Alternative remedies used by resource-limited farmers in the treatment and manipulation of the reproductive system of non-descript goats in the Eastern Cape Province, South Africa

By

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Together in Excellence

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DECLARATION

I, Maxwell Rwodzi hereby declare that this work is original (input from other authors is acknowledged) and it has not previously been submitted at this or any other university.

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Maxwell Rwodzi                      Date

Approved as to style and content by:

_______________________________    _________________________
Prof P.J. Masika (Main supervisor)   Dr V. Maphosa (Co-supervisor)
ABSTRACT

This study was carried out to determine alternative approaches used by resource-limited farmers to enhance fertility and in the treatment of reproduction ailments of goats in Mount Frere area, Eastern Cape Province of South Africa. Furthermore, information on farmer perceptions pertaining to these practices was also gathered. A questionnaire survey was conducted to determine materials used by resource-limited farmers to enhance fertility and treat reproductive ailments. The survey revealed 10 plant species that were commonly used for reproductive health in goats. *Elephantorrhiza elephantina* (37.1%) and *Rhoicissus tomentosa* (25.7%) were the most frequently cited plant species. *Elephantorrhiza elephantina* and *R. tomentosa* recorded the highest Fidelity Level (FL) values of 92.86% and 75% respectively, for their use in the treatment of reproductive ailments and these were the plants on which further studies were conducted. The study also revealed that farmers had different perceptions on the effectiveness of the traditional medicines.

An in-vivo assessment was done to determine efficacy of *E. elephantina* and *R. tomentosa* using serum estradiol and progesterone profiles. The highest (P<0.05) serum estradiol and progesterone concentration was observed in animals administered with aqueous extracts of *E. elephantina* and *R. tomentosa* mixture while the least (P<0.05) concentrations were observed in the negative control group. Does administered with a mixture of aqueous *E. elephantina* and *R. tomentosa* extract had the highest (P<0.05) body weight (65 kg) as well as body condition scores, while the negative control which were administered with distilled water had the least body weights (54 kg) and body condition scores. Does that were administered with a mixture of *E.
*elephantina* and *R. tomentosa* were all pregnant with twins (100%), confirmed by a doppler ultrasound scan.

**Key words:** Does, Efficacy, Fertility, Plants, Reproductive system
DEDICATION

I fully dedicate the success of this study to God, for granting me health and strength during my studies. I would like to pass my special dedications to my grandmother and grandfather, Mrs and Mr Kamusisi and as well as my mother, Mrs Rwodzi for their moral support.
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<tr>
<td>%</td>
<td>Percentage</td>
</tr>
<tr>
<td>AI</td>
<td>Artificial Insemination</td>
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<td>ARDRI</td>
<td>Agriculture and Rural Development Research Institute</td>
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<tr>
<td>BCS</td>
<td>Body Condition Score</td>
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<td>EVM</td>
<td>Ethno-veterinary medicine</td>
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<td>FL</td>
<td>Fidelity Level</td>
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<td>MOET</td>
<td>Multiple Ovulation and Embryo Transfer</td>
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CHAPTER 1: General introduction

1.1. Background

Goats account for about 30% of Africa’s ruminant livestock and contribute 17 and 12% to meat and milk production, respectively (Lebbie, 2004). Goat keeping is widely practiced in the rural areas, contributing to household income and food security (Masika and Mafu, 2004). For example goats are used for milk, meat, fibre, skin and draught under various conditions (Dubeuf et al., 2004). Furthermore, easy handling and the effective conversion of limited food resources into meat and milk are also important factors favouring goats as a stock animal for resource-limited farmers (Harper and Penzhorn, 1999). However, in many communal areas reproduction performance is one of the important constraints faced in improving goat productivity.

Goat fertility is one of the fundamental areas that affect reproductive performance of goats (Yakubu and Afolayan, 2009). There are several modern methods developed to improve reproduction performance but these options are expensive, not easily available and have serious side effects (Mellado et al., 2006). On the other hand goat farmers in the communal areas, who are not aware of modern technologies, use a wider range of alternative remedies to enhance fertility and treat livestock reproductive ailments. Some farmers use medicinal plants, which are known to have a positive influence on the estrous cycle, ovary function, ovulation, pregnancy and semen function (Rong-zhen and Dao-wei, 2013). Furthermore, some of these plants are known to have phyoestrogens, which are compounds with structures similar to female hormones.
and these are perceived to improve the reproductive performance of livestock (Edouard et al., 2014). Besides fertility enhancement, problems of retained placenta and their accompanying secondary infections can also be treated using these alternative remedies. Masika and Afolayan (2002) reported that a variety of medicinal plants are used to treat various diseases including reproductive ailments in both human and domestic animals. More than 700 indigenous plant species are still being used by approximately 72% of Black South Africans for medicinal purposes (Meyer and Afolayan, 1995; Williams et al., 2013). The treatment of infectious reproductive ailments is important because they can result into infertility and cause problems like abortion in livestock (Tibary et al., 2006). In the process of treating and controlling the effects of these ailments farmers could use synthetic drugs but these options are expensive and not readily available especially to resource-limited farmers. Hence farmers resort to the use of cost-effective and reliable alternatives such as medicinal plants (Geyid et al., 2005).

The medicinal properties of these plants are reported to be due to bioactive compounds such as tannins, triterpenes, isoflavonoids, anthroquinones, flavonoids, saponins, and lignans (Kim et al., 2008; Rochfort et al., 2008). Some of these compounds are known to have an influence on the reproductive system of livestock and are responsible for the antimicrobial activity of some medicinal plants. The treatment of reproductive ailments and enhancement of animal fertility increases the productivity of livestock (Pivoda et al., 2008) since reproduction and production performances play an important role in animal husbandry and profitability of farming (Sabapara et al., 2010). However, there are very few studies that have been conducted to identify the factors responsible for reproductive failure and how they can be overcome through the use of ethnoveterinary medicines (Ravinder et al., 2010).
1.2. Problem statement

Farmers widely use alternative remedies to address reproduction issues in their livestock however, little is known of what is used and their effect on fertility and reproduction ailments. Although, several modern methods of controlling reproduction have been developed, these options are expensive, not easily available and have serious side effects (Mellado et al., 2006). Therefore, in the process of increasing animal productivity, farmers use plants to prepare remedies which are administered to animals for various purposes even though, little is known about what is used and whether they are effective or not. Use of materials with unknown efficacy may result into losses to the farmers. It was therefore important to conduct research on these alternatives used by farmers.

1.3. Justification

Presently, contribution of livestock in the communal farming system, to the gross domestic product (GDP) of the province and country is very low. However, if health and reproduction management are improved, farmers will realize better returns from their farming endeavours. Therefore, documenting alternative practices used to improve health and reproduction will preserve the knowledge, which to start with is disappearing due to civilization (Nfi et al., 2001), because it is transferred orally. There is need to validate the materials farmers use to address reproduction losses and determine their efficacy. Results from efficacy tests, maybe be used to inform the farmers which of the remedies they use are effective. The research may provide a
starting point for the search of new drugs/remedies since the efficacy of the alternative remedies used by farmers were determined. Therefore, research on the alternative remedies used by farmers for health and reproduction purposes is of paramount importance if livestock production is to be improved in the communal farming system.

1.4. Objectives

The main objective was to document and validate alternatives used to manipulate reproduction and treat ailments related to reproduction in livestock by resource-limited farmers in the Eastern Cape Province of South Africa.

Specific objectives

The specific objectives were to:

1. To document materials used to enhance fertility and treat reproductive ailments of goats; and

2. To determine the efficacy of the two plants that are most frequently used to influence reproduction in goats.
1.5. Hypotheses

The following null hypotheses tested:

1. Materials used to enhance fertility and treat reproductive ailments of goats are not known; and

2. The two frequently used plants have no influence on the reproductive system of goats.

1.6. References


CHAPTER 2: Literature review

2.1. Introduction

Farmers use a wide range of traditional medicines for various purposes such as the control and treatment of diseases and parasites (Mwale and Masika, 2009). Before the introduction of western medicines, stock keepers relied on traditional medicines to treat livestock diseases and ailments. Traditional animal health care, which includes knowledge, skills, methods and beliefs about animal care, is referred to as ethnoveterinary medicine (EVM) (McKorkle and Mathias-Mundy, 1992). This information is passed down orally from generation to generation (Toyang et al., 2007). Although, farmers use these traditional medicines, there is insufficient scientific documentation of the practices (Masika and Afolayan, 2003). Ethnoveterinary medicines are gaining popularity due to their greater accessibility, lower costs and apparent effectiveness (Mwale et al., 2005). According to Gurib-Fakim (2006), farmers who use medicinal plants may not understand the scientific explanation of how their medicines work but they use personal experience to identify medicinal plants that are effective if used at therapeutic doses.

However, it is difficult for the younger generation to appreciate the beliefs and practices of their ancestors because of civilization and introduction of conventional methods of disease control. Moreover, there is a risk that when the elder members of the community pass away, there might be a complete loss of information concerning ethnoveterinary medicine (Mathias, 2005). The situation is further worsened by the high rates of deforestation in the world. Africa is reported to
have the highest rate of deforestation in the world (Gurib-Fakim, 2006), hence ethnoveterinary knowledge is gradually being lost.

2.2. History of ethnoveterinary medicines

Many nations have been using medicinal plants since at least 7000 years ago (Iqbal et al., 2005). Most ethnoveterinary medicines originated from Europe, India, Africa and South America (Lans and Brown, 1998). However, Pakistan, China and India are regarded to have much knowledge about medicinal plants used in ethnoveterinary medicine since they are the greatest users of medicinal plants (Iqbal et al., 2005). Currently, ethnoveterinary medicine has won growing attention and respect within conventional science and in the international development community (McCorkle and Mathias-Mundy, 1992) such that some countries are involved in the manufacturing of herbal remedies (Wanzala et al., 2005). However, Singh et al. (2014) argues that the development of medicinal plants for commercial use seems to be less due to the high costs of the techniques used in developing medicinal plant based drugs. The other issue is of Intellectual Property Rights (IPR) and benefit sharing as knowledge holders want to be recognized. IPR not well defined, leading to delay in formation of patents, due to secrecy.

2.3. Comparison between ethnoveterinary medicines and conventional medicines

There are several reasons why ethnoveterinary medicines are gaining popularity. The high cost, residual effects in meat and parasite resistance to drugs of the conventional methods to control livestock diseases compel farmers to resort to ethnoveterinary methods (Mathius et al., 1996;
Luseba and van der Merwe, 2006; Mwale and Masika, 2009). Drug resistance makes it difficult to control diseases (Hemalatha et al., 2013). The use of modern medicines is associated with drug resistance problems, but there is reduced drug resistance when EVMs are used (Tekwu et al., 2012). Communal farmers believe that conventional drugs have more damaging adverse effects and produce more toxic residues in food than the naturally produced remedies (Wanzala et al., 2005).

When traditional medicines are used, there are no restrictions or withdrawal periods for consumption of meat from treated animals (Luseba and van der Merwe, 2006). Furthermore, some resource-limited farmers do not have access to conventional drugs which could be expensive and not available in times of need (Ghotge et al., 2002). According to Iqbal et al. (2005), poverty can be reduced through the use of ethnoveterinary medicines. This is because generally medicinal plants are said to be cheaper, accessible and acceptable in the communities (Maphosa and Masika, 2010). Modern conventional medicines usually aims to develop a patentable single compound which can treat specific ailment, but traditional medicines contain a wide range of bioactive substances and when different plants are mixed together, a synergistic effect is maximized and more than one ailment can be treated with the same remedy (Gurib-Fakim, 2006). Ethnoveterinary medicine has been the starting point for the discovery of many drugs now used in modern animal medicine (Barboza et al., 2007). Traditional remedies are easier for livestock keepers to prepare from plants at a lower cost compared to allopathic medicines (Iqbal et al., 2005). However, Haverkort et al. (1996) argued that traditional remedies are totally ineffective. Ethnoveterinary medicines do not cause environmental pollution compared to conventional medicines (Fajimi and Taiwo, 2005).
There are several challenges that can be faced with the use of traditional remedies. The disadvantages of ethnoveterinary medicine include risk of incorrect diagnoses, imprecise dosages, low hygiene standards, secrecy of some healing practices and some treatments may be harmful or ineffective (Toyang et al., 2007). In addition, according to Wanzala et al. (2005), there are some inconveniences associated with the use of traditional remedies in terms of preparation, ineffectiveness, poor diagnoses, toxicity and unavailability of ingredients to prepare medicines.

