Determining the potential of a LoRa technology approach to measure methane emission in sheep

by

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Summary

As a result of their contribution to global methane (CH_4) emissions, ruminants are under scrutiny, with research focusing on quantifying CH_4 production to contribute to the development of CH_4 mitigation strategies. Previous studies have quantified CH_4 emissions from ruminants; however, these studies were carried out under controlled conditions, and therefore the results cannot be extrapolated to animals under extensive (free range) production conditions. Despite the various studies on CH_4 emissions in ruminants, there is a lack of data regarding CH_4 emissions in sheep under extensive production conditions. Agriculture as an industry is in a unique state of transformation, as new technologies provide the opportunity to create automated and data-driven agricultural practices. A prominent technology available to the industry is LoRa (Long Range), a sub-technology of the 'Internet of Things' (IoT). LoRa technology presents an opportunity for the development of a low-power, affordable, and simple CH_4 measurement technique, which can measure CH_4 emissions with little to no human input. This study aimed to determine the potential of a novel LoRa CH_4 detection unit to measure CH_4 emissions in sheep under South African grazing conditions. The CH_4 emissions of ten intact Dohne Merino rams grazing kikuyu pasture were determined using the LoRa CH_4 detection units, a hand-held Laser methane detector, and an Australian-adapted Tier 2 approach. Three LoRa CH_4 detection units were installed in a 0.07 ha camp, and set to take CH_4 measurements for ten days, i.e. two days where background CH_4 concentrations were measured, and eight days where sheep CH_4 emissions were recorded. The LMD was used to take daily enteric CH_4 emission measurements from each ram for ten days. The objectives of this study included determining the potential of the LoRa CH_4 detection units to measure sheep CH_4 emissions under grazing conditions, and to compare emissions measured by the LoRa devices with that recorded by a laser methane detector (LMD). Both devices were used to establish diurnal CH_4 emissions in sheep, and to compare the recorded levels with the calculated IPCC Tier 2 levels for sheep under grazing conditions. The effect of ambient conditions on the CH_4 concentrations measured by the LoRa detection units was investigated. Relative humidity had a significant positive correlation with the CH_4 concentrations measured by the LoRa detection units, while air temperature, wind speed and solar radiation had a negative correlation with the CH_4 concentrations measured by the LoRa detection units. Significant correlations were reported for Device 3 only. The LoRa detection units and LMD compared favourably in terms of the characterization of the diurnal fluctuation in CH_4 concentration. The CH_4 levels measured per ram by LoRa Devices 1 (24.0 ppm) and Device 2 (52.9 ppm) were significantly higher than the levels detected by the LMD (13.9 ppm), while the CH_4 levels measured per ram by LoRa Device 3 (11.9 ppm) were similar to the LMD detected levels. The IPCC Tier 2 approach (10.3 g/day) underestimated the CH_4 emissions per ram compared to the LMD (27.6 g/day). It was not possible to compare the CH_4 emissions data obtained using the LoRa technology and Tier 2 approach in this study as their emission estimates had different units (ppm versus g/day, respectively). The LoRa CH_4 detection device developed for this study, has the potential to be a low-cost and practical measurement technique to quantify CH_4 emissions from sheep under grazing conditions, limited to use in small, controlled camps. Once the device design is refined to overcome the few limitations identified in this study, the LoRa technology can assist with the generation of sheep CH_4 emission data under various production conditions to improve emission inventories and verify mitigation strategies, on a national and international scale.

Opsomming

As gevolg van hul bydrae tot globale metaanvrystellings (CH_4) word herkouers onder die loep geneem, met navorsing wat fokus op die kwantifisering van CH_4-produksie om by te dra tot die ontwikkeling van strategieë wat sal bydra tot verlaging van CH_4 geproduseer deur herkouers. Vorige studies het CH_4-emissies van herkouers gekwantifiseer; hierdie studies is egter onder beheerde toestande uitgevoer en daarom kan die resultate nie geëkstrapoleer word na diere onder ekstensiewe (vrylopende) produksietoestande nie. Ten spyte van die verskeie studies oor CH_4 vrystellings by herkouers, is daar 'n gebrek aan data rakende CH_4 emissies by skape onder ekstensiewe produksietoestande. Landbou as 'n bedryf is in 'n unieke toestand van transformasie, aangesien nuwe tegnologie die geleentheid bied om geoutomatiseerde en data-gedrewe landboupraktyke te skep. 'n Prominente tegnologie wat vir die bedryf beskikbaar is, is LoRa (*Long Range*), 'n sub-tegnologie van die 'Internet of Things' (IoT). LoRa tegnologie bied 'n geleentheid vir die ontwikkeling van 'n lae-krag, bekostigbare en eenvoudige CH_4 metingstegniek, wat CH_4 emissies kan meet met minimale menslike insette. Hierdie studie het ten doel gehad om die potensiaal van 'n nuwe LoRa CH_4 metingseenheid te bepaal om CH_4 emissies in skape onder Suid-Afrikaanse weidingstoestande te meet. Die CH_4 vrystellings van tien intakte Dohne Merino ramme wat kikoejoe weiding bewei het, is bepaal deur gebruik te maak van die LoRa CH_4 metingseenhede, 'n laser-metaan metingtoestel (LMD) en 'n Australies-aangepaste Vlak 2-benadering. Drie LoRa CH_4 metingseenhede is in 'n kamp van 0,07 ha geïnstalleer en gestel om CH_4 metings vir tien dae te neem, dit wil sê twee dae waar agtergrond CH_4 konsentrasies gemeet is, en agt dae waar skaap CH_4 vrystellings aangeteken is. Die LMD is gebruik om daaglikse enteriese CH_4 emissiemetings van elke ram vir tien dae te neem. Die doelwitte van hierdie studie het ingesluit die bepaling van die potensiaal van die LoRa CH_4 metingseenhede om skaap CH_4 vrystellings onder weidingstoestande te meet, en om emissies gemeet deur die LoRa eenhede te vergelyk met dié wat deur 'n LMD aangeteken is. Beide toestelle is gebruik om daaglikse CH_4 emissies by skape vas te stel, en om die aangetekende vlakke te vergelyk met die berekende IPCC Vlak 2-vlakke vir skape onder weidingstoestande. Die effek van omgewingstoestande op die CH_4 konsentrasies gemeet deur die LoRa metingseenhede is ondersoek. Relatiewe humiditeit het 'n beduidende positiewe korrelasie gehad met die CH_4 konsentrasies gemeet deur die LoRa metingseenhede, terwyl lugtemperatuur, windspoed en sonstraling 'n negatiewe korrelasie gehad het met die CH_4 konsentrasies gemeet deur die LoRa metingseenhede. Beduidende korrelasies is slegs vir Toestel 3 aangemeld. Die LoRa metingseenhede en LMD het gunstig vergelyk in terme van die karakterisering van die daaglikse fluktuasie in CH_4 konsentrasie. Die CH_4 vlakke gemeet per ram deur LoRa metingseenheid 1 (24.0 dpm) en metingseenheid 2 (52.9 dpm) was aansienlik hoër as die vlakke wat deur die LMD opgespoor is (13.9 dpm), terwyl die CH_4 vlakke gemeet per ram deur LoRa metingseenheid 3 (11.9 dpm)) soortgelyk was aan die LMD-bespeurde vlakke. Die IPCC Vlak 2-benadering (10,3 g/dag) het die CH_4 emissies per ram onderskat in vergelyking met die LMD (27,6 g/dag). Dit was nie moontlik om die CH_4 emissiedata wat verkry is met behulp van die LoRa tegnologie en Vlak 2-benadering in hierdie studie te vergelyk nie, aangesien hul emissieskattings verskillende eenhede gehad het (onderskeidelik dpm versus g/dag). Die LoRa CH_4 metingstoestel wat vir hierdie studie ontwikkel is, het die potensiaal om 'n lae-koste en praktiese metingstegniek te wees om CH_4 emissies van skape onder weidingstoestande te kwantifiseer, beperk tot gebruik in klein, beheerde kampe. Sodra die toestel-ontwerp verfyn is om die paar beperkings wat in hierdie studie geïdentifiseer is, te oorkom, kan die LoRa tegnologie help met die generering van skape

CH_4 emissiedata onder verskeie produksietoestande om emissie-databasisse te verbeter en strategieë om metaanproduksie te verlaag te verifieer, op nasionale en internasionale skaal.

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List of Abbreviations

4IR	Fourth Industrial Revolution
µL/L	Microliter per litre
ADC	Analog to Digital Convertor
ANIR	Australian National Inventory Report
CH_2O	Formaldehyde
CH_4	Methane
CO	Carbon monoxide
CO_2	Carbon dioxide
CO_2-eq	Carbon dioxide equivalent
CV	Coefficient of Variation
D_1	Device 1
D_2	Device 2
D_3	Device 3
DE	Digestible Energy
DMD	Dry Matter Digestibility
g/d	grams/day
GC	Gas chromatograph
GDP	Gross Domestic Product
GF	GreenFeed
GHG	Greenhouse gas
GPS	Global Positioning System
GWP	Global warming potential
ha	Hectare
IDE	Integrated Development Environment
IoT	Internet of Things
IPCC	Intergovernmental Panel on Climate Change
IR	Infrared radiation
ISM	Industrial, Scientific, and Medical
IVGPT	*in vitro* gas production technique
LEDS	Low-Emission Development Strategy
LMD	Laser Methane Detector
LoRa	Long Range
LoRaWAN	Long Range wide area network
M	Methane emission (kg/head/day)
MAC	Media access control

MAD	Median Absolute Deviation
ME	Metabolisable Energy
MEF	Methane Emission Factor
NDIR	Non-dispersive infrared
O_3	Ozone
OH^-	Hydroxyl
PI	Potential intake
ppb	parts per billion
ppm	parts per million
ppm-m	parts per million per meter
QGIS	Quantum Geographic Information System
q_m	Metabolisability of a diet (ME/DE)
RC	Respiration chamber
RH	Relative humidity
RSSI	Received signal strength indication
SACRS	South African Coordinate Reference System
SADC	Southern African Development Community
SDG	Sustainable Development Goal
SF_6	Sulphur hexafluoride
SQL	Structured Query Language
TLU	Tropical Livestock Unit
UNFCCC	United Nations Framework Convention on Climate Change
W	Live weight
WAN	Wide Area Network
Y	Methane Production (g/day)

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Chapter 1
General Introduction

1.1 Background and problem statement

The contribution of ruminant livestock to global methane (CH_4) emissions and their resulting influence on climate change has placed these animals at the centre of climate research. Recently, research has been dedicated to identifying factors driving enteric CH_4 production and devising strategies to reduce overall emissions, aiming for more sustainable and environmentally friendly ruminant livestock production practices. Progress is, however, limited because of the lack of an affordable, user-friendly and readily available measurement technique that will enable researchers and producers to measure *in situ* CH_4 emissions in animal production systems consistently. The ability of researchers and producers to measure CH_4 emissions under various production conditions and to develop and implement the most appropriate mitigation strategies may assist in the overall lessening of the carbon and water footprint of livestock production systems.

Climate change has been at the forefront of global environmental discussions for over two decades. Changes in the mean global temperature and weather patterns that persist over extended periods characterise climate change (IPCC, 2018). The third industrial revolution in the 1950s saw drastic increases in atmospheric greenhouse gas (GHG) concentrations due to changing anthropogenic activities. The increase in GHG emissions and their effect on climate change is the consequence of the varying abilities of GHGs to absorb solar infrared (IR) radiation. Such GHGs effectively trap the outgoing terrestrial radiation within the earth's atmosphere, resulting in a rise in global surface temperature (Lelieveld *et al.*, 1993). This phenomenon is known as the greenhouse effect. Numerous gases significantly contribute to the greenhouse effect, with CH_4 considered one of the most significant contributors, second to CO_2.

