperatives to more easily purchase and administer trypanocidal drugs and pour-on (Kemp, 2006). The use of pour-on in the area reduced trypanosomosis with 63 % and the number of needed trypanocidal treatments with 50 % (Ayalew & Mulatu, 2004).

In the year 2000, ILRI and EIAR started a breed comparison project located in the Ghibe valley (Lemecha *et al.*, 2006). The breeds compared were Abigar, Gurage, Horro and Sheko. The aim of the project was to investigate whether or not trypanotolerant cattle breeds exists in Ethiopia.

# Aims of the thesis

The ultimate aim of this study was to increase the possibility to improve household welfare in tsetse infested areas in South-Western Ethiopia, by investigating the possibility of a better and more effective use of trypanotolerant cattle. To achieve this, the studies and thesis research were designed to:

- investigate livestock keeping practices in four different tsetse infested areas, including aspects on management, herding, production and breeding.

- acquire a better understanding of livestock keeper's knowledge and perceptions regarding constraints for livestock production, diseases and trypanosomosis in particular.

- objectively determine the levels of trypanosomosis and PCV among the four Ethiopian cattle breeds in their respective home areas.

- compare the four Ethiopian cattle breeds regarding production, reproduction, survival, and measures of trypanotolerance such as parasitaemia and PCV, when kept jointly in the same tsetse infested area to avoid confounding between environmental effects and breed effects.

- advice on the possible use of a breed or breeds that show a high degree of trypanotolerance and yet have acceptable production characteristics.

If trypanotolerant breeds are identified, multiplied and spread in tsetse infested areas, this could greatly improve cattle health and productivity, and subsequently improve the household welfare for people living in tsetse infested areas.

# Summary of studies

The four cattle breeds studied were Abigar, Gurage, Horro and Sheko. The breeds are kept in different parts of South-Western Ethiopia (figure 3), which are all subject to tsetse and trypanosomosis infestation.



*Figure 3.* The four areas in Ethiopa where the breeds Abigar, Gurage, Horro and Sheko are normally kept. The Ghibe/Tolley experimental station was the location for the comparative trypanotolerance study, where all breeds were held in the same tsetse infested environment (published with permission from Lemecha *et al.* (2006))



Figure 4. Abigar cattle at the Ghibe/Tolley station. (photo: Jennie Stein)



Figure 5. Gurage cattle at the Ghibe/Tolley station. (photo: Jennie Stein)

# Abigar

The Abigar breed (figure 4) is from the Gambella region of western Ethiopia, close to the border of Sudan. The breed belongs to the Nilotic Sanga group. The Sanga cattle are believed to have evolved as a result of interbreeding between Longhorn-, Shorthorn- and zebu type cattle over 3000 years ago (Payne & Wilson, 1999). The Abigar breed has kept some characteristics of the traditional Sanga, such as the large horns and a relatively small hump. The estimated population size of the Abigar cattle is close to 550,000 (Rege, 1999). The main uses of Abigar cattle include milk and meat production and different types of work (DAGRIS, 2007).

# Gurage

The Gurage breed (figure 5) is found in the Gurage zone in close proximity to the tsetse infested Ghibe valley (DAGRIS, 2007). The breed is of the Small East African Zebu type and belongs to the sub-group Abyssinian Shorthorned Zebu (Rege, 1999). Zebu cattle are trypano-susceptible but in areas with absence of tsetse flies and trypanosomes, they can compete with trypanotolerant breeds regarding production and reproduction. The main uses are for draught and milk production but due to tsetse infestation and poor pastures they are perceived not to perform well in their natural home area (Rege & Tawah, 1999). There is an estimated number of 583,000 cattle in the Gurage Zone (Gurage Zone Government, 2008).



Figure 6. Horro cattle at the Ghibe/Tolley station. (photo: Jennie Stein)



Figure 7. Sheko cattle at the Ghibe/Tolley station. (photo: Jennie Stein)



# Horro

The Horro breed (figure 6) is a Zenga type of cattle, i.e. an intermediate breed type between the Sanga and the Zebu (DAGRIS, 2007; Rege, 1999). Zenga breeds are often found in areas between the typical Zebu areas in northern Africa and Sanga areas in the south. The Horro cattle are found in the western parts of Ethiopia, mainly in the Eastern Wollega and Western Shoa Zones of the Oromia Region. The estimated total population size reaches almost 3.3 million animals (Rege, 1999) and are thus one of the more common breeds in Ethiopia. Horro cattle are mainly used for milk and meat production and for draught purposes (Rege & Tawah, 1999).

# Sheko

The Sheko breed (figure 7) classifies as a humpless Shorthorn and is the only known breed of taurine type in eastern Africa (Rege, 1999). The breed is found in the Bench-Maji zone of Southern Region in the south-western parts of Ethiopia. Sheko cattle are mainly used for meat production and work (DAGRIS, 2007). Early information claims that the Sheko possess trypanotolerant attributes (Alberro & Haile-Mariam, 1982; Epstein, 1971) and later research has supported this (Taye *et al.*, 2007; Lemecha *et al.*, 2006).

The population number varies greatly depending on the time and source of information. Numbers range from as high as 31,000 (*DAD-IS*, 1999; Rege, 1999) to 4,040 animals in a field assessment (Taye *et al.*, 2007) and 2,400 animals according to a recent census (Dadi *et al.*, 2008). Sheko cattle are classified as endangered, because many livestock keepers are indiscriminately interbreeding them with zebu cattle (DAGRIS, 2007; Rege, 1999).

# Materials and Methods

#### Livestock keeper interviews

As a first part of this study it was considered important to learn from the livestock keepers what their perceptions about constraints for livestock production were and in particular the role of cattle diseases. A questionnaire was constructed and pre-tested before visiting livestock keepers in all the four breed areas. From each area 60 livestock keepers were randomly

selected and interviewed, resulting in a total of 240 interviews. The questionnaire covered issues regarding livestock production, reproduction, breeding practices, constraints for cattle production, diseases, with emphasis on trypanosomosis, as well as socio-economic characteristics. It was of particular interest to find out when the peak challenge of trypanosomosis occurred in each home area of the four breeds. All interviews were carried out in local language by assistance of enumerators. The questionnaire was semi-structured and answers resulted in either continuous numerical values or categorical responses.