The other major problems associated with traditional remedies are that, methods used are geographically localized and there is poor dissemination of information concerning traditional remedies (Andrews and Blowey, 2008). Since most of the medicinal plants are wild and only a few are cultivated, there is a great risk of total forest destruction when a plant is identified to be of medicinal use, resulting in extinction of some plant species (Yirga et al., 2012). When using traditional remedies there is a problem of seasonal variation in the availability of some plants, making treatment impossible if they must be used fresh (Haverkort et al., 1996). According to Haverkort et al. (1996), determining optimal dosages and even treatment schedules can be difficult when using traditional medicines. But Fielding, (1999) argues that it is needless to have formal validation since the stock raisers have been using ethnoveterinary medicines for long time and have already validated in their own ways, through many trials. This shows that there is need to validate the efficacy of ethnoveterinary medicines. Nevertheless, the quality and efficacy may vary due to the same type of plant medicine coming from varying environments and when different parts of a plant are used during remedy preparation (Obomsawin, 2008).
When using conventional drugs the problems of toxicity are minimal as compared to the use of traditional remedies because conventional drugs have standardised dosages. However, Soforowa (1996) stated that the possibility of overdose from traditional medicines is probably insignificant because the concentration of the active ingredient in a decoction is usually far less than that in a conventional drug. Nevertheless, some plants like comfrey root contain alkaloids which are known to be toxic when fed in large quantity to animals (Obomsawin, 2008). Despite some potential challenges of traditional medicines, they still play a vital role in the control and treatment of diseases, improving reproduction and the treatment of reproductive ailments.

2.4. The role of ethnoveterinary medicines on reproduction

2.4.1. Reproductive Performance

Reproductive performance is an important key component in goat production. According to Dobson and Smith (2000), reproduction is a vital physiological system because it assists in the furtherance of a species. According to Mellado et al. (2006) reproductive performance of goats has an effect on productivity and economic viability of a goat enterprise. For instance, at farm level, the number of offspring produced by the does in a given period of time determines the gross income from goats, which means that animals of high fertility and prolificacy are desirable (Akingbade, 2001). When fertility is poor there are losses due to low kidding rate, low kid sales, lower milk sales, higher semen costs, higher veterinary costs as well as slow genetic progress (Mellado, 2011).
There are several processes that determine reproduction efficiency in does and these include ovulation rate, the length of the cyclic activity, fertilisation rate, breeding season, the post-partum anoestrous period and the growth and viability of the offspring (Greyling, 2000). The same author pointed out that the number of ova liberated and eventually expressed as the number of kids born per doe kidding is referred to as ovulation rate and it is an important characteristic in goat production. When there are no drastic shortcomings with regard to health and husbandry system, goats are amongst the most fertile of the domestic species, with conception rates in the range of up to 90% (Holtz, 2005).

2.4.2. Factors affecting reproductive performance

Reproductive performance of goats is affected by both the environment and the genetic make-up of the animals. Reproduction in goats is mainly influenced by latitude and climate, breed, physiological stage, presence of a male, nutritional status, genetic potential, breeding system and photoperiod (Fatet et al., 2011; Mellado, 2011). However, photoperiod is the main environmental factor affecting seasonal breeding in small ruminants (Blache et al., 2008) but it has less effect on the reproductive performance of goats. Reproductive disorders, such as abortion can seriously affect the reproductive performance of goats (Mellado et al, 2006). The causes of abortion include diseases, stress and poor nutrition.

2.4.3. Methods to improve reproductive performance
There are several modern methods of controlling reproduction in goats which include the administration of hormones that modify the physiological chain of events involved in the sexual cycle (Abecia et al., 2012). Lehloenyia et al. (2005) citing Wildt (1992) stated that assisted reproductive techniques such as artificial insemination (AI), multiple ovulation and embryo transfer (MOET) are considered to be useful in goat reproduction. Most pharmaceutical controls of reproduction are done on female animals but it is also important to confirm fertility of male animals (Abecia et al., 2012). Currently, artificial synthetic estrogen is regarded as the most effective agent for hormone replacement therapy but there are fears of negative side effects such that alternatives are becoming more preferred (Mellado et al., 2006; Hsu et al., 2008). Countries belonging to the European Union have restricted the use of synthetic hormones on livestock to meet the demands of consumers who are now demanding natural ways of manipulating reproduction in livestock (Delgadillo et al., 2014). According to Patel et al., (2011) there is need to search for natural supplement from medicinal plants because of the serious side effects and high cost of conventional medicines. Dobson and Smith (2000) stated that the persistent genetic development in animal species in order to meet food supply has resulted in reduced fertility in animals. Therefore, there is need for more pharmacological studies on cheaper, effective and safe natural treatment options (Abdillahi and Van Staden, 2012). Problems and constraints in animal’s reproduction can be addressed by using traditional reproductive management practices for maximum economic return (Ravinder et al., 2010).

Despite the documentation of some traditional medicines, very little scientific information on efficacy and phytochemistry has become available on medicinal plants (van Vuuren, 2008). In South Africa, few researches have been done on pregnancy-related herbal medicines (Varga and
Veale, 1997). Furthermore, the economic impact of plant remedies on goat reproduction function is poorly understood (Mellado et al., 2006). Therefore, there is need to carry out studies on the efficacy of these alternatives on animal reproduction. There are also plants such as *Panax ginsengs* that are used to treat sexual dysfunction in China while *Lepidium meyenii* is used as a nutrient, energizer, aphrodisiac and fertility-enhancing agent (Patel et al., 2011). Viegi et al. (2003) reported that sprouting seeds of *Avena sativa* and leaves of *Prunus avium* are used to promote estrus in animals. These plants are perceived to contain hormonal substances but not yet confirmed scientifically (Viegi et al., 2003). Some medicinal plants have been used as hormone replacements, for example the root extracts of *Dioscorea villosa* have been applied in hormone replacement therapy (Hsu et al., 2008). This is because some of the medicinal plants are reported to contain phytoestrogens such as diethylstilbestrol which has similar structure and function to the synthetic estrogens (Ahmad et al., 2013). There are several plants that have been identified to contain phytoestrogens (Khorshidi and Sherafatmandjour, 2013), among these is soya bean (Jing Cheung, 2000).

Farmers in South Africa sometimes buy prepared medicines from traditional healers. The most often purchased prepared herbal medicine is *isihlambezo* which is used to reduce vaginal discharge and reduce placental size (Varga and Veale, 1997). Previous researches have shown that decoctions of *Agapanthus africanus*, *Gunnera perpensa* and *Pentanisia prunelloides* had direct effect on the uterine contractions in response to oxytocin and acetylcholine on the isolated rat uterus (Varga and Veale, 1997).
2.4.4. Efficacy of medicinal plants on the reproductive system

Medicinal plants may have negative or positive effects on the male and female reproductive systems, affecting almost every stage of the reproductive system in goats (Durmic and Blache, 2012). According to Patel et al. (2011), some medical plants are aphrodisiac and are responsible for stimulating the expression of male reproductive behaviors such as courtship and mating behavior. The extract of *Tribulus terrestris* is known to be important in improving semen quality in terms of sperm motility and ejaculatory volume (Gauthaman and Ganesan, 2008). The same author also indicated that this plant extract can also stimulate secretion of testosterone.

Farmers use medicinal plants singly or in combinations to improve fertility and to induce oestrus in ruminants (Lans and Brown, 1998). However, it is important to note that when traditional healers prepare remedies, they alter dosages depending on the severity of the ailment (Stark et al., 2013). Since some plants are used to increase egg production in chickens, it is suggested that they have an effect on ovulation (Viegi et al., 2003). Several plants have been used in traditional veterinary medicine to help deal with problems such as dystocia and retained placenta (Lans and Brown, 1998; Viegi et al., 2003). The activity of the plants is due to compounds like saponins, tannins and alkaloids but the mechanisms of action of these compounds are mostly unknown (Viegi et al., 2003). However, saponin extracts are reported to be used in the manufacturing of steroidal drugs and this is made possible by chemically altering saponins to sapogenins which can then be used as precursors for steroid drugs such as androgens, oestrogens and progestins (Gurib-Fakim, 2006). These steroid drugs have got the same functions as of the natural hormones (Skipor et al., 2012; Ahmad et al, 2013). A variety of medicinal plants were reported to have an
aphrodisiac effect and these include *Tribulus terrestris, Aframomum melegueta, Ferula harmonis, Eurycoma longifolia, Lepidium meyenii, Passiflora incarnate* and *Mununa pruriens* (Patel, 2011). Furthermore, plants like *Leonurus cardiac* and *Linumusita tissimum* can be used for milk production and pregnancy support as well as conception respectively (Lans *et al.*, 2009). Although several plants have been found to be effective, there are also a number of plants that have been found to be ineffective after scientific validation, despite the continual use of these plants for reproduction purposes by farmers. For instance plants high in phytoestrogen content were found to cause temporal infertility in ewes (Adams, 1995). But some of the plants used by farmers are reported to be rich in phytoestrogens and farmers assert that such plants are effective. However, the results from scientific validation may differ with the claims of farmers due to use of unsuitable models to assess the plant efficacies.

In South Africa, *Elephantorrhiza elephantina* is used as an aphrodisiac in males (Abdillahi and Van Staden, 2012). Furthermore, it is used as a remedy for irregular menstruation in women and for cleansing the womb after abortion in livestock (Moyo *et al.*, 1999). *Elephantorrhiza elephantina* is a plant used as a remedy for a wide range of ailments in both animals and human (Maphosa, 2010). The ability of this plant to work on a wide range of ailments might be due to its bioactive compounds which include flavonoids, gallic acid derivatives, esterified sugars and simple phenolic derivatives (Moyo *et al.*, 1999).
2.5. The role of ethnoveterinary medicines on reproductive ailments

2.5.1. Reproductive ailments in goats

There are several bacteria species reported to affect the reproductive system of goats either directly or indirectly and these include *Staphylococcus aureus*, *Enterobacteraerogenes*, *Brucella abortus*, *Brucella meletensi*, *Pseudomonas aeruginosa*, *Campylobacter fetus*, and *Tritrichomonas fetus* (Tibary *et al.*, 2006). These bacteria are also associated with secondary infection, especially in cases of abortion, retained placenta and endometritis. According to Ouweltjes *et al.* (1996) abortion, retained placenta, endometritis and irregular estrus are the most common reproductive disorders of livestock. Female animals that have previous cases of abortion, dystocia and retained placenta have got high chances of having uterine infection (Vaughan and Tibary, 2006; Tibary *et al.*, 2006). These infections reduce the productivity of goats. The role of infectious diseases in abortion and their prevention should be researched on so that the impact of these infectious diseases will be minimized (Vaughan and Tibary, 2006). For instance, infectious diseases can lead to abortion in pregnant does (Tibary *et al.*, 2006). Resource-limited farmers are the ones who suffer most and that is the reason why Kumar *et al.* (2003) mentioned that losses suffered by resource-limited farmers on account of diseases in goat production indicate that researches on health aspects should be given a priority. This is because rural households own over 90% of the goats in the world (Lebbie, 2004). However, mortality and morbidity losses due to diseases in goats put a heavy burden on resource-limited farmers and hence large losses to the agricultural economy of most developing countries like South Africa.
(Kumar et al., 2003). Currently, in many developing countries, people rely heavily on traditional medicines to meet primary health care needs (Upadhyay et al., 2011).

There is increased resistance to antimicrobial drugs by microorganisms even though a number of new antimicrobial drugs have been developed in recent years (Tekwu et al., 2012). This is because pathogenic bacteria evolved new strains which are resistant to antibiotics and drugs (Dubey et al., 2012). Antimicrobial resistance is a growing problem that is said to complicate the treatment of economically important infectious diseases (Tekwu et al., 2012). Staphylococcus aureus and Escherichia coli are some of the bacteria strains that have developed multiple resistances to a wider range of antibiotics (Dubey et al., 2012). Therefore, medicinal plants can be used as alternative remedies since approximately 7000 species of higher plants are reported to have medicinal properties in South Africa (Kalayou et al., 2012). Plant based antimicrobials have huge therapeutic potential and this makes them to be a potential solution to many of the side effects of synthetic antimicrobials (Kokoska et al., 2002).