There are various natural and anthropogenic sources of CH_4 (Karakurt *et al.*, 2012). Atmospheric CH_4 concentrations have tripled since 1750, with most of this increase occurring in the past century (Khalil & Rasmussen, 1994; Peng *et al.*, 2016). Globally, the three primary anthropogenic sources of CH_4 include the agricultural, waste, and energy industries. These anthropogenic sources contribute about 68% of total CH_4 emissions (Xiaoli *et al.*, 2016). Agriculture is a top emitter of GHGs, with atmospheric CH_4 emission increases primarily linked to agricultural growth (Lassey, 2007; Karakurt *et al.*, 2012; Yusuf *et al.*, 2012). The global domestic ruminant population is a leading contributor to the agricultural sector's CH_4 emissions (Lassey, 2007). Methane originates from ruminant animals as a by-product of enteric fermentation. Ruminant enteric CH_4 emissions significantly impact the acceleration of climate change and are estimated to contribute 28-30% of anthropogenic CH_4 emissions (Yusuf *et al.*, 2012; FAO, 2017) and 59.84% of agricultural emissions (Karakurt *et al.*, 2012). Along with its contribution to climate

change, enteric CH_4 production also represents a 2-12% loss of dietary energy, showing sizeable inefficiencies in the ruminant digestion process (Hristov *et al.*, 2013; Henderson *et al.*, 2015).

Sheep significantly contribute to the domestic livestock industry's CH_4 emissions. Zervas & Tsiplakou (2012) state that the average carbon footprint of 1 kg of lamb meat is equal to 19 kg CO_2-eq, with 80% originating from on-farm practises, predominantly as enteric CH_4 emissions. Herrero *et al.* (2008) estimated CH_4 emissions per tropical livestock unit (TLU) for Southern Africa to be 32.7 kg CH_4/year/TLU. They also estimated that, in 2030, sheep in Africa would emit a total of 854.2 million kg of CH_4. Of this, 829.3 million kg would be from enteric fermentation and 24.9 million kg from manure.

There are over 22 million sheep that make up the South African sheep industry (FAOSTAT, 2019). In South Africa, sheep are farmed in arid and semi-arid zones where their aridity and poor soil quality limit the potential for intensive or semi-intensive production. Most of the farming land available in South Africa, about 86.2 million ha, is situated either in arid or semi-arid zones (Cloete & Olivier, 2010). The majority of this land, approximately 71.9 million ha, is suitable only for extensive livestock farming (Abstract of Agricultural Statistics, 2018). Sheep farming provides a source of sustainable production in areas where no alternative farming ventures are feasible. Due to their prominence in South African agriculture, it is essential to consider the contribution of sheep to domestic ruminant livestock CH_4 emissions.

Numerous countries are working together to initiate a global response to climate change, developing various policies and protocols that define goals and actionable steps toward reducing GHG emissions. The two main climate-focusing goals are the Paris Agreement, adopted in 2015 (UNFCCC, 2016), and the United Nations Sustainable Development Goal 13 (FAO, 2018). Both encourage quantification and reduction of GHG emissions from the various responsible sectors. In recognition of climate change and the abovementioned agreements, South Africa submitted the Low-Emission Development Strategy (LEDS) to the United Nations Framework Convention on Climate Change. The LEDS aims to be the first step in South Africa's journey toward a net zero carbon economy by 2050 (RSA, 2020).

GHG inventory development is vital in evaluating the country's progress toward meeting Paris Agreement and Sustainable Development Goal targets. However, due to insufficient data, South Africa faces a challenge in GHG inventory compilation (DFFE, 2021). Accurate enteric CH_4 emission measurement is crucial in compiling reliable national emission inventories, developing mitigation plans and successfully implementing these mitigation strategies by providing accurate and up-to-date data (DEA, 2012). It is also needed to accurately determine the sector's emission contributions and set benchmarks against which mitigation actions are evaluated (DEA, 2012). There are numerous CH_4 measurement techniques available, the most popular being the respiration chamber (RC), the sulphur hexafluoride (SF_6) tracer technique and the GreenFeed (GF) technique (Hristov *et al.*, 2018). Each available technique has advantages and disadvantages that make it suitable for different applications. They are accurate and efficient in evaluating ruminant

CH_4 emission when applied at the experimental level. However, limited by their inability to be used for large-scale, on-farm applications, their use in the applied and participatory research level, often completed on commercial farms, is restricted (Chagunda *et al.*, 2009). LoRa (Long range) is a technology quickly gaining interest for application in agriculture. LoRa is a wide-area network (WAN) technology that can extend large geographical distances and relay data over these long ranges using low-power wireless networks (Charania & Li, 2020). The deployment of the LoRa network is usually in a star-of-star topology, which enables gateways to relay messages between the devices and the central network (Charania & Li, 2020). LoRa has the potential for use as a wireless CH_4 sensor, which researchers and farmers can employ.

Currently, South African ruminant emissions estimates are derived using prediction equations based on emission values obtained in other countries with different production systems, using measurement techniques unsuitable for commercial farming environments. Tongwane and Moeletsi (2020) identify the limited emission factors available to determine South African enteric CH_4 emissions using these prediction equations. There can be some difficulties in using extant models through the limited availability and difficulty in obtaining reliable input variables for the models. The lack of suitable input variables, for example regarding the characteristics of feed, animal physiological factors, or environmental factors, to use in the models can result in considerable inaccuracies in estimated emissions values (Ellis *et al.*, 2007). It can potentially lead to emissions data that is vastly unreliable. Collecting data from the various South African ruminant production systems and accurate emissions quantification will allow for a reliable CH_4 emission inventory to be compiled and assist with determining baseline emission values. Knowledge of these factors will assist policymakers in setting attainable emission reduction goals based on these emission inventories. Tubiello *et al.* (2014) express the importance of obtaining reliable data to quantify emissions and develop national emission inventories so that country specific policies can be drawn up, allowing for the optimal mitigation of emissions.

Compiling reliable national inventories and identifying mitigation strategies suitable for various combinations of production systems, animal species and breed, geographical location, and available resources require an affordable and accurate CH_4 emission measurement technique. Developing a measurement technique that researchers and producers can apply on commercial farms will enable data collection and assist the journey toward a zero or negative net CH_4 production from ruminant production systems. There is a need for a simple and affordable measurement technique that is ideal for commercial farms, can be used by producers to analyse their farm's emissions, and can be used to create national and global GHG emission inventories. LoRa technology combined with an appropriate CH_4 sensor shows promise in fulfilling the earlier specifications as a CH_4 measurement technique that researchers and producers can use. It also provides more opportunities for lower-cost research. This study will determine this technology's effectiveness, accuracy and efficiency in measuring on-farm CH_4 emissions.

1.2 Research aim and objectives

This study aims to establish the potential of LoRa technology to measure CH_4 emissions in sheep under grazing conditions in South Africa.

The objectives of this study are:

1. To determine the potential of a novel LoRa methane detection device to measure methane emissions of sheep under extensive grazing conditions.
2. To compare the LMD and LoRa techniques in terms of the measurement of methane emission in sheep under grazing conditions.
3. To compare the LoRa, LMD and IPCC Tier 2 approach in terms of determining methane emissions in sheep under grazing conditions.

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Chapter 2
Literature review

2.1 Climate change and the contribution of greenhouse gases

The IPCC defines climate change as long-term changes in the mean global temperature and weather patterns (2018). Climate change has become a crucial part of the global agenda, dominating global discussions due to its sudden emergence and rapid effects (FAO, 2019a). The adverse impact of climate change on natural resources, such as changing or extinction of flora and fauna in some areas, changing or termination of livestock production systems, as well as the negative impact on people and the global economy, is a cause for the growing concern (Yusuf *et al.*, 2012).

Since pre-industrial times, the global surface temperature has increased by an average of 1ºC. The Intergovernmental Panel for Climate Change (IPCC, 2022) states that if the increase above the 1ºC increase remains in the lower limits of 0.5 to 1ºC, by 2100, there should not be too many unpredictable and adverse effects, and adaptation will be achievable. However, the current rate at which changes are happening is too fast for adaptation. Recent estimates have found that a further increase of 0.5ºC will already occur by 2030. The IPCC's fifth assessment report (AR5) estimates that the average global temperature could increase by 3 to 4.8ºC by the end of the decade compared to the temperatures recorded at the end of the 20th century (IPCC, 2022). This extreme temperature increase was already documented in 2015, recording the highest average global temperature of 0.75ºC above the average yearly global temperatures between 1961 and 1990 (IPCC, 2013). If temperature increases reach the upper limit of the IPCC (2022) estimates, global food production will decrease, ultimately compromising global food security. Inaction could result in imminent catastrophe, with the effects of climate change already harming many countries across the globe. The World Economic Forum's 2022 Global Risks Report labels "climate action failure" as the most significant long-term threat to the world (McLennan *et al.*, 2022).

2.1.1 The role of GHG, in particular, methane

Greenhouse gases (GHG) contribute significantly to global warming and increased climate variability through their varying abilities to absorb solar infrared (IR) radiation. They absorb a portion of the radiative forcing reflected from the Earth's surface, re-emit it and, by doing so, trap the outgoing terrestrial radiation within the Earth's atmosphere, resulting in a rise in global surface temperature (Lelieveld *et al.*, 1993). Therefore, as the atmospheric concentration of GHGs increases, the overall capacity to capture radiative forcing increases, leading to continued drastic changes in global temperature and, ultimately, more climate variability. This process is commonly

known as the greenhouse effect. Without this effect, the global surface temperature would be below freezing (Kasting, 2005). The greenhouse effect is, therefore, important for maintaining a habitable environment for life.

The rate at which global warming is currently occurring threatens the long-term vitality and habitability of the earth. Greenhouse gas levels remained relatively constant following the natural gas cycles of the environment. However, anthropogenic activities have disrupted this balance by increasing GHG emissions beyond the capacity of natural carbon sinks.

Each GHG has a specific global warming potential (GWP), which indicates its potential radiative forcing. The GWP concept was developed to determine the radiative forcing of each gas relative to that of CO_2. The amount of energy absorbed by a particular GHG over a given time is compared to the amount of energy absorbed by the same mass of CO_2 over the same time (IPCC, 1994). The larger the GWP value, the greater the potential of the specific GHG to capture radiative forcing and accelerate global warming. Allen *et al.* (2018) further developed GWP* that is used to estimate the climate impact of short-lived climate pollutants (SLCPs) such as CH_4. The GWP* considers both the short- and long-term effects of the changing emission rates of these SLCPs (Allen *et al.*, 2018). The GWP of CH_4 is estimated to be 28 over 100 years and 84 over 20 years (IPCC, 2014). However, when these values are corrected using the GWP* the effect of CH_4 is reduced to about 7.8 over 100 years (Costa *et al.*, 2021). Methane is relatively stable and remains in the atmosphere for approximately 9.8 years before it is oxidised in the troposphere by hydroxyl (OH^-) to form formaldehyde (CH_2O), carbon monoxide (CO), and ozone (O_3), which augment its radiative forcing (SPARC, 2013, Hill *et al.*, 2016; Skytt *et al.*, 2020). Methane is the most abundant organic trace gas in the atmosphere, and its reactivity and presence in the atmosphere are essential for various processes in and composition of the troposphere and stratosphere (Badr *et al.*, 1991; Wuebbles and Hayhoe, 2002). However, its presence significantly reduces biosphere integrity (Springmann *et al.*, 2018). The GWP* of CH_4, coupled with its oxidation by hydroxyl (OH-), make it a significant contributor to climate change while playing an important role in chemical feedback (Lelieveld *et al.*, 1993; Hill *et al.*, 2016).

Methane is a complicated gas that cannot be synthesised in the atmosphere due to its high internal energy (Badr *et al.*, 1991). Its only entry into the atmosphere is through various natural and anthropogenic sources on the earth's surface. Methane currently contributes 17% to radiative forcing (Bodelier & Steenbergh, 2014) and 16% of total anthropogenic GHG emissions (Center for Climate and Energy Solutions, 2020).

Atmospheric CH_4 concentrations remained relatively stable for thousands of years before the 19th century, after which the concentration started steadily increasing (Yusuf *et al.*, 2012). In the 1750s atmospheric CH_4 concentrations were an estimated 676-716 ppb and have increased to around 1,880 to 1,960 ppb in about 2018, depending on latitude (Yusuf *et al.*, 2012; Glikson, 2018; Our World in Data, 2018a). Atmospheric CH_4 concentrations increased significantly over the past 70 years, and levels are still rising at radical rates. Anthropogenic activities such as burning fossil

fuels, more intensive agricultural practices, and waste disposal are responsible for this increase (Wuebbles and Hayhoe, 2002; Pérez-Barbería, 2017). Khalil and Rasmussen (1994) calculated estimates of combined natural and anthropogenic global CH_4 emissions over the past 500 years to identify how emissions have changed. They found that in the 15th century, emissions were an estimated 180 Tg/year, moving to 200 Tg/year in the early parts of the 18th century and are now estimated to be about 450 Tg/year. This increase in global CH_4 emissions mostly happened in the past 60 years following the onset of the third industrial revolution.