## Bleeding Survey

From the first questionnaire survey it was found that, according to the livestock keepers, the trypanosomosis challenge varied during the year and between the four breed areas. At the time of the year when trypanosomosis challenge peaked in an area, bleeding and measuring of the animals took place. The purpose was to get objective measures of several indicators of trypanosomosis of the four breeds in their home areas. Samplings took place in April for Gurage, in May for Sheko and in August for the Horro breed. The Abigar breed was supposed to be sampled during September-October but due to civil unrest and flooding of the area the sampling was postponed until May the following year. Consequently, a second round of sampling was carried out in the Abigar breed in October to ensure data from the peak challenge period. Results from the samplings during the high challenge period are referred to as "Abigar", whereas results from the low challenge period are referred to as "Abigar(II)".

A group of livestock keepers in each selected area were invited to gather with their cattle at a suitable site for blood sampling. Whenever it was possible local Animal Health representatives were invited to attend, and to take the opportunity to treat animals in need. Approximately 100 animals of each breed were blood sampled from the ear vein into heparinized capillary tubes. The tubes were centrifuged for 5 minutes for immediate determination of PCV and parasiteamia. Three body measurements (body length, heart girth and height at withers) were noted for each animal using a plastic coated textile measuring tape. Additionally, each animal received a subjective Body Condition Score (BCS) between 1 and 5, where the value 1 characterized a poorly developed, thin animal and 5 characterized an animal in very good condition.

At the time of sampling the livestock keepers provided additional information about the animal regarding age, current health status and previous trypanocidal treatments.

## **Ghibe/Tolley Station**

The last steps of the study aimed at objectively compare the four breeds when they were kept in the same tsetse infested environment, thus excluding confounding effects with the environment. The Ghibe/Tolley station is located on the eastern bank of the Ghibe River about 230 km southwest of Addis Abeba (figure 3). The Ghibe valley is located in a major tsetse infested area with large number of *G. pallidipes*, *G. fuscipes* and *G. morsitans submorsitans*. The area has a sub-humid climate with a moderately high temperature. The rainy season occurs between June and September (Leak *et al.*, 1993).

Animals from all four breeds were selected, purchased and brought to the Ghibe valley in the year 2000, as part of a large research project led by ILRI and EIAR. After two months of quarantine the animals were brought to the Ghibe/Tolley research station. The number of purchased animals as well as the number of calves born at the station until May 2007 is shown in table 1. A total of 375 animals were included in the study. Approximately 2000 ha of the surrounding land were used for grazing and for harvesting hay. (Lemecha *et al.*, 2006)

Table 1. Herd structure (number of purchased animals and calves born at the Ghibe experimental station 2000-2007).

	Purchased animals		Calves born	n at station		
Breed	Female	Male	Female	Male	Total number	
Abigar	64	7	4	5	80	
Gurage	41	5	12	6	64	
Horro	49	5	22	38	114	
Sheko	62	5	23	27	117	
Total number	216	22	61	76	375	

Heifers, cows and calves were housed in four separate sheds, one for each breed. Bulls of all breeds were kept in a joint shed. During daytime all females were herded jointly and separated from the bulls. Matings occurred during evenings or early mornings when selected bulls were brought to the cow shed. Calves born at the station were kept at the station when adult animals were out grazing. Feeding consisted of natural grazing in the surrounding areas and a supplement of hay at the sheds. Concentrates (wheat bran) were given to calves, lactating cows and sick animals. Water for all animals was supplied at the adjacent Ghibe River. Recordings on all animals were carried out monthly, including live weight, trypanosomosis infection, PCV and occurrence of other diseases, as well as medical treatments.

To determine parasitaemia and PCV, all animals were blood-sampled from the ear-vein into heparinized capillary tubes. The buffy-coat technique was used to determine parasitaemia (Murray *et al.*, 1977). Also, for animals with a PCV below 20 %, thin blood smears were prepared to search for other haemoparasites such as *Anaplasma*, *Babesia* and *Theleria*. Animals showing a PCV of 20 % or below were treated with Berenil® (3.5 mg/kg body weight) either if they were found to be parasitaemic or if no parasites could be detected but the animals showed clinical signs of trypanosomosis.

Weight of the animal was recorded monthly by the use of a weighing bridge and a digital reader. Additionally, all calves were weighed at birth.

### Statistical analyses

The statistical analyses were carried out using the SAS software (SAS, 2009). All data were edited and cleaned from unrealistic figures. Numerical values were analyzed using a fixed linear model always including the effect of area/breed, whereas categorical data were analyzed using Chi-square.

## Main findings

#### Trypanosomosis

Almost all livestock keepers in the four areas, where the breeds are normally kept, considered trypanosomosis to be a major problem and 59 % of them ranked it as the number one disease (Paper I-II). The disease was considered to be one of the top four diseases by over 90 % of the livestock keepers. However, Abigar and Sheko livestock keepers did not find trypanosomosis to be as big a problem as Gurage and Horro keepers did.

There was a knowledge gap among the livestock keepers concerning the cause of disease and the transmission of trypanosomosis. Some livestock keepers correctly correlated the disease with the tsetse fly, while others believed that the disease occurred due to bad weather, animal urine or animal to animal contact (Paper II). The main reported symptoms of the disease were the characteristic rough hair of the animals and emaciation. Among Abigar, Gurage and Horro livestock keepers there was also a large knowledge gap concerning the trait trypanotolerance. It is worth noting, however, that even though the knowledge of trypanosomosis was low, a vast majority of the livestock keepers were interested in purchasing



trypanotolerant animals or even replace the complete herd with such animals. As many livestock keepers generally tend to stick to the breed they already have, this highlights the severity of the trypanosomosis disease even more.

