THE EFFECT OF SUPPLEMENTATION OF PLANT AND ANIMAL-DERIVED DIETARY OILS IN PRE- AND POST-PARTURITION DÖHNE MERINO EWES ON THE GROWTH PERFORMANCE OF THE LAMBS

by

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DECLARATION

I, Tebogo Sedupane, declare that this thesis titled “The supplementation of plant and animal-derived dietary oils on milk production and growth performance in Döhne Merino sheep” is my work, has not been submitted before for any degree or examination at any other university, and all sources I have used or quoted have been indicated and acknowledged by complete references.

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Tebogo Sedupane

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Date

DEDICATION

I dedicate this to the loving memories of my late brother, Thabiso Sedupane, and uncles, Buru Edga Sedupane and Motseothata Sima, who have all passed on. You will remain forever in our hearts.

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ABSTRACT

The objective of this study was to investigate the effect of dietary supplementation of sunflower oil, olive oil, fish oil, and palm oil to pre- and post-parturition Döhne Merino ewes on lamb growth performance. Birth weight, post-natal growth rate, and weaning weight of lambs suckling on supplemented ewes were compared. Fifty South African Döhne Merino ewes, 2–4 years old (second parity), weighing 46–55 kg, with a mean body condition score (BCS) of 3.5 ± 0.4, were randomly divided into groups of 10 animals each. Each group (n = 10 per group) received a daily dose of 30 ml of either sunflower, olive, fish, or palm oil, while the control group received no oil. The study was conducted over a period of 210 days (i.e., 7 months). Oestrus was synchronised through the use of controlled internal drug release (CIDR) devices inserted intravaginally for a period of 15 days. Oestrus synchronisation and oil supplementation commenced simultaneously. At CIDR withdrawal, ewes were injected intramuscularly with 200 IU Pregnant Mare Serum Gonadotrophin (PMSG). Fixed-time laparoscopic artificial insemination (LAI) was performed with diluted Döhne Merino semen 48 hours following CIDR removal. Two weeks after LAI, follow-up Döhne Merino rams were introduced to all 50 ewes for 2 weeks to mate ewes that exhibited any signs of oestrus, as this is standard procedure of the experimental farm, and it was also importatnt that all selected ewes for the experiment conceive, although not part of the objectives of the present research. The production parameters were expressed as an average per group. The mean conception rate and lambing rate of ewes across all groups were 88%. The olive oil treatment recorded the highest conception rate and lambing rate (100% for both), but did not differ significantly (P> 0.05) from other groups. followed by sunflower oil (90%), fish oil (90%), palm oil (70%); the control group had rates of 90%. post-hoc test using Tukey's HSD to identify significant differences between treatment groups at specific time points at a 95% confidence interval. The IBM Statistical Package for the Social Sciences (SPSS) 27 software programme was utilised.

The shortest gestation length of 150 days was recorded for the control group. The gestation lengths of the sunflower oil, olive oil, fish oil, and palm oil groups were 153, 156, 155, and 156 days, respectively, with no significant differences ($P > 0.05$) between any groups. The birth weights of female lambs from the control, sunflower oil, olive oil, fish oil, and palm oil groups were 5.41 ± 0.87 kg, 5.70 ± 0.61 kg, 5.8 ± 0.70 kg, 5.8 ± 0.74 kg, and 4.9 ± 0.50 kg, respectively. The birth weights of male lambs from the control, sunflower oil, olive oil, fish oil, and palm oil groups were 5.6 ± 1.19 kg, 4.85 ± 0.70 kg, 5.6 ± 0.84 kg, 5.5 ± 0.49 kg, and 5.4 ± 0.33 kg, respectively. The results indicate no significant difference ($P > 0.05$) between different treatment groups for birth weights for both male and female lambs. There was improvement or significance ($P>0.05$) on percentage increment in weight over time in weeks 5,9 and 11. There was significant difference experienced in ADG of lambs among groups in weeks 5,6,10,11 and 13. There was no significant difference ($P > 0.05$) observed in the final body weight at weaning for both males from the control, sunflower oil, olive oil, fish oil, and palm oil groups (28.8 ± 4.93 kg, 25.5 ± 8.13 kg, 29.0 ± 4.88 kg, 30.2 ± 4.59 kg, and 28.8 ± 5.03, respectively) and females from the control, sunflower oil, olive oil, fish oil, and palm oil groups (28.5 ± 4.58 kg, 28.3 ± 3.81 kg, 25.8 ± 6.93 kg, 28.9 ± 4.51 kg, and 27.6 ± 2.46 kg, respectively) for the lambs of ewes supplemented with different experimental dietary oils. There was difference in the effect of plant and animal-derived dietary oils – namely sunflower oil, olive oil, fish oil, and palm oil – on the percentage increment in weight over time and ADG of lambs among groups. The growth performance of the lambs was similar to the control group. It can thus be concluded that, in this study, diets enriched with plant and animal-derived dietary oils pre- and post-mating had an effect on growth performance in Döhne Merino sheep.

Keywords: Döhne Merino, dietary oils, reproductive performance, growth performance

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ACRONYMS AND ABBREVIATIONS

ADG	average daily gain
AI	artificial insemination
ANOVA	analysis of variance
BCS	body condition score
BW	body weight
CIDR	controlled internal drug release
DHA	docosahexaenoic acid
eCG	equine chorionic gonadotropin
ECM	energy-corrected milk
EPA	eicosapentaenoic acid
FGA	fluogestone acetate
FTAI	fixed-time artificial insemination
GLM	General Linear Model
HFA	hard fatty acid
HSD	honest significant difference
LAI	laparoscopic artificial insemination
LSU	large stock unit
MAP	medroxyprogesterone acetate
MUFA	monounsaturated fatty acid
MUN	milk urea nitrogen
NE_1	net energy
NRF	National Research Fund

NS	no significance
PAG	pregnancy-associated glycoprotein
PFAD	palm fatty acid distillate
PMSG	Pregnant Mare Serum Gonadotropin
PUFA	polyunsaturated fatty acid
SANS	South African National Standard
SBO	soya bean oil
SCC	somatic cell count
SD	standard deviation
SFA	saturated fatty acid

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1. CHAPTER ONE: INTRODUCTION

1.1. GENERAL INTRODUCTION

Sheep farming plays an integral role in agriculture around the world. In 2019, the total global sheep population was 1.24 billion – of which 408 million were in Africa and 25.5 million in South Africa (Cloete, 2021). There is much variation in the density of the sheep population in the various provinces of South Africa, but generally, the highest numbers of sheep are found in the Eastern Cape, followed by the Northern Cape and Free State (DAFF, 2020). According to Cloete and Olivier (2010), extensive small stock production occurs mostly in the drier western and north western parts of the country, which have a low grazing capacity of below 12 ha per large stock unit (LSU). In addition, the population of extensively reared beef increases in the eastern and northern directions, as the average precipitation rises. Extensive beef production has replaced much of the extensive small stock production in the northern provinces (Cloete *et al.*, 2014).

Only 16.5% of the approximately 86.2 million ha of commercial agricultural land in South Africa can be considered arable (DAFF, 2012). Extensive farming practices play an important role in agriculture and are mainly determined by environmental conditions such as climate, soil type, and availability of grown pasture species. However, animals kept intensively grow faster than animals kept extensively. Extensively kept animals' delayed growth reduces production efficiency and the ability to achieve slaughter and carcass weight at the required time (De Brito *et al.*, 2017).

Feed additives are often used in animal feed rations, especially in extensive animal production systems. Feed additives are included to enrich animal diets and enhance digestive and metabolic processes (Zaitsev *et al.*, 2020). These feed additives are also used to stimulate growth, health, and animal production performance. Phytogenic feed, reduction licks, energy licks, and ready-mixed protein/energy/mineral supplements (e.g., Voermol's Supermol and Molovite supplements [Voermol, 2024]) form part of feed additives. Feed additives comprise various spices, herbs, plants (e.g., sunflower and palm oil), and animal (fish oil) products and contain essential dietary oils (Windisch *et al.*, 2008).

Antibiotics have been used to prevent and cure diseases as well as to improve growth and growth efficiency (Benchaar *et al*., 2009). However, the emergence of antibiotic-resistant bacteria is one of the main threats to global health (Coimbra *et al*., 2022). The extensive use of antibiotics in ruminants can also leave antibiotic residues in meat and milk, which negatively affects human health when these products are consumed (Treiber & Beranek-Knauer, 2021). Recently, interest in the use of natural products to improve the health and productivity of livestock has grown (Dorantes-Iturbide *et al*., 2022). Examples of these natural products are plant and animal-derived oils.

There are innovative bio refinery and oil-extraction methods in chemistry, biology, and processing that are sustainable and environmentally friendly. These extractions include green extraction, hydistillation, and cold-pressing methods. Green extraction is mostly used to extract oil from fish waste through both traditional and hydraulic pressing. The extraction process is relatively new and innovative, and there are various environmentally friendly methods such as supercritical fluid extraction, enzyme extraction, microwave-assisted extraction, and ultrasound-assisted extraction that can be used (Rubio-Rodriguez *et al*., 2010; Adeoti & Hawboldt, 2014).

Hydistillation includes steam distillation, which is a standard method for pure essential-oil extraction. The process involves breaking down the plant's trichomes with steam and vaporising the volatile compound; within the process of condensation and collection, the essential oil can be separated. This method of extraction can take approximately 30 minutes to 22 hours, with the plant matter generally breaking down at 100ºC (Enterprise Ethanol, 2024).