The primary anthropogenic CH_4 sources are divided into three sectors: agriculture, waste, and energy. The agriculture sector is estimated to have the largest total GHG emissions, with the increase in atmospheric CH_4 concentration primarily linked to agricultural growth (Lassey, 2007; Karakurt *et al.*, 2012; Yusuf *et al.*, 2012). The increase in agricultural CH_4 emissions mirrored the trend of global increases in CH_4 emissions (Khalil and Rasmussen, 1994). Domestic ruminant production is mainly responsible for the agricultural sector's contribution to CH_4 emissions and the increase in CH_4 emissions over the past few decades (Lassey, 2007).

2.2 The importance of studying climate change

Babel *et al.* (2020) found a complex link between water and climate change, stating that water is the primary channel through which climate change will be felt. Climate change has caused rising sea levels and more frequent and severe droughts and floods, and it is expected to increase the severity of these phenomena further. As the global surface temperature rises, sea water expands through thermal expansion resulting in rising sea levels. Warming of the oceans and atmosphere further results in melting polar ice caps in the Arctic and Antarctic regions (which also store massive amounts of methane), causing an even greater rise in sea levels (Moss *et al.*, 2000). Like polar ice caps, many other land and ocean reservoirs contain approximately 15,400 GtC CH_4, which become more susceptible to release as global temperatures rise (Glikson, 2018). Glikson (2018) states that this release of CH_4 would have catastrophic effects on the biosphere (drastically decreasing biosphere integrity) and cause large ice sheets to melt, resulting in a rise in sea levels. There has already been evidence of rises in temperatures causing the release of billions of tons of CH_4 into the atmosphere from sources such as permafrost, lakes, shallow seas and sediments. Between 2015 and 2018, the Artic temperature rose an average of 3 to 8ºC, causing the release of CH_4 (Glikson, 2018). The effects of climate change are expected to be seen through global water shortages, coastal flooding and decreased global food security.

Climate change affects the environment and plays a considerable role in moulding modern society, a concept known as ‘environmental determinism’ (Chen *et al.*, 2020). As climate change alters the environment, society will have to develop ways to reduce its contribution to GHG emissions while learning to adapt to the manifestations of climate change. The increase in frequency and intensity of natural disasters will have a crippling effect on societies and economies.

The 2019 FAO report, discussing their current work on climate change, expresses a few challenges for the future concerning agriculture and climate change. They state that if climate change is allowed to persist and aggravate, it could push 122 million people into extreme poverty by 2030, most of whom are farmers (FAO, 2019a). They state that water scarcity is becoming an increasingly prominent problem. With every 1ºC increase in average global temperature, an extra 500 million people will experience a 20% dip in renewable water sources.

Many countries in Sub-Saharan Africa, including South Africa, already experience frequent and severe droughts, along with a high average annual temperature. Climate events such as higher average temperatures and extended periods of high temperatures, coupled with low, varied and unpredictable precipitation, are predicted to become more commonplace in these countries (Elum *et al.*, 2017). South Africa is already a dry country with an average annual rainfall of less than 500 mm (Elum *et al.*, 2017). Climate change will ultimately exacerbate the effects of the already high temperatures and regular droughts typical for South Africa and surrounding countries, affecting the farming potential of the region.

Lucas (2021) points out how the IPCC has used linear projections for a nonlinear climate system, and policymakers have been using these projections to develop policies for achieving carbon neutrality. He states that the nonlinearity of the climate system makes it much more sensitive to global warming than previously predicted, meaning the predicted effects of climate change are likely underestimated. This emphasises the importance of and the need to take drastic and immediate action toward reducing emissions. Reducing emissions can, however, only be accomplished once global and national inventories are accurately quantified.

2.2.1 Global climate change mitigation policies and strategies

Various policies and protocols, such as the Kyoto Protocol, Paris Agreement and 2030 Agenda of Sustainable Development, have been developed to initiate a global response to climate change. However, despite the overwhelming global focus on climate change, GHG emissions still show no sign of decreasing.

The Kyoto Protocol was signed in the 1990s, adopted in 1997, and entered into force in 2005. The Kyoto Protocol aims to set binding targets for Annex 1 countries to reduce GHG emissions. The majority of developed countries have not accomplished the stated goals. Emissions have increased drastically since the Protocol entered into force, with 73% of the growth arising from developing and transition countries (van Beek *et al.*, 2010).

The Paris Agreement was adopted in 2015 under the United Nations Framework Convention on Climate Change (UNFCCC). The Paris Agreement policy aims to prevent the increase of the average global temperatures to more than 1.5ºC above pre-industrial global temperatures (UNFCCC, 2016). However, Olhoff (2018) states that exceeding the 1.5ºC mark of the Paris Agreement by 2030 is unavoidable if the current action is not improved and made more ambitious.

Along with maintaining global temperature, the Paris Agreement also aims to increase the ability of nations to adapt to adverse climate impacts resulting from climate change, as well as encourage climate resilience.

In 2015 the United Nations set out the 17 Sustainable Development Goals (SDGs) with 169 specific targets and 232 indicators (FAO, 2019b). This review focuses on SDG 13, which is 'to take urgent action to combat climate change and impacts' (FAO, 2018). This goal aims to improve the adaptive capacity and resilience of nations to natural disasters and the changes caused by climate change. It also aims to encourage countries to develop national policies and strategies to decrease GHG emissions in all sectors while ensuring countries are simultaneously adapting to the adverse impacts of climate change (FAO, 2018). Another aim of SDG 13 is to raise awareness about climate change, allowing all people to understand the problem and do what they can to accomplish this goal (FAO, 2018). A leading factor that will play a role in the ability of nations to realise the SDGs is to ensure there are effective ways of measuring the necessary parameters by collecting data, monitoring targets and measuring the progress (FAO, 2017). Therefore, accurate and efficient measurement techniques are needed.

In recognition of the above goals, South Africa submitted the Low-Emission Development Strategy (LEDS) to the UNFCCC as the first step in the country's journey toward a net zero carbon economy by 2050 (RSA, 2020). The success of this strategy depends on collecting accurate emissions data against which emissions targets can be set and measured.

2.2.2 The demand for livestock products

The global demand for livestock products is increasing in a phenomenon known as the 'livestock revolution', resulting from drastic population growth, socio-economic development, increased affluence predominantly in developing countries, and rising urbanisation (Delgado *et al.*, 1999; Herrero *et al.*, 2008; Thornton, 2010; Rojas-Downing *et al.*, 2017; Lan & Yang, 2019). These factors are drivers of an estimated doubling the demand for livestock products in low- and middle-income countries by 2050 (Rojas-Downing *et al.*, 2017; FAO, 2018; FAO, 2019a). In South Africa, the population increase has been accompanied by an increase in the country's middle-class population, proof of increased affluence in the country paired with changing preference toward incorporating more expensive livestock products into the diet (Meissner *et al.*, 2013). As the worldwide standard of living improves, the demand for livestock products increases. Global meat production has quadrupled in the past 50 years, producing 320 million tonnes yearly (Ritchie & Roser, 2017). The demand for sheep products, primarily meat and wool, is expected to increase due to these factors. The demand for lamb and mutton is expected to increase at a rate of 1.5% between 2006 and 2050 (Opio *et al.*, 2013). Increases in animal numbers to meet growing demands in the past were simple, and technology and science contributed to improving production efficiency. However, future production increases will have to consider the environmental impact of

the industry, specifically the industry's carbon footprint, and is likely to be significantly limited by this factor (Thornton, 2010). Developing and transition countries will be responsible for most of the increase in livestock production to meet growing demands, as they still have low production levels and the most capacity for increase (van Beek *et al.*, 2010).

2.3 The contribution of livestock production to methane emissions

Considerable attention lies in the agricultural sectors' contribution to anthropogenic CH_4 emissions, particularly focusing on ruminant enteric fermentation. Recent research has focused on this topic, intending to quantify the industry's contribution, specifically of enteric fermentation, to global CH_4 emissions. Various studies have estimated the contribution of enteric fermentation to global emissions, but these values have little uniformity.

Enteric fermentation contributes approximately 24-28% of global anthropogenic CH_4 emissions and 39% of global agricultural CH_4 emissions (Yusuf *et al.*, 2012; Glasson *et al.*, 2022). Opio *et al.* (2013) state that the global ruminant supply chain is responsible for producing 80% of the GHG emissions, primarily CH_4, amounting to 5.8 $GtCO_2$-equivalent per annum. They further state that enteric fermentation contributes 47% of the livestock sector's total GHG emissions and over 90% of its CH_4 emissions (Opio *et al.*, 2013). Africa produced approximately 7.8 million tonnes of CH_4 from the livestock industry in 2000, and emissions are predicted to increase to 11.1 million tonnes/year by 2030 (Herrero *et al.*, 2008). This 42% emissions increase is caused predominantly by increases in livestock numbers. The IPCC AR5 states that emissions have increased the most in Africa, with a 2.4 %/yr average increase (IPCC, 2014).

Small ruminants, goats and sheep contribute 6.5% of the livestock sector's global emissions, equating to about 475 million tonnes CO_2-eq (Gerber *et al.*, 2013; Opio *et al.*, 2013). Marino *et al.* (2016) state that small ruminants make up 56% of the global domestic ruminant population. Small ruminants are, therefore, necessary for developing CH_4 measurement techniques and mitigation strategies. Gerber *et al.* (2013) state that the sheep population in Africa and Sub-Saharan Africa contribute significantly to CH_4 emissions due to high sheep population numbers in these areas. Numerous studies have investigated the contribution of individual sheep and different sheep production systems to global CH_4 emissions. Browne *et al.* (2011) found that sheep in Australia contribute 2.8-4.3 t CO_2-eq/ha. Wool sheep were responsible for 18.1-18.7 t CO_2-eq/t clean fleece and prime lamb for 1.4-12.0 t CO_2-eq/t carcass weight (Browne *et al.*, 2011). Wiedemann *et al.* (2016) completed a multiple impact life cycle assessment (LCA) on three wool types in three geographically different regions of Australia. They found that the GHG emissions for these sheep were between 20.1 ± 3.1 and 21.3 ± 3.4 kg CO_2-eq/kg of wool. These values formed a basis upon which mitigation strategies could be developed. This focus has allowed Australia to reach their goals of producing a more sustainable industry and working toward carbon neutrality (Mayberry *et al.*, 2019). Ripoll-Bosch *et al.* (2013) examined the differences in GHG emissions from sheep in

three production systems in Spain: pasture-based, mixed sheep-cereal and industrial (zero-grazing) systems. The calculated GHG emissions for these three systems varied from 19.5 to 25.9 kg CO_2-eq/kg of lamb live-weight or 39.0 to 51.7 kg CO_2-eq/kg of lamb meat. Zervas & Tsiplakou (2012) state that the average carbon footprint of 1 kg of lamb meat is equal to 19 kg CO_2-eq, with 80% originating from on-farm practices. Herrero *et al.* (2008) estimated CH_4 emissions per tropical livestock unit (TLU) for Southern Africa to be 32.7 kg CH_4/year/TLU. They also estimated that, in 2030, sheep in Africa would emit a total of 854.2 million kg of CH_4. Of this, 829.3 million kg would be from enteric fermentation and 24.9 million kg from manure.

2.3.1 Enteric methane production

Methane originates in sheep from the rumen as a non-utilisable by-product of enteric fermentation and from manure. Of the CH_4 produced by sheep, 97% is eructated or respired, and only 3% is from the manure (Herrero *et al.*, 2008). Murray *et al.* (1976) found that approximately 87% of enteric CH_4 originates from the rumen and the remaining 13% from hindgut fermentation. The ability of ruminants to digest fibre gives them an important role in the food chain. However, this ability is paired with CH_4 production. As CH_4 is a GHG and an indicator of the ruminant's loss of ingested gross energy, it presents an ecological and economic problem (Kahraman *et al.*, 2015).
Ruminant animals have four stomach compartments, namely the non-glandular reticulum, rumen and omasum of the forestomach, and glandular abomasum. Over time the rumen has evolved to create an anaerobic environment, ideal for the growth and development of a microbial community. Ruminants have developed a symbiotic environment with their gut microbiome. These microbes assist the animal with the breakdown of consumed food by synthesising microbial enzymes that can digest cellulose and hemicellulose (Knapp *et al.*, 2014). They enable ruminants to ferment and use cellulose and hemicellulose (complex carbohydrates), making various nutrients available to the animal as their primary energy source. The main focus is on *methanogens* responsible for CH_4 production. Methane is produced and expelled to maintain a balance within the rumen, ensuring normal chemical processes can continue. The emission of CH_4 follows a clear diurnal pattern related to intake patterns (Hammond *et al.*, 2016).