2.5.2. Anti-microbial efficacy of medicinal plants

Plants are known to produce several secondary metabolites to protect themselves against microorganisms and this might show that compounds in these plants are potentially therapeutic against microorganisms (Chung et al., 2011). There are several studies that were done to determine the antimicrobial activity of some medicinal plants. A variety of plants are reported to have antibacterial properties and these include Achyranthes aspera, Calpurinia aurea, Croton
macrostachys, Cestrum auriculatum, Iryanthera lancifolia, Lepechinia meyenii and Ophryosporus peruvianus (Kalayou et al., 2012; Rojas et al., 2003). In a study by Al-Bakri and Afifi (2007) crude plant extracts of Hypericum triquetrifolium, Ononis natrix, Ruta chalepensis, Ballota undulata and Marrubium vulgare showed great antimicrobial activity against bacteria species: Bacillus subtilis, Escherichia coli, Staphylococcus aureus and Pseudomonas aeruginosa. Some of these bacteria species were isolated from animals having endometritis (Tibary et al., 2006). Extracts from Punica protopunica, Boswellia species, Commiphora parvifolia, Buxushilde brandtii, Jatropha unicosatata, Kalanchoem farinacea and Withania species were also found to have antibacterial properties especially against all tested Gram positive bacteria (Mothana et al., 2005). The antimicrobial activity of these plants is due to the secondary metabolites such as flavonoids, volatile oils, alkaloids and polyphenols present in these plants (Herna´ndez et al., 2005; Al-Bakri and Afifi, 2007). However, there are some plant species that showed no antimicrobial activity against tested microorganisms. For instance in a study by Kalayou et al. (2012), Solanum hastifolium showed no antimicrobial activity against all tested microorganisms while Ficus caria was also found to be ineffective against Escherichia coli and Staphylococcus aureus.

In another study by Mabona et al. (2013), Pentanisia prunelloides did not show any noteworthy antimicrobial activity when it was used independently, although it only showed antimicrobial activity when it was used in combination with Elephantorrhiza elephantina. The activity of the combination of these plants is perceived to be due to synergistic interaction of the bioactive compounds in these plants (Mabona et al., 2013). However, the antimicrobial activity may vary from one species of plant to another. This variation in the antimicrobial activity of different
plants might be due to differences in terms of plant parts used, climatic condition in which the plant was growing and the solvent used for extraction because plant extracts may have different constituents based on these factors (Tekwu et al., 2012). These differences in the antimicrobial activity of medicinal plants might be considered for further studies on the isolation of bioactive compounds from crude extracts.

2.6. Summary

There are several potential benefits of using traditional medicines in goat reproduction. Traditional remedies can reduce livestock farmers' input costs and improve reproductive performance of goats. Ethnoveterinary medicines can be one of the best options for small-scale livestock farmers who cannot afford allopathic drugs or even commercial farmers who are specializing in organic animal products. Despite all these benefits, there are so many gaps concerning ethnoveterinary medicines. Therefore, there is great need to document information about ethnoveterinary medicines to prevent total loss of such knowledge. It is also imperative to continue validating the efficacy of the various materials that farmers use in traditional medicines.

2.7. References


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CHAPTER 3: Alternatives used by resource-limited farmers to enhance fertility and treat reproductive ailments of goats

Abstract

Modern technologies such as artificial hormones, artificial insemination (AI) and multiple ovulation and embryo transfer (MOET) have been developed to improve the reproductive performance of livestock but these methods are costly and inaccessible to resource-limited farmers. This therefore, leads farmers to seek cheaper and affordable alternatives such as medicinal plants and any other alternatives. This study was, therefore, conducted using a questionnaire, to document alternative approaches that resource-limited farmers use to enhance fertility and in treatment of reproduction ailments of goats. Furthermore, information on farmer perceptions pertaining to these practices was gathered. The survey revealed 10 plant species that are commonly used for reproductive health in goats. *Elephantorrhiza elephantina* (37.1%) and *Rhoicissus tomentosa* (25.7%) were the most frequently cited plant species. *Elephantorrhiza elephantina* and *R. tomentosa* recorded the highest Fidelity Level (FL) values of 92.86 and 75% respectively, for their use in the treatment of reproductive ailments. Roots were the most frequently used plant part (76.9%), and decoctions (69.2%) were used most. Farmers administered remedies to goats orally, in quantities of 275 ml using small bottles. Perceptions of the farmers on the effectiveness of the remedies they used varied from moderate to very effective. A small proportion of the farmers lacked an opinion of the effectiveness of the medicines. Validation of these remedies will provide information on efficacy and safety of alternatives used by resource-limited farmers.
**Key words:** Medicinal plants, non-plant remedies, reproductive performance.

### 3.1. Introduction

Small ruminant production is very important to livelihoods of most communal farmers and over 90% of the world goat population is raised in developing countries (Rashidi et al., 2011). However, productivity of the goat enterprise depends on the goats’ reproductive performance (Dobson and Smith, 2000). There are several reproductive ailments that affect livestock production which include: retained placenta, abortion, irregular heat and endometritis (Ouweltjes et al., 1996). Some of these ailments can result in secondary infection if not treated. Conventional drugs are important in controlling and treating infectious reproductive ailments but farmers in Eastern Cape find it difficult to purchase these drugs due to high costs since these farmers have low incomes (Masika and Afolayan, 2003; Kalayou et al., 2012). There are modern reproductive techniques such as artificial insemination (AI), multiple ovulation and embryo transfer (MOET) and estrus synchronization that have been developed to manipulate the reproductive system of goats (Lehloenya et al., 2005).

Besides, several problems have emerged with use of antibiotics and these include development of drug resistance and chemical residue in meat products (Rojas et al., 2003; Al-Fatimi et al., 2007; Kalayou et al., 2012). These problems have necessitated a search for alternative antimicrobial remedies from medicinal plants (Ahmad and Beg, 2001). There are several materials used by farmers to improve the reproductive performance of goats and to treat
reproductive ailments. Plants used by farmers are presumed to have hormonal substances and this is the reason why *Aven sativa* is used to promote estrus in animals (Viegi *et al*., 2003). According to Al-Fatim *et al*. (2007) plant extracts can be used to search for new drugs. This shows that medicinal plants still play a vital role in livestock production.

Livestock farmers try to treat reproductive ailments because they understand that reproductive problems result in loss of offspring, increased kidding interval and reduced milk production. However, the association of infectious ailments with reproductive failures is largely unknown (Gebremedhin *et al*., 2013). Livestock farmers in the Eastern Cape of South Africa have been using medicinal plants to treat livestock ailments for quite a long time. Masika *et al*. (2000) estimated that approximately 75% of resource-limited farmers who keep livestock in the Eastern Cape Province use traditional medicines to treat their animals.

Since information about traditional medicines is transmitted orally from generation to generation, there is a danger that some of the knowledge may be distorted, and this might lead to the total loss of the information as time progresses (Tshibangu *et al*., 2002; Maphosa, 2010). Hence, there is great need to document ethno-veterinary medicines and practices used by resource-limited so that medicinal plants and knowledge is preserved for the coming generations. The objective of the study was to document materials used by resource-limited farmers to manipulate reproduction system in goats and to treat reproductive ailments as well as to assess farmers’ perception on the use of these materials.
3.2. Materials and Methods

3.2.1. Description of study site

The study was conducted in Mount Frere area found in Alfred Nzo District Municipality in the Eastern Cape Province of South Africa. Mount Frere is situated 30° 54′ S 28° 59′ E at an altitude of 1153 m above sea level. The survey was conducted in two villages namely: Qanqu and Mhlotsheni. These two villages are characterized by extensive (communal) goat production.

3.2.2. Data collection

A snowballing technique was used to select respondents, based on knowledge of traditional remedies. According to Illenberger and Flötteröd (2012), snowball sampling is a technique for locating an initial set of information-rich respondents, denoted as seeds, who are interviewed, after which they in-turn direct the interviewer to other potential respondents. A total of 60 participants were selected purposively from Mount Frere and all participated in the questionnaire survey. During the survey, data was captured on stock-owner household demographics, livestock figures, common diseases, and materials used to manipulate livestock reproduction in Mount Frere area. Information on the methods of preparation (including quantities used during preparation) and application of plant remedies was also collected from livestock farmers. Trained enumerators assisted in interviewing the farmers in isiXhosa which is the vernacular language in the study area. Following the interviews, plant samples were collected, pressed and
identified by their vernacular names. The plants samples were taken to Giffen Herbarium at the University of Fort Hare for identification. The study protocol was approved by the ethics committee at the University of Fort Hare and the collection of plant materials was done with the assistance of a community traditional healer so that the biodiversity rights of the local people is protected.

3.2.3. Statistical analysis

PROC FREQ procedures of Statistical Analyses Systems (SAS) (2003) were used to determine frequencies for gender, household head, marital status, age, level of education, and occupation of the farmers, as well as plants species and farmers’ perceptions in terms of effectiveness of the remedies. The healing potential of the cited medicinal plant species was calculated according to the method of Friedman et al. (1986) which involves the use of an index called fidelity level (FL) which is based on the proportion of respondents who cited the use of a given medicinal plant species against a major ailment category. The formula for calculating the index is given as FL = Ip/Iµ x 100, where Ip is the number of respondents who independently cited the use of plant species for the same major ailment and Iµ represents the total number of respondents who cited the plant species for any major ailment.
3.3. Results

3.3.1. Demographic information and characteristics of farmers

There were more males (56.67%) than females (43.33%); the majority were above 70 years of age (46.67%), attended primary education as their highest level of education (66.67%), were married (51.67%), and relied on government grants and/or pension (69.49%) for survival (Table 3.1).
<table>
<thead>
<tr>
<th>Gender</th>
<th>Proportion %</th>
<th>Household head</th>
<th>Proportion %</th>
<th>Marital status</th>
<th>Proportion %</th>
<th>Age (Years)</th>
<th>Proportion %</th>
<th>Level of education</th>
<th>Proportion %</th>
<th>Occupation</th>
<th>Proportion %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Female</td>
<td>43.33</td>
<td>Father</td>
<td>58.33</td>
<td>Single</td>
<td>5.00</td>
<td>25 and below</td>
<td>0</td>
<td>Never attended school</td>
<td>3.33</td>
<td>Teacher</td>
<td>3.39</td>
</tr>
<tr>
<td>Male</td>
<td>56.67</td>
<td>Mother</td>
<td>40.00</td>
<td>Married</td>
<td>51.67</td>
<td>26-40</td>
<td>10</td>
<td>Primary level</td>
<td>66.67</td>
<td>Traditional healer</td>
<td>1.69</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Child</td>
<td>1.67</td>
<td>Widowed</td>
<td>40.00</td>
<td>41-55</td>
<td>23.33</td>
<td>Secondary level</td>
<td>16.67</td>
<td>Police officer</td>
<td>1.69</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Separated</td>
<td>3.33</td>
<td>56-70</td>
<td>20.00</td>
<td>University</td>
<td>5.00</td>
<td>Self employed</td>
<td>23.73</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Above 70</td>
<td>46.67</td>
<td>Grant/Pension</td>
<td>69.49</td>
</tr>
<tr>
<td>Total</td>
<td>100</td>
<td></td>
<td>100</td>
<td></td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td></td>
<td>100</td>
</tr>
</tbody>
</table>
3.3.2. Farmers’ perceptions and knowledge on kidding characteristics and use ethno-veterinary medicines for reproduction purposes

Of the farmers interviewed 39.13% reported their goats kid in winter, 24% in summer, 14.49% in autumn, 11.59% in spring and 10.44% all year round (Figure 3.1). In the majority of cases (76.4%) farmers reported having 1-5 does in a flock, which had previously produced twins. However, a small proportion (1.8%) of the farmers reported having more than 15 does in their flocks that produced twins (Figure 3.2).