Cold pressing is an older process used to obtain essential oils. Cold pressing was used long before humans discovered the process of distillation and has the advantage of little or no heat generation during the process; however, it provides ow yield (van Doosselaere, 2013). Cold pressing is used for the isolation of peel oils of citrus fruit due to the relative thermal instability of the aldehydes contained in them (Kubeczka, 2010). Mechanical cold pressing is mostly used for the extraction of oil from citrus peels by cracking the oil glands and applying pressure so that the oil can be discharged and washed away with a water spray (Reyes-Jurad *et al.*, 2014).

Oils have different fatty acid profiles and ratios of fatty acids (n:3, n:6, n:9) with different effects on the reproduction and general health of an animal. Fish oil is rich in n:3 fatty acids, sunflower is rich in n:6 fatty acids, and palm oil and olive oil are both rich in n:9 fatty acids. In a study by Suseno *et al*. (2014), fatty acids of softshell turtle oil, freshwater eel oil, cod liver oil, tuna oil, and lemur oil were analysed. The showed that saturated fatty acids (SFAs) ranged from 1.76–37.99%, monounsaturated fatty acids (MUFAs) ranged from 3.17–38.34%, and polyunsaturated fatty acids (PUFAs) ranged from 0.70–34.99%.

Previous research reports that different feed sources and/or dietary inclusion of different oil sources may alter the milk composition of a lactating animal. Nudda *et al.* (2020) conducted a study comparing the fatty acid composition of sheep and goat fat when including herbage rich in PUFAs or vegetable, marine, or essential oils in the diet of lactating animals. Sheep had a more positive response than goats to the use of fresh herbage and nutritional approaches, producing milk with increased concentrations of c9, t11-conjugated linoleic acid (c9, t111-CLA), and α-linolenic acid. The study found that dietary polyphenols can impact milk fatty acid profile, hindering the activity and growth of some strains of rumen microbes involved in the biohydrogenation of PUFAs (Nudda *et al.*, 2020).

Similarly, it has been found that increased consumption of dietary oil without negatively affecting dry-matter intake is advantageous for growth and milk production. Turner *et al.* (2014) report that it is imperative to supplement sheep to enhance growth and carcass production. Furthermore, lambs that are supplemented with a concentrate-rich diet perform better in growth compared to those consuming only grazing-pasture-based diets (Bessa *et al.*, 2008; Armero & Falagan, 2015; De Brito *et al.*, 2017). Other research shows that the genetic makeup and gender of an animal have an impact on meat characteristics (Gruffat *et al.*, 2013; Malau-Aduli *et al.*, 2016), and that a concentrate-rich diet plays a significant role in the nutritional value and sensory properties of meat (Asadollahi *et al.*, 2017; Chikwanha *et al.*, 2017).

The practice of incorporating a lamb finishing diet in intensive farming has become common to improve both animal and paddock productivity to meet a broader range of markets (Watkins *et al.*, 2013; Frank *et al.*, 2017). The selection of the right supplementary feed for either pregnant, lactating, or dry animals is important; it must not only be cost-effective but also take into consideration factors such as competing with humans for food resources and the impact on the environment and on animal health and welfare.

A meta-analysis by Dorantes-Iturbide *et al*. (2022) found that the dietary inclusion of essential oils increases dry-matter intake and feed-conversion ratio (FCR). In addition, the inclusion of essential oils in small ruminants' diets decreased enteric methane emissions and increased ruminal propionate concentration. The researchers also observed higher milk production and increased lactose content in the milk of small ruminants.

This study aims to investigate whether supplementation of plant or animals-derived oils to ewes will improve reproductive performance and their suckling lambs' growth rates.

1.2. PROBLEM STATEMENT

The direct effects of poor nutrition include reduced conception, embryonic losses, and reduced lambing percentages in ewes. Poor nutrition may also cause delays in the onset of puberty, loss in body condition of pregnant ewes that will lead to low birth weights of lambs, and poor milk production in lactating ewes that results in retarded lamb growth and poor weaning weight. To date, results have been conflicting about which dietary oils are effective in improving reproduction efficiency, ewe milk production, and weaning weights of lambs. Some researchers found that oil supplementation decreases feed digestibility (Bhatt *et al.*, 2011), while others found the contrary (Yusuf *et al.*, 2009; Bhatt *et al.*, 2013).

1.3. MOTIVATION

Small stock plays a significant role in human food security and income at the household level. Production methods can be substantially improved from the point of breeding until the time of sale of the offspring to the abattoir. Many South African farmers endeavour to increase the lambing frequency, number of lambs born, and weaning percentage, as these affect the profitability of their enterprise. Low lamb output affects the efficiency of production and results in low profit margins.

To increase productivity, the sheep farmer must improve the nutrient intake of their sheep. Dietary supplementation or flushing of sheep can be expensive but may positively improve the reproduction parameters in ewes and the growth of lambs. Using oils as a supplement to ewes may increase the economic return through lambs that are weaned. In terms of carcass quality, some researchers have indicated that lambs' PUFA content can be improved by supplementing PUFA-rich plant oils or fish oils in the lamb finishing diet (Ferreira *et al*., 2014; Francisco *et al*., 2015).

There is no literature on dietary oil supplementation in South African Döhne Merino sheep. Mirzaei-Alamouti *et al*. (2018) report an increased number of medium follicles and size of the ovulatory follicle in fat-tailed Iranian Afshari ewes supplemented with fish oil and sunflower oil before mating. During mating, the number of follicles was found to be significantly higher ($P < 0.05$) in the ewes supplemented with fish oil.

This study attempted to investigate the impact of plant and animal-derived dietary oils on the productivity of Döhne Merino sheep during a 210-day feeding trial. Findings from this project will give farmers a guideline on which oil supplement (if any) to use to improve production efficiency.

1.4. AIMS AND OBJECTIVES

1.4.1. Aim

This study aimed to investigate the effect of supplementation with various dietary oils on reproduction efficiency and lamb growth performance in Döhne Merino sheep.

1.4.2. Objective

To investigate and compare the reproductive performance of Döhne Merino ewes as well as the birth weight, growth, and weaning weight of lambs that were born from and suckled on ewes supplemented with different plant and animal-derived oils.

1.4.3. Hypothesis

H0: Supplementation with plant or animal-derived dietary oils will have a significant positive effect on the reproductive performance of Döhne Merino ewes and also have a positive effect on the growth performance of lambs born from and that suckled on supplemented ewes, leading to a productive and profitable farming enterprise.

2. CHAPTER TWO: LITERATURE REVIEW

2.1. THE DÖHNE MERINO BREED

The Döhne Merino sheep is a South African breed that was developed by the Department of Agriculture in the late 1930s by breeding Peppin-style Merino ewes and German Mutton Merino rams. It is a dual-purpose breed bred for the production of meat and wool (Sheep101, 2021). Mature Döhne Merino rams weigh approximately 100 kg and ewes weigh 50–65 kg. The Döhne Merino produces a fleece of 3.5–5.0 kg, with an average diameter of 17–21 microns per annum. This breed is tough (known for its ability to survive extreme weather and its resistance to diseases), produces good-quality slaughter lambs and high-quality fine apparel wool, and performs well under both intensive and extensive production systems, with fast lamb growth rates (Agribook, n.d.).

2.2. REPRODUCTIVE PERFORMANCE IN SHEEP

In developing countries, sheep are one of the most important domesticated ruminant species and are of great interest around the world (Asgari Safdar *et al*., 2017). One of the most economically important traits in sheep is reproductive performance, particularly when lamb and mutton are the main products (Fourie & Hendenrych, 1982; Van Haandel & Visscher, 1995; Snyman *et al.*, 1998; Cloete *et al.*, 2000; Abegaz *et al.*, 2002; Van Wyk *et al.*, 2003; Senger, 2012; Ali *et al.*, 2020).

Reproductive performance is measured by a number of parameters. These parameters include (but are not limited to) the proportion of ewes bred that lamb (a measure of fertility), the percentage of lambs born (dead or alive) per ewe, as well as the percentage of lambs weaned (weaning percentage). Pre-weaning mortality rates are incorporated as an indication of pre-weaning lamb losses (Rosati *et al.*, 2002).

Fertility and reproductive performance are dependent on a number of factors and interaction between these factors. These factors include the environment, genetic factors, and interaction between environment and genetics for both ewes and rams (Rosati *et al.*, 2002). Ewe nutrition, supplementation, body condition score (BCS), and age at breeding are salient factors that play pivotal roles in the chances of successful conception during fertilisation. Nutrition is among the most important environmental factors affecting fertility and reproductive performance (Kia & Safdar 2015; Asgari Safdar *et al*., 2017). Nutrition has a stimulatory impact on follicle development and ovulation, and flush feeding is often applied as a management method a few weeks before the breeding season (Asgari Safdar *et al*., 2017). In gestating and lactating ewes, PUFAs play a pivotal role in foetal programming, lambing ease, colostrum and milk production, udder health, and offspring development (Nudda *et al*., 2015; Macías-Cruz *et al*., 2017; Nickles *et al*., 2019).

2.3. MANAGEMENT TO IMPROVE REPRODUCTIVE EFFICICIENCY

2.3.1. Oestrus synchronisation

Reproductive management of sheep is often based on the induction and synchronisation of oestrus and ovulation by the use of pharmacological treatment – either in the reproductive or non-reproductive season and for natural mating or artificial insemination (AI) (Abecia *et al*., 2012).

Synchronisation of oestrus and ovulation for fixed-time artificial insemination (FTAI) in sheep is mainly based on inserting intravaginal devices holding either control internal drug release (CIDR®) inserts or sponges impregnated with fluogestone acetate (FGA) or medroxyprogesterone acetate (MAP). During device removal, a single intramuscular injection of equine chorionic gonadotrophin (eCG) is administered for inducing ovulation and synchronising its time of appearance among animals in the same lot (Abecia *et al.*, 2012).