2.3.2 Potential mitigation strategies

A part of improving the sustainability of ruminant livestock production systems would be implementing practices to mitigate CH_4 emissions from the animals. Knowledge and understanding of enteric CH_4 production and CH_4 emission values are needed to accomplish this.

Enteric CH_4 production is impacted by the amount of feed consumed, the quality and type of feed, the composition of the feed (namely concentrate to roughage ratio), feed additives, the rumen microbial community, and the digestibility of the feed (Herrero *et al.*, 2008; Grossi *et al.*, 2019; van Gastelen *et al.*, 2019). High-roughage diets increase acetate and butyrate production, while high-

concentrate diets increase propionate production. Hydrogen is a by-product of acetate and butyrate production and is used for propionate production. Therefore, high-roughage diets result in higher CH_4 production than high-concentrate diets. The relative digestibility of a diet can be improved by increasing the amount of concentrate in the diet, reducing emissions by about 35 to 40% (Gerber *et al.*, 2013; Knapp *et al.*, 2014). However, concentrate levels in the diet must be monitored to avoid levels becoming too high, causing metabolic diseases such as rumen acidosis (Grossi *et al.*, 2019). Ruminant CH_4 production is also affected by diurnal fluctuations in feed intake, feed passage rate and mean retention time, and animal genetics (van Gastelen *et al.*, 2019). Region, production system and product type also affect the quantity of CH_4 emitted from livestock systems (Herrero *et al.*, 2013). Many of these factors can be used to increase ruminant productivity while reducing each ruminant's overall methane production.

As the global concern about climate change and global warming grows, pressure from consumers and governments to reduce the negative environmental impact of the livestock production industry becomes more pronounced. Many governments, including the South African government, are developing policies and mitigation strategies to reduce national CH_4 emissions. Decisions for livestock farms previously focused on increasing production efficiency, decreasing costs and maximising profits. The focus has now shifted to include environmental considerations by using fewer natural resources, improving animal welfare, decreasing the carbon footprint of production, and handling food safety issues (Zervas & Tsiplakou, 2012).

Response to climate change occurs through mitigation and adaptation strategies. Adaptation strategies are more focused on dealing with change that has already occurred, while mitigation strategies involve applying changes to reduce GHG emissions (Elum *et al.*, 2017). Mitigation strategies can be implemented to decrease CH_4 emissions to a constant rate. Constant emissions will result in the impact of CH_4 on global warming reaching a steady state and eventually will no longer contribute to an increase in global warming (Skytt *et al.*, 2020). Adaptation and mitigation policies need to be developed by local governments to assist farmers in adapting to changing climate and farming more efficiently to reduce the environmental impacts of farming.

Two enteric CH_4 mitigation strategies are available. The first is indirectly reducing overall emissions by improving production efficiency and subsequently increasing animal productivity, and the second is directly reducing the amount of CH_4 produced and emitted (Grossi *et al.*, 2019). Decreasing the amount of CH_4 produced by enteric fermentation will improve feed efficiency, redirecting intake energy, usually lost as CH_4, to other bodily functions (Patra, 2014). Enteric CH_4 production can be reduced by directly altering feed, animal genetics or the rumen microbiome (Kahraman *et al.*, 2015). It can also be indirectly reduced through better land management, technology application, and animal husbandry and management, resulting in increased production efficiency and decreased animal numbers (Mayberry *et al.*, 2019).

2.4 Global population growth and the link to methane production

Along with facing the challenge of global warming and adapting to its effects, the world is also under pressure to increase and improve global food production. The continuously growing population has resulted in a drastic increase in demand for food. Producers and nations are challenged by the reduced food security of millions of people, changes in dietary preferences of the growing population and decreased availability of natural resources (FAO, 2019b). As the ruminant livestock sector grows and the effects of climate change worsen, the sector is challenged with the need to increase production to meet growing global demands and increase average food security. While simultaneously reducing GHG emissions intensity per unit of product and the net environmental impact of the sector to achieve national emission targets implemented by numerous new climate change mitigation policies (Gerber *et al.*, 2013; Grossi *et al.*, 2019).

2.4.1 Global population growth

The global population has experienced exponential growth over the past few decades and is predicted to continue increasing. Between 2005 and 2017, a growth of about 1 billion people was recorded, resulting in a population total of 7.6 billion people. The expected average annual global population growth is 83 million people, resulting in an estimated population size of 8.6 billion in 2030 and 9.8 billion by 2050 (Grossi *et al.*, 2019; United Nations, 2019). If this growth materialises, the population size will have increased by 33% by 2050, with more than 1 billion of this increase expected to occur in Africa (Thornton *et al.*, 2009). The estimated population growth rate of Sub-Saharan Africa is at 1.6% for the remainder of the century (Thornton & Herrero, 2014). The African continent is experiencing rapid population growth, with a substantial portion of this growing population being undernourished. As population growth continues, the need for increased food supply to ensure global food security rises.

2.4.2 The link to methane production

In response to the increased demand for livestock products, livestock numbers are rising, especially in developing countries, with evident negative impacts on the environment through increased GHG emissions and land degradation (Prakash & Stigler, 2012). The livestock industry is one of the fastest growing subsectors in developing countries, contributing 33% to the agricultural GDP and rapidly increasing its share in the agricultural GDP (Thornton, 2010). The global sheep population increased from 780 million in 1950 to 1.266 billion in 2021 (Ritchie & Roser, 2017; IWTO, 2022). Delgado *et al.* (1999) state that there will be an increase in the number of ruminant animals in Africa to satiate the growing population's demand for meat and milk. Ruminant livestock production systems are experiencing swift changes in structure and function to meet these growing demands (Herrero *et al.*, 2008; Kumari *et al.*, 2020). Without careful

consideration of the environmental impact, these changes could potentially lead to drastic increases in the already over-flowing atmospheric CH_4 concentrations. Global enteric CH_4 emissions are estimated to increase by 70%, by 2055, compared to 1995 (Popp *et al.*, 2010). van Beek *et al.* (2010) suggest that increases in global livestock production and populations are possible without increasing CH_4 emissions. However, for this to be true, many changes must be made, mitigation strategies applied, and technological advancements developed and utilised.

2.5 The South African sheep industry

The South African sheep industry has an essential role in the country. It is a source of livelihood for many people, has an important economic contribution, is a popular food source, contributes significantly to the textile industry, makes use of land unsuitable for other types of farming and has potential use for bioenergy. South Africa has over 22 million sheep (FAOSTAT, 2019). Extensive production systems dominate the industry, and many of these sheep are owned by subsistence or emerging small-scale farmers.

2.5.1 The proportion of land used for sheep farming

Most of the farming land available in South Africa, about 86.2 million ha, is situated either in arid or semi-arid zones (Cloete & Olivier, 2010). The potential of these areas for intensive and semi-intensive plant production is limited due to the aridity of these areas and poor soil quality, resulting in only 16.5% of this land having cropping potential (Abstract of Agricultural Statistics, 2018). Therefore, the majority of this land, 71.9 million ha, is suitable only for extensive livestock production (Abstract of Agricultural Statistics, 2018). Sheep farming is a source of sustainable production in areas where no alternative farming ventures are viable. Meissner *et al.* (2013) state that of the total agricultural land in South Africa, 70% is suitable only for utilisation by livestock and game, making up about 80% of the land resources.

2.5.2 The importance of the South African sheep industry

The South African livestock industry provides immense opportunities for permanent employment. Considering the amount of money invested in the industry, it has the second largest employment multiplier after construction (Cloete & Olivier, 2010). However, as the South African livestock industry undergoes rapid changes in response to climate change and to increase production while ensuring sustainability, the resource-poor people who rely on livestock production for their livelihoods could be adversely affected (Thornton *et al.*, 2009). Many rural towns in South Africa rely on sheep for their livelihoods. Meissner *et al.* (2013) state that the livestock sector employs 245 000 people, and 1.45 million depend on the industry as a source of income. This excludes people dependent on communal and emerging farms, where the industry plays an important socio-

economic role, contributing to the sustenance and livelihoods of many in rural communities. The wages of these farm employees amount to R 6 100 million (Meissner *et al.*, 2013).

The agriculture sector contributes approximately 4-27% of the Southern African Development Community (SADC) GDP (Elum *et al.*, 2017) and 2.6% to the total South African GDP (DAFF, 2013). South Africa is one of the largest meat producers in Africa, producing between 2.5 to 5 million tonnes of meat in 2018 (Our world in data, 2018b). The lamb, mutton and wool industries play a vital role in the South African economy. In 2006/2007, the South African mutton industry was worth about R 2.8 billion, which increased significantly to about R 6 billion by 2015/2016 (DAFF, 2017). Despite being a net importer of mutton, South Africa is an important exporter to SADC countries (DAFF, 2018). The gross value of the mutton industry between 2007 and 2017 amounted to R 4.57 billion per annum (DAFF, 2018). The sector contributes 8.2% of the total GDP of animal products in SA (Schoeman *et al.*, 2010). In the past, environmental factors such as the drought in 2015/2016 have caused a decrease in mutton prices, indicating the potential future effects of climate change on the industry. Along with the lamb and mutton industry, the wool industry also has a significant role in the South African economy. Most of the wool produced in South Africa originates from areas with harsher climates and low rainfall, such as the Karoo (DAFF, 2016). Wool is an important commodity for trade in South Africa, with more than 90% of the total wool produced being an export product, either as greasy wool or in a semi-processed form such as lanolin (DAFF, 2016).

As sheep are ruminants, they can utilise fibrous plant material that humans cannot digest and produce products such as meat, which is useful in the human diet. The lamb and mutton industry is an important protein source in many South Africans' diets. In February 2020, Knorr completed a study to determine the average South African eating habits. They found that South Africans have a keen meat-eating culture, eating it an average of 4 times a week (Knorr, 2020). The study found that 84% of South Africans are meat eaters and that red meat is highly favoured (Knorr, 2020). South Africa has the highest annual per-person meat consumption in Africa, recording an average of 60.02 kg per person in 2017 (Ritchie & Roser, 2017).

Sheep farming is dominant in many parts of the country, as sheep can be farmed in more unfavourable conditions where crops and cattle cannot. Sheep farming complements cropping, utilising the by-products of this farming or serving as the primary source of livelihood when droughts arise, and crops fail (Cloete & Olivier, 2010). The 80% of land dominated by arid climate is suitable for extensive sheep production, providing little opportunity for intensive small stock farming, agricultural crop production, and beef and dairy production (Schoeman *et al.*, 2010).

2.6 Methane measurement techniques

Measuring CH_4 emissions from sheep and other ruminant animals can assist with the development of an emissions profile for animals in certain areas and production systems, which will provide a

baseline against which emission targets can be set and their progress measured (Jones *et al.*, 2014). Hammond *et al.* (2016) describe a list of criteria for appropriate and acceptable CH_4 measurement techniques, pointing out non-invasive and non-intrusive characteristics as most important for measurements from animals in their 'normal' environment. The method should also be able to be applied under conditions relevant to commercial production and be rapid, cost-effective and, ideally, automated.

Ruminant CH_4 emissions can be determined using various measurement techniques developed over the last 100 years. These methods are either direct, indirect, or short-term measurement techniques. Currently, the most commonly applied CH_4 emission measurement techniques are the respiration chamber (RC), Sulphur Hexafluoride (SF_6) tracer technique and GreenFeed (GF) system (Hristov *et al.*, 2018). The decision as to which measurement technique to use is made according to the study's objectives and the available resources (Hammond *et al.*, 2016). Although the majority of these techniques are widely used, there is still potential for them to estimate values with low accuracy and precision, or produce misleading results, owing to improper implementation of these techniques. Each technique is subject to experimental variation and random errors and has its benefits and limitations, which should be reviewed to meet the experiment's needs best.

2.6.1 Direct measurement techniques

Direct measurement techniques measure emissions directly from an individual animal or a group of animals.