As the pressure of trypanosomes fluctuates throughout the year, it was important to learn when the main tsetse and trypanosomosis challenge period occurred in each of the four areas. According to the livestock keepers the problems concerning trypanosomosis peaked in April for Gurage, in May for Sheko, in June for Horro and in September–October for the Abigar breed (figure 8).



*Figure 8.* Seasonal variation in tsetse and trypanosomosis challenge as perceived by the livestock keepers in the different breed areas.

#### Trypanosomosis infection rate and treatments

The trypanosomosis infection rate was measured both in the home area with a onetime sampling of the animals during the high challenge period (Paper II), and monthly at the Ghibe experimental station during 2000-2007 (Paper III).

As seen in figure 9 the prevalence of trypanosomosis varied greatly between the breeds, where Sheko consistently had the lowest infection levels both in its home area and at the Ghibe station. Abigar and Gurage had the highest infection levels both in their respective home area and at the Ghibe station. In the figure it is also clearly shown that the Abigar, Gurage and Horro livestock keepers greatly overestimated the number of infected animals in their home areas as compared to the results from objective detection of trypanosomosis in blood samples (Paper II).

The difference in infection rate for Abigar between the high and low challenge season shows the importance of investigating trypanosomosis during the correct time of the year, unless records are collected continuously during the year.



*Figure 9.* Trypanosomosis prevalence in cattle (Paper II-III): *i*. as perceived by the livestock keepers in the respective home areas during the high trypanosomosis challenge period, *ii*. objectively detected from blood samples of animals in the home areas, and *iii*. overall detected trypanosomosis prevalence at the Ghibe experimental station measured from 2000 to 2007. Abigar(II) refers to the survey carried out in the home area during the low challenge period.

In general almost all livestock keepers treated their animals against trypanosomosis and many used traditional methods, especially in the Abigar and Gurage areas (Paper II). The number of trypanocidal treatments varied greatly between the breed areas. Sheko received the fewest number of treatments (1 treatment per animal and year), whereas Gurage in average got as many as 24 trypanocidal treatments per animal and year. Also at the Ghibe experimental station, Gurage needed the highest number of treatments, but the difference was much less pronounced (Paper III).

## Packed red Cell Volume

Even though PCV depends on the general health status of the animals and can be affected by many different reasons, it is a commonly accepted measure and used as an indicator of trypanosomosis infection. Consistently, and as expected, the PCV was lower among infected animals compared to non-infected animals, both in the home areas of the breeds and on the experimental station (Paper II-III). It was also found that for all breeds except Abigar the PCV was higher in their home areas during the trypanosomosis high challenge period, compared to the PCV obtained at the Ghibe experimental station (table 2). There was a re-ranking between the breeds for PCV on-station compared to the home areas; Horro and Sheko had the highest PCV in the home areas, whereas Sheko and Abigar had the highest PCV at the experimental station. In Paper III it was also shown that, over the year, PCV had a seasonal pattern of change, which was associated with changes of the animal live weights.

	Overall				Infected		Non-infected	
	No. of animals	No. of obs.	Mean	SD	Mean	SD	Mean	SD
In breed hom	ie areas							
Abigar	103		20.0	4.5	17.5	3.8	20.8	4.4
Abigar(II)	101		23.1	3.9	17.5	4.9	23.2	3.9
Gurage	100		22.7	4.8	21.2	4.8	23.1	4.7
Horro	93		26.2	4.7	21.3	4.6	27.2	4.1
Sheko	109		25.1	4.7	19.5	3.3	25.4	4.6
On experime	ental station							
Abigar	75	2861	24.1	4.2	22.4	4.0	24.6	4.2
Gurage	53	2096	22.5	4.6	20.3	4.2	23.7	4.5
Horro	102	4613	23.0	4.3	20.4	4.3	23.7	4.0
Sheko	111	4372	24.6	4.4	21.3	4.8	24.9	4.3

Table 2. Packed red Cell Volume of animals measured once in their normal home areas and animals measured monthly at the Ghibe experimental station between 2000 and 2007 (overall mean and SD).

#### Survival rate

According to the livestock keepers, more cattle had died due to trypanosomosis in the Abigar and Gurage area (12-13 %) compared to Horro (6 %) and Sheko (4 %) during the last year prior to our survey (Paper II). The same breed pattern was shown at the Ghibe experimental station (Paper III). Sheko animals born at the Ghibe station had the highest overall survival (78 %), followed by the Horro breed. Out of the purchased animals,

Horro had the highest overall survival (76 %), followed by the Sheko breed. Abigar and Gurage had the lowest survival rates at the Ghibe station, just like in their home areas.

## Reproduction

The age at first calving (AFC) was lower in the home areas for all breeds, except Gurage, compared to the results at the Ghibe experimental station (Paper I & III). In the survey on livestock keeper perceptions in the breed areas, Abigar had the best reproductive characteristics with the lowest AFC, shortest calving interval and the highest number of calves per cow. At the station, however, Abigar performed the worst and had the fewest total number of calves and the highest AFC. On the other hand, Horro, that performed intermediate in the home area, had the best reproductive characteristics at the experimental station. Sheko cows had, together with Abigar, the highest mean number of calves born (8.5-8.8) in the reproductive lifetime of a cow, compared to Horro and Gurage (5.6-5.7) (Paper I). At the Ghibe station, Horro and Sheko cows got the clearly highest number of calves. The pregnancy rate was overall very low in all the breeds at the station. The highest pregnancy rates were found among Horro (34 %) and Sheko (25 %) cows. Once again Abigar showed the least favorable results with only 5 % of the females being pregnant at any year (Paper III). Overall, the results show a better reproductive ability of the animals when kept in their home areas compared to the animals kept at the Ghibe station.

Concerning selection of animals in the breed home areas a majority of the livestock keepers said that they purposely selected animals for breeding. However, fewer controlled the actual mating. Less than half of the livestock keepers (16-45 %) claimed that they knew the sire of their calves (Paper I).