Martinez-Ros *et al.* (2018) investigated the pre-ovulatory and ovulatory events (in terms of timing of onset of oestrus and subsequent ovulation) and the yield obtained (in terms of rate, progesterone secretion, and fertility) after inserting CIDR for 5, 6, 7, or 14 days, with or without eCG. All treatment groups resulted in above 80% of ewes showing oestrus, but the onset of oestrus was earlier and more synchronised when using eCG. The eCG group's oestrus onset was found to be earlier in the sheep treated for fourteen days than in short-term treatment ($P < 0.05$ for all). Furthermore, ovulatory success without eCG was found to be independent of treatment length, with a high percentage of animals ovulating after 5 days of treatment (83.3%) and a very low percentage after treatment for 6 or 7 days (40% and 20%, respectively). The best results were produced after 14 days plus eCG and after 5 days without eCG (83.3% for both groups; $P < 0.05$ when compared to other groups with different lengths and with or without eCG) (Martinez-Ros *et al.*, 2018).

2.3.2. Laparoscopic artificial insemination (LAI)

Artificial insemination involves manual deposition of semen into the uterus of a ewe during oestrus by non-natural methods. This method can offer many valuable benefits, such as prevention and control of disease through the elimination of direct contact between males and females and enhanced progress of genetic exchange through selective breeding (Alexander *et al.*, 2010). A higher fertilisation percentage (91.5% versus 44.8%, respectively; $P < 0.05$) was achieved with LAI versus natural mating (Azawi & Al-Mola, 2011).

This lower fertilisation rate during natural mating compared to LAI may be due to failure in the transport of semen through the tightly folded cervix and into the uterus or due to sperm quality (Alavares *et al.*, 2020). Dovenski *et al.* (2012) used intrauterine insemination in sheep with frozen semen and discovered higher fertility during the oestrus season (60%) than in the anoestrus season (45%). The highest rates of fertility with frozen semen were achieved during LAI and lower rates with natural insemination (Bancheva *et al.*, 2021).

A higher fertilisation percentage (54%) has been reported in ewes with fertilised oocytes inserted through LAI compared to the percentage (19%) achieved via cervical insemination (Fair *et al.*, 2005). Najafi *et al*. (2014) compared laparoscopic and cervical AI and reported about 83.3% and 60% pregnancy rates, respectively.

2.4. FEEDING TO IMPROVE REPRODUCTIVE EFFICIENCY

According to Nieto *et al.* (2015), the inclusion of long-chain n-3 and n-6 fatty acids has a positive impact on ruminant reproduction. These two fatty acids are synthesised by the ruminant through desaturation and elongation of short-chain fatty acids into n-3 linoleic acid (LA, 18:3n-3) and n-6 linoleic acid (LA, C18:2n-6) (Nieto *et al.*, 2015). Cerri *et al.* (2009) report that supplementary fatty acids in ruminant feed can enhance fertility and embryo development.

The advantage of dietary supplementation of fat, and PUFAs in particular, is known to enhance reproduction in dairy cows. Apart from improving conception rate, n-3 PUFAs have also been found to have a positive effect on various reproductive processes like the size of ovulatory follicles (Mendoza *et al.*, 2011), quality of oocytes (Zeron *et al.*, 2002), corpus luteum development (Petit *et al.*, 2002), and embryo development (Childs *et al.*, 2008).

Kiliçalp and Yücel (2019) established that the addition of PUFAs enhanced fertility of flush-fed ewes. According to Khotijah *et al*. (2015), the inclusion of 4% sunflower oil in the ration of ewes increases reproductive performance (lamb size and male-female ratio) and cholesterol concentration, and it results in a pregnancy rate that is 12.5% higher than other groups. Kiliçalp and Yücel (2019) found that n-3 PUFA supplementation in ewes during flush feeding increases the fertility of Karayaka sheep. Furthermore, higher percentages of twin births were observed in ewes supplemented with n-3 PUFAs. A higher lambing percentage was obtained in Afshari ewes supplemented with n-3 fatty acids compared to ewes supplemented with n-6 fatty acids during the flushing period (Kia *et al.*, 2015).

2.4.1. Physiological role of fatty acids in reproduction

The physiological effects of fatty acids on reproductive performance are disputable and possibly involve the effects of prostaglandin. Dietary oils with n-3 PUFAs were established to reduce the luteolytic $PGF_{2\alpha}$ and increase the pregnancy rate in dairy cows. However, there is less information on the effect of dietary oils on goat fertility (Mahla *et al.*, 2017). It was observed that the fertility percentage of all ewe lambs and birth weight of those fed olive oil cake were higher than those fed HFA and the control group (EL-Tarabany *et al.*, 2017). Olive oil cake also influenced the stage of puberty in female lambs; dietary supplement oils made lambs reach puberty faster and their thyroid and progesterone developed earlier than lambs that were fed hard fatty acid (HFA) ration, including the control group (EL-Tarabany *et al.*, 2017). Dos Santos *et al.* (2019) found no difference ($P > 0.05$) in body weight (BW), BCS, and average daily gain (ADG) during pregnancy between ewes fed protected fat from palm oil compared and those in the control group.

Essential fatty acids are key in a ration, as they function as a membrane compound of prostaglandin synthesis, which is important for growth, lactation, and reproduction (Cheng *et al.*, 2001; Encinias *et al*., 2004; Yaqoob & Calder, 2007; Palmquist, 2010). Fatty acids play a pivotal role in stimulating follicles (prostaglandin) to increase chances of conception and reduce prostaglandin during pregnancy. At a later stage, they support progesterone production to maintain the pregnancy (Wiryawan, 2015).

The study conducted by Kılıçalp and Yücel (2019) revealed that there was a significant impact on the oestrus of sheep fed barley. Furthermore, a higher ovulation rate was found in sheep fed pasture, barley, and omega-3 compared to sheep from the control group. It was also discovered that sheep fed n-3 had a higher fertility percentage compared to other groups.

Kiliçalp and Yücel (2019) reported supplementing ewes at the finishing stage with omega-3 PUFAs, which increased the fertility of Karayaka sheep. Furthermore, a higher percentage of twin births was observed in ewes supplemented with omega-3 PUFAs. It was also observed that a higher lambing percentage was obtained in Afshari ewes supplemented with omega-3 fatty acids compared to ewes supplemented with omega-6 fatty acids during the flushing period (Kia *et al.*, 2015).

2.5. NEONATAL AND PERINATAL LAMB VIGOUR IN SHEEP

The weight of weaned lambs is another economically important trait, as it directly influences the viability of a sheep production system. Capper *et al*. (2007) state that the weight of lambs weaned per ewe and lamb growth rate must be maximised to decrease mortality and thereby maintain the profitability of a unit. One of the most important factors that influences the mortality rate in neonatal lambs is hypothermia due to the depletion of brown fat reserves and possible delayed suckling (Slee, 1981). Lively and vigorous lambs at birth are more likely to survive during the perinatal period. According to Koletzko (1992), long-chain PUFAs, arachidonic acid (C20:4 (n-6)), and docosahexaenoic acid (DHA) (C22:6 (n-3)) are essential for the development of neural tissue within the neonatal and suckling animal.

2.6. MANAGEMENT TO IMPROVE LAMB VIGOUR

Capper *et al*. (2006) state that fatty acids can improve a lamb's ability to suckle after lambing. He further reports that the inclusion of PUFAs in the diet of pregnant ewes resulted in improved neonatal lamb vigour. However, there is little and conflicting information available on the effects of continuing dietary PUFA supplementation to a ewe on her lamb's growth rate (Capper *et al*., 2006).

2.6.1. Physiological role of fatty acids in ewes' milk production

Results obtained from studies on the effect of continuous supplementation of fatty acids to a lactating ewe on milk composition and lamb vigour are conflicting and inconclusive. A study by Khotijah *et al.* (2017) found a positive effect, where the addition of 6% sunflower into the ration enhanced fermentability in the rumen and had a positive effect on the lactating ewe's productivity through the improvement of body condition and improved body recovery postpartum. On the contrary, a study by Abeer *et al*. (2019) detected that supplementary oils had no effect on ewes' BW during late pregnancy and lactation when compared with a control group. In fact, all ewes involved in the experimental trial lost weight during the suckling period. Similar results were found in cows, where a decrease in BCS and BW were observed in dairy cows when SFAs or calcium salts of PUFAs were supplemented at 1.7% and 2.0%, respectively (Antonacci *et al.*, 2018). Supplementation with dietary fish oils reduced milk concentration and yield in the study by Capper *et al.* (2007). There was no significant difference observed in milk yield between ewes fed eicosapentaenoic acid (EPA) and DHA at 30 days lactation period after supplementation was ended (Coleman *et al.*, 2018). It was also observed that there was no difference in fat, protein, lactose, or solid percentage yield (P > 0.10). There was also no difference in somatic cell count (SCC), milk urea nitrogen (MUN), energy-corrected milk (ECM), and net energy (NE_1) (Coleman *et al.*, 2018). Feeding ewes with enriched sources of EPA and DHA during late pregnancy did not have an influence on ewe performance, metabolic status, or milk production and composition at 30 days in lactation (Coleman *et al.*, 2018). Other researchers also found no changes in milk yield with supplementation of various sources of EPA and DHA during lactation (Kitessa *et al.*, 2003; Reynolds *et al.*, 2006; Capper *et al.*, 2007; Gallardo *et al.*, 2014).