2.6.1.1 The Respiration Chamber

The whole animal open-circuit indirect-respiration chamber (RC) is currently the most common measurement technique. When operated correctly, the RC is considered the 'gold' standard for enteric CH_4 measurement from individual ruminant animals as its results are proven to be accurate with a low coefficient of variation (Blaxter & Clapperton, 1965; Grainger *et al.*, 2007; Hristov *et al.*, 2018). Since the RC is considered the most reliable CH_4 measurement technique, it is one of the primary data sources on CH_4 emissions from livestock (Gardiner *et al.*, 2015). Moreover, models that estimate national and global CH_4 emissions are primarily based on RC measurements (Johnson & Johnson, 1995).

The RC technique has been around for over 120 years as an indirect calorimeter for measuring ruminant respiratory exchange and CH_4 energy losses (Hammond *et al.*, 2016). This technique was initially used for ruminant energy metabolism studies but has since been adopted for studies measuring ruminant CH_4 emissions, following growing concerns regarding global CH_4 emissions. There has been an exchange and accumulation of knowledge concerning the construction specifications and use of the RC for ruminant energy metabolism research. In the past

two decades, the focus has shifted toward CH_4 emission research, and a publication of the 'Technical Manual on Respiration Chamber Designs' (Global Research Alliance, 2018) provides examples of the use of the RC for ruminant CH_4 measurement around the world (Hammond *et al.*, 2016).

The RC is typically used to obtain CH_4 emission measurements over 24h periods for 1-7 sequential days to account for between-day variation (Hammond *et al.*, 2016). The animal is housed inside the chamber for the duration of the measurement period. Respiration chamber estimates of enteric CH_4 emissions are highly accurate, and their design enables the estimation of emissions from all enclosed orifices. The basic principle of the RC relies on air (controlled airflow) passed into the chamber through an inlet, circulated around the animals' head, nose, and mouth, mixing with the animals' eructated and expired air, and extracted through an outlet. The total airflow through the chamber and the difference in CH_4 concentration measured between the inlet and outlet air indicate the animal's CH_4 emission (Johnson & Johnson, 1995; Storm *et al.*, 2012). Circulated air is pumped through a flow meter and passed through a gas sensor, either a gas chromatograph (GC) or infrared analyser, directly connected to the chamber (Gupta *et al.*, 2018). For this system to provide accurate estimates, it needs to be properly sealed, with slight negative pressure, to ensure leaks are inwards and that there is no net loss of CH_4 (Johnson & Johnson, 1995).

The RC is a favoured CH_4 emission measurement technique, providing precise, accurate and reliable results with low within- and between-animal variation (Blaxter & Clapperton, 1965; Grainger *et al.*, 2007). It provides valuable estimates of individual animals' daily CH_4 emissions from ruminal and hindgut fermentation and information on diurnal emission patterns (Johnson & Johnson, 1995; Storm *et al.*, 2012; Gupta *et al.*, 2018). These factors, paired with the continuous measurement period and the ability to control the RC environment, make the chamber suitable for analysing various treatment effects on CH_4 emissions and testing mitigation strategies (Gardiner *et al.*, 2015; Goopy *et al.*, 2016). However, the RC is limited by high investment, running and maintenance costs, an inability to test many animals, and animal confinement. The high costs limit the number of chambers available to a particular research facility and, in some cases, prevent some facilities from having even one. Furthermore, the RC requires a trained technician; it has high labour requirements and is a time-consuming technique (Johnson & Johnson, 1995; Hammond *et al.*, 2016). It also requires trained animals (Johnson & Johnson, 1995). These factors limit the number of animals that can be experimentally examined at one time, making it less practical for applied research, which evaluates large groups of animals under commercial production (Storm *et al.*, 2012; Hristov *et al.*, 2015; Hammond *et al.*, 2016). Animal confinement limits animal movement, preventing it from exhibiting natural behaviour, inhibiting normal social interaction with peers and the environment, restricting diet selection, and ultimately lowering energy expenditure compared to loose housing or grazing environments (Pinares-Patiño *et al.*, 2011; Hammond *et al.*, 2016). There is concern that this could result in CH_4 emission values that

do not accurately represent those that would arise from free-ranging animals allowed to express their natural behaviour and be able to select their diet. Therefore, the values obtained using the RC cannot be applied to free-ranging animals on pasture, who have an overall higher intake and diet selection compared to animals housed in RC (Grainger *et al.*, 2007; Storm *et al.*, 2012; Li *et al.*, 2014). The limited capacity of the respiration chambers and the need for trained animals make this technique unsuitable for screening animals to assess the heredity of CH_4 production (Storm *et al.*, 2012).

2.6.1.2 The Sulphur Hexafluoride tracer technique

The sulphur hexafluoride (SF_6) tracer technique is an estimation technique, which relies on a measured and constant release rate of a tracer gas (SF_6) from a permeation tube within the reticulo-rumen to determine daily CH_4 production. This technique is primarily used for free-ranging grazing animals. It was developed in 1993 by Zimmerman (1993) and first used experimentally by Johnson *et al.* (1994) to determine CH_4 emissions from cattle. It has since been applied globally, becoming a favourable method for assessing enteric CH_4 emissions from various ruminant animals under grazing conditions.

Sulphur hexafluoride is an ideal tracer gas. It is chemically and biologically inert, undergoing no interactions with substances in the reticulo-rumen. It is a gas at standard temperature and pressure, non-toxic, cheap, and stable, mixing with the rumen air similar to CH_4 and co-released with CH_4 from the rumen (Johnson *et al.*, 1994; Berndt *et al.*, 2014; Gupta *et al.*, 2018). The eructation of the SF_6 and CH_4 into the breath is, therefore, correlated, resulting in CH_4 emission estimation based on a known release rate of the tracer gas.

The basic principle of the SF_6 tracer technique relies on inserting a SF_6 charged permeation tube, with a pre-determined SF_6 release rate, into the rumen of each animal (Johnson *et al.*, 1994). The permeation tube releases SF_6 at a rate determined through gravimetric calibration before insertion of the tube into the rumen. A sampling line is placed above the animal's nose and is connected to an evacuated collection canister carried on the animal's back. Each animal has a halter that holds the sampling line near its nasal cavity for the sampling period. Time-integrated breath samples are collected over 24 hours (a complete feeding cycle) as the animal respires or eructates air. The eructated or respired air is passively collected via the sampling line into the collection canister, and the sample is analysed using GC to determine CH_4 and SF_6 concentrations and mixing ratios (Johnson *et al.*, 1994; Berndt *et al.*, 2014; Deighton *et al.*, 2014). Background air samples are collected simultaneously for the measurement period's duration using the same sampling line and canister system. This background air is considered the CH_4 and SF_6 concentration in the breath sample with a source other than the animal (Berndt *et al.*, 2014). Each animal's daily CH_4 release rate is determined by equating the ratio of CH_4 to SF_6 release rate to the concentration ratio of these two gases in the breath sample, corrected for measured CH_4 and SF_6

background concentrations and the duration of sample collection (Johnson *et al.*, 1994; Berndt *et al.*, 2014).

Various studies evaluated the accuracy and precision of the SF_6 tracer technique, comparing it to the RC (Johnson *et al.*, 1994; Grainger *et al.*, 2007; Pinares-Patiño *et al.*, 2008; Pinares-Patiño *et al.*, 2011; Muñoz *et al.*, 2012). These studies found the comparisons to be generally favourable. However, CH_4 emission estimates using the SF_6 tracer technique were subject to greater among- and within-animal variability than the chamber technique (Pinares-Patiño *et al.*, 2011). Johnson *et al.* (1994), Grainger *et al.* (2007), and Pinares-Patiño *et al.* (2008) found that CH_4 emission estimates determined using the SF_6 tracer technique were lower than those measured by the chamber. In contrast, Pinares-Patiño *et al.* (2011) and Muñoz *et al.* (2012) found that the SF_6 tracer technique overestimated the mean CH_4 emission compared to the chamber technique. However, comparing the RC and SF_6 tracer techniques is complex and could result in significant inaccuracies due to the drastic differences in the conditions under which the techniques are applied.

There are three main advantages of this technique. Namely, it can be used on freely-grazing ruminants, applied to a large group of animals, and does not restrict the animal's natural behaviour (Swainson *et al.*, 2011; Hill *et al.*, 2016; Gupta *et al.*, 2018). This technique can be performed under natural, free-ranging or confined, more controlled conditions where factors such as feed intake can be regulated or measured (Goopy *et al.*, 2016). This method is suitable for comparing and evaluating CH_4 mitigation strategies, different treatment effects and the genetic screening of animals, ranking them according to daily CH_4 emissions. Compared to the RC, the equipment cost per animal is much lower, enabling larger groups of animals to be sampled at one time (Berndt *et al.*, 2014), as well as making the method more accessible for use where funds are not as freely available. However, the SF_6 tracer technique is less precise than chamber techniques, the equipment is physically less robust, with equipment failure being common for this method, and it is labour intensive (Goopy *et al.*, 2016; Gupta *et al.*, 2018). Like the chamber technique, the SF_6 technique requires animal training. Before the SF_6 tracer technique can be used, animals are first selected based on temperament and handling – as this technique requires frequent animal handling, and the animal requires training to wear the halter and collection canister (Gupta *et al.*, 2018). Despite its limitations, the SF_6 tracer technique is still a favoured measurement technique because it is suitable for estimating emissions from individual grazing animals, allowing the animals to express natural behaviour while being able to sample a large number of animals simultaneously.

2.6.2 Indirect measurement techniques

2.6.2.1 *In vitro* measurement of methane

In vitro fermentation studies have been used in research for many years. This technique was initially developed to evaluate forages and the nutritive value of feeds by determining the degradation of different types of feedstuffs (Cone *et al.*, 1996). As the interest in more efficient use of feedstuffs has grown, this technique has been used more widely in studying fermentation kinetics, with gas measurements providing valuable data on the digestion of both soluble and insoluble fractions (Getachew *et al.*, 1998). More recently, this method has been used to determine the effect of various feeds, additives, and diet compositions on the production of CH_4. The basic principle of the *in vitro* gas production technique (IVGPT) is fermenting ruminant feeds under controlled laboratory conditions using natural rumen microbes (Storm *et al.*, 2012). This method provides a starting point for determining the potential of dietary CH_4 mitigation strategies. As *in vivo* gas measurement techniques are costly, laborious, time-consuming, require large quantities of feed, and unsuitable for large-scale feed evaluation, the IVGPT has become a favourable method to determine the potential of substrates and additives for reducing overall CH_4 production (Getachew *et al.*, 1998).

McBee (1953) explained the initial *in vitro* batch fermentation. Tilley and Terry (1963) described an *in vitro* method of forage evaluation used to determine the organic matter digestibility of feedstuffs with microorganisms. This technique became an extremely important tool for evaluating ruminant feeds and was the standard on which future methods were based. This method became a popular technique used in laboratories for forage evaluation due to its convenience when large-scale testing was required (Getachew *et al.*, 1998). It was, however, limited in that, as an end-point digestibility method, it could not provide any information on the kinetics of forage digestion without applying lengthier and more labour-intensive studies (Tilley & Terry, 1963; Theodorou *et al.*, 1994). Goering and Van Soest (1970) further developed this technique, which is now the basis of modern IVGPT used for *in vitro* ruminant CH_4 emission studies. These methods were not yet developed to a point where they could be used to determine gas production, specifically CH_4 production resulting from the fermentation of various feedstuffs. They were time-consuming and labour-intensive, and residue determination destroyed the sample, meaning many replicates were needed (Getachew *et al.*, 1998). Researchers have modified the original batch fermentation technique for gas production measurement (Patra & Yu, 2013). The four main gas measuring techniques are the Hohenheim gas method described by Menke *et al.* (1979), the Liquid displacement system (Beuvink *et al.*, 1992), the Manometric method (Waghorn and Stafford, 1993) and the various pressure transducer systems: manual (Theodorou *et al.*, 1994), computerised (Pell & Schofield, 1993), and combination of pressure transducer and gas release system (Cone *et al.*, 1996).