## Production

#### Live weight and Body Condition Score

It was not possible to weigh animals in the home areas of the breeds, as done at the Ghibe station. However, a subjectively given BCS between 1 and 5 was given to all animals for evaluation of the animal's condition (Paper II). The BCS was highest in Horro cattle (3.6) meaning that they were in a better condition than the other breeds. Abigar and Gurage had the lowest BCS (2.9 - 3.1) indicating a worse condition and more poorly developed animals. The BCS was lower among infected animals compared to noninfected ones in all breeds. Among purchased animals at the experimental station, the Horro and Abigar animals had the largest live weights (Paper III). However, when considering the animals born at the station, Sheko and Horro had the highest live weights, up to three years of age. Animals born at the station had, in general, a higher live weight at three years of age, compared to purchased animals of the same age, indicating good living conditions during the first years of life.

The live weight showed a seasonal fluctuating pattern (figure 10) through all years of data recording at the experimental station (Paper III). The variations in live weight followed the same pattern annually. Two distinct seasons could be seen, a "weight gain" season (May-October) and a "weight loss" season (November-April). Put aside the seasonal pattern, the first years of the study were characterized by a general weight gain in all breeds, as the animals were only about one year of age when purchased and brought to the Ghibe station.



*Figure 10.* Mean monthly live weights of purchased animals at the Ghibe experimental station (October 2000 – May 2007).

#### Milk production

In the respective home areas of the breeds, Sheko and Abigar had the highest reported daily milk yield and lactation length, resulting in highest estimated lactation yields, 630 and 530 liters respectively per lactation (Paper I). Gurage had the lowest lactation yield (160 liters) followed by the Horro (330 liters). The differences between the breeds were less pronounced at the Ghibe experimental station, where Horro and Sheko had the highest milk production, whereas Abigar had the lowest (Paper III).

The methods for milk collection varied between the home areas of the breeds and the experimental station. In the breed areas, the cows were used to milking and the livestock keepers regularly milked the animals for home consumption or sales of milk. At the Ghibe experimental station, the cows were only milked for data registrations once a week and the remaining time the calf was allowed to suckle. These differences make it difficult to compare the results.
# General Discussion

This thesis has investigated the four indigenous Ethiopian cattle breeds, Abigar, Gurage, Horro and Sheko regarding their trypanosomosis infection rate, PCV, survival, production and reproduction. Livestock keepers' perceptions about their cattle and trypanosomosis have been captured as well as objective measures have been taken of the animals in their home areas and when they were jointly kept at the Ghibe/Tolley experimental station. Thus, extensive data are available for an assessment of the trypanotolerance and phenotypic characteristics of the four breeds. Such an assessment provides guidance in the choice of breed(s) for future expanded use in tsetse infested areas of Ethiopia.

Apart from the specific characteristics for each individual breed, there were some general aspects that were valid for all the four breeds studied.

A clear seasonal pattern in live weight was detected at the Ghibe experimental station. The pattern was characterized by a weight loss season between November and April, and a weight gain season between May and October. The rainy season in the Ghibe valley takes place between June and September (Leak *et al.*, 1993) and could be a contributor to the results of the weight gain season.

Lowered PCV is considered to be one of the most clear and detectable effects of trypanosomosis. In accordance with this, and as expected, it was in this study found that infected animals of all breeds had a lower PCV compared to non-infected animals. It was also shown that the BCS was lower among infected animals compared to non-infected animals. The lower BCS is also in agreement with the fact that live weight and production is affected by trypanosomosis infection (Eisler *et al.*, 2004). The PCV was also better during the weight gain season, compared to the weight loss season. As PCV can be used as a measure of the animals' health it is logical that the animals are in a better health status as they are gaining weight.

Overall the mean trypanosomosis infection rate was comparable in size between the field study and the figures from the Ghibe experimental station. Also the PCV values corresponded well between the analyses in the field and at the experimental station.

## Abigar

The Abigar is a Sanga breed and it is sometimes claimed to possess some trypanotolerant attributes, as it is an intermediate breed type between taurine and indicus (Naessens *et al.*, 2001).

The Abigar breed was, due to certain circumstances, bled twice in the home area, once during the high trypanosomosis challenge period and once during the low challenge period. There was a notable difference between the two different bleeding times. The level of infection was, as expected, much higher during the high challenge period. The PCV was higher during the low challenge period indicating animals with a better health status. Worth noting is the lower PCV among non-infected animals in the high challenge period (20.8 %) compared to non-infected animals during the low challenge period (23.2 %). This difference can be a result of previous infections during the high challenge period, from which the PCV not yet has recovered. It is also possible that other diseases and infections follow the same yearly pattern as trypanosomosis and are the cause of the lower PCV. The clear differences in infection rate and PCV between the two time periods recorded show the importance of selecting the appropriate time and method for investigations on trypanosomosis.

The survival of Abigar animals at the Ghibe station were the lowest among purchased animals and, although a small group to compare with, had the second lowest survival rate of the animals born at the station.

In the home area, Abigar cattle had the best reproductive performance compared to the other breeds, with the lowest AFC, shortest calving interval and the highest number of calves per cow in a reproductive lifetime. However, at the Ghibe station, Abigar was instead the breed with the lowest reproduction showing the highest age at first calving, the least number of calves born and the lowest rate of pregnancies and calvings. It was never investigated whether the reproduction failures were dependent on poor reproduction of bulls or cows. Reproduction and PCV is known to have a strong link, but the mean PCV for Abigar was better at the station than in the home area (both for the bleeding at the high and low challenge period) and can thus not be suggested as a reason for the poor reproduction at the Ghibe station.

The milk production in the home areas was the best for Abigar cows with the highest lactation yield, comparable in size to reported figures of another indigenous Ethiopian cattle breed, the Barca (Tadesse & Dessie, 2003). At the station, Abigar had a good daily off-take, but the lactation lengths were short, resulting in a low total lactation yield.

Abigar cattle, followed by Horro, were the biggest in the home area. However, their BCS were the lowest indicating bigger animals in a less favorable condition. At the station, Horro, followed by Abigar, had the highest live weight among the purchased animals. The few Abigar animals that were born at the Ghibe station had the second lowest live weights.