Close to 28% of the farmers frequently used medicinal plants whereas 22% used zootherapeutic (animal derived) remedies to manipulate goat reproduction (Figure 3.3). Most farmers (76.36%) reported that 1 to 5 does in their flock gave birth to twins or triplets (Figure 3.4). Although the majority of the farmers used conventional drugs (38%), relatively high proportion 32 and 26% used medicinal plants and non-plant remedies, respectively. Farmers perceived the effectiveness of the various remedies to be different; 54.7, 32.1 and 25% considered conventional drugs, medicinal plants and non-plant materials to be very effective, respectively. On the other hand, 37.7, 34 and 33% reported these remedies to be moderately effective. About 25% of the respondents reported non-plant remedies to be ineffective. Small proportions of farmers did not know any differences in the effectiveness of the remedies (Figure 3.5).
Figure 3.1: Percentages of different kidding seasons
Figure 3.2: Farmers’ experiences with multiple births in their flocks
Figure 3.3: Type of remedy and frequency of use for manipulation of the reproductive system of goat
Figure 3.4: Type of remedy and frequency of use for reproductive ailments in goats
Figure 3.5: Farmers’ perceptions on the effectiveness of type of remedy used for reproduction purposes and treatment of reproductive ailments
3.3.3. Medicinal plants used by farmers to manipulate the reproductive system of goats and to treat reproductive ailments

A total of 10 plant species were reported to be used by farmers for the manipulation of reproduction and treatment of reproductive ailments (Table 3.2). *Elephantorrhiza elephantina* (37.1%) and *Rhoicissus tomentosa* (25.7%) were most used, and these two plants had higher fidelity level values than others (Table 3.3). All respondents frequently purchased prepared medicines called *Isihlambezo* which is a mixture of different plants, only known by the herbalist. A few farmers (3.33%) used animal parts in mixing their remedies, which included the intestines of a porcupine and a mole. They are dried, crushed, mixed with water and drenched to the goats. All the respondents (100%) obtained knowledge on the use of medicinal plants from their elders, which mean information was passed orally from generation to generation. However, they indicated that the younger generation was not interested to learn about these traditional medicines.

Different plant parts were used to prepare medication; roots (76.9%), bark (15.4%) and leaves (6.7%) (Table 3.2). In the majority of cases (69.2%) decoctions were used, 23.1% used infusions and only 7.7% ground the material into powder. All the respondents administered medicines to goats orally using a 275 ml bottle. The frequency of treatment varied from one farmer to another. However, a larger proportion of farmers (70%) administered the remedies during summer.
Farmers’ experience with the use of plant remedies ranged from 20 to 70 years. Sharing of information on the plants used in the formulation of remedies is not readily done, 40% of the respondents indicated that they do not want to share the knowledge about medicinal plants with others, while 60% of the respondents would want the knowledge to be shared. Furthermore, the majority (70%) of the farmers were of the opinion that ethno-veterinary medicine should be included in the curricula of schools, colleges and universities. Most farmers who had knowledge about ethno-veterinary medicines did not support the idea of sharing the knowledge about medicinal plants.
Table 3.2: Medicinal plants used for reproduction purposes in goats

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Local name</th>
<th>Common names</th>
<th>No of citations</th>
<th>Part used</th>
<th>Preparation methods</th>
<th>Usage</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Elephantorrhiza elephantine</em></td>
<td>Intolwane</td>
<td>Elephant’s root</td>
<td>13</td>
<td>Roots</td>
<td>Decoction</td>
<td>Retained placenta; abortion; womb cleansing; fertility</td>
</tr>
<tr>
<td><em>Rhoicissus tomentosa</em></td>
<td>Isaqoni</td>
<td>Wild grape</td>
<td>9</td>
<td>Roots</td>
<td>Infusion</td>
<td>Fertility; abortion; retained placenta</td>
</tr>
<tr>
<td><em>Asparagus macowani</em></td>
<td>Umviti</td>
<td>Asparagus</td>
<td>4</td>
<td>Bark</td>
<td>Decoction</td>
<td>Fertility; induce estrus</td>
</tr>
<tr>
<td><em>Noltea Africana</em></td>
<td>Maluleka</td>
<td>Soap bush</td>
<td>2</td>
<td>Roots</td>
<td>Grinding into powder</td>
<td>Womb cleansing; fertility</td>
</tr>
<tr>
<td><em>Rhamnus prinoids</em></td>
<td>Unyenye</td>
<td>Dog wood</td>
<td>2</td>
<td>Roots</td>
<td>Infusion</td>
<td>Twin or triplets production</td>
</tr>
<tr>
<td><em>Bidens pilosa</em></td>
<td>Uladolo</td>
<td>Black Jack</td>
<td>1</td>
<td>Roots</td>
<td>Decoction</td>
<td>Fertility; induce estrus</td>
</tr>
<tr>
<td><em>Laportea peduncularis</em></td>
<td>Ubusi</td>
<td>River nettle</td>
<td>1</td>
<td>Roots</td>
<td>Decoction</td>
<td>Twin or triplet production</td>
</tr>
<tr>
<td><em>Duvernoia adhatodoides</em></td>
<td>Ihlehlwe</td>
<td>Pistol bush</td>
<td>1</td>
<td>Leaves</td>
<td>Decoction</td>
<td>Fertility; milk let down</td>
</tr>
<tr>
<td><em>Pentanisia prunelloides</em></td>
<td>Icimamlilo</td>
<td>Wild verbena</td>
<td>1</td>
<td>Roots</td>
<td>Decoction</td>
<td>Abortion</td>
</tr>
<tr>
<td><em>Dicoma zeyheri</em></td>
<td>Umlunge</td>
<td>Toy sugar bush</td>
<td>1</td>
<td>Roots</td>
<td>Infusion</td>
<td>Abortion</td>
</tr>
</tbody>
</table>

Decoction = heating in water to boiling point

Infusion = soaking in water at ambient temperature overnight
Table 3.3: Fidelity Level values of medicinal plants cited by two or more informants for being used for reproductive purposes

<table>
<thead>
<tr>
<th>Plant name</th>
<th>Major ailment group</th>
<th>(I_p)</th>
<th>(I_\mu)</th>
<th>FL value (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Elephantorrhiza</em></td>
<td>Retained placenta; abortion; womb cleansing and fertility</td>
<td>13</td>
<td>14</td>
<td>92.86</td>
</tr>
<tr>
<td><em>elephantina</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Rhoicussus</em></td>
<td>Fertility; abortion; retained placenta</td>
<td>9</td>
<td>12</td>
<td>75</td>
</tr>
<tr>
<td><em>tomentosa</em></td>
<td>placenta</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Asparagus</em></td>
<td>Fertility</td>
<td>4</td>
<td>7</td>
<td>57.14</td>
</tr>
<tr>
<td><em>macowani</em></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Rhamnus prinoids</em></td>
<td>Twin or triplet production</td>
<td>2</td>
<td>6</td>
<td>33.33</td>
</tr>
<tr>
<td><em>Noltea Africana</em></td>
<td>Womb cleansing; fertility</td>
<td>2</td>
<td>7</td>
<td>28.57</td>
</tr>
</tbody>
</table>

\(I_p\) = the number of respondents who independently cited the use of plant species for the same major ailment

\(I_\mu\) = represents the total number of respondents who cited the plant species for any major ailment.
3.4. Discussion

The observation that farmers’ experiences in using medicinal plants ranged from 20 to 70 years, with 46.8% of the farmers with this knowledge being above 70 years is in line with the previous study of Maphosa (2010). The author reported that the majority of people with knowledge about traditional medicines were elderly people. This presents a threat to the knowledge concerning traditional medicines since the elders of the communities are passing on (Mathias, 2005). The situation is worsened by the fact that the younger generations are finding it difficult to appreciate traditional medicines.

The study revealed that most people used traditional medicines in one way or another in livestock. This might be due to the fact that traditional medicines are readily available and cheaper as compared to conventional drugs (Mwale et al., 2005). Considering that the majority of the farmers were not employed but depended on government grants, the low cost of these medicines presents a very attractive option to them. Farmers regarded traditional medicines to be less toxic and as such are considered not to have residual effects in animal products such as meat and milk. This agrees with the study of Wanzala et al. (2005) who reported that medicinal plants do not produce toxic residues in animal products. In a study by Luseba and Van der Merwe (2006) farmers also reported that there are no restrictions or withdrawal periods for the consumption of animal products such as meat and milk. Generally traditional medicines are perceived to have no harmful effects (Balaji and Chakrarthi, 2010). Medicinal plants are known to contain saponins which are converted through various metabolic processes to sapogenins,
which are precursors of steroid hormones (Gurib-Fakim, 2006). However, it is not known if these compounds may end up in animal products. Hence this could be further researched on.

Farmers in the study area, did not have a fixed breeding season for their goats, 39.1% of those who gave traditional medicines to their animals had goats kidding in winter. Ideally, goats should kid in spring time, which is around September – October, in South Africa. The season of kidding has an influence on puberty (Rekik et al., 2012). In the study by Greyling (2000), the recorded mean age at the onset of puberty in the Boer goat does was 191.1 and 157.2 days for does born in late winter and mid-summer, respectively.

Farmers administered remedies to their animals during summer to ensure that their animals are generally healthy, as they prepare to conceive. According to Viegi et al. (2003) farmers administer prepared remedies to promote estrus in their livestock. Animals treated with medicinal plants are expected by farmers to produce twins or triplets. These medicinal plants are perceived to contain hormonal substances (Viegi et al., 2003; Gurib-Fakim, 2006) that induce multiple ovulation which can subsequently lead to twins and triplets. Furthermore, farmers administer remedies during summer because there are a lot of plants available, which is also the rain season in the Eastern Cape Province, of South Africa. Some of the plants might not be available during the dry season for instance in winter.
Most of the farmers (76.4%) reported that a range of 1 -5 does in a flock give birth to twins or triplets. Ovulation rate has a major influence on litter size and reproductive efficiency, but it is affected by stage of the breeding season, parity, nutrition and genotype (Rekik et al., 2012). The current study revealed that, 45.3% of the respondents perceived that traditional medicines (both medicinal plants and non-plants medicines) are very effective while 54.7% of the respondents reported that conventional drugs are very effective. But the majority of the farmers perceived that traditional medicines are moderately effective. This shows that most farmers believe that traditional remedies are moderately effective. In another study, Fielding (1999) reported that farmers perceived that traditional medicines are effective since they validated them in their own ways through many trials. Farmers have been using traditional medicines over a long period of time, (Kalayou et al., 2012) that is the reason why they believe that they are effective. There were, however, more farmers who did not know the effectiveness of traditional remedies compared to the effectiveness of conventional drugs. Researchers also have different perceptions on the effectiveness of traditional medicines, Haverkort et al. 1996 argues that traditional remedies are ineffective, but on the other hand, Fielding (1999) states that traditional medicines are totally effective. Effectiveness could differ depending on the measure used to assess it, and efficacy of the same plant may differ due to its source (Obomsawin, 2008). This means there is need to carry out scientific validation of traditional medicines. Furthermore, efficacy can be influenced by several other factors such as season, time of the day, area, methods of preparation, concentration used, frequency of administration and parts used. Farmers might also do wrong diagnosis such that treatment would not bring about the effectiveness. This is why it is important to carry out efficacy test on these medicines so that farmers will know whether they are effective or not.
Overall 10 plant species used by resource-limited farmers, to address reproduction and in treatment of reproductive ailments of goats were recorded. One of the plants, *E. elephantina*, was frequently mentioned, and which has previously been recorded to be used for medicinal purposes in humans and livestock (Maphosa *et al.*, 2010). Farmers also buy prepared decoction from traditional healers, the most popular one was *isihlambezo* which means “that which cleans” (Medical Association of South Africa, 1997). This remedy is perceived to be effective in both humans and livestock because it is a mixture of different medicinal plants, which seem to have synergistic effect. Its popularity might be a result of its recommendation by several traditional healers (Varga and Veale, 1997). The same remedy is used by many Zulu women for reproductive health during pregnancy (Varga and Veale, 1997). The study was conducted in an area with people from the Xhosa and Zulu tribes, so the use of *isihlambezo* by the Zulu people could have influenced this wide use in the study area. Although all farmers who bought *isihlambezo* did not know the ingredients used in it, but only known by the herbalist, a study by Varga and Veale (1997) revealed some medicinal plants which are used to prepare this remedy and these include; *Asclepias fruticosa*, *Scadoxus puniceus*, *Typha capensis*, *Veronia neocorymbosa*, *Rhoicissus tridentata*, *Pentanisia prunelloides*, *Clivia miniata*, *Gunnera perpensa*, *Combretum erythrophyllum* and *Callilepis laureola*. Plants of the genus *Asclepias* are reported to contain glycosides (Warashina *et al.*, 2011), and this might be also the case of *Asclepias fruticosa*. Lin *et al.* (1999) citing Hutching *et al.* (1996) reported that *Typha capensis* is used during pregnancy to ensure easy delivery. This same plant is also reported to contain several novel compounds such as thyphaphthalide, typharin, sitosterol and flavonoids (Lin *et al.*, 1999). *Rhoicissus tridentata* belongs to the Vitacea family together with *Rhoicissus tomentosa*. 
*Rhoicissus tridentata* is known to have anti-microbial and anti-inflammatory properties (Lin *et al*., 1999). McGaw *et al*. (2005) perceives that the use of *Gunnera perpensa* for treating reproductive ailments in humans and animals is due to its anti-microbial effects. Among these medicinal plants two plants were also cited by farmers to be of use for reproductive health and these are *Rhoicissus spp* and *Pentanisia prunelloides*.