The IVGPT is inexpensive and does not require specialised facilities and resources (Getachew *et al.*, 2004). It is less laborious and time-consuming compared to *in vivo* methods of CH_4 measurement and easier to standardise as separate feedstuffs can be tested rather than whole rations as in *in vivo* techniques (Cone *et al.*, 1996). This method is also extremely valuable in evaluating mitigation strategies. *In vitro* studies can simulate *in vivo* conditions and the mitigation strategies tested to determine how they would affect CH_4 production *in vivo*. IVGPT has a large capacity for fast screening of many different feedstuffs, additives and diet types and for maintaining a more constant fermentation environment than *in vivo* techniques (Storm *et al.*, 2012; Gupta *et al.*, 2018). However, this method cannot take into account physical factors such as the passage rate of digesta or the physical form of the feed and cannot evaluate the long-term effects of various treatments on CH_4 production (Storm *et al.*, 2012). *In vitro* degradation is very different compared to *in vivo* degradation, where the level of feeding and the physical and chemical nature of the feed affect digestion (Hill *et al.*, 2016). Emissions and digestibility by the whole animal cannot be simulated (Gupta *et al.*, 2018). These limitations could ultimately result in CH_4 production that will differ significantly between *in vivo* and *in vitro* evaluations of the same treatment (Hill *et al.*, 2016). The IVGPT is the first step to evaluating the effect of various feedstuffs and additives on CH_4 emissions and is useful for studies where controlled incubation conditions are needed (Storm *et al.*, 2012).

2.6.2.2 Prediction models and equations

Indirect methods, such as prediction equations and models, are generally used when enteric CH_4 emission data are needed for larger areas, such as when estimating regional, national or international emissions values. This method is mainly used to develop national, continental and international CH_4 emission inventories for domestic livestock. These CH_4 prediction models are based on data obtained from *in vivo* studies such as animal characteristics, feed characteristics, feed intake or digested nutrients (Gupta *et al.*, 2018).

Several published mathematical models are available for ruminant CH_4 emission prediction; however, most are limited to cattle in developed countries (Benaouda *et al.*, 2020). Different models estimate enteric CH_4 emissions based on the animal and feed in question. The IPCC released their Tier (1, 2, and 3) methodology for determining national, continental, and global CH_4 emission inventories from livestock, in 2006 (IPCC, 2006). Each successive tier provides increased accuracy with the cost of increasing complexity (Eugène *et al.*, 2019). The IPCC Tier models are the standard for estimating ruminant CH_4 emissions. They have, since then, made refinements to these 2006 guidelines (IPCC, 2019). The IPCC Tier models are the most commonly used to estimate enteric CH_4 emissions, as they have been designed for broad, global use and can therefore be used by many different countries. These methods are widely used to develop GHG inventories for various livestock production systems in different regions and countries (Aljaloud *et*

al., 2011). The choice of model to use depends on the amount of data available and the area for which estimations are needed (Aljaloud *et al.*, 2011). Tier 1 and 2 are typically used for larger areas, whereas Tier 3 is more suited for smaller, restricted areas (Aljaloud *et al.*, 2011). The IPCC Tier 1 or Tier 2 methodologies are used in many countries to report their national GHG emission inventories (Benaouda *et al.*, 2019). Tier 1 is the simplest of the three models, used when only the number of animals is accurately known, assuming an average emissions factor obtained from literature (Tongwane & Moeletsi, 2020). It is recommended for use by countries or studies that do not yet have local emission values (Tongwane & Moeletsi, 2020). The tier 2 model is slightly more advanced, requiring more information about animal categories, feeding and production systems, and manure management (Eugène *et al.*, 2019). This model is specific to the location or country and compares the required gross energy to the actual feed intake of the animal (Tongwane & Moeletsi, 2020). Tier 3 is the most data-dependent and complex of the three tier methods, requiring knowledge of the feeding system and the animal's energy requirements (Storm *et al.*, 2012; Eugène *et al.*, 2019).

These models provide the means to create national and global GHG inventories and assist with developing environmental policies and strategies to reduce overall emissions (IPCC, 2006). They are easy to apply and accessible, enabling all countries to estimate the average total emissions of different ruminant livestock species and breeds and the ruminant livestock population, keeping track of the increase in emissions (Aljaloud *et al.*, 2011; Storm *et al.*, 2012). However, compared to direct methods, enteric CH_4 emissions estimated by these models usually have lower accuracy and are associated with more uncertainty (Hristov *et al.*, 2018). These inaccuracies are partly linked to the small data sets used for model development and parameterisation (Hristov *et al.*, 2018). For this method to be accurate, much information must be collected regarding the animal, DMI, diet composition and nutrient content, production system, location, and weather factors. Models are tested by comparing collected CH_4 emission data to the calculated values. In this regard, expensive methods such as the respiration chamber or the SF_6 tracer technique are first required to collect emission data for various production situations. Regarding South Africa, Tongwane and Moeletsi (2020) point out the limited availability of emission factors available to determine SA enteric CH_4 emissions. There can be some difficulties in using extant models through the limited availability and difficulty obtaining reliable input variables for the model. The lack of suitable input variables to use in the models can result in significant inaccuracies in estimated emissions values (Ellis *et al.*, 2007) and lead to unreliable emissions data. Many countries (mainly developing) have only recently started measuring enteric CH_4 emissions due to the high cost of equipment and the specialised methodologies required to obtain measurements from live animals (Benaouda *et al.*, 2020). Therefore, a limited amount of data specific to these countries can be used as emission factors in prediction models. Most models that are available are more specific for developed countries. When these models are applied to areas where little research has been done, it could lead to inaccurate CH_4 emission estimates unsuitable for the region, which can differ

in genetic potential, diet, climate, and management practices (Benaouda *et al.*, 2020). For highly accurate prediction models to be developed, more CH_4 emissions data must be obtained for different countries, regions, production systems and species.

2.6.3 Short-term measurement techniques

The increasing interest in estimating CH_4 emissions from a large number of animals in their natural environment has led to the development of numerous short-term measurement techniques.

2.6.3.1 The GreenFeed system

The GreenFeed (GF) CH_4 measurement system is a direct, short-term, individual measurement technique. It is a newer method of determining ruminant enteric CH_4 emissions and has been used for the past ten years. The GF system is an automated head-chamber system developed for spot sampling respired and eructated gas (Zimmerman, 2011). The SF_6 tracer technique and the GF system are the two most widely used methods for determining ruminant enteric CH_4 emissions on pasture (Hristov *et al.*, 2016). Zimmerman (2011) patented the GF measurement system with the initial design for cattle. Hegarty (2014) was the first to develop a GreenFeed emissions system specifically for sheep.

The GF system collects numerous short-term CH_4 emission samples from an individual animal multiple times a day over several days or weeks. These samples estimate the animal's average daily CH_4 emissions (Hammond *et al.*, 2016). The GF system (C-Lock Inc., Rapid City, South Dakota, USA) consists of a feed dispenser, an extractor fan that creates a measured airflow, and a gas analyser close to where the ruminant puts its head. Supplemental feed (or sometimes water) is placed in the feed dispenser to entice the animal to use the unit. Each unit is fitted with a tag reader to determine which animal has visited the unit. Based on which animal visits the unit, an operating feed dispenser provides a ration of supplement feed (Zimmerman, 2011). The GF system relies on the animal's voluntary visits to the unit. The animal can visit at any time, but a feed reward is only given, and a CH_4 measurement is taken if a specified amount of time has passed between visits (Hammond *et al.*, 2016). Software provided with the system allows the investigator to control the timing of feed availability and distribute CH_4 measurements at various times throughout the day (Hammond *et al.*, 2016). The air collected at each valid visit is filtered, the airflow rate measured, and the sampled air analysed for CH_4 using non-dispersive infrared (NDIR) analysis (Coppa *et al.*, 2021). The active airflow, CH_4 concentration and the animal's head position are recorded, and through an internal algorithm, the 24h CH_4 production is estimated (Hammond *et al.*, 2016; Hristov *et al.*, 2016). The timing and duration of animal visits to the GF unit can affect the estimated CH_4 emissions values. The best results are therefore obtained when the investigator controls the number and timing of animal visits to the unit to be appropriately distributed over a 24-h feeding cycle to account for diurnal variation (Hammond *et al.*, 2015; Hristov *et al.*, 2015; Hristov

et al., 2018). A single GF unit can be used for 15-20 animals when grazing and 20-25 animals when housed in free stalls (Hammond *et al.*, 2015; Sorg *et al.*, 2018).

Hammond *et al*. (2015) evaluated the validity of the GF system by comparing it to the RC and the SF_6 tracer technique. The average CH_4 emissions determined using the GF system were similar to that of the RC. However, a poor agreement between the two methods is said to be attributable to the small number of measurements obtained using the GF system (Hammond *et al.*, 2015). When comparing the GF and SF_6 techniques, the SF_6 obtained a higher average than the GF system, but with a significant agreement between the two methods (Hammond *et al.*, 2015). In these comparisons, the GF system did not measure the significant treatment effects measured by the RC and SF_6 techniques, and the GF system ranked treatments and individual animals differently (Hammond *et al.*, 2015). These differences are said to be due, in part, to a small number of visits to the GF unit and the timing of measurements obtained relative to daily patterns of CH_4 emissions (Hammond *et al.*, 2015). Velazco *et al.* (2016) compared daily CH_4 production estimates of the GF system to those of the RC. They found no difference between the daily CH_4 production averages and CH_4 yield averages of the two methods and a high correlation between them (Velazco *et al.*, 2016). Hristov *et al.* (2016) compared the GF system to the SF_6 tracer technique. In this comparison, the SF_6 technique produced greater variability in daily CH_4 emissions than the GF system, with low concordance and correlation between the two methods. However, the overall average CH_4 production throughout the experiment was agreeable, with small, inconsistent differences. These results are said to result from ventilation in the barn where the experiment was conducted and the background CH_4 and SF_6 concentrations (Hristov *et al.*, 2016).

The GF system can analyse CH_4 emissions from many animals in free-ranging conditions on the field and free- or tie-stall barns (Hristov *et al.*, 2015). This system allows animals to remain in their natural environment on pasture or move freely in a free-stall barn, enabling them to exhibit normal behaviour and select their forages (Hristov *et al.*, 2015). Compared to the RC, the GF system has lower investment costs, is cheaper to assemble and maintain, and is easier to operate (Hristov *et al.*, 2015). Compared to the SF_6 tracer technique, which requires specialised equipment, comprehensive training and animal handling, the GF system is simpler and easier to operate, non-invasive, less expensive, and does not require complex and expensive analytical equipment (Hristov *et al.*, 2015). The GF system takes automated CH_4 emissions measurements in real-time with minimal disturbance to cow behaviour (Guinguina *et al.*, 2021). The GF system is the most suitable measurement technique for commercial farm conditions, allowing many animals to be sampled simultaneously (Coppa *et al.*, 2021). The main limitations of this method are the unrepresentative sampling, resulting from irregular and infrequent visits to the unit, and the need for an enticement supplemental feed, which could end up representing 5% of the animal's DMI at the time of a gas measurement event (Hristov *et al.*, 2015). The supplement provided may introduce between-day variations in supplement consumption and thus influence or interact with the assessed treatments (Hammond *et al.*, 2016). This system is dependent on voluntarily visits to

the unit. Therefore, the gas measurements are reliant on animal visits, and one animal's visit to the unit may prevent or delay another animal's visit. Therefore, CH_4 measurements are not entirely independent, as each animal is not a completely independent experimental unit (Hammond *et al.*, 2015). Also, the number, timing and duration of the animal's visits may not represent the diurnal pattern of CH_4 production, resulting in biased CH_4 emission estimates (Hristov *et al.*, 2015; Hammond *et al.*, 2015). Like the RC, the animal must be trained to use the units. However, certain animals do not become accustomed to using it and can, therefore, not be included in the experimental population. Hammond *et al.* (2015) found that the GF system could not rank according to treatments. Hammond *et al.* (2015) and Coppa *et al.* (2021) also found that the GF system could not rank individual animals according to the CH_4 emission rate. This method is, therefore, not suitable for the genetic screening of individual animals but is good for average herd CH_4 emission evaluation.