The Abigar breed displayed in general favorable characteristics in their home areas, but at the Ghibe station a drastic drop happened in reproductive performance which unabled a solid assessment of the breed. However, due to the contradictory results between the two environments Abigar cattle cannot at this stage be recommended for expansion to improve the trypanotolerance characteristics of cattle populations in tsetse infested areas.

### Gurage

The Gurage is a Zebu type breed and they are generally believed to be trypano-susceptible (Murray *et al.*, 2004; Rege, 1999). In this study the Gurage breed had a consistently poor performance both in their home area and at the Ghibe station.

The Gurage cattle had the highest infection rate at the Ghibe station and the second highest in the home area, despite the fact that they also received the highest number of trypanocidal treatments in both surveys. Possible reasons for the high number of treatments in the Gurage area could be a high number of preventive treatments, difficulties in correctly diagnosing trypanosomosis or poor response to treatment. At the Ghibe station, however, all trypanocidal treatments were given at the same premises to all the breeds, showing a higher need for the treatments in the Gurage breed. Likewise, the Gurage breed also showed a low PCV both in the home area and at the Ghibe station.

It is often described that cattle of taurine type (like the Sheko) is smaller in body size compared to zebu cattle (Rege, 1999). However, in this study Gurage was found to be smaller than Sheko cattle, to have a less favorable BCS (Paper II), and to have a sometimes lower live weight (Paper III). Additionally the milk production was the most unfavorable of the four breeds both in the home area (Paper I) and at the Ghibe station (Paper III). The reproduction was together with Horro the least favorable in the home area (Paper I), and the second least favorable at the station. When investigating East African Zebu in the Ghibe valley, Rowlands *et al.* (1994) found an average AFC of 3.4 years, and a calving interval of just under 1.3 years. These findings are largely deviating from the current study, where Gurage had a much higher AFC of 5.5 years in both surveys, and longer calving intervals (2.2 years in the home area).

Altogether, Gurage showed many unfavorable characteristics, like a high parasitaemia, low PCV, small size and low production and reproduction. The presumption that Zebu cattle often are trypano-susceptible is thereby strengthened by their poor performance in this study. No particularly good characteristics were found in the Gurage breed, and the home area would probably benefit from a change of breed or crossbreeding with a more trypanotolerant breed.

### Horro

The Horro breed is a Zenga breed and it may possess some level of trypanotolerance. The trypanotolerance characteristics are not altogether displayed by the Horro cattle in this study. Instead, they had a relatively high parasitaemia (17-20 %). The lower level of trypanosomosis that were found in their home area was also followed by a higher PCV, compared to the results from the Ghibe station. The required number of treatments at the station was also relatively high.

However, despite the higher level of parasitaemia at the Ghibe station, the Horro breed showed the best survival rate, production and reproduction. In the home area, where infection rate, PCV and number of treatments were better compared to the other breeds, the Horro instead had a lower reproduction and milk production. The lactation yield was higher on station compared to the home area. However, still, both results were lower than the earlier reported lactation yield of approximately 500 liters/lactation (Alberro & Haile-Mariam, 1982). The AFC was lower in the home area (4.7 years, Paper I) compared to the Ghibe station (5.4 years, Paper III). The finding in the home area is comparable to results from a study on Highland/Shorthorn Zebu, where the AFC was found to be 4.4 years (Mukasa-Mugerwa *et al.*, 1989).

Although the Horro breed had, relative to Sheko, a high level of trypanosomosis and needed more trypanocidal treatments, they still had a good, and sometimes better, performance, both in the home area and at the

station. These results show that if trypanocidal drugs are readily available, the Horro breed could be a good choice in tsetse infested areas.

## Sheko

The Sheko breed is the only remaining breed of taurine type that can be found in East Africa (Rege, 1999) and the breed is considered to possess trypanotolerant attributes (Taye *et al.*, 2007; Lemecha *et al.*, 2006; Alberro & Haile-Mariam, 1982; Epstein, 1971).

In the present studies Sheko was found to have a low level of trypanosome infections (6-8 %), both in the home area and at the Ghibe experimental station (Paper II-III). Compared to the other breeds investigated, Sheko had the clearly lowest infection rate. The Sheko also needed the fewest number of trypanocidal treatments in both surveys, and were able to maintain a good PCV. Additionally, livestock keepers in the Sheko area considered trypanosomosis to be less of a problem than livestock keepers in the other breed areas (Paper II).

Sheko cattle performed well concerning survival rate, reproduction and production in both parts of the study. The total lactation yield was reported to be higher in the home area than was estimated at the Ghibe experimental station. However, in comparison to the taurine trypanotolerant N'Dama in West Africa, the current results are comparable or better (Fall *et al.*, 1999; Agyemang *et al.*, 1991). Compared to the same studies, Sheko cattle in this study also had a shorter calving interval (1.3 vs. 1.7 years) in the home area, than the N'Dama.

Reported numbers of the Sheko population size vary greatly between different sources. The higher numbers of approximately 31,000 Sheko animals, were estimated based on the presumption that cattle in the Sheko area were predominantly Sheko (Rege, 2011, Personal communication). Later, lower estimates (Dadi *et al.*, 2008; Taye *et al.*, 2007), were based on a more critical review of the actual breed of the animals. When interviewing livestock keepers, Desta *et al.* (2011) found that only 3 % had a pure Sheko herd. It was also found that less than half of the livestock keepers preferred the Sheko breed over local Zebus or crosses thereof (Desta *et al.*, 2011). The decline in number of Sheko cattle in the area was, according to the livestock keepers partly due to lack of feed in this coffee producing area for the higher feed demanding and producing Sheko cattle. The same study showed that part the decline was reported to be due to reduced labor force as children in the area more often attend school (Sheko is considered needing more labor for herding compared to tethered Zebu).

Throughout this study Sheko has consistently shown very favorable characteristics and it is clearly the most trypanotolerant breed out of the four breeds compared. The Sheko breed should be considered a breed of interest to keep in tsetse infested areas in Ethiopia.