Different plant parts were used in the formulation of remedies. Roots were mostly cited, however, these findings differ with those in studies by Masika and Afolayan (2003) and Maphosa and Masika (2010), who reported that most farmers used leaves in the preparation of herbal remedies. They also differed with findings of the study by Beníteza *et al*. (2011) who reported wide use of stems. The differences in parts used could be attributed to the medicinal plants used and nature of ailments to be treated, for instance the above mentioned authors were focusing on internal and external parasites whereas the current study focused on traditional remedies used for reproduction purposes. Use of roots have got advantages in the sense that they can be available all year round, but the only challenge is that the use of roots has destructive effects because sometimes the whole plant has to be uprooted when the farmers are harvesting the roots.

According to Maphosa and Masika (2010) there are conservational advantages that are associated with the use of leaves, though they might be seasonally available. The reason why farmers use roots frequently is that they believe that roots are close to the soil and they have got more bioactive compounds than any other plant part. However, this disagrees with the results of
Sulaiman and Balachandran (2013) where roots were reported to contain less secondary metabolites such as phenolics, flavonoids and alkaloids as compared to the stem. In another study by Menghini et al. (2014) revealed that the highest amount of secondary metabolites are present in flowers and leaves. This explains why people collect blooming aerial plant parts to prepare traditional remedies (Menghini et al., 2014). The availability of roots is not also affected by the season like leaves, which might not be available during the winter season. But once a root is harvested it will take a long time for a plant of the same type to grow. However, another strategy that could be used to maintain a sustainable harvesting of medicinal plants without damaging the whole plant could be to substitute roots with stem or leaves of the same plant provided they result in same healing effects (Sulaiman and Balachandran, 2013).

Farmers used a combination of medicinal plants when preparing decoctions because they perceived this to result into a stronger medicine, than one from a single plant. When more than one medicinal plant is used during the preparation of herbal remedy, it is believed that more than one ailment can be treated with same remedy due to the synergistic effect (Gurib-Fakim, 2006). For instance farmers indicated that they mix *E. elephantina* and *R. tomentosa* to make a remedy they use to enhance fertility in livestock and they believe that the plants produce better results when they are used in combination. These two plants are reported to contain polyphenolic compounds, which possess high anti-oxidative activity (Naidoo, 2004). Plants with anti-oxidative properties are of paramount importance in improving the reproductive performance of livestock because excess free radicals have got serious adverse effects on the reproductive system of livestock (Agarwal et al., 2004). In male animals, an imbalance of free radical generation and detoxification can result in oxidative stress which can damage sperm and
testicular tissues resulting in poor semen quality (Rong-zhen and Dao-wei, 2013). Therefore, the use of plants with anti-oxidative compounds will improve semen quality and reproductive performance of animals. In females, oxidative stress might result in irregular estrus cycle as a result of ovary dysfunction (Gupta et al., 2011). Generally, reproductive losses can be reduced by using plants containing anti-oxidative compounds. This might be the reason why farmers use these plants for reproductive purposes since they contain anti-oxidants (Gupta et al., 2011), a fact they might not know.

The FL values were calculated for all the cited plants. Elephantorrhiza elephantina (92.86%) and R. tomentosa (75%) had the highest FL values for their use to treat reproductive ailments such as abortion, retained placenta and fertility. On the other hand, medicinal plants such as Rhamnus prinoids (33.33%) and Noltea Africana (28.57%) scored lower FL values for their use on twin or triplet production and fertility enhancement. This could be due to the fact that few people use these plants for reproductive purposes although they are used to treat other ailments. Medicinal plants scoring higher FL values are perceived to be more potent than those plants with less FL values (Trotter and Logan, 1986). Therefore, the high FL values scored by E. elephantina and R. tomentosa indicate that these plants have high healing potential towards reproductive problems, and according to Giday and Teklehaymanot (2013) medicinal plants that scores the highest FL values should be given much attention. A plant like E. elephantina is used as an aphrodisiac in human males (Abdillahi and Van Staden, 2012). This suggests that this plant might have hormonal substances and this might also be the reason why farmers use it for reproduction purposes in livestock. In this study, E. elephantina was used for cleansing the womb after animals abort and also to enhance fertility. This agrees with Moyo et al. (1999) who reported that
the same plant is used to prepare remedies for problems related to reproductive organs such as the penis, vulva and womb problems as well as for cleansing the womb after abortion. The medicinal properties of *E. elephantina* could be due to the flavonoids, gallic derivatives and phenolic derivatives it contains and these compounds have got anti-microbial activity (Naidoo, 2004). Farmers also used the same plant to treat reproductive ailments especially secondary infections which might result after abortion. This suggests that this possesses antibiotic properties (Masika and Afolayan, 2003; Maphosa and Masika 2010), which are due to condensed tannins and triterpenoids compounds (Naidoo, 2004). Farmers reported that they commonly use *R. tomentosa* for treatment of animals that regularly abort and also to cleanse the womb after abortion, which agrees with the findings of McGaw and Eloff (2008). The same medicinal plant was identified to be one of the ingredients for *isihlambezo* which is a mixture of medicinal plants used to treat reproductive ailments in Zulu females (Varga and Veale, 1997).

Plants that belong to the Ebenaceae family for example *N. africana* were also reported to be used by farmers for womb cleansing after abortion. These plants have been reported to treat several diseases since they are believed to produce naphthoquinones in the form of dimmers and triterpenes. These naphthoquinones are known for their antimicrobials activities (Tangmouo *et al.*, 2009). *Bidens pilosa* is known to contain essential oil that have antibacterial, antifungal and antioxidant properties (Deba *et al.*, 2008). Besides detoxification of free radicals, anti-oxidants such as polyphenols are known to stimulate cell proliferation (Naidoo, 2004). These essential oils that are found in this plant are responsible for the strong smell that is produced by this plant. In South Africa this plant is used to treat a wide range of ailments in both humans and livestock (Deba *et al.*, 2008). The same plant is also reported to contain alkaloids, saponins, triterpens,
polyacetylenes and flavonoids, and in the rest of Africa it is also used to treat malaria (Abajo et al., 2004). Since *B. pilosa* is reported to have essential oils that have antimicrobial properties, this might suggest why farmers use this plant for cleansing the womb after abortion in goats. *Laportea penduncularis* comes from the Urticaceae family and plants from this family are known to be used to treat ailments like jaundice, dysentery and diarrhoea (Sharma et al., 2001). The antihelmintic properties of *R. tomentosa* have been reported by McGaw and Eloff (2008). However, farmers in the Eastern Cape province of South Africa also use it to treat reproductive ailments like vaginitis and endometritis. *Pentanisia prunelloides* is reported to have antimicrobial activities and it has synergistic interactions with *E. elephantina* when they are mixed together (Mabona et al., 2013). In addition, this plant is also known to have anti-viral properties and also contains substances that stimulate uterine contractions during labour. Therefore, this is important especially in the case of retained placenta.

Most plants are reported to possess various medicinal properties that include antimicrobial, anti-inflammatory and immunoregulation, however it is important to narrow down to those medicinal plants that can be used for reproductive purposes and to treat reproductive ailments. The study revealed that farmers use medicinal plants for fertility, cleansing of the womb after abortion, induce estrus and also to stimulate milk production in goats. However, there is limited research done on medicinal plants used by farmers for reproduction purposes although farmers indicated that they use medicinal plants for reproduction. Beside the use of medicinal plants, farmers also used animal parts as medicines; findings of this study are in agreement with Sharma *et al.* (2001). However, the morphological and behavioral characteristics of the animal used for
medicinal purposes are perceived to determine the therapeutic use of that animal in question (Alves and Alves, 2011).

Farmers reported that they use porcupine and mole intestines for fertility on their goats. This is in agreement with Alves and Rosa (2005) who reported that people use animal viscera as medicines. There are various animal parts and products that are used to prepare traditional medicines and these include: horns, leather, dermal plates, heart, blood, bile, viscera, bone, eyes, fat and meat (Alves and Alves, 2011; Souto et al., 2011). The treatment of human or animal ailments with remedies made from animals and their products is referred to as zootherapy (Alves and Alves, 2011). Zootherapeutic remedies are used to treat a wider range of ailments, in the study of Souto et al. (2011) farmers mentioned that they use zootherapeutic remedies to treat uterine prolapsed, retained placenta and to tame angry animals. This agrees with the current study where the intestines of the porcupines and moles were used for enhancing fertility and treat retained placenta. Alves and Rosa (2005) reported that animals can be an important source of drugs for modern science since currently 8.7% of the essential chemicals selected by the World Health Organization come from animals and 11.1% from plants. However, from a study of Souto et al (2011), 89% of the remedies were prepared from dead animals. Since most zootherapeutic remedies are prepared from wild animals (Alves and Alves 2011) there is a danger of over exploitation of these animals which can subsequently lead to extinction of some animal species. There are also some chances of transmission of diseases to the patient being treated using therapeutic remedies because organs and tissues can be a source of Salmonella infections causing diarrhoea and endotoxic shock (Alves and Alves, 2011). Hence, there is great need to carry out research on the safety of using these therapeutic remedies. According to Alves and Alves (2011)
the effectiveness of most of the medicines from wild animals has not been scientifically proven and their potency can be questioned. There is, therefore need to carry out studies on the pharmacological constituents of these animal parts used by farmers.

3.5. Conclusion

The study revealed the role traditional remedies play in goat farming of resource-limited farmers in Mount Frere. Goat farmers medicate their animals using medicinal plants and animal parts to improve reproductive performance and treat reproductive ailments in goats. Furthermore, farmers had different perceptions on the use of traditional medicines especially on their effectiveness. If their efficacy could be determined, traditional medicines can be the best alternatives that could be used for reproduction purposes since they are cheap, readily available and environmentally friendly. According to Souto et al. 2011 attention should be given to traditional medicines due to their socio-economic, conservationist and cultural components. Therefore work still needs to be done to validate the safety and efficacy of the cited plants.

3.6. References


Warashina T., Umehara K., Miyase T. and Noro T., 2011. 8,12;8,20-Diepoxy-8,14-secopregnane glycosides from roots of *Asclepias tuberosa* and their effect on proliferation of human skin fibroblasts. *Phytochemistry*, **72**: 1865 – 1875.
CHAPTER 4: Effects of Elephantorrhiza elephantina and Rhoicissus tomentosa on hormone profiles and pregnancy rate in non-descript goats

Abstract

Small-scale livestock farmers in rural areas in the Eastern Cape Province widely use medicinal plants to influence reproduction in goats. However, information on the efficacy of plants used is lacking. This study was conducted to determine the efficacy of Elephantorrhiza elephantina and Rhoicissus tomentosa on manipulating the reproduction system of non-descript goats. Elephantorrhiza elephantina and R. tomentosa were selected for validation because they were the most widely used plants. An in-vivo assessment was done to determine efficacy using serum estradiol and progesterone profiles. Does were divided into 4 groups of 6 does each, to which respective aqueous extracts of; E. elephantina, R. tomentosa, E. elephantina and R. tomentosa mixture (1:1 ratio) and distilled water, were administered orally. Blood was collected before administration of extracts and at intervals of 6, 18, 30 and 42 hours after administration of plant extracts and analysed for serum estradiol and progesterone concentration. Animal body weights and body condition scores (BCS) were recorded 2 hours before administration of the plant extracts and then after every fort-night for 16 weeks. After 42 hours, does administered with aqueous extracts of E. elephantina and R. tomentosa mixture had the highest (P<0.05) serum estradiol and progesterone concentration and goats administered with distilled water had the least (P<0.05). Does administered with a mixture of aqueous E. elephantina and R. tomentosa extract had the highest (P<0.05) body weight (65 kg) and those administered with distilled water had the least body weights (54 kg). Animals administered with E. elephantina and the mixture of E.
**elephantina** and *R. tomentosa* had the highest body condition (P>0.05) scores while the control group had the least body condition scores (BCS). Does that were administered with a mixture of *E. elephantina* and *R. tomentosa* were all pregnant with twins (100%), confirmed by a doppler ultrasound scan. The study revealed that medicinal plants used by farmers to influence reproduction had an influence on the hormonal concentrations, conception and body weights of does, and therefore is effective for the purpose for which they are used.