2.6.3.2 The Laser Methane Detector

Chagunda *et al.* (2009) identified the proprietary laser methane detector (LMD) as an alternative measurement technique to determine enteric CH_4 emissions from ruminant animals in their natural environment under normal husbandry practices (Chagunda *et al.*, 2009; Grobler *et al.*, 2014). Most enteric CH_4 measurement techniques can only be applied at the controlled experimental level, whereas the LMD can be used at the applied research level on commercial farms (Chagunda *et al.*, 2009). The LMD was initially developed for detection applications in the natural gas industry (Iseki, 2004; Crowcon Detection Instruments, 2019). Knowledge of the benefits and abilities of the LMD led Chagunda *et al.* (2009) to suggest it for measuring cattle enteric CH_4 emissions. This method has since been applied to sheep (Chagunda *et al.*, 2013; Ricci *et al.*, 2014) and goats (Roessler *et al.*, 2018).

The LMD is a portable, hand-held, open-path laser measuring device used for remote measurement of column density for CH_4-containing gases (Tokyo Gas Engineering Co. Ltd., 2006). LMD measurements are based on infrared-absorption spectroscopy (Chagunda *et al.*, 2009). Measurements are taken manually, with the operator holding the LMD about 1-3m away from the animal's nose or mouth. A visible HeNe collimated laser beam is directed at the animal's nose to measure the concentration of CH_4 in the air between the hand-held LMD and the animal's nose or mouth, allowing estimation of exhaled enteric CH_4 emissions (Chagunda, 2013; Chagunda *et al.*, 2013; Ricci *et al.*, 2014). Methane measurements are expressed in parts per million per metre (ppm-m), relating to the CH_4 concentration and the depth of the CH_4 respiratory plume (Chagunda *et al.*, 2013). The LMD is a short-term method, providing estimates of intervals of enteric CH_4 flux and scaling the estimates up to represent the daily CH_4 emissions of each animal (Goopy *et al.*, 2016).

Chagunda & Yan, (2011), Chagunda *et al.* (2013) and Ricci *et al.* (2014) compared the LMD and RC. Chagunda and Yan (2011) reported a strong positive correlation (r = 0.8) between the LMD and RC, indicating that these techniques were measuring the same trait. Despite the high correlation, measurements obtained using the LMD were, on average higher than those of the chamber (Chagunda & Yan, 2011). In this study, the LMD measured CH_4 concentrations from the outflow of the chambers rather than the animals. The higher values obtained using the LMD could therefore be a result of the LMD measuring both respired and eructated CH_4 of the cattle and the background CH_4 concentration of the chamber. Chagunda *et al*. (2013) and Ricci *et al.* (2014) reported a positive but weak correlation between the LMD and RC. Chagunda *et al.* (2013) found that CH_4 measurements of the LMD had greater variation and a higher mean than the chamber. Ricci *et al.* (2014) state that the LMD showed promise in comparing and ranking the effects of various feeding routines and diets.

The LMD is inexpensive compared to the RC, convenient and practical to use, and its portability makes it easy to apply in many different environments (Grobler *et al.*, 2014). The LMD can obtain on-field CH_4 measurements from animals in their natural environment without disturbing their normal activity (Roessler *et al.*, 2018). LMD measurements can differentiate between CH_4 concentrations produced while the animal performs different physiological activities (Chagunda *et al.*, 2013). The LMD can be used for on-farm monitoring of ruminant emissions and, subsequently, for decision support for CH_4 mitigation strategies (Chagunda *et al.*, 2013). The LMD is a non-invasive, non-destructive and non-contact technique that quickly collects real-time measurements from individual animals (Chagunda, 2013; Ricci *et al.*, 2014). These characteristics allow the maintenance of the animal's welfare and the handler's safety (Chagunda, 2013; Ricci *et al.*, 2014). The LMD is, however, limited in that it only takes measurements for short periods within the day, extrapolating daily CH_4 emission estimates from these short measurement periods, and animals are required to remain still for the duration of the measurement period. Although this method has been proven reliable in estimating enteric CH_4 emissions in controlled research environments and respiration chambers, few peer-reviewed publications have analysed its efficacy under uncontrolled field conditions (Roessler *et al.*, 2018). Various studies that have taken measurements under outdoor field conditions found that ambient conditions such as wind speed, relative humidity, and atmospheric pressure significantly affect the accuracy and precision of LMD recorded concentrations (Chagunda, 2013; Chagunda *et al.*, 2013; van Wyngaard, 2018). The angle at which the LMD is held and the operator influence the measured CH_4 values (Sorg *et al.*, 2018). As the LMD is hand-held, it is labour intensive and introduces variation (Hammond *et al.*, 2016). Roessler *et al.* (2018) pointed out the lack of a standard measurement protocol for each livestock species, which could significantly increase the variability of results obtained in different studies, making the comparison of results challenging and inaccurate. Once the measurement protocol of the LMD is refined and standardised, the few limitations of this technique can be eliminated, and the LMD will have the potential to be applied more broadly.

2.7 Application of technology in the livestock industry

The livestock industry currently faces the dilemma of increasing production to meet growing demands as it works to maintain global food security while improving the sustainability of production. The problem arises with the lack of the ability of traditional farming techniques to keep up with the rising demands, coupled with the many deleterious effects of traditional farming practices on environmental sustainability. These trends force the agricultural industry into a position where there is an increasing need to transition away from the industrial era's traditional farming practices towards a new era that facilitates significantly increased productivity while effectively dealing with the increasing scarcity of resources to achieve sustainable production (Charania & Li, 2020). A potential solution is in technology application in agriculture, with a particular focus on the Internet of Things (IoT). The application of the IoT in agriculture has become increasingly popular over the past decade, drastically rising since 2016 (Tzounis *et al.*, 2017).

Technological advancements can significantly impact the ruminant livestock sector. Technology improvement provides a tremendous opportunity within the industry to meet growing demands while reducing GHG emissions and overall environmental impact, creating the opportunity for the industry to achieve UN SDG 13. The combination of the need for increased food production and the Fourth Industrial Revolution (4IR) places agriculture in the unique precipice of the next agricultural evolution, with the ability to affect not only variety and crop yields and production efficiency but also environmental factors and climatological and social outcomes (Charania & Li, 2020). This revolution sees the integration of various information and communication technologies such as remote sensing IoTs, Unmanned Aerial Vehicles (UAVs), Big Data Analytics, and machine learning (Boursianis *et al.*, 2020). The application and advancements of these technologies will enable an era of data-driven and automated agriculture (Charania & Li, 2020). The IoT and UAVs stand out as the two enabling technologies that could lead the transformation of traditional agricultural practices through the fourth revolution (Boursianis *et al.*, 2020). The correct development and implementation of these technologies aim to transform the agriculture industry by improving production efficiency, optimising resource use, increasing profits and improving sustainability. Technological advancements and innovation are crucial for the progression of the livestock industry. They assist in achieving goals, improving efficiency and creating job opportunities. Data-driven agriculture will assist producers with real-time decision-making and informed management practices, reducing inefficiencies and uncertainties and reducing environmental impact (Villa-Henriksen *et al.*, 2020).

As the agriculture industry evolves, the concept of smart farming or Agriculture 4.0 is becoming more prominent. The technologies driving agriculture advancement in the new age of smart farming can collect information about numerous different factors such as the quality of the land and changes in quality of the land and soil over the years, determining pH, temperature, weed

seeking, and wind speed (Boursianis *et al.*, 2020). The development of smart farming aims to assist producers in making wiser and timelier decisions based on collected data, to assist with the implementation of adaptation techniques and as a green technology approach, to reduce the ecological footprint of current farming (Boursianis *et al.*, 2020). These new technologies can collect large volumes of data. This data collection from many farms allows data sharing among farming communities and governments to assist with policy-making and let the government know where producers need assistance.

Within this surge of technology application in agriculture is the opportunity to develop a CH_4 measurement technique that is easy to use, accurate, affordable, has low labour requirements and can be purchased and used by producers. This technique could assist in developing, testing, and applying effective mitigation strategies within the industry, which are favourable for livestock farmers.

2.8 LoRa (Long Range)

LoRa (Long range) is a sub-technology of the 'Internet of Things' (IoT). The IoT is a concept of connecting a network of physical 'things,' such as sensors, to or through the internet without any direct human intervention (Villa-Henriksen *et al.*, 2020). It is a network of physical devices enhanced by electronics, software and connectivity (Tzounis *et al.*, 2017; Charania & Li, 2020). LoRa is a technology that is quickly gaining interest for application in agriculture. Its potential uses are broad and include assistance with monitoring various factors such as weather conditions, soil quality and moisture, and animal monitoring, to name a few, with the potential to assist in meeting the requirements of traceability throughout the industry supply chain (Mahbub, 2020; Villa-Henriksen *et al.*, 2020). LoRa is a wide-area network (WAN) technology that can extend large geographical distances and relay data over these long ranges using low-power wireless networks (Charania & Li, 2020). It was developed in 2009 by Cycleo (Grenoble, France) and bought in 2012 by Semtech Corporation (USA) (Miles *et al.*, 2020). LoRa is the physical layer, and LoRaWAN (Long Range wide area network) is the communication protocol, standardised by the LoRa Alliance, allowing the transmission of small amounts of data over long distances (Peña Queralta *et al.*, 2019; Saban *et al.*, 2021). The LoRa network is usually deployed in a star-of-star topology, which enables gateways to relay messages between the devices and the central network (Charania & Li, 2020). It can be used in areas with no network coverage and transmits small amounts of data over distances of about 5km in urban and 20km in rural areas (Miles *et al.*, 2020). The LoRa network consists of three layers, namely the physical perception (sensing) layer, the network layer (data transfer) and the application layer (where data is stored, processed, manipulated and analysed) (Tzounis *et al.*, 2017; Villa-Henriksen *et al.*, 2020).

LoRa technology already has several applications in the livestock industry. It has been applied in poultry farming, providing real-time measurements of the birds' environmental conditions

and increasing hatching results by monitoring egg incubator conditions. Swine farming has applied LoRa technology in monitoring pig health, production, and environmental conditions and supply chain tracking and monitoring. This technology has also been applied in cattle farming for remote herd monitoring and management, environmental monitoring on dairy farms, optimisation of cattle health and well-being, animal tracking, and internal body condition tracking (LoRa Alliance, 2020). Mohteshamuddin *et al.* (2023) recently tested the use of an orally ingested bio-capsule to monitor cattle health, using a customised LoRa network and gateway server installed on the farm for communication of animal conditions to a cloud server and mobile device.

LoRa technology can potentially be used with a wireless CH_4 sensor as an on-farm enteric CH_4 measurement technique. The main benefits of LoRa technology are its low energy consumption, relatively low cost, low power transmission, and reduced amount of data transferred through the network (Gomes *et al.*, 2019). It can transmit data over long distances in areas with no network coverage. Compared to the direct CH_4 measurement techniques listed above, it is much less time-consuming and has low labour requirements, as, after calibration and installation, data is collected without human input. This technique would provide the long-term measurement of CH_4 emissions from ruminant animals, allowing the effects of various mitigation strategies to be determined. This method enables real-time data acquisition. Producers can share databases and work together to make decisions toward more ecologically friendly farming practices, improved production, and good animal health and welfare. As many farms are located in areas with poor network coverage, the use of LoRa provides an opportunity to collect and transfer data through LoRa gateways to the network. LoRa provides a distant monitoring system, and solar panels can power it due to its low power consumption. Like the LMD, LoRa has the potential for on-farm emission monitoring, providing data for decision support regarding CH_4 mitigation strategies. It is also able to take measurements without disturbing the animals. Producers can easily monitor implemented CH_4 mitigation strategies to determine which fits their business best. This method could be the solution to obtaining more accurate values for compiling national and international GHG inventories.

The use of LoRa technology in livestock farming provides numerous opportunities in the industry. Not only for determining methane emissions values from animals but also for collecting weather data. Weather data is critical in times of climate change. As described, climate change can affect normal climate patterns, such as wind, precipitation and temperatures, making them unpredictable. This unpredictability of climate patterns over time could result in difficulty in knowing when optimal planting time is for cattle feed and changes in breeding patterns. The climate data collection by LoRa can be used to make predictions each year and analyse the pattern in which the changes are occurring to remove some of the unpredictability that accompanies climate change.