2.6.2. Physiological role of fatty acids on suckling lamb performance

Coleman *et al*. (2018) report that there is no difference in lamb performance for those fed both treatment (EPA + DHA) and palm fatty acid distillate (PFAD) for BW ($P = 0.22$) and ADG ($P = 0.21$). At weaning (60 days), lambs from different treatments had the same BW ($P = 14$). However, Garcia *et al. (*2014) indicate that dairy calves that were born from multiparous dams and supplemented with rumen-inert essential PUFAs within 8 weeks of pregnancy had a significantly higher birth weight compared to calves born to multiparous dams fed no fat. Dos Santos *et al*. (2019) observed no significant difference in lamb weight from birth to weaning between lambs suckled from ewes supplemented with protected fat from palm oil and a control group.

2.6.3. Physiological role of fatty acids on suckling lamb time of weaning

Dos Santos *et al*. (2019) found a significant difference ($P < 0.05$) at the time of weaning between lambs that suckled from ewes supplemented with protected fat from palm oil and a control group. Lambs from a control group weaned at 64.53 days and those from the palm oil treatment group weaned at 49.57 days of age (Dos Santos *et al*., 2019). Dos Santos *et al.* (2019) further state that ewes supplemented with dietary oils are more productive, have a shorter weaning period, and come on heat faster. However, Da Costa *et al.* (2011), who supplemented Santa Ines sheep with 30g of protected fat per sheep per day, did not observe a decrease in postpartum anoestrus.

3. CHAPTER THREE: MATERIALS AND METHODS

3.1. INTRODUCTION

This trial was carried out on fifty (50) South African Döhne Merino ewes ranging in age from 2 to 4 years (4–6 tooth), weighing 46–55kg, and with a mean BCS of 3.5 ± 0.4. The ewes were housed in a sheltered environment but had access to outside paddocks. The animals were quarantined for a period of 21 days and inspected for possible diseases and infections before the start of the trial.

The ewes were synchronised during the normal breeding season (i.e., March–April) using 200 UI kg of Pregnant Mare Serum Gonadotropin (PMSG) and kept away from rams to prevent voluntary natural mating. In the meantime, rams were kept close enough for use in the collection of semen and later use in the detection of oestrus cyclicity among the ewes.

The animals used in the research trial were managed and treated by the Paradys experimental farm's veterinarian in accordance with the standard farm management practices as dictated by the South African National Standard (SANS 10386:2008) on care and use of animals for scientific purposes. The types of dietary oils used in the study are commonly utilised on farms to optimise productivity and improve physiological processes of livestock.

3.2. PROJECT LOCATION / STUDY AREA

This project was conducted on the University of Free State farm (Paradys experimental farm) situated in the city of Bloemfontein, Free State Province, South Africa (latitude: 29.221511; longitude: 26.206606). The study commenced in January 2021 and ended in December 2021. The mean average rainfall in Bloemfontein is 548 mm per annum, with most of the rainfall occurring during summer (i.e., December–February). The Free State receives the lowest average rainfall (9mm) in June and the highest (84mm) in February. The mean annual temperature of Bloemfontein is 17.1°C, with an average summer temperature of 23ºC and an average winter temperature of 8ºC. January is the hottest month, with a temperature range of 15–32ºC, while June is the coldest, with a range of 1ºC–17ºC (SA-Venues, n.d.).

Mucina and Rutherford (2006) mapped nine biomes around South Africa – namely, Grassland, Savannah, Succulent Karoo, Nama Karoo, Forest, Fynbos, Desert, Indian Ocean Coastal Belt, and Thicket. They describe biomes as being the dominant forms of plant life and climatic factors in a specific region. The study area (Bloemfontein, Free State Province) forms part of the Grassland biome (Mucina & Rutherford, 2006), with mixed crop-livestock production dominated by dryland crop farming and sheep-cattle as the principal production systems. The dominant vegetation in the area includes *Themeda triandra* and *Eragrostis lehmanniana* as well as other continuously available grasses such as *Eragrostis curvula* and *Digitaria eriantha* (Dingaan & Du Preez, 2013).

The study protocol was reviewed and approved by the Animal Research Ethics Committee of the University of the Free State (ethics number UFS-AED2020/0020/63). This study is a culmination of two concurrent experimental trials that were carried out on the South African Döhne Merino sheep.

3.3. MANAGEMENT AND TRIAL DESIGN OF THE PROJECT

3.3.1. Housing

All 50 selected Döhne Merino ewes were housed in a sheltered paddock of 5 ha with adequate space for grazing. The paddock consisted of a mixture of natural pasture and unrestricted water access in troughs that were connected to permanent water supply (tanks) and were cleaned regularly. Each evening, the ewes were kept in an overnight sheltered kraal for protection from unfavourable weather conditions and predators.

3.3.2. Feeding of ewes

The Döhne Merino ewes were grazed daily from 09:00 to 16:00 on the pastures and, in addition, received 2 kg of pellets per day. The pellets consisted of a ration of crushed yellow maize, lucerne, and molasses. The feed composition is explained in Table **3.1** below. Ewes that experienced diarrhoea were fed 1 kg pellets and 1 kg crushed lucerne, and water was provided *ad libitum*.

Table 3.1 Nutrient composition of feed for Döhne Merino ewes

Mixing instructions (kg)					
	Main-trace lick	Lambing pen mixture	Licks for producing animals on different types of grazing		
			1	2	3
Voermol Maxiwol concentrate	400	150	150	200	250
Maize meal/whole grain	-	400	300	250	200
Salt	100	-	50	50	50
Lucerne	-	400	-	-	-
Procon 33	-	50	-	-	-
Total	**500**	**1000**	**500**	**500**	**500**
Crude protein g/kg (min)	280	160	153	180	200

Source: Barnard, J., 2021. Nutrient composition of feed for Döhne Merino ewes. Interview 21 February 2021, University of the Free State, Bloemfontein.

3.3.3. Health management

In this trial, inspections were conducted on the Döhne Merino ewes by the farm's veterinarian and farm management for any signs of disease and infection during the selection of ewes. The experimental animals were dosed for internal parasites using Prodose Orange, dipped with Prodip, and vaccinated against ovine pasteurellosis at the beginning of the trial. Sick animals were separated from healthy animals after daily routine inspections. Sick animals were isolated and kept aside in sick bays, where they were fed enough hay, had access to water, and were given appropriate treatment.

3.3.4. Experimental design

Fifty South African Döhne Merino ewes were selected based on a weight of approximately 55 kg. The ewes that weighed between 35 and 45 kg were considered underweight and those that weighed above 60 kg were considered overweight. Mature, second-parity ewes (4–6 tooth) were used in order to ensure fertility. The 50 ewes were randomly allocated to five dietary oil treatment groups; each group consisted of 10 ewes as indicated in Table **3.2** below.

Ewes were tagged, and each treatment group had its own specific tag colour. The animals were tagged on the right-hand side, and the ear tag clearly indicated the animal's number and its group.

Table 3.2 Experimental design

Treatment	Tag colours	Oil supplement
Treatment 1	Grey	Control group (n = 10)
Treatment 2	Yellow	Sunflower (n = 10)
Treatment 3	Green	Olive oil (n = 10)
Treatment 4	Blue	Fish oil (n = 10)
Treatment 5	Orange	Palm oil (n = 10)

3.4. DRENCHING OF EWES WITH DIETARY OILS

The drenching of the Döhne Merino ewes with the dietary oils commenced on 15 March 2021 and ended on 31 March 2021. Each ewe received 30 ml of oil that corresponded with the colour of the ear tag. The ewes received 30 ml of dietary oil each day throughout the two-week period. The voluntary feed intake of sheep is around 2–3 g/kg live mass or 2–3% of live mass (McDonald *et al*., 1995). The ewes in this study weighed between 50–60 kg and therefore consumed roughly 1–1.8 kg per day. It is common practice in the nutrition of ruminants to provide no more than 6% lipids in the diet, with around 3% coming from the forages and grains and the rest added as supplemental fat (Bionaz *et al*., 2020). Therefore, in order not to have a negative impact on fibre fermentation, oil was supplemented at 3% of the predicted minimum daily intake of the ewes, which was 30 ml per ewe once per day.

Drenching took place in the morning from 07:00 to 09:00, and this was performed in crush pens (as shown in Figure **3.1** below). The four different oils, as per their different colour-coded treatment groups (shown in Table **3.2**), were each allocated to their own drench gun (i.e., each treatment group had its own drench gun). For the drenching, the ewe's head was lifted in a slightly upward position, holding it towards the administrator's hip to gain full control of the animal's head. With the other hand, the drench gun was slowly inserted into the ewe's mouth (illustrated in Figure **3.2** below). Oils were gently drenched in the animal's mouth, and the animal was allowed to swallow before releasing. After drenching, the animals were taken to pasture camps for grazing and returned to the kraal at 16:30, where they were sheltered for overnight resting.

Figure 3.1 Ewes lined in a crush pen for drenching

Source: Private collection, *Tebogo Sedupane*

Figure 3.2 Drenching ewes with dietary oils

Source: Private collection, *Tebogo Sedupane*

3.5. SYNCHRONISATION OF OESTRUS USING CIDR®

The synchronisation of oestrus was done by a veterinarian, with students' assistance, on 15 March 2021. Oestrus was synchronised in the 50 South African Döhne Merino second-parity ewes using a CIDR (Pharmacia & Upjohn, Auckland, New Zealand) with 0.6 g progesterone to assist in the administering of exogenous progesterone (Plate 3.17). The CIDR was placed with an applicator and an antiseptic cream applied to act as a lubricant and to prevent bacterial infections during insertion into the vagina. The CIDRs were in place for a period of 15 days (from 15 March until 29 March 2021). All ewes were checked daily to ensure that the CIDRs remained in place for the duration of the treatment period. At CIDR removal (i.e., after 15 days), all ewes were administered 200 IU/kg of PMSG (Pharmacia Upjohn) for stimulation of oestrus. Following progestagen withdrawal, all does were stimulated using two Döhne Merino breeding rams that were introduced in the adjacent kraal for a period of 48 hours. A visual oestrus detection was performed twice daily (08:00 and 15:00) for approximately 30 minutes until the day of LAI, which took place on 1 April 2021.