**Key word**: Serum estradiol, progesterone, body weights, body condition score

### 4.1. Introduction

Small ruminant animals are expected to contribute large proportions of meat to the global market in the coming future (Fuah and Priyanto, 2013). In order to enhance productivity of goats, it is important to have some adjustments in terms of goat reproduction, but the problem is that little information is available on the reproduction aspects in goats (Chentouf et al., 2011). Poor reproductive performance is a serious problem in goats raised by resource-limited farmers (Fuah and Priyanto, 2013). According to Notter (2012), in most parts of the world where goats are raised under extensive production system, twinning is rare. Although there are modern techniques that have been developed to increase the incidence of twinning using synthetic hormones, most resource-limited farmers cannot afford them due to their high costs and also due to lack of knowledge about these options. The situation is also worsened by the fact that most resource-limited farmers who raise goats lack capital and credit facilities (Fuah and Priyanto,
2013), and this makes it difficult for them to access modern methods of manipulating reproduction in goats.

In addition, some countries belonging to the European Union adopted restrictions on the use of synthetic hormones on livestock because of consumer demand for natural ways of manipulating reproduction in livestock (Delgadillo et al., 2014). Several medicinal plants are used by resource-limited farmers to improve reproductive performance in livestock, specifically goats. Farmers use medicinal plants singly or in combination to improve fertility and to induce estrus in livestock (Lans and Brown, 1998). Although farmers use medicinal plants, there is less attention that has been given to plant bioactive compounds which enhance female sexual behaviour (Lans and Brown, 1998). Furthermore, the economic impact of plant remedies on the reproduction system of goats is not well understood (van Vuuren, 2008).

Some plant species like Dioscorea villosa have been used for hormone replacement therapy (Hsu et al., 2008). Others such as Loranthus species are used to improve kidding rate in rabbits while Pauzzozia mixta is used to treat reproductive ailments such as retained placenta (Matekaire and Bwakure, 2004). Plants that are used to treat retained placenta are said to have both mechanical and biological properties (Luseba et al., 2007). However, the same author outlined that when treating retained placenta, the changes that occur during digestion when the plant mixture is given orally is not known. Some medicinal plants are given to induce estrus in livestock, for example Avena sativa and Prunus avium (Viegi et al., 2003). Elephantorrhiza elephantina is another plant species which is used for reproduction purposes as reported in the previous chapter (Abdillahi and van Staden, 2012).
Elephantorrhiza elephantina is a non-climbing shrub and low growing suffrutex with red tubers (Quattrocchi, 2012). This plant is found in Southern Africa in countries including Namibia, Botswana, Swaziland, Mozambique, Zimbabwe and South Africa (Jansen, 2005). Elephantorrhiza elephantina has also been used in humans to treat painful menstruations and it is also taken by women against infertility as well as an aphrodisiac (Quattrocchi, 2012). As reported in the previous chapter, Rhoicissus tomentosa is another plant that is used for medicinal purposes. This plant is a fast growing climber with ranked grape-like leaves which can later produce wine-red grape-like berries (Kirsten, 2001). The roots of R. tomentosa are used to treat ailments like hernia, stomach ache and bilharzia (Quattrocchi, 2012). Although these plants have been used for reproduction purposes, validation of their efficacy is still lacking. The objective of the study was to determine the efficacy of E. elephantina and R. tomentosa on manipulating the reproduction system of non-descript goats by using serum estradiol and progesterone profiles as indicators.

4.2. Materials and Methods

4.2.1. Study site description

The study was done at the University of Fort Hare Honeydale Farm. It is situated 32.8° S and 26.9° E at an altitude of 520m above the sea level. The mean annual rainfall and temperature is 480mm and 18.7°C, respectively. The area is generally flat with few steep slopes (Ngambu,
2011). The farm is found in the False Thornveld of the Eastern Cape of South Africa (Acocks, 1975).

4.2.2 Collection of plant material

The tubers and roots of *E. elephantina* and *R. tomentosa* were collected in November 2013 at Mount Frere area, Alfred Nzo District Municipality, in the Eastern Cape Province. The village in which the plants were collected is situated 30° 54′ S 28° 59′ E at an altitude of 1153m above sea level. The plant parts were collected with the assistance of a herbalist. Plant identification was done at the University of Fort Hare herberium and plant specimen were deposited at the University of Fort Hare herberium.

4.2.3 Preparation of plant extract and dosage

The tubers of *E. elephantina* and the roots of *R. tomentosa* were air dried at room temperature to get constant weights. The dried materials were ground separately into powder, using a laboratory grinder with a 2 mm sieve. Weights of 300 g for each plant powder were mixed with 3000 ml of distilled water and boiled for 10 minutes under atmospheric pressure. The obtained decoction was filtered under reduced pressure using Whatman No 1 filter paper. The filtrate was lyophilized using a freeze dryer (Alpha 1 – 2 LD plus, Christ Lasec, German) for 48 hours, yielding 58 g and 52 g of *E. elephantina* and *R. tomentosa*, respectively. The extract powder was
stored in sealed bottles and kept in a desiccator until testing (Maphosa, 2010). Dilutions were prepared from the initial extract using distilled water to obtain extract solutions at concentrations of 100 mg/ml. Farmers mixed 1 table spoon of *Elephantorrhiza elephantina* and 1 table spoon *Rhoicissus tomentosa* in 275 ml of water, which translated to 600 mg/kg of body weight of the animal. The dose used in the study was similar to that used by farmers.

### 4.2.4. Animals

A total of 24 non-descript does at 2.5± 0.2 years of age, weighing 43.5±4.5 kg and all in the anoestrus period were used in the study. Animals used in this study were never dosed against worms. The animals were reared at Honeydale farm and were identified using numbered ear tags. Goats were grazed and browsed on the natural pastures and *Acacia karroo*, respectively, for 7 hours under the supervision of a farm worker. The animals were kept in pens during the night and were allowed free access to fresh water (unrestricted access to water). The ethical issues were considered to make sure that the animals are not overcrowded in pens and that they are given freedom to access feed and water. The study was approved by the ethics committee at the University of Fort Hare and the certificate reference number was MUC0121SRW001.
Figure 4.1: Administration of the plant extracts to the goat

4.2.5. Experimental design

Does were completely randomized into 4 groups of 6 each. A dose of 600 mg/kg was used for the aqueous extracts of *Elephantorhiza elephantina*, *Rhoicissus tomentosa*, mixture of *Elephantorhiza elephantina* and *Rhoicissus tomentosa* (1:1 ratio). Animals in the negative control group were given 275 ml of distilled water per animal per day once. The aqueous plant
extracts and distilled water were given to the animals orally using a dozing gun (Figure 4.1). After administration of plant extracts the does were allowed to move together with bucks to allow natural mating.

4.2.6. Female fertility study

All animals used in the study were in anoestrus period, and not pregnant, as determined by a Doppler ultrasound scan (Figure 4.3). Animals were randomly distributed into groups A, B, C, and D, which were administered with the aqueous extracts of *E. elephantina*, *R. tomentosa*, mixture of *E. elephantina* and *R. tomentosa* (1:1 ratio) and distilled water, respectively. The animals were weighed and blood samples collected 2 hours before administration of the plant extracts and distilled water. Thereafter, blood samples were collected at intervals of 6, 18, 30, 42 hours after administration of the plant extracts and distilled water. Blood was collected from each animal by jugular veni-puncture using heparized vacutainers (Figure 4.2). The blood was centrifuged at 3000 rpm for 10 minutes; serum was collected and used for hormone profile assay. The Coat-A-Count estradiol radioimmunoassay procedure was used to determine the quantitative measurement of estradiol in serum, while the Coat-A-Count progesterone radioimmunoassay was used to measure the levels of progesterone in serum (Khanum et al., 2008). After 16 weeks, does were tested again for pregnancy using a Doppler ultrasound scan.

Body condition scoring and weighing of the animals was done after every 2 weeks over a period of 16 weeks. The animals were weighed using a small stock balance (Kattleway Calf Mass Meter, Serial Number A13) and body condition scoring was done by visual assessment of the
animal’s body, as described by Ngwa et al. (2007). Scoring was done on a scale of 1 – 5, using the backbone, flank, ribs, and spinous processes of the lumbar vertebrae as reference points.

Figure 4.2: Blood collection from the jugular vein of the goat
4.2.7. Statistical analyses

Data were checked for normality using PROC UNIVARIATE. Body condition scores were square root transformed since the data were not following the normal distribution. The effects of time and treatment on the hormone concentration, body weights and body condition scores of goats were analyzed as repeated measures using PROC General Linear Model (GLM) of SAS (SAS 2003).

4.3. Results

4.3.1. Effects of different remedies on hormone concentration

The effects of different aqueous plant extracts and distilled water on serum estradiol in does are shown on Figure 4.5. There was a difference (p<0.05) in the estradiol concentration between the control group and other treatment groups during the study period. Does that were given a mixture of *E. elephantina* and *R. tomentosa* aqueous extracts had the highest (p<0.05) increase in the mean estradiol concentration and peaked (7.74±1.360 pg/ml) after 18 hours while those given distilled water showed the lowest (p<0.05) increase in the mean estradiol concentration which had mean concentration of 3.9±1.360 pg/ml after 18 hours. However, after 30 hours, does treated with aqueous extracts of *E. elephantina* showed a sharp drop (P<0.05) in estradiol concentration. Then after 42 hours, does administered with aqueous extracts of a mixture of *E. elephantina* and *R. tomentosa* had the highest (P<0.05) serum estradiol concentration (5.67 pg/ml).
At time zero, the does had almost similar serum progesterone concentration (14.11±0.615 nmol/l) (Figure 4.6). After administration of *E. elephantina*, *R. tomentosa* and a mixture of *E. elephantina* and *R. tomentosa* aqueous extracts to respective groups there was a rise in serum progesterone concentration in all the treated animals. After 6 hours, does administered with a mixture of *E. elephantina* and *R. tomentosa* had the highest progesterone concentration (31.4±3.243 nmol/l) followed by those treated with *E. elephantina* (23.03±3.243 nmol/l) and the lowest (17.56±3.243 nmol/l) in the control group (Figure 4.6). Even though there was a fall in progesterone concentration after 6 hours in does administered with a mixture of *E. elephantina* and *R. tomentosa*; does treated with *E. elephantina* showed a continuous rise in progesterone concentration, but which dropped after 18 hours while those administered with *R. tomentosa* started to show a rise again in progesterone concentration up to 42 hours. After 30 hours the concentration of progesterone in does administered with *E. elephantina* continued to drop while those administered with a mixture of *E. elephantina* and *R. tomentosa* showed a rise in progesterone concentration and after 42 hours this group of animals had the highest (P<0.05) progesterone concentration (35.6±3.243 nmol/l), while the control group had the lowest (22.59±3.243 nmol/l).
Figure 4.3: Pregnancy diagnosis using Doppler ultrasound scan
Figure 4.4: Animals that were confirmed to be all pregnant with twins after administration of an aqueous mixture of *E. elephatina* and *R. tomentosa* extract.
Figure 4.5: Effect of different treatments on estradiol concentration (pg/ml)
Figure 4.6: Effect of different treatments on serum progesterone concentration (nmol/l)
4.3.2. Effects of different remedies on goat body weights and body condition score

Figure 4.7 shows the influence of different treatments on body weight. The weights of does administered with *E. elephantina* and a mixture of aqueous *E. elephantina* and *R. tomentosa* were higher than those of the does administered with *R. tomentosa* and distilled water throughout the study period, showing a sharp increase between week 6 and 12. After 16 weeks, does administered with a mixture of aqueous *E. elephantina* and *R. tomentosa* extract had the highest (P<0.05) mean body weight (64±3.729 kg) and those in the control group had the least body weights (57±3.729 kg). Generally, does administered with the aqueous extract of *E. elephantina* and aqueous extract of a mixture of *E. elephantina* and *R. tomentosa* had almost similar body weights. The average weight gains of does treated with a mixture of *E. elephantina* and *R. tomentosa*, *E. elephantina*, *R. tomentosa* and distilled water after 16 weeks were 19.13±0.402, 18.66±0.402, 14.09±0.402 and 8.08±0.402 kg, respectively.