### Genotype x Environment interaction

This thesis comprises comparisons between the four breeds both in their respective home area and in the same tsetse infested environment at the Ghibe station. Between the breeds, or between certain phenotypic characteristics of the breeds, a re-ranking took place when the environmental conditions changed. It is, for instance, shown that the reproductive characteristics for Abigar in the home area were good and the best out of the four breeds. However, nothing of that was repeated at the Ghibe station, where they performed very poorly. One could argue that the difference was due to a higher trypanosomosis challenge at the station, but in fact, Abigar had almost the same infection rate in the two studies, and a better PCV at the station. Horro, on the other hand, had the opposite situation to Abigar, with more favorable levels of infection, PCV and treatments in the home area compared to the station. At the same time, the reproductive performance was less favorable in the home area compared to the Ghibe station results.

A certain re-ranking of health and performance of the breeds might be expected, and elimination of confounding effects with the environment was part of the purpose of investigating the breeds in the same tsetse infested environment. When the performance of different traits do not change in the same direction when changing the environment it is difficult to establish and determine the potential of the breeds. Consequently, it is of high importance to investigate breeds of interest, in the environment where they are supposed to be kept, and not only compare breeds from their home environment or from a station that possibly do not have the same environmental conditions as the final target area.

Yet, it was clearly found that the Gurage breed performed poorly in both environments. In contrast, the Sheko breed performed best in both environments and constantly showed favorable trypanotolerant characteristics.

### Diagnosis and treatments

In study II it was found that, except in the Sheko area, many livestock keepers had difficulties in correctly diagnosing trypanosomosis and to differentiate it from other health problems their cattle might have. This has implications on the number of unnecessary trypanocidal treatments that are administered. For instance, livestock keepers in the Gurage area greatly overestimated the number of cattle infected by trypanosomes (98 % perceived vs. 20 % detected) and the animals were also treated much more frequently compared to animals in the other home areas. A too extensive use of trypanocidal drugs (i.e. over-dosage or unnecessary treatments) has negative impacts such as increased cost for livestock keepers, higher risk for drug residues in meat and milk, and higher risk for environmental impacts. Further implications of a wrong diagnosis arise, as the undetected disease remains untreated, when trypanosomosis is assumed to be the cause of the animals' bad health. Additionally, at the same time as livestock keepers overestimate the number of infected animals, they also overestimate the magnitude of the trypanosomosis problem in the area. To avoid consequences of wrongly administered trypanocidal drugs, livestock keepers need to be better informed about clinical signs of trypanosomosis. If possible, animals without a clear clinical diagnosis should be tested for PCV and parasitaemia. Otherwise, the use of more trypanotolerant animals would decrease the need for trypanocidal treatments and consequently reduce the negative effects of medical over-use.

## Options for use of promising breeds

The Sheko breed shows such strong favorable trypanotolerant attributes, that it would be of everybody's interest to multiply the breed. There are today areas where it is almost impossible to keep livestock due to trypanosomosis and yet many other areas where the cattle are performing poorly due to the disease. A multiplication and spread of the Sheko, or its genes, into some of these areas could greatly improve many livelihoods as well as making a better use of existing resources. Both today and for the future focus must be on having as productive and functional animals as possible to meet the future food demands without making a too large impact on the environment. This means that the number of sick or unproductive animals must be minimized and the full potential of all breeds must be better utilized.

### Purebreeding for conservation

The Sheko breed is currently on decline due to interbreeding with zebu cattle. A pure-breeding strategy is necessary for breed conservation and it may be accomplished by a well organized community based breeding program supported by a nucleus herd of purebred Sheko animals. As the Sheko breed already displays such favorable trypanotolerant characteristics the overall aim of the pure-breeding must not necessarily be further genetic improvements of the breed, but instead focus on breed conservation, by increasing the number of purebred Sheko, especially females. If and when possible, artificial insemination (AI) and sexed semen should be used in the nucleus herd for production of both replacements and animals for sale.

#### Crossbreeding for dissemination of Sheko genes

There is an urgent matter to spread the Sheko genes to animals in other tsetse infested areas. It may not be possible to completely replace any herd with Sheko cattle due to the small number of Sheko cattle available. It is probably more realistic to vision a cross-breeding scheme, where Sheko genes could be widely spread. Crossbreeding could take place between purebred Sheko bulls and cows of the current breed in the area. In such a case bulls could be transported to the new area for natural service. A more effective option, which could be practiced in areas where the infrastructure allows, is to use AI with semen from selected bulls either from the nucleus herd or the community population. In the case of AI it may in the future be possible to use sexed semen to increase the number of female calves being born.

If there are no incentives for keeping the current breed in an area, there is the option of selecting yet another breed, Horro, to crossbreed with the Sheko. The Horro breed, although not trypanotolerant, has displayed many favorable characteristics, as it is able to survive, produce and reproduce under tsetse and trypanosomosis challenge. The total number of Horro cattle is much larger than that of the Sheko, which, if a crossbreeding between the two breeds could take place, would simplify a more rapid increase in the number of animals with desired genes. It would therefore be an interesting option to crossbreed Sheko and Horro to produce bulls for dissemination of trypanotolerant attributes to a wider range of areas.

#### General aspects

A crucial factor for the success of any type of program is the active involvement from all stakeholders and there is a need of communication with livestock keepers in tsetse infested areas, their advisors and animal health officers. Livestock keepers' willingness to use a new breed, that is trypanotolerant, has to be clarified, as does the commitments from all other involved parties, such as the local and government authorities. If all parties are in agreement and working in the same direction with the same ultimate goal, there is hope for success. The individual livestock keepers, as well as officials, have to feel ownership of the program in order to fully commit to it. It is the livestock keepers, and their advisors together with local and national officials that eventually need to manage any potential breeding program aiming at a better use of trypanotolerant animals.