3.6. LAPAROSCOPIC ARTIFICIAL INSEMINATION (LAI)

Laparoscopy is an intrauterine technique of insemination, especially used in small species to go around their unique tortuous cervix (Sathe, 2018). The laparoscopic technique for AI is regarded to be less dangerous than the traditional surgical approach (Evans *et al.*, 2004; Anel *et al.*, 2006). This technique is widely regarded by scientists to produce improved conception rates and as a highly effective method of semen administration at the point of reception compared to other surgical techniques (Stefani *et al.*, 1990; Fantinati *et al.*, 2005; Shipley *et al.*, 2007).

The procedure involves animals being restricted and sedated for a short period to achieve optimum sedation. The animal is then lifted and restrained on a custom-made LAI cradle with the fore- and hind feet tied up to restrict movement. The excess wool on the ventral abdomen up to the umbilicus is carefully sheared, allowing the area to be disinfected with 70% isopropyl alcohol (Sathe, 2018).

The LAI cradle is then elevated up to 45° for proper positioning of the ewe for the LAI procedure. A surgical blade is utilised to create a 12.5 mm incision for the insertion of a sterile, flexible cannula into the abdomen. Air insufflation into the stomach follows until it is sufficiently tense. A 5 mm trocar and cannula are inserted into the abdomen through the incision, and the trocar is then removed to give way for the 5 mm telescope to remain in the cannula in order to clearly see the caudal abdomen (Sathe, 2018).

Once the reproductive tract is seen, the second trocar and cannula are inserted near the teat cannula. A loaded LAI gun with semen straw is inserted through the port and lined up opposite the greater curvature of the uterine horn under laparoscopic eye guidance (Sathe, 2018). The semen is injected in the uterine horn. The AI gun and other instruments are withdrawn and followed by a thorough observation for bleeding. Wound spray is applied to avoid infection by flies and other parasites (Sathe, 2018).

Figure 3.3 Local anaesthetic is administered

Source: Sathe (2018)

Figure 3.4 Trocar and cannula inserted into the abdominal cavity

Source: Sathe (2018)

Figure 3.5 Laparoscopy AI gun and needle used for intrauterine injection of semen

Source: Sathe (2018)

Figure 3.6 Intra-abdominal view of the reproductive tract
Source: Sathe (2018)

For this study, LAI was conducted on 1 April 2021. All does were laparoscopically inseminated with 0.1 ml fresh diluted Döhne Merino ram semen (150X 10^6) at a fixed-time (i.e., 48 hours following CIDR withdrawal). The semen was collected from rams of proven fertility with the aid of an artificial vagina and using a ewe in heat. The semen was then evaluated microscopically for progressive motility and diluted at a ratio of 1:2 with sterilised skimmed milk. Only semen samples with 3+ motility score were utilised for AI. Laparoscopy was performed by Dr Johan Steyn from RAMSEM Pty (Ltd) – a company based in Bloemfontein that offers reproductive technology services for sheep, goats, and cattle.

3.7. FOLLOW-UP RAMS UNTIL PREGNANCY

After LAI, animals were not handled for 14 days, after which follow-up Döhne Merino rams were introduced to ewes on 15 April 2021 and removed on 30 April 2021 to observe ewes that exhibited any return to oestrus and which were then mated. The introduction of follow-up rams to the ewes is a standard procedure on the experimental farm and, for this trial, was not part of the objectives and parameters measured. Two South African Döhne Merino rams were introduced to the ewes, which were divided into two groups of 25 ewes. There was no drenching of ewes for a period of 45 days to ensure proper embryo attachment and limit their stress levels. Drenching of animals with dietary oils in different treatment groups resumed on 15 May 2021 and took place up until the end of the experimental trial on 27 November 2021.

3.8. PREGNANCY DIAGNOSIS

The importance of pregnancy diagnosis is to detect ewes that are pregnant those that are not pregnant on time. The procedure of pregnancy diagnosis was done by a farm-allocated veterinarian using a sonar scanner. The nursing facility consisted of 60 single holding pens and 12 twin pens. Ewes were placed in holding pens soon after parturition. The lactating ewes and lambs remained in the holding pens for a period of one week; if lambs were not found to be strong enough, then three more days in the pens were allowed before being introduced onto the pasture. The holding pens were covered with straw bedding that was changed every day, as shown in Figure **3.7** below.

Figure 3.7 Holding pens for lambs and ewes soon after parturition

Source: Private collection, *Tebogo Sedupane*

3.9. MEASUREMENTS AND EXPERIMENTAL PROTOCOLS

Before lambing, conception rate and pregnancy rate data of the Döhne Merino ewes were captured. Within a day after lambing, the data captured included lambs born alive, singletons versus multiple birth rate, lamb birth weight, lamb mortality rate, lamb average weight gain (weekly), ADG of lambs, time taken until weaning (determined by date)0 and, weight of the lambs at weaning.

3.10. DATA COLLECTED AND STATISTICAL ANALYSIS

3.10.1. Data collection

Lambs were separated from the flock and placed in smaller holding pens. Lambs were then weighed using a 50 kg mechanical hanging scale (see Figure **3.8**). During weighing, lambs were put inside a half bag with long handles to allow the bag to hang from a scale (see Figure **3.9**).

Weights of all lambs were recorded immediately after birth to obtain birth weight and they were shortly returned to their respective dams after weighing to avoid stress. Lambs were then weighed each week in order to obtain ADG. The last weights of lambs were obtained after 13 weeks, on 26 November 2021, during weaning.

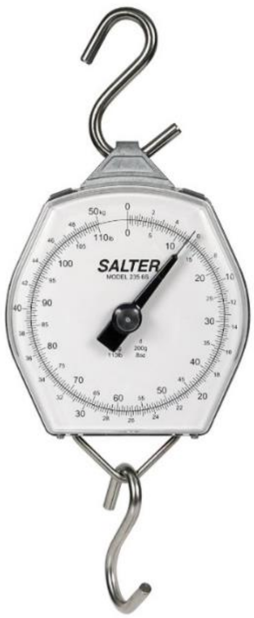

Figure 3.8 A 50kg mechanical weighing scale used to weigh lambs

Source: Weigh-Comm (n.d.)

Figure 3.9 Weighing of lambs

Source: Private collection, *Tebogo Sedupane*

3.11. PARAMETERS AND STATISTICAL ANALYSIS

3.11.1. Statistical analysis

The following calculations were performed to get the average production performance per group (Smith, 2011):

$$\text{Lambing percentage} = (\text{number of females lambing} \; / \; \text{total number of mated females}) \text{ x } 100$$

$$\text{Weaning percentage} = (\text{number of lambs weaned} \; / \; \text{number of lambs born}) \text{ x } 100$$

For the mean ± SD litter size and gestation length and birth and weaning weights between males and females, the study employed a one-way analysis of variance (ANOVA) to examine the effect of treatment on the outcome variable, followed by a post-hoc test using Tukey’s Honest Significant Difference (HSD) to identify significant differences between treatment groups (IBM SPSS statistics 27 programme) at a 95% confidence interval.

For the growth at specific points in time, ADG over time, growth rate, and % increment over time, the study utilised a repeated-measures ANOVA to examine the effect of treatment on the outcome variable over time, followed by a post-hoc test using Tukey’s HSD to identify significant differences between treatment groups at specific time points at a 95% confidence interval. The IBM Statistical Package for the Social Sciences (SPSS) 27 software programme was utilised.

4. CHAPTER FOUR: RESULTS AND DISCUSSION

4.1. PRODUCTION PARAMATERS OF DÖHNE MERINO EWES SUPPLEMENTED WITH DIFFERENT DIETARY OILS

A review by Kenyon *et al.* (2014) suggests a positive relationship between BCS and breeding and ewe reproductive traits (pregnancy rate and lambing percentage) in ewes supplemented with dietary oils. Generally, these parameters are influenced by an increase in BCS from 2.5–3.0 (Keyon *et al*., 2004; Abdel-Mageed, 2009; Yilmaz *et al.*, 2011).

This study evaluated the growth performance of lambs suckling on ewes that were supplemented with plant or animal-derived dietary oils (sunflower, olive, fish, and palm oil). It is important to determine any positive effects of such supplementation (if any are present at significant levels) and also to ensure that the production performance of the ewes is not negatively affected by treatment. The results obtained in this study in relation to different production parameters of the ewes receiving different types of supplementation (or no supplementation) are discussed in detail in this chapter.

4.1.1. Conception and lambing rates of Döhne Merino ewes supplemented with different dietary oils

The fertility evaluation of Döhne Merino ewes in this study was an important tool to verify *in vivo* results of the various oil treatments and to ensure that oil supplementation did not have a negative effect. All the ewes in the five allocated treatment groups showed oestrus behaviour within 48 hours following CIDR withdrawal, indicating the effectiveness of oestrus synchronisation. In total, 44 of the 50 ewes conceived and lambed; six ewes from four treatment groups did not conceive (control group = 1 out of 10, olive oil = 1 out of 10, Fish oil = 1 out of 10, and palm oil = 3 out of 10). A conception rate of 100% (10 out of 10) was experienced in the olive oil group. CIDRs accompanied by PMSG have been extensively used in many countries for oestrus synchronisation of small ruminants to improve ovulation rate (Najafi *et al*., 2014). The reproductive performance in the present study demonstrated that a good ovarian and conception response could be achieved by using CIDR plus PMSG in Döhne Merino sheep. Vilariño *et al.* (2013) conducted a study to determine ovarian responses in sheep inseminated following a short-term protocol (six days of treatment) with previously used controlled CIDR-G devices. They concluded that a short-term CIDR protocol was effective for oestrous synchronisation and ovulation. In another study, PMSG treatment was found to result in rapid follicle growth, an increased number of ovulated follicles, and synchronisation of oestrous and ovulation (Banchera *et al.*, 2021). The average conception and lambing rates of the Döhne Merino ewes across all treatment groups in this study were both 88%. These results are in the same range as those of Jainudeem *et al.* (2000), who found an average conception rate of 85% in mature sheep.