Different remedies used in the study caused variations (P>0.05) in body condition scores (BCS) of does. On week 2, the BCS of all animal groups except for the control group were the same (1.73±0.024), but after week 2 the BCS for the animals administered with aqueous extract of *E. elephantina* and aqueous extract of *E. elephantina* and *R. tomentosa* mixture were higher than those of animals administered with aqueous extract of *R. tomentosa* and the control group throughout the study period. Animals administered *E. elephantina* and the mixture of *E. elephantina* and *R. tomentosa* had the highest BCS (1.94±0.024) throughout the study period followed by those administered aqueous extract of *R. tomentosa* (1.91±0.024) and the control group had the least BCS (1.89±0.024). (Figure 4.8)
Does in the different groups exhibited significant (P<0.05) pregnancy rates (Table 4.1). Does that were administered with the mixture of *E. elephantina* and *R. tomentosa* resulted in 100% twins (Figure 4.4). Whereas, those administered with *E. elephantina* had 100% pregnancy with 50% twins and 50% singletons. However, animals in each of the control and *R. tomentosa* treated groups, resulted in 50% pregnancy rates with singletons.
Figure 4.7: Body weights of does under different treatments.
Figure 4.8: Body condition scores of does under different treatments.
Table 4.1: Pregnancy and twinning rate of non-descript goats administered with different treatments

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<thead>
<tr>
<th>Treatment</th>
<th>Pregnancy and Twinning (%)</th>
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<tr>
<td></td>
<td>Non-pregnant</td>
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<tr>
<td><em>E. elephantina</em> aqueous extract</td>
<td>0</td>
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<tr>
<td><em>E. elephantina</em> and <em>R. tomentosa</em> aqueous extract</td>
<td>0</td>
</tr>
<tr>
<td><em>R. tomentosa</em> aqueous extract</td>
<td>50</td>
</tr>
<tr>
<td>Distilled water</td>
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</tr>
</tbody>
</table>
4.4. Discussion

Aqueous extracts of *E. elephantina* and a mixture of *E. elephantina* and *R. tomentosa* showed a great increase in serum estradiol and progesterone. This agrees with a study by Dilshad *et al.* (2012) who found that the extracts of flax seeds increased the levels of serum estradiol. The rise in serum estrogen was also observed after treating animals with *Drynaria quercifolia* rhizome extracts (Das *et al.*, 2014). This is due to the phytoestrogenic properties of plant extracts (Dilshad *et al.*, 2012). Phytoestrogens are natural phytochemicals found in plants and plant products that have similar structure and function to the synthetic estrogens such as diethylstilbesterol (Ahmad *et al.*, 2013). The major phytoestrogens found in plants include flavones, isoflavonoids and coumestans (Jing Cheung, 2000). Phytohormones can be classified as isoflavonoids, flavonoids, triterpenes, saponins, anthroquinones and lignans based on their structure (Kim *et al.*, 2008). Several phytochemicals have been isolated from *E. elephantina*, and these include β-sitosterol, phenolic compounds, flavones, catechin, flavonoid glycosides and epicatechin (Mthembu, 2007). Some of these compounds such as flavones are part of phytoestrogens that have an effect on the levels of estradiol. A mixture of flavonoid glycosides, flavones, catechin and epicatechin was reported to inhibit steroid 5α reductase enzyme (Mthembu, 2007). This action results in increased levels of testosterone. Several plants were identified to have estrogenic activities and several mammalian sex hormones were isolated from the various plant species (Khorshidi and Sherafatmandjouir, 2013). However, the possible effects of these phytoestrogens on reproductive function can differ due to age, kind of animal, sex and stage of reproduction (Ahmad *et al.*, 2013).
*Rhoicissus* species were reported to contain several compounds and these include: polyphones (gallic acid), epicatechin, proanthocyanidins, procyanidin B3 and B4, triterpenoids, siterol and sitosterolin (Opoku *et al.*, 2002; Brookes and Katsoulis, 2006). Among these compounds found in the *Rhoicissus* species, only siterol and sitosterolin were reported to cause slight estrogenic activity when the extracts of these plants are administered to animals (Brookes and Katsoulis, 2006). This agrees with the current study, in which the aqueous extract of *Rhoicissus tomentosa* showed slight estrogenic activity. In the previous study by Jing Cheung (2000) soya beans were reported to be rich in isoflavonoids, known to be phytoestrogens with structure similar to mammalian estradiol. Phytoestrogens can also be detected in the blood during hormone assays (Skipor *et al.*, 2012). Furthermore, phytoestrogens are able to trigger estrogenic activity by binding to the estrogen receptors (Guerrero-Bosagna and Skinner, 2014). However, the estrogenic and antiestrogenic effects of phytoestrogens are determined by the ratio of phytoestrogens to estrogen since phytoestrogens compete with endogenous sterooids (Adams, 1995). This might be the reason why does administered with aqueous extracts of *E. elephantina* and a mixture of *E. elephantina* and *R. tomentosa* showed a rise in estradiol levels shortly after administration of the extracts to the does. The estrogenic effects are mainly detected in ruminants such as sheep, goats and cattle since these species have relatively low levels of estradiol circulating (Adams, 1995). Estradiol is known to stimulate the growth of the uterine lining, causing it to thicken before the ovulatory phase of the oestrus cycle (Yakubu *et al.*, 2008). This may be the reason why farmers use *E. elephantina* for stimulating ovulation.

Does administered with the aqueous extracts of *E. elephantina* and a mixture of *E. elephantina* and *R. tomentosa* had the highest increase in progesterone levels. Previous studies have shown
that treating animals with plants containing phytoestrogens stimulated the release of luteinizing hormone which subsequently causes luteinisation of Graafian follicles in the ovaries resulting in the increased levels of serum progesterone (Ahmad et al., 2013). Previous studies have shown that some plant extracts have an effect on the pituitary gland resulting in decreased follicle stimulating hormone and increased luteinizing hormone which subsequently stimulate secretion of progesterone hormone into the blood (Farhoodi et al., 2011 citing Lucy, 2011). Progesterone and estrogen are vital hormones in maintaining pregnancy (Asuquo et al., 2013 citing Mukherjee, 2002).

Alkaloids in plants have been reported to reduce the concentration of estradiol and progesterone (Basin et al., 2007). These alkaloids might reduce the levels of estradiol by inhibiting aromatase (Yakubu et al., 2008). This might suggest the presence of endocrine disrupting photochemicals in R. tomentosa (Yakubu et al., 2008). A mixture of E. elephantina and R. tomentosa caused fluctuations in the levels of estradiol and progesterone. This might be due to the fact that some chemical agents in plant extracts can cause hormone imbalances (Yakubu et al., 2008). The fluctuations might also be due to antagonistic effects of compounds in E. elephantina and R. tomentosa because individual results of these two plants species showed that E. elephantina caused a significant increase in hormones while R. tomentosa caused slight fluctuations in progesterone. It is unknown whether a single type of phytohormone or rather the additive effect of more than one type contributes to the hormone dependent activities (Kim et al., 2008).
Phytoestrogens are known to increase or decrease estrogen levels and, have an influence on gonadotropin production (Johari et al., 2011). Since some phytoestrogens regulate aromatase and the activity of vital enzymes in the synthesis of estrogen, it means the transformation of androgens to estrogens in tissues will be hindered resulting in reduced estrogen production (Johari et al., 2011). The same author also reported that some phytoestrogens such as 3- and 7-dihydroxyflavone, 3- alpha 7- hydroxyl flavone and flavone inhibit 20-al-phahydroxy steroid dehydranase enzyme which is important in deactivating and transforming progesterone to 3- α5- α tetrahydroprogesterone. This causes increased level of serum progesterone (Brozic et al., 2006). Excessive consumption of plants containing high concentrations of phytoestrogens has resulted in cystic ovaries and infertility (Adams, 1995). Therefore, caution should be taken to make sure that animals treated with medicinal plants containing phytoestrogens receive the right dosages to prevent these side effects. This is because high estradiol concentrations are reported to cause pregnancy loss due to its harmful effects on oviductal, follicular and uterine environments (Johnson et al., 1996).

Animals administered with aqueous extracts of E. elephantina and a mixture of E. elephantina and R. tomentosa extract showed the highest body weight gains as compared to the aqueous extracts of R. tomentosa. This disagrees with the result of Maphosa (2010) who reported insignificant differences between the goats treated with E. elephantina and the control. This might be due to the fact that Maphosa (2010) did the study for only 9 days, such that the differences in weights could not be detected but in the current study body weights were recorded over a period of 16 weeks. The body weight gains observed in does administered with aqueous extracts of E. elephantina and a mixture of E. elephantina and R. tomentosa extracts might be
primarily due to animals having been pregnant which resulted in more weight being added by foetuses. In addition, *E. elephantina* is known to have anthelmintic properties (Maphosa, 2010), which could have improved the feed conversion efficiency such that more nutrients were available to the animals, resulting in weight gain. The anthelmintic properties of *E. elephantina* are reported to be due to flavonoids, tannins and some phenolic compounds (Naidoo, 2004). The animals in the control group had the lowest live weight gains, which could have been due to the fact that they did not receive the anthelmintic remedies. Furthermore, this could also be attributed to the fact that only 50% of the does in the control group were pregnant, thus the weight of empty does led to low average weight compared to other treatment groups.

Lower body condition scores were observed in the does treated with aqueous extract of *R. tomentosa* and distilled water than those treated with a mixture of *E. elephatina* and *R. tomentosa* (1:1) and *E. elephantina* extracts. The lower body condition scores observed in the study might be due to the presence of phytohormones present in *R. tomentosa*, which are known to suppress fat accumulation (Schilling *et al.*, 2014). Polyphenols and flavonoids have anti-obesity properties (Williams *et al.*, 2013), this could be the reason why animals treated with aqueous plant extract of *R. tomentosa* had lower body condition scores. According to Mellado (2011), body condition scores are very important in evaluating the nutritional status of goats to be used for breeding since poor conception rates are associated with does having body condition score below 2.5 and above 4.
Animals administered with a mixture of *E. elephantina* and *R. tomentosa* showed high pregnancy rates as compared to those treated with single aqueous plant extracts. This agrees with Lans and Brown (1998), who reported that medicinal plants are used in combination to improve fertility and induce estrus in ruminants. According to Patel *et al.* (2011), several plants are used as fertility-enhancing agents. The high twinning percentages observed in does administered with the aqueous extract of *E. elephantina* and *R. tomentosa* mixture might be due to the phytochemicals in these plants that might stimulate follicular growth leading to high ovulation rate (Zarkawi *et al.*, 1999 citing Greyling and van Niekerk, 1999). The conception rate (100%) observed in does treated with *E. elephantina* and *R. tomentosa* mixture was also higher than (65.8%) recorded by Zarkawi *et al.* (1999) when artificial hormones were used. High conception rates were observed in does which had high concentrations of progesterone and this concurs with the study by Johnson *et al.* (1996). The high twinning rate observed in goats administered with a mixture of *E. elephantina* and *R. tomentosa* aqueous extract might be due to the influence of the phytochemicals on gonadotropins, which can result in ovarian over-stimulation leading to twinning (Mutiga and Mukasa-Mugerwa, 1992). Besides gonadotropins, there are other factors such as ovarian proteins and growth factors that control the ovarian function (Driancourt, 1991 citing Gorosje *et al.*, 1988), therefore knowledge of these factors might provide helpful clues on how these factors can be manipulated to improve reproduction performance by altering the ovarian function (Driancourt, 1991). However, further studies need to be done to determine the effects of these plant bioactive compounds on follicle stimulating hormone (FSH) and luteinizing hormone (LH) the foetus survival.
4.5. Conclusion

The medicinal plants commonly used by farmers to improve reproductive performance had an influence on the hormonal concentrations, conception and body weights of the goats. A mixture of aqueous extract of *E. elephantina* and *R. tomentosa* was most influential on estradiol and progesterone levels. Average body weight gains were higher in animals treated with medicinal plants than the control. A mixture of *E. elephantina* and *R. tomentosa* caused highest pregnancy rate and twinning rate in goats. However, further studies need to be done on the toxicity of these plants and their effect on gonadotropins and foetuses.