To be able to make justified and sound decisions it is important to have correct and relevant information and research facts at hand. However, no research of this kind is finished until it is spread to the relevant recipients. Therefore, the importance and urgency for a rapid dissemination of the information and results now available should be stressed.

# Conclusion

Trypanosomosis was considered to be a major constraint in all four investigated areas where Abigar, Gurage, Horro and Sheko cattle are kept.

Sheko cattle showed the most favorable characteristics both in their home environment and at the Ghibe station.

Horro cattle showed many favorable characteristics, such as reproduction and survival rate, although they required more trypanocidal treatments than the Sheko and had higher trypanosomosis prevalence.

Abigar cattle showed some favorable characteristics in their home environment especially concerning reproduction and milk production, but performed poorly at the Ghibe station.

Gurage cattle showed overall unfavorable characteristics as regards trypanotolerance, reproduction and survival rate, both in their home environment and at the Ghibe station.

Overall, the most promising of the four breeds in this study is the Sheko breed. As the breed shows the highest level of trypanotolerance but is threatened by extinction, there is a need for immediate actions to save the breed and disseminate its favorable characteristics.

Between the breeds, or between certain phenotypic characteristics in the breeds, a re-ranking took place when the environmental conditions changed. This shows the importance of evaluating breeds in an appropriate environment where the animals are going to be used.

Difficulties among livestock keepers to correctly diagnose trypanosomosis were found, and there is a need to further educate livestock keepers in this matter to ensure a correct use of trypanocidal drugs.

It is of high importance to spread knowledge among livestock keepers and other stakeholders about trypanotolerance and trypanotolerant breeds for possible future use in tsetse infested areas.

There is a need for conservation of the Sheko breed by a community based breeding program supported by a nucleus herd. For multiplication of the Sheko breed, or dissemination of its genes, Sheko should be used for crossbreeding with zebu breeds, possibly with the Horro, as it has shown many favorable characteristics. Artificial insemination, possibly with use of sexed semen, would be the most effective way of producing crossbred animals.

# **Future Research**

Trypanotolerance is a very complex trait and therefore there are also many different aspects and angles for future research. At the same time, research on trypanosomosis and trypanotolerance has already been carried out for decades, and a lot has been done.

It would be of great interest to record performance and trypanotolerance, especially for Sheko and Horro cattle, over multiple generations to be able to estimate the heritability for the trait. With such estimates it would be possible to predict what the selection response would be when selecting trypanotolerant animals within the breeds.

Studies on crossbred animals between Sheko and Horro are also of great need and interest in order to determine how trypanotolerance is passed on to the offspring. It is suggested, based on this study, that Sheko and Horro might be a suitable choice of breeds to use in a crossbreeding scheme. However, the trypanotolerance of these crossbreds needs to be researched.

The feasibility of using sexed semen in AI for the Sheko breed needs to be investigated for a future possible use for effective dissemination of the genes of the breed. Although sexed semen currently is commercially used in parts of the developed world, information regarding prospects of the technique in developing countries and their breeds is lacking.

Further work is needed to set up properly designed breeding programs, both for a community based breeding program supported by a nucleus herd, and for multiplying the Sheko breed or dissemination of its genes through crossbreeding with Horro and/or other suitable zebu breeds. Investigations

of available infrastructure, and possibly also improvements, are needed to further ensure the success of the programs.

Although a lot has already been done, molecular investigations of trypanotolerance is still a matter for future research as there are many questions to answer for efficient selection of trypanotolerant animals. Molecular research has so far focused especially on mice and the N'Dama cattle breed, but many possible trypanotolerant species and breeds remain.

## Svensk sammanfattning

Trypanosomosis är den enskilt allvarligaste och största djursjukdomen i Afrika söder om Sahara och hela 37 länder är drabbade. Det uppskattas att nästan 9 miljoner km<sup>2</sup> av Afrikas landyta har potentiell infektionsrisk. Runt 60 miljoner nötkreatur lever i områden med ständig risk för infektion och man räknar med att 3 miljoner nötkreatur årligen dör av sjukdomen.

## Bakgrund

Trypanosomosis orsakas av parasiten *Trypanosoma* som överförs till djur via tsetseflugan. När samma parasit infekterar en människa kallas sjukdomen sömnsjuka. Parasiten har en del av sin livscykel i värddjuret (i denna studie nötkreatur) och en del i flugan. Tre arter av trypanosomer är virulenta för nötkreatur: *Trypanosoma congolense, Trypanosoma brucei* och *Trypanosoma vivax*. Infekterade djur drabbas ofta av blodbrist, viktnedgång, missfall, svullna lymfkörtlar och minskad mjölkproduktion. Om sjukdomen inte behandlas dör djuren oftast. Trypanosomosis gör det svårt, och ibland helt omöjligt, att hålla produktiva och friska djur i angripna områden.

Vissa raser kan hantera och kontrollera parasiten och dess effekter bättre än andra. Dessa raser kallas trypanotoleranta och de kan överleva, producera och reproducera sig även i områden med hög risk för infektion. Dessa trypanotoleranta raser är också kända för att kräva färre behandlingar med antibiotika om de drabbas än mer mottagliga raser. Det är främst raser av *Bos taurus*-typ (vår tamboskap) som anses vara toleranta medan *Bos indicus*-raser (zebu) är mer benägna att vara mottagliga för trypanosomosis. N'Dama från Västafrika är den mest kända trypanotoleranta rasen och har samma ursprung som vår tamboskap. Även i Etiopien finns det vissa raser som lever i områden med högt infektionstryck och det är därför möjligt att även de kan vara trypanotoleranta. Den sista kvarlevande rasen i östra Afrika med samma

ursprung som vår boskap, Sheko, finns i Etiopien och den anses vara tolerant.

En bättre användning av trypanotoleranta raser kan:

- förbättra nötkreaturs produktion och reproduktion
- öka nötkreaturens överlevnad och välmående
- minska kostnaderna för medicinska behandlingar
- öka nyttjandet av befintliga eller nya områden för djurhållning, där det tidigare var omöjligt att ha djur på grund av trypanosomosis
- förbättra människors ekonomi och välfärd i tsetse- och trypanosomosisangripna områden.