The average conception and lambing rate of the palm oil treatment group was found to be 70%; although this is lower than the other treatment groups in the present study, it is higher than that found by Masoudi *et al.* (2016), who observed a pregnancy rate of 44% and a lambing rate of 46% in Zandi ewes that were supplemented with palm oil (n-3). The lower conception and lambing rate found in the palm oil treatment group may be caused by follicle-enclosed oocytes that prefer to gather SFAs, and high levels of palmitic (c16:0) and stearic (c18:0) acid damage progression and post-fertilisation development (Sinclair, 2010). Possibly, this may be due to selective uptake mechanisms in the ovary and high levels of relatively saturated free fatty acids in peripheral circulation (Sinclair, 2010). On the other hand, the olive oil treatment recorded conception and lambing rates of 100%, while the rest of the treatment groups and the control group all recorded conception and lambing rates of 90%.

4.1.2. Gestation length of Döhne Merino ewes supplemented with different dietary oils

Gestation length is generally defined as time from recorded date of insemination or mating until the date of lambing. Gestation involves changes in physiological parameters, including hormones and enzymes, and functions of the gastrointestinal and cardiovascular system (Aydin *et al.*, 2010). These developmental changes are essential for the growing foetus and for preparing the dam for parturition. Maternal nutrition during pregnancy is instrumental to foetal growth regulation and has an impact on the offspring's health and productivity throughout their lifespan (Belakcemi *et al.*, 2010). The results obtained in relation to the gestation length (in days) of Döhne Merino ewes supplemented with different dietary oils are presented in terms the mean and standard deviation (SD) in Table **4.1**.

Table 4.1 The mean ± SD gestation length (days) of ewes supplemented with different dietary oils

	Treatment groups					
	Control group*	**Sunflower oil**	**Olive oil**	**Fish oil**	**Palm oil**	**Difference**
Gestation period (days)	150 ± 1.36	155 ± 7.78	154 ± 8.22	154 ± 6.56	154 ± 6.75	NS

*Control: no oil (sunflower oil, olive oil, fish oil, or palm oil)

The gestation length in the current study ranged between 150 and 155 days, with an overall mean of 153 ± 6.13 days. There was no significant difference ($P > 0.05$) between the groups.

The length of gestation in sheep is generally variable. Alshdaifat (2017) found that the addition of fish oil and palm oil resulted in shorter gestational length in Awassi ewes (palm oil 145 days and fish oil 149 days). Fogarty *et al*. (2005) found a gestation length that ranged between 147.1 (Finnsheep cross lambs) and 150.3 days (Corridale sired lambs). A review by Cardoso and Padmanabha (2019) report a mean gestation period of 147 days for ewes, with a range of 142 to 155 days. De Carolis *et al. (*2020) found the average gestation period of ewes to be 149 days in Sarda and Lacaune sheep. The gestation lengths in the current study was found to be longer than the means of other studies. This could be due to factors such as dam age, as older animals may have a lower BCS and live weight – both of which affect gestation length (Cam *et al*., 2018; Tozlu Celik *et al*., 2021). Furthermore, litter size (singleton births versus twins or triplets) and nutrition can affect the weight of the lamb, as inadequate nutrition supplied to pregnant ewes due to poor-quality feed may result in slow growth of the foetus, while heavier lambs may result in longer gestation (Öztűrk & Aktaş, 1996).

4.1.3. Mortality and weaning rates of lambs from Döhne Merino ewes supplemented with different dietary oils

A mortality rate of 11% and 14% was observed in groups that were supplemented with fish oil and palm oil, respectively. The cause of mortality for two lambs can be attributed to starvation due to poor mothering ability of ewes, Although the records of mothering ability at the parity is not known. In this current study, a 100% weaning rate was observed in the control group, sunflower oil group, and olive oil group. A weaning rate of 88% was found in the fish oil group and the lowest rate (85.70%) was found in the palm oil treatment group.

4.1.4. Fecundity of Döhne Merino ewes supplemented with different dietary oils

The average fecundity rates (lambs born/ewes lambing) for the different treatment groups ranged from 100% to 133%, with the control group at 122%, sunflower treatment 133%, olive and fish oils both 100%, and palm oil 114%. The sunflower treatment group had the highest fecundity rate. Brand *et al*. (2014) report an annual fecundity of 262% on Dohne Merino ewes supplemeted with self formulated ration and licks. Supplementing with Bersem clover throughout seasonal anoestrus disrupts behavioural estrus by decreasing fecundity (Hashem et al., 2018).

Figure **4.1** below indicates the litter size of Döhne Merino ewes supplemented with dietary oils. The ewes in this experimental study only had single and twin births; there were no triplet births. There were 38 single births, 6 twin births, and 6 ewes that did not lamb. The different treatments administered had no significant ($P > 0.05$) effect on litter size in the current study.

Marais (2011) conducted a study supplementing 144 ewes with dietary oils; 77 (1.26 ± 0.64) gave birth to single lambs and 67 (1.45 ± 0.58) gave birth to twins. Lévy *et al.* (2020) conducted a study in which 22 Ile de France ewes were divided into a control group and a group supplemented with sunflower meal. The group of 9 ewes that were supplemented with sunflower gave birth to 15 lambs – 8 singles, 2 twins, and 3 triplets. The 13 ewes from the control group gave birth to 15 lambs – 11 singles and 4 twins.

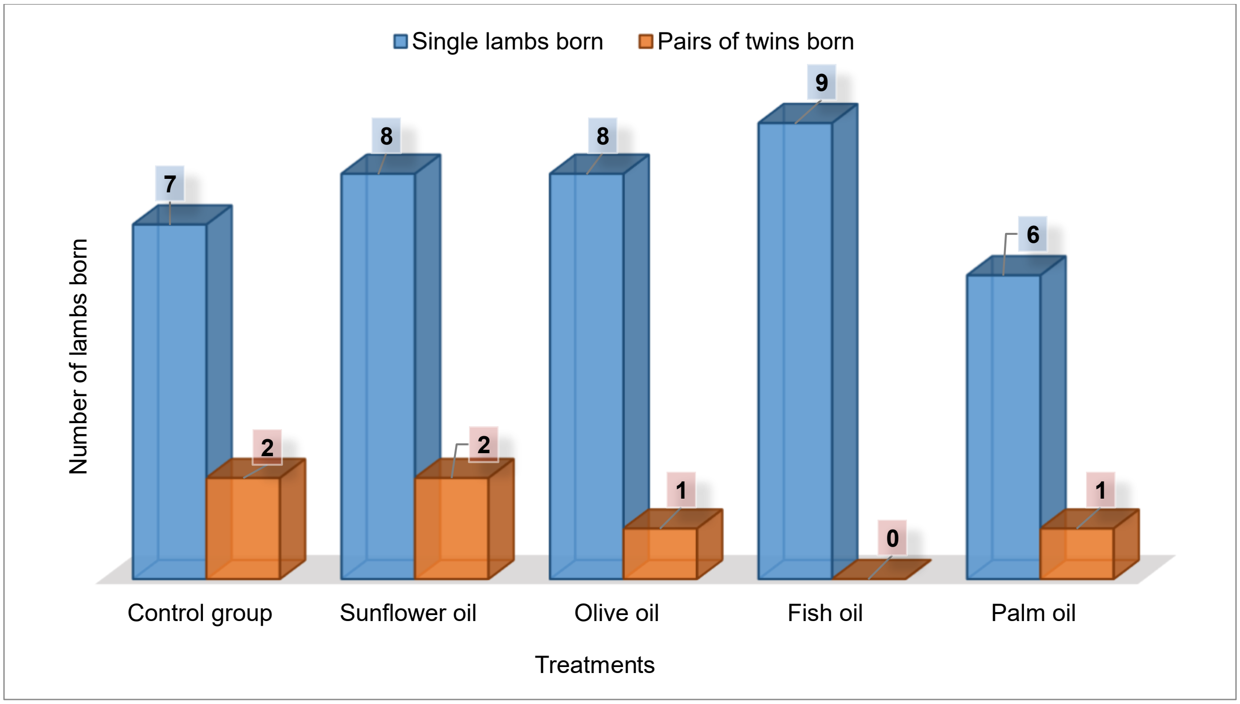

Figure 4.1 Number of single and pairs of twin lambs born from ewes supplemented with no oil, sunflower oil, olive oil, fish oil, and palm oil

4.2. GROWTH PERFORMANCE OF LAMBS BORN FROM DÖHNE MERINO EWES SUPPLEMENTED WITH DIFFERENT DIETARY OILS

4.2.1. Weight at different points in time of lambs born from Döhne Merino ewes supplemented with different dietary oils

Previous research on sheep did not conclusively demonstrate that lamb birth weight is affected by prepartum supplementation of protected SFAs, PUFAs (Palmquist *et al.*, 1977), or fish oil (Capper *et al.*, 2006). However, dairy calves born from multiparous dams fed rumen-inert SFAs or essential PUFAs during the last 8 weeks of pregnancy had a significantly higher birth weight compared to calves born to multiparous dams fed no fatty acids (Garcia *et al.*, 2014). Gardner *et al*. (2007) indicated advanced factors that influence birth weight include environmental effects, nutrition, BCS, genetics, gestation length, and sex of a lamb.