4.6. References


Farhoodi M., Khorshid M. and Eyvani D., 2011. *Vitex agnus-castus* effects on inter estrus interval in dairy cows. Faculty of Veterinary Medicine, Karaj Branch, Islamic Azad University, Karaj, Iran.


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Mthembu X.S., 2007. A phytochemical study of *Schefflera umbellifera* and *Elephantorrhiza elephantina*. Dissertation submitted in fulfilment of the academic requirements of the degree of Master of Science in the Discipline Chemistry, School of Chemistry, University of KwaZulu-Natal.


CHAPTER 5: General Discussion, Conclusion and Recommendation

5.1. General discussion

Goats play a major role to resource-limited farmers (Maphosa, 2010) because they can be a source of food, manure, milk and income. However, goat production is faced with several challenges and one of the major constraints is that the reproduction potential of these livestock species is not fully exploited by farmers (Fuah and Priyanto, 2013). Therefore in order to improve reproductive performance of goats, resource-limited farmers end up using medicinal plants to enhance fertility and to treat reproductive ailments. However, it is important to note that, in order to increase production of goats, it is important to optimize the reproductive performance by enhancing reproduction without incurring high cost in production (Notter, 2012). The objective of the study was to document and validate the efficacy of medicinal plants used to influence reproduction and treat infectious reproductive ailments in goats.

In Chapter 3, alternative remedies used by resource-limited farmers to manipulate reproduction and treat reproductive ailments were recorded. A total of ten medicinal plants were reported, with *Elephantorrhiza elephantina* and *Rhoicissus tomentosa* being most frequently cited. The preparation methods and plant parts used were also recorded and the findings of the current study concurred with the study by Ribeiro et al. (2010) who reported that *E. elephantina* was mostly cited and 38.8% of the respondents used roots during remedy preparation. The same author also indicated that decoctions were mostly used in remedy preparation and this agrees with the
current study. Farmers had different perceptions on the efficacy of traditional remedies. In South Africa, most resource-limited farmers use traditional medicines because they are readily available and cheaper than conventional methods (Mwale et al., 2005). However, the incorporation of more districts in the study could have led to the documentation of a wide range of traditional medicines used for reproduction purposes since people from different ethnic groups and cultures have a variety of knowledge on traditional medicines.

In Chapter 4, there was a significant influence noted of the mixture for aqueous extract of *E. elephantina* and *R. tomentosa* on the reproduction system of goats. This mixture caused high increase in concentrations of serum estradiol and progesterone as well as increase in body weights of goats. This is perceived to be a result of phytoestrogenic compounds found in these plants (Dilshad et al., 2012). Phytoestrogens were reported to decrease secretion of follicle stimulating hormone and increased secretion of luteinizing hormone from the pituitary gland which subsequently lead to increased progesterone and estrogen levels (Farhoodi et al., 2011 citing Lucy, 2011). The high body weights observed in the animals administered with a mixture of *E. elephantina* and *R. tomentosa* were due to animals having been pregnant and possibly anthelmintic properties of *E. elephantina* (Naidoo, 2004). It is also possible that these plants had nutritive properties, which were not investigated. Goats administered with a mixture of *E. elephantina* and *R. tomentosa* had a pregnancy rate of 100%, but lower rates were observed in animals given aqueous extracts of *R. tomentosa* and in the negative control. This goes on to show that when *R. tomentosa* is used independently its efficacy is compromised than when it is used in combination. When plants are combined during remedy preparation their efficacy tends to
improve due to the synergistic effect (Chung et al., 2011). This might be the probable reason why farmers use these plants in combination.

5.2. Conclusions

Resource-limited farmers in Mount Frere area in the Eastern Cape Province, South Africa use both medicinal plants and zoo therapeutic remedies to enhance fertility and treat reproductive ailments. In the in vivo studies, medicinal plants used by farmers to influence reproduction showed an influence on the hormonal concentrations, body weights as well as pregnancy rate of the goats, therefore are effective for the purpose for which they are used.

5.3. Recommendations

Although findings from the study have shown that *E. elephantina* and *R. tomentosa* both influences reproduction in goats, there is need to conduct the study for a longer period, in different seasons and with a larger group of animals. Furthermore, it is also important to grow these plants on a massive basis if they are to be explored for commercial purposes.

It can be recommended that further studies should be done in the following areas:

1. Investigate the effect of administering the remedies at different stages of animal’s estrus cycle and using different age groups in both males and females.
2. Determine the potential of medicinal plants to cause residual effect in animal products, when used for prolonged periods in goats.

3. Isolation of the active compounds responsible for influencing reproduction in goats from plant extract fractions.

5.4. References


Farhoodi M., Khorshid M. and Eyvani D., 2011. Vitex agnus-castus effects on inter estrus interval in dairy cows. Faculty of Veterinary Medicine, Karaj Branch, Islamic Azad University, Karaj, Iran.


APPENDICES

Appendix 1: Questionnaire

Assessing alternative remedies used to influence reproduction and treat reproductive ailments in goats in the small-holder sector.

Household number

Date

Year Month Day

Province

Enumerater name

District

Respondents name

Ward

Sex of respondent

Village

SECTION A: HOUSEHOLD DEMOGRAPHY

1. Who is the head of the household?  
   1) Father 2) Mother 3) Children 4) other*  
   *Specify
2. Table on household demographics

<table>
<thead>
<tr>
<th>Household member</th>
<th>Relation with household head&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Marital status&lt;sup&gt;b&lt;/sup&gt;</th>
<th>Gender</th>
<th>Age&lt;sup&gt;c&lt;/sup&gt;</th>
<th>Highest level of education&lt;sup&gt;d&lt;/sup&gt;</th>
<th>Animal husbandry training&lt;sup&gt;e&lt;/sup&gt;</th>
<th>Occupation&lt;sup&gt;f&lt;/sup&gt;</th>
</tr>
</thead>
</table>

<sup>a</sup>: (1) Father, (2) mother, (3) son, (4) daughter, (5) daughter in law, (6) son in law, (7) wife, (8) husband, (9) self, (10) specify

<sup>b</sup>: (1) single, (2) married, (3) divorced, (4) widowed, (5) separated, (6) other (Sp, 7) N/A

<sup>c</sup>: (1) ≤ 25 years, (2) 26 – 40 years, (3) 41 – 55 years, (4) 56 – 70 years, (5) > 70

<sup>d</sup>: (1) never attended school, (2) Primary level, (3) Secondary, (4) Tertiary level, (5) University, (5) N/A
3. Sources of income? (1) Crops ☐ (2) livestock ☐ (3) salaries/wages ☐
(4) Other (specify) ____________________ (5) None ☐ (6) N/A ☐

SECTION B: LIVESTOCK INVENTORY

4. Fill in the table below for the livestock owned

<table>
<thead>
<tr>
<th>Class</th>
<th>Goats</th>
<th>Other stock</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goats</td>
<td>Does</td>
<td>Bucks</td>
</tr>
<tr>
<td>Number</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

5. Who owns the goats? (1) Father, (2) Mother, (3) Grandfather (4) Grandmother (5) child
(6) other (specify) ________________

6. What functions do goats play in your household? Complete the table below
<table>
<thead>
<tr>
<th>Role</th>
<th>Rank according to importance from 1 to 4 which will be the least important</th>
<th>Rank subfunctions according to importance within major role</th>
</tr>
</thead>
<tbody>
<tr>
<td>Relating to crop production</td>
<td>Provision of manure</td>
<td>Transport (of inputs, water etc)</td>
</tr>
<tr>
<td>Consumption</td>
<td>Milk for domestic consumption and sale</td>
<td>Meat for local use or sale</td>
</tr>
<tr>
<td>Household finance</td>
<td>Investment from crop income</td>
<td>School fees</td>
</tr>
<tr>
<td>Social</td>
<td>Installation of ancestral spirits</td>
<td>Bride wealth</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Social status (pleasure in ownership)</td>
</tr>
</tbody>
</table>
7. Do you give your goats feed supplements? Yes □ No □

8. If you give supplements what do you give them? (1) silage □ (2) Stover □ (3) other (specify)

9. When do you give them supplements? Summer □ Autumn □ Winter □ Spring □

10. Specify the amount of feed supplements you give..........................................................

SECTION C: GOAT REPRODUCTION SITUATION

11. When do your goats kid? Summer □ Autumn □ Winter □ Spring □

12. How many of the does produce twins? 1-5 □ 6-10 □ 10-15 □ 15+ □

13. Is there anything you give them to produce twins? Yes □ No □
14. If your answer is Yes, then what do you give them?

........................................................................................................................................................................

15. How do you prepare it?

........................................................................................................................................................................

16. How do you administer it?

........................................................................................................................................................................

17. How often do you use it? Every 1 – 2 weeks □  Every 3 - 4 weeks □  Every 5 - 26 weeks □  Every 27 – 52 weeks □

18. Is there anything you give to the bucks to improve its fertility or libido? Yes □  No □

19. If your answer is Yes, then what do you give them?

........................................................................................................................................................................

111
20. How do you prepare it?


21. How do you administer it?


22. How often do you use it? Every 1 – 2 weeks

   Every 3 - 4 weeks

   Every 5 - 26 weeks

   Every 27 – 52 weeks

23. If plants are used which ones are those and how do you rank their importance, complete table below

<table>
<thead>
<tr>
<th>Disease, Purpose, Use</th>
<th>Plant (local name)</th>
<th>Part of plant used</th>
<th>How prepared</th>
<th>How* applied</th>
<th>Effective for how long</th>
<th>Rank**</th>
</tr>
</thead>
<tbody>
<tr>
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</tbody>
</table>
*include frequency of use, *(1) most important (2) moderately important (3) not important

24. Of the conditions that affect goats fill in the following table and rank them according to the importance from 1 (very important) to 7 or greater (least important)

<table>
<thead>
<tr>
<th>Condition/Disease</th>
<th>Season of the year</th>
<th>Rank</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infertility</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Abortion</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Behavioural abnormality</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Irregular estrus cycles</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Retained placenta</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Venereal diseases</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td></td>
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</tr>
</tbody>
</table>
SECTION D: GOAT REPRODUCTIVE AILMENTS

25. Do you treat any reproductive ailments or conditions affecting your goats?  
   (1) Yes □  
   (2) No □

26. Where do you get the remedies/treatment you use? Pharmacy □ traditional healers □  
   Use own knowledge □

27. If bought, how much does it cost? R1 – R25 □  R 26 – R50 □  R 51 – R75 □  R  
   76 – R100 □  Above R100 □

28. Is there any indigenous knowledge you use/local remedies?  Yes □  No □

29. If your answer is YES, what is the name of the remedy you use

   ………………………………………………………………………………………………………………………………………

30. How do you prepare it?

   ……………………………………………………………………………………………………………………………
31. How do you administer it to animals? Drench ☐ Spray ☐ Inject ☐ Any other way (specify) ☐

32. For what do you use them for (tick any or all)  twin/triplet production ☐ reproductive ailments ☐ increase libido ☐ Any other way (specify) ☐

33. How often do you use the following practices for reproduction in goats?; use key below.

   Plants ☐ Commercial drugs ☐ non-plant remedies ☐

   (1) most often (monthly) (2) (seasonally) (3) never

34. How often do you use the following practices for treating reproductive diseases; use key below.

   Plants ☐ Commercial drugs ☐ non-plant remedies ☐

   (2) most often (monthly) (2) few occasions (seasonally) (3) never

35. How do you rate the effectiveness of the treatment methods used in 33 and 34? Use keys below.

   Plants ☐ Commercial drugs ☐ non-plant remedies ☐

   (1) very effective (2) moderately effective (3) not effective (4) do not know

36. Is your choice of treatment method based on effectiveness  Yes ☐ No ☐
If your answer is NO what determines your choice of treatment method

...................................................................................................................................................

37. If for the reproduction you use alternative methods such as plants or non-plant remedies, how did you obtain the knowledge? (Circle the most important source).

(1) Oral tradition (2) from other farmers (3) read somewhere (4) local elders.

38. Are you allowed to share the knowledge of alternative control methods mentioned above?
Yes ☐ No ☐

39. Do you think that students should be taught the use of plants in schools, colleges and Universities Yes ☐ No ☐

40. Are alternative methods used, better than the use of commercial products, Yes ☐ No ☐

41. Do the extension officers or animal health technicians know this alternative remedies? Yes ☐ No ☐

42. If your answer is YES where did they learn it?
43. If your answer is NO why don’t they know it?

THANK YOU – SIYABULELA - ENKOSI