Syftet med den här studien var att undersöka och jämföra fyra etiopiska nötkreatursraser med avseende på olika egenskaper som rör trypanotolerans. Raserna som studerades var Abigar, Gurage, Horro och Sheko. Dessa raser återfinns i fyra olika delar av sydvästra Etiopien (figur 3). Vi ville undersöka raserna både i de områden där de normalt finns (studie I-II) och på en fältstation i ett tsetse-område där alla djur utsattes för samma miljö (studie III).

## Sammanfattning av studierna

Den första delen av studien bestod av intervjuer av djurägare i de områden där raserna normalt hålls. Undersökningen bestod av frågor om produktion, reproduktion, sjukdomar och avel. Totalt intervjuades 60 djurägare i varje område (studie I-II).

Den beräknade avkastningen var högst för Sheko och Abigar med ca 600 liter mjölk per laktation. Gurage hade den lägsta mjölkproduktionen med endast runt 180 liter mjölk under en hel laktation. Abigar, Sheko och Horro hade klart bättre reproduktionsegenskaper än Gurage. Gurage hade en högre ålder vid första kalvningen (>1 år äldre) och nästan dubbla kalvningsintervallet (2,2 år) jämfört med de övriga tre raserna. Abigar och Sheko hade det största antalet kalvar under kons livstid med i genomsnitt 8-9 kalvar, medan Gurage och Horro i genomsnitt producerade 5-6 kalvar per ko. Sjukdomar angavs som det största problemet för produktion med nötkreatur i alla områdena utom i Sheko-området där bristen på betesareal ansågs vara den allvarligaste restriktionen för djurhållning. Bland angivna sjukdomar var trypanosomosis den mest angivna och allvarligaste sjukdomen i alla områden.



Vid den tiden på året då djurägarna ansåg att risken för trypanosomosis var som störst, utfördes nästa del av studien (studie II). Blodprov togs på ca 100 djur av varje ras för att med hjälp av mikroskop bestämma infektionsstatus på djuren. Samtidigt mättes nivån av hematokrit (på svenska känt som sänkan), det vill säga de röda blodkropparnas andel av blodet (Packed red Cell Volume, PCV) vilket kan användas som ett indirekt mått på djurens hälsotillstånd. Tre kroppsmått togs på djuren (kroppslängd, mankhöjd och bröstomfång) och en subjektiv bedömning gjordes av kroppskonditionen (Body Condition Score, BCS). Djurägaren intervjuades om djurens hälsotillstånd och tidigare medicinska behandlingar.

Sheko var minst infekterade av trypanosomer (6%), medan de andra raserna var infekterade i mycket högre grad (17-23%). Horro hade tillsammans med Sheko de mest fördelaktiga nivåerna på PCV och BCS. Som väntat hade infekterade djur lägre PCV och BCS jämfört med icke infekterade djur. Antalet behandlingar varierade stort mellan de olika raserna, där Sheko hade lägst antal behandlingar med i genomsnitt 1 behandling per djur och år, medan Gurage-djuren i genomsnitt fick 24 behandlingar per djur och år.

Den sista delen av studien utfördes på en fältstation i Ghibe-dalen 23 mil sydväst om Addis Abeba (studie III). Området är känt för högt infektionstryck. Totalt 238 djur (ungefär 1 år gamla) av de fyra raserna köptes in från sina hemtrakter. Dessa djur hölls sedan under 2000-2007 på fältstationen, där alla utsattes för samma tsetse-angripna miljö. Detta möjliggjorde en jämförelse mellan raserna utan att de olika miljö- och produktionsförhållandena i deras hemtrakter behövde beaktas. Totalt föddes 137 kalvar på fältstationen, vilket medförde att ett totalt antal på 375 djur ingick i studien. Varje månad utfördes registreringar på djuren där infektion, PCV och vikt angavs. Utöver dessa registreringar, noterades även fruktsamhet och mjölkproduktion.

Sheko hade återigen den lägsta infektionsnivån (9 %) jämfört med de andra raserna (21-26%). Sheko behövde också minsta antalet medicinska behandlingar mot trypanosomosis. Sheko och Abigar hade de högsta PCVvärdena under hela studien. Abigar och Gurage hade mycket sämre fruktsamhet då endast 9 respektive 18 kalvar föddes under de 7 år studien varade. Horro och Sheko fick 50-60 kalvar var under samma period. Överlevnaden hos både inköpta djur och de som fötts på stationen var också bäst hos dessa två raser. Även mjölkproduktionen var högst hos Horro och Sheko. Levande vikten hos de inköpta djuren följde ett säsongsmönster (figur 10) som utgjordes av en viktminskning under november till april och

viktökning under maj till oktober. PCV var ogynnsamt lägre under viktminskningssäsongen och bättre när djuren istället gick upp i vikt.

## Kortfattade slutsatser

När alla faktorer beaktas, både från områdena där raserna normalt hålls och från fältstationen i Ghibe, står det klart att Sheko är den mest trypanotoleranta rasen av de fyra som jämfördes. Sheko blev sällan infekterade av trypanosomer och de lyckades hålla PCV, hull, mjölkavkastning, fruktsamhet och överlevnadsförmåga på bra nivåer. Även Horro visade upp många bra egenskaper men de var oftare infekterade och behövde fler medicinska behandlingar än Sheko. Trots detta kan Horro också tänkas vara en bra alternativ ras genom sin goda fruktsamhet och överlevnadsförmåga, att öka användningen av. Ett hållbart avelsprogram för förökning och större spridning av Sheko, eller korsningar mellan Sheko och andra raser skulle medföra att rasen och dess gener för trypanotolerans kan få större spridning. Det är dock angeläget att kontrollera vad, framförallt korsningen mellan Sheko och Horro, har för egenskaper när de används i områden med stort infektionstryck. En bättre användning av rasen Sheko i tsetse-angripna områden skulle främja djurens hälsa och hushållens ekonomi och välfärd.



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