In this study, supplementing pregnant and lactating ewes up until weaning of lambs with diets containing PUFA was found to positively enhance post-lambing performance, as the

There was improvement or significance (*P*>0.05) on percentage increment in weight over time and ADG. Previous studies found that during later stages of gestation, intake in pregnant dams is due to reduced ruminal capacity; on the other hand, nutrient requirements increase for foetal growth, udder development, and lactogenesis (Vicente-Pérez *et al.*, 2015).

The birth weight and weight at different points in time of lambs from ewes supplemented with different dietary oils and a control group are indicated in Table **4.2**. In the current study, there were no significant (P > 0.05) differences between different treatment groups in terms of birth weights.

Table 4.2 The mean ± SD birth weight and weight at different points in time of lambs from ewes supplemented with different dietary oils

	Treatments groups					
	Control group*	**Sunflower oil**	**Olive oil**	**Fish oil**	**Palm oil**	**Difference**
Initial body weight (birth)	5.49 ± 0.94	5.49 ± 0.67	5.71 ± 0.81	5.74 ± 0.65	5.46 ± 0.28	NS
Body weight (kg) Week 1	7.20 ± 1.62	7.83 ± 0.78	8.10 ± 1.30	7.35 ± 0.80	7.66 ± 1.00	NS
Body weight (kg) Week 2	9.96 ± 1.94	10.52 ± 1.07	10.98 ± 1.82	10.18 ± 0.53	10.46 ± 1.27	NS
Body weight (kg) Week 3	12.79 ± 2.34	13.16 ± 1.28	13.51 ± 2.53	12.81 ± 0.64	12.72 ± 1.82	NS
Body weight (kg) Week 4	18.40 ± 2.70	19.37 ± 1.84	19.92 ± 3.10	19.28 ± 1.25	19.00 ± 2.37	NS
Body weight (kg) Week 5	17.63 ± 3.11	20.56 ± 2.32	19.92 ± 4.09	18.92 ± 1.83	18.80 ± 2.88	NS
Body weight (kg) Week 6	19.40 ± 3.52	20.43 ± 2.62	20.92 ± 4.07	20.50 ± 2.04	20.10 ± 2.90	NS
Body weight (kg) Week 7	20.45 ± 3.34	21.37 ± 2.51	21.85 ± 4.58	21.35 ± 2.07	20.80 ± 2.68	NS
Body weight (kg) Week 8	21.90 ± 3.68	23.18 ± 2.72	23.92 ± 5.15	23.14 ± 2.93	23.10 ± 3.18	NS
Body weight (kg) Week 9	22.86 ± 3.70	25.25 ± 3.04	25.00 ± 5.45	24.14 ± 2.67	24.80 ± 3.25	NS
Body weight (kg) Week 10	24.31 ± 3.93	26.43 ± 2.98	26.35 ± 5.55	26.50 ± 2.88	25.90 ± 3.71	NS
Body weight (kg) Week 11	26.00 ± 4.26	27.18 ± 3.62	28.07 ± 5.36	27.64. ± 3.02	27.10 ± 3.48	NS
Body weight (kg) Week 12	26.68 ± 4.08	27.87 ± 3.83	28.00 ± 4.80	28.42 ± 2.76	27.40 ± 4.05	NS
Body weight (kg) Week 13	28.63 ± 4.46	26.91 ± 6.23	28.10 ± 5.38	29.50 ± 4.23	28.35 ± 3.89	NS

*Control: no oil (sunflower oil, olive oil, fish oil, or palm oil)

In a study by Ghoniem and Atia (2020), 0.4% and 6% of calcium soap as protected fatty acid were offered to Sufflolk X Ossimi crossed ewes in the experimental ration. Improved birth weights of 7.19% and 19.93% for inclusion of 0.4% and 6% were observed, respectively. Similar results were reported by Gabr *et al*. (2008), who found that lambs born from ewes supplemented with fish oil displayed higher ($P < 0.05$) birth weight and growth rate than the control group. Shakweer *et al.* (2010) recorded similar results for kids of Damcus goats at birth. The ruminant placenta indicates the larger degree of disconnection between maternal and foetal physiology, requiring a direct route by which microbes my access the foetal gut (Gwendolynn & Cunningham-Hollinger, 2022). The birth weights of Döhne Merino lambs in this study ranged from 4.85 ± 0.07 kg to 5.8 ± 0.74 kg.

4.2.2. Birth and weaning weights of male and female lambs from Döhne Merino ewes supplemented with different dietary oils

Table **4.3** below indicates the birth and weaning weights of male and female lambs from ewes supplemented with sunflower oil, olive oil, fish oil, palm oil, and no oil. For male lambs, the birth weights (kg) were 4.85 ± 0.70, 4.85 ± 0.70, 5.5 ± 0.49, 5.4 ± 0.33, and 5.6 ± 1.19, respectively; for females the birth weights were 5.70 ± 0.61, 5.8 ± 0.70, 5.8 ± 0.74, 4.9 ± 0.50, and 5.41 ± 0.87, respectively, for the sunflower oil, olive oil, fish oil, palm oil, and control groups. No significant difference ($P > 0.05$) was observed in the birth weights of different groups and genders.

The current study does not align with the study by Alshdaifat (2017), which found that the inclusion of fish oil during the late stages of gestation improved the birth weight of lambs. Hamada *et al*. (2013) also reported that supplementing ewes with fish oil during late gestation improves lamb birth weight. Improved lamb birth weight may be due to a longer gestation period. Alshdaifat (2017) reported a relationship between foetal growth rates and gestation length of 0.15–0.2 kg weight gain per day during late pregnancy.

The weaning weights (kg) for male lambs that suckled on ewes that were supplemented with sunflower oil, olive oil, fish oil, palm oil, and in the control group were as follows: 25.5 ± 8.13, 29.0 ± 4.88, 30.2 ± 4.59, 28.8 ± 5.03, and 28.8 ± 4.93, respectively. The weaning weights (kg) for female lambs within the aforementioned groups were as follows: 28.3 ± 3.81, 25.8 ± 6.93, 28.9 ± 4.51, 27.6 ± 2.46, and 28.5 ± 4.58, respectively. No significant difference ($P > 0.05$) was observed in weaning weights of different groups and genders. Results from Manso *et al*. (2011) and Berthelot *et al*. (2012) are consistent with the present study, as different dietary oils supplemented to ewes did not influence the weaning weights of suckling lambs or their ADG before weaning. Similar results have been reported by Bampidis *et al*. (2005), who found that oregano essential, when used as a supplement, did not positively stimulate lamb growth performance and muscle tissue growth.

Table 4.3 Mean ± SD birth and weaning weights of male and female lambs born from ewes supplemented with different dietary oils

	Sunflower oil	Olive oil	Fish oil	Palm oil	Control*	Difference
Birth weight (kg) Male	4.85 ± 0.70	5.6 ± 0.84	5.5 ± 0.49	5.4 ± 0.33	5.6 ± 1.19	NS
Birth weight (kg) Females	5.70 ± 0.61	5.8 ± 0.70	5.8 ± 0.74	4.9 ± 0.50	5.41 ± 0.87	NS
Weaning weight (kg) Male	25.5 ± 8.13	29.0 ± 4.88	30.2 ± 4.59	28.8 ± 5.03	28.8 ± 4.93	NS
Weaning weight (kg) Females	28.3 ± 3.81	25.8 ± 6.93	28.9 ± 4.51	27.6 ± 2.46	28.5 ± 4.58	NS

*Control: no oil (sunflower oil, olive oil, fish oil, or palm oil)

4.2.3. Growth rate (% increment and ADG) over time of lambs born from Döhne Merino ewes supplemented with different dietary oils

When an animal grows up, two things are experienced: firstly, BW increases until maturity is reached, and this is called growth; secondly, the body reaches its full function and shape, and this is called development. Growth is demonstrated in quantitative terms phenotypic characteristics such as height, length, girth, and volume. Development consists of qualitative characteristic; these characteristics may also be referred to as genotypic characteristics.

Growth and development of the entire animal rely on processes taking place at a tissue and cellular level (Batt, 1980). These processes include: (a) increase in cell size (hypertrophy); (b) net increase in cell number (hyperplasia) by motorised cell division; (c) differentiation of cells into type, which gives rise to tissues of differing structure and function (including extra-cellular material such as collagen and elastin); and (d) organisation of hypertrophy, hyperplasia, and differentiation. Development is thus linked closely with the growth of tissue (Batt, 1980).

Batt (1980) indicates that the growth rate differs in various tissues and regions of the body. Graphically, these rates tend to go up in regular succession known as growth waves. Growth waves are there for the placement of tissue, for tissue is not laid down at a uniform rate throughout. Kidney fat is the first to be developed and intramuscular (marbling) fat the last. The deterioration in animal growth curve is due to reduced cell number in tissues rather than to a difference in cell size. Animals and children will continue to develop even when presence of food is insufficient for growth (Batt, 1980).

Table **4.4** below illustrates lambs' percentage increment in weight over time, from birth to weaning. This percentage increment showed improvement or significance ($P < 0.05$) in weeks 5, 9, and 11. Fast growth (growth spurt) may follow sufficient feeding, depending on the level to which the animal's growth has deteriorated and the stage at which deterioration occurred, which varies with species. A growth spurt entails an increase in mitotic rate of cells. This may be adequate to lead to complete compensation for the deterioration; however, a growth spurt cannot compensate adequately where there has been early and continuous underfeeding (Batt, 1980).

A growth spurt was experienced in week four, during which time it was important to feed lambs sufficiently. This helps to sustain the growth of lambs, which results in better weaning weights.

Table 4.4 The mean ± SD percentage (%) increment in weight over time (in weeks) from birth until weaning of lambs from ewes supplemented differed dietary oils

	Treatments groups					
	Control group	**Sunflower oil**	**Olive oil**	**Fish oil**	**Palm oil**	**Difference**
% Increment Week 1	30.93 ± 19.71	44.68 ± 13.19	45.98 ± 18.59	28.62 ± 11.63	39.83 ± 11.31	NS
% Increment Week 2	39.68 ± 9.31	34.34 ± 5.01	35.54 ± 4.26	39.48 ± 12.77	36.70 ± 5.02	NS
% Increment Week 3	28.86 ± 10.07	26.11 ± 7.14	20.53 ± 4.97	25.85 ± 3.35	39.21 ± 39.04	NS
% Increment Week 4	45.82 ± 16.23	54.14 ± 14.92	59.50 ± 19.80	50.44 ± 4.59	60.42 ± 20.71	NS
% Increment Week 5	-4.60 ± 4.52^{b}	15.39 ± 28.87^{a}	0.29 ± 7.14	-1.99 ± 4.45	-1.93 ± 7.17	0.05
% Increment Week 6	10.07 ± 3.98	0.99 ± 12.55	6.49 ± 9.33	8.45 ± 6.58	10.79 ± 7.84	NS
% Increment Week 7	5.75 ± 4.16	5.74 ± 5.18	4.39 ± 3.04	4.25 ± 3.05	4.09 ± 4.10	NS
% Increment Week 8	7.03 ± 2.38	10.52 ± 6.02	10.26 ± 3.39	8.19 ± 5.88	11.39 ± 3.62	NS
% Increment Week 9	4.60 ± 5.16^{b}	9.14 ± 3.83^{a}	4.59 ± 2.44	4.56 ± 3.97^{b}	8.09 ± 1.65^{b}	0.05
% Increment Week 10	6.48 ± 4.29	8.24 ± 9.27	9.13 ± 6.18	9.84 ± 3.48	5.49 ± 8.24	NS
% Increment Week 11	6.99 ± 4.64^{a}	2.13 ± 5.01^{b}	6.07 ± 2.95	4.33 ± 2.58	5.92 ± 4.51	0.05
% Increment Week 12	2.81 ± 21.92	2.59 ± 2.54	1.00 ± 2.24	3.00 ± 3.44	1.64 ± 3.89	NS
% Increment Week 13	7.33 ± 3.06	7.11 ± 2.89	7.56 ± 2.38	6.43 ± 2.06	7.67 ± 2.42	NS

*Control: no oil (sunflower oil, olive oil, fish oil, or palm oil)

$^{a, b}$ Values with different superscripts in the same row differ significantly ($P < 0.05$)

Table **4.5** below illustrates the ADG of lambs from ewes supplemented with different dietary oils and a control group. There were significance differences ($P < 0.05$) in ADG of lambs among the groups in weeks 5, 6, 10, 11, and 13. Changes in performance of suckling lambs are mostly associated to variance in milk yield as well as milk fat and protein levels (Manso *et al.*, 2011). Some studies (Zhou *et al.*, 2020; Wang *et al.*, 2022) report that essential oils expand the proportional abundance of bacterial families (*Lachnospiraceae*, *Rikenellaceae,* and *Chistensenellaceae*) that are positively correlated with ADG and negatively correlated with FCR (Yang *et al.*, 2018; Li *et al.*, 2022). Wu *et al*. (2021) report that dietary inclusion of low doses (50, 80, and 250mg/kg DM) of essential oils improves serum levels of IGF-1 (insulin-like growth factor 1) in lambs, which is positively correlated with ADG in sheep. Supplementation of essential oils improves the abundance of *Lachnospiraceae* bacteria in bovine rumen fluid (Zhou *et al.*, 2020), which has a positive correlation with the length of papillae in sheep (Yang *et al.*, 2018; Mao *et al.*, 2021).

Table 4.5 The mean ± SD average daily gain (kg) over time (in weeks) from birth until weaning of lambs from ewes supplemented with different dietary oils

	Treatments groups					
	Control group*	**Sunflower oil**	**Olive oil**	**Fish oil**	**Palm oil**	**Difference (P-Value)**
ADG Week 1	0.24 ± 0.15	0.34 ± 0.09	0.36 ± 0.14	0.23 ± 0.09	0.31 ± 0.10	NS
ADG Week 2	0.39 ± 0.79	0.47 ± 0.19	0.54 ± 0.23	0.40 ± 0.09	0.40 ± 0.7	NS
ADG Week 3	0.40 ± 0.13	0.35 ± 0.8	0.30 ± 0.14	0.38 ± 0.05	0.36 ± 0.14	NS
ADG Week 4	0.80 ± 0.19	0.79 ± 0.24	0.89 ± 0.10	0.92 ± 0.10	0.89 ± 0.83	NS
ADG Week 5	-0.11 ± 0.10[b]	0.20 ± 0.35[a]	0.01 ± 0.17	-0.05 ± 0.12	-0.04 ± 0.18	0.01
ADG Week 6	0.25 ± 0.12[a]	0.01 ± 0.29[b]	0.14 ± 0.10	0.22 ± 0.18	0.24 ± 0.13	0.05
ADG Week 7	0.15 ± 0.10	0.14 ± 0.12	0.12 ± 0.09	0.12 ± 0.89	0.10 ± 0.12	NS
ADG Week 8	0.20 ± 0.08	0.26 ± 0.89	0.29 ± 0.10	0.26 ± 0.19	0.31 ± 0.12	NS
ADG Week 9	0.14 ± 0.17	0.27 ± 0.10	0.14 ± 0.09	0.14 ± 0.12	0.25 ± 0.04	NS
ADG Week 10	0.21 ± 0.14	0.22 ± 0.18	0.26 ± 0.14	0.34 ± 0.12[a]	0.17 ± 0.22[b]	0.05
ADG Week 11	0.24 ± 0.16	0.08 ± 0.16[b]	0.21 ± 0.09[a]	0.16 ± 0.10	0.19 ± 0.12	0.05
ADG Week 12	0.09 ± 0.07	0.10 ± 0.08	0.02 ± 0.18	0.11 ± 0.12	0.06 ± 0.15	NS
ADG Week 13	0.38 ± 0.12	0.35 ± 0.17	0.3 ± 0.17[b]	0.38 ± 0.10[a]	0.35 ± 0.17	0.05

*Control: no oil (sunflower oil, olive oil, fish oil, or palm oil)

[a, b] Values with different superscripts in the same row differ significantly (*P* < 0.05)

ADG: average daily gain (kg)

5. CHAPTER FIVE: CONCLUSION AND RECOMMENDATIONS

5.1. CONCLUSION

Globally, livestock producers, including sheep producers, face various challenges such as high input costs, diseases, and global warming and thus are under pressure in all fronts to farm sustainably and viably. In order to maximise the economic return from sheep farming, farmers have to optimise their reproductive efficiency. The use of dietary oils has been shown to improve reproductive efficiency and fecundity and accelerate lamb growth in different species.

The aim of this study was to investigate the effect of various dietary oils (sunflower oil, olive oil, fish oil, palm oil) on the reproductive efficiency and lamb growth performance in Döhne Merino ewes as compared to a control group where no oil was supplemented. The Döhne Merino is known to be fertile and have a high libido (Delport, 2019).

The conception rate through LAI and the use of a follow-up ram was 88%.The aim of the current study was not to compare the LAI and natural mating; thus, it is impossible to recommend to farmers which of these methods to use. There was no significant difference ($P > 0.05$) in gestation length between any of the groups. Therefore, the oral supplementation of dietary oils had no effect on shortening or extending the gestation length of Döhne Merino ewes. Gestation length was also not affected by birth status (single vs twins), sex of the lambs, age of ewes, management, or nutrition.

There were no differences in fecundity, litter size, lamb sex, and ratio of male to female lambs between any of the groups in the present study. The percentage increment in lambs' weight over time, from birth to weaning, showed improvement or significance ($P < 0.05$) in weeks 5, 9 and 11. Brand *et al*. (2014) report an annual fecundity (lambs born/ewes lambing) of 262%, whereas the average fecundity rates for the different treatment groups in this study ranged from 100% to 133%.

Furthermore, the oral supplementation of dietary oils to ewes used in this study made no positive contribution to birth weight, weaning weight, or mortality rate of lambs born from supplemented ewes. However, significant differences ($P < 0.05$) in lamb ADG were observed in weeks 5, 6, 10, 11, and 13. The results of this study were obtained from supplementation of ewes before mating until weaning of lambs.

The overall and general conclusion that can be made based on the results of this study is that oral supplementation of dietary oils – namely, sunflower, olive oil, fish oil, and palm oil – did not significantly affect the reproductive performance of ewes. In addition, oral supplementation of dietary oil to ewes had no positive effect on the growth performance of lambs, as all treatment groups performed similarly to the control group.

It should be noted that the variation between groups is an indication that significant differences may have been found between groups if more uniform or more animals were used in the study.

5.2. RECOMMENDATIONS

- Due to increased pressure to produce high-yielding lambs in a shorter period, farmers are often advised on supplementation. Preliminary results from this study shows that oral supplementation of dietary oils to Döhne Merino ewes is expensive and does not appear to be an effective way to improve the reproductive performance of ewes or the growth performance of their offspring.
- It is recommended that farmers follow good farming practices that include a proper feeding programme, management of mating season, and a vaccination programme in order to ensure the profitability of their farming enterprise.
- It is recommended that a follow-up study be undertaken, which will investigate puberty in small rams and milk solids in ewes.

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