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Chapter 3

The design and placement of LoRa sensors to measure methane emissions in sheep under grazing conditions

Abstract

As the pressure on the ruminant livestock industry to reduce enteric methane emissions increases, the need for an enteric methane measurement technique, which can be used under extensive grazing conditions to verify global livestock methane emission inventories and assess various mitigation strategies, increases. The agricultural industry is in a unique space of transformation as the fourth industrial revolution encourages the use of technology to create automated and data-driven production. This study investigates the potential of a methane sensor in combination with LoRa technology to measure enteric methane emissions from sheep under extensive grazing conditions and analyses the design and placement of these devices. Measurements were taken over three periods with three devices installed for the first and second and one for the third. Measurements were taken every 50 seconds and recorded in parts per million. The first period measured methane when no animals were present to determine the background methane concentration and the effect of ambient conditions on the device. Relative humidity was found to be significantly positively correlated to the observed methane measurements, while air temperature, wind speed and solar radiation were negatively correlated with the measured methane concentrations. Ten Dohne Merino rams with GPS tracker collars were placed into a camp for the second and third measurement periods. The methane levels measured in Periods 2 and 3 were compared to Period 1 and corrected for background methane concentrations. A significant difference in methane concentration was found between the first and second measurement periods, indicating the potential impact of the rams on the increased methane concentrations measured. Analysis of the relative distance of each ram from the three devices and the subsequent methane observation was used to determine whether there was an association between the distance of the ram from the device and methane measurement. No association was observed between distance and methane observation, revealing that in this study, the proximity of the sheep had no effect on methane concentrations measured. However, a higher methane concentration was measured when sheep were in the camp. These results indicate that through further development, the device could have the potential to determine methane emissions from sheep under grazing conditions.

Keywords: CH_4 measurement, sheep, LoRa

3.1 Introduction

Over the past 30 years, climate change has become an important research topic and a prominent global concern. Climate change is characterised by long-term changes in mean global temperatures and increased variability of weather patterns (IPCC, 2018). It is driven by changes in human activities that have led to considerable increases in anthropogenic greenhouse gas (GHG) emissions, disrupting the natural carbon cycle. Methane (CH_4) is one of the most important GHGs, intensifying the impact of climate change, as it is an extremely potent GHG and the most abundant organic trace gas in the atmosphere (Wuebbles & Hayhoe, 2002). Methane has a global warming potential* (GWP*) of about 7.8 over 100 years (Costa *et al.*, 2021). The atmospheric CH_4 concentration has increased considerably since the onset of the 19th century industrial age, increasing from a steady 617-716 parts per billion (ppb) to between 1,880 and 1,960 ppb in 2018, depending on latitude (Yusuf *et al.*, 2012; Glikson, 2018; Our World in Data, 2018).

Ruminants contribute significantly to global CH_4 emissions, with enteric fermentation contributing approximately 24-28% of global CH_4 emissions and 39% of the agricultural sector's CH_4 emissions (Yusuf *et al.*, 2012; Glasson *et al.*, 2022). *Methanogenic archaea* in the reticulo-rumen of ruminant animals produce CH_4 as a by-product of fibre fermentation (Li *et al.*, 2014). Methane accumulated in the rumen is removed by eructation and expiration into the atmosphere. Because CH_4 is a highly concentrated form of energy, the CH_4 produced represents a 2-12% loss of dietary energy, revealing an inefficiency in the ruminant fibre digestion process (Johnson & Johnson, 1995). Despite the considerable contribution to emissions, ruminants are valuable in sustainable agricultural systems due to their ability to convert feed sources unsuitable for human consumption into highly nutritious and human-edible products (van Lingen *et al.*, 2021).

Small ruminants comprise 56% of the global ruminant population and can play an essential role in the development of CH_4 measurement techniques and mitigation strategies (Marino *et al.*, 2016). The domesticated small ruminant population, i.e. sheep and goats, produce 6.5% of the global livestock industry GHG emissions, emitting approximately 475 million tonnes of CO_2-equivalents per year (Gerber *et al.*, 2013; van Lingen *et al.*, 2021). In South Africa, small ruminants are responsible for approximately 15.6% of total national livestock emissions (du Toit *et al.*, 2013). In the South African context, sheep production is an important component of South African agriculture. South Africa has over 18 million commercial sheep (Abstract of Agricultural Statistics, 2022). The South African sheep industry is a source of livelihood for many South Africans and is important for economic growth (Herrero *et al.*, 2013; Elum *et al.*, 2017). Sheep production also makes use of land unsuitable for more intensive farming practices. Of the 86.2 million ha of farming land available in South Africa, approximately 71.9 million ha is suitable only for extensive livestock farming, lying within arid and semi-arid zones (Schoeman *et al.*, 2010; Abstract of Agricultural Statistics, 2018). Sheep farming represents a source of sustainable production in these areas, and most of South Africa's sheep are therefore reared in extensive production systems.

Due to the drastic increase in anthropogenic global GHG emissions, various climate change mitigation policies and strategies have been developed. The Kyoto Protocol (UNFCCC, 1997), the Paris Agreement (UNFCCC, 2016), and the 13th sustainable development goal (SDG) of the 2030 Agenda of Sustainable Development (United Nations, 2022) are the leading global initiatives focusing on climate change reduction. In recognition of climate change and the abovementioned agreements, South Africa submitted the Low-Emission Development Strategy (LEDS) to the United Nations Framework Convention on Climate Change (UNFCCC). The LEDS aims to be the first step in South Africa's journey toward a net zero-carbon economy by 2050 (RSA, 2020). New climate change policies and global movements toward increased sustainability have focused on livestock production as an ideal area for GHG emission reduction. Ruminants are at the forefront of CH_4 mitigation studies because of their significant contribution to anthropogenic CH_4 emissions through enteric fermentation. Consequently, the ruminant livestock industry is now under increasing pressure to make changes in production strategies and feed management to reduce its contribution to CH_4 emissions and increase the sustainability of production. Methane's short atmospheric lifespan and high global warming potential make it an important avenue for emission reductions, creating the potential for the short-term slowing of climate change (Cain *et al.*, 2019; Glasson *et al.*, 2022). Successful CH_4 emission mitigation requires the development of effective CH_4 mitigation techniques and valid livestock emission inventories. There are many uncertainties in ruminant CH_4 emission studies, especially regarding livestock emission estimates and the contribution of livestock to global CH_4 emissions (Hristov *et al.*, 2018). These uncertainties are predominantly a result of very little knowledge of domestic ruminant CH_4 emissions in extensive production systems (Pérez-Barbería *et al.*, 2020). Uncertainties in CH_4 emission quantification lead to uncertainties in compiling inventories and determining baselines against which emission reduction progress is measured.

The ability to accurately measure ruminant CH_4 emissions is crucial for developing and accessing mitigation strategies, compiling, and validating national GHG inventories, developing quantification protocols, and assisting with genetic selection (Swainson *et al.*, 2011; Velazco *et al.*, 2016; Hammond *et al.*, 2016). The abovementioned factors require an accurate and precise measurement technique to measure enteric CH_4 emissions from ruminant animals in their different production environments (Johnson & Johnson, 1995; Velazco *et al.*, 2016). The measured emission values will assist in developing emission profiles for animals in certain areas and production systems, providing an emissions baseline important for setting emissions targets and measuring their progress (Jones *et al.*, 2014). Accurate quantification of ruminant CH_4 emissions under their natural production environment and precise measurement of the changes in emissions will assist with refining livestock emission estimates and analysing the effectiveness of mitigation strategies under different production conditions (Phillips, 2012). As ruminant production systems and regions in South Africa are incredibly diverse, mitigation strategies should be specific to the

region and production system. Understanding how region, feed, and production systems affect enteric CH_4 production is vital for production efficiency and mitigation strategy development.

Despite the numerous enteric CH_4 emission measurement techniques available, many are expensive, labour-intensive, and limited in their application on commercial farms with a large number of animals (Ricci *et al.*, 2014). As most sheep are reared under extensive grazing conditions in South Africa, a CH_4 emission measurement technique is needed that can be used under these production conditions. The most widely used direct, individual animal CH_4 emission measurement techniques are the respiration chamber (RC), Sulphur Hexafluoride (SF_6) tracer technique and GreenFeed (GF) system (Hristov *et al.*, 2018). The laser methane detector (LMD) is also gaining popularity as an enteric CH_4 emission measurement technique. The SF_6 tracer technique and RC are presently the most reliable techniques available; however, their application on commercial farms, where applied research is performed, is less practical and extremely limited (Chagunda & Yan, 2011; Hammond *et al.*, 2016).

The RC is currently the most common measurement technique. Due to its reliability, it is the primary source of emissions data on which national and global domestic ruminant emissions are based (Johnson & Johnson, 1995; Gardiner *et al.*, 2015). When operated correctly, it is a precise technique that provides accurate CH_4 emission values and information on diurnal emission variations and is effective in comparing various enteric CH_4 mitigation strategies (Storm *et al.*, 2012; Goopy *et al.*, 2016; Hammond *et al.*, 2016). However, the RC is limited by the need for trained and acclimatised animals, and its confinement of these animals in an unnatural environment limiting animal movement, natural behaviour, diet selection, and interaction with peers and the environment (Johnson & Johnson, 1995; Pinares-Patiño *et al.*, 2011; Storm *et al.*, 2012; Berndt *et al.*, 2014). Consequently, the CH_4 emission values obtained using the RC do not provide reliable estimates that can be applied to grazing animals, as grazing animals select their diet and have a higher maximum intake compared to animals housed in chambers (Johnson & Johnson, 1995; Storm *et al.*, 2012; Li *et al.*, 2014; Hammond *et al.*, 2015; Hammond *et al.*, 2016). Furthermore, the number of animals that can be monitored using the RC are limited as the chamber requires a skilled operator (i.e., creating a high margin for human error), is characterized by high equipment and maintenance costs, and requires trained animals (Johnson & Johnson, 1995; Storm *et al.*, 2012; Deighton *et al.*, 2013; Goopy *et al.*, 2016; Huhtanen *et al.*, 2019).

Zimmerman (1993) and a research team at Washington State University developed the SF_6 tracer technique, and Johnson *et al.* (1994) first applied it experimentally. This technique has a lower cost than the RC and can determine enteric CH_4 emissions from a large number of individual ruminants simultaneously under grazing conditions (Swainson *et al.*, 2011; Deighton *et al.*, 2013; Berndt *et al.*, 2014). The SF_6 tracer technique is still the most reliable technique for measuring CH_4 emissions from free-ranging animals (Hammond *et al.*, 2016). However, some studies have observed high among- and within-animal variation in CH_4 emission estimates caused by differing permeation rates for individual permeation tubes (Grainger *et al.*, 2007; Pinares-Patiño *et al.*, 2008;

Pinares-Patiño *et al.*, 2011). Pinares-Patiño *et al.* (2011) also found that the SF_6 tracer technique overestimated emissions compared to the RC. Compared to the RC, the SF_6 tracer technique is less precise, more labour intensive, and the equipment is less physically robust (Hammond *et al.*, 2016; Goopy *et al.*, 2016).

The GF system was patented by Zimmerman (2011). It is a static, automated head chamber that spot-samples eructated and exhaled air (Zimmerman, 2011; Hammond *et al.*, 2016). It consists of a feed dispenser that provides supplements to lure the animals and a gas analyser proximate to where the animal places its head (Zimmerman, 2011). It is non-intrusive, less expensive compared to the RC and SF_6 tracer technique, is easy to operate, does not restrict animals, and can be used for large groups of animals in their normal production system for extended periods (Hristov *et al.*, 2015; Velazco *et al.*, 2016; Guinguina *et al.*, 2021). The GF system is, however, limited by the need for supplemental feed or water as an enticement to ensure the animal uses the system (Goopy *et al.*, 2016; Hammond *et al.*, 2016). It is a concern that the enticement feed contributes excessively to the animal's daily intake, which will negatively affect mitigation and treatment effect studies, and cause between-day variation in supplement consumption (Hammond *et al.*, 2015; Hammond *et al.*, 2016). It is also possible that the enticement feed affects basal ration or pasture intake, altering overall daily CH_4 production (Velazco *et al.*, 2016). Measurements are only valid when the animal's head is in an acceptable position relative to the analyser and the measurement period is uninterrupted (Hammond *et al.*, 2016). The success of the GF system requires animal visitation, and the number and timing of visitations obtained relative to diurnal CH_4 emissions may not represent true daily CH_4 emission fluctuations, biasing estimated CH_4 emissions (Storm *et al.*, 2012; Hammond *et al.*, 2015; Hammond *et al.*, 2016). Animals need to be trained to use the GF system, and some may not become frequent users (Hammond *et al.*, 2016).

The LMD was initially developed to detect leaks from natural gas pipelines and emissions from garbage landfills (Iseki, 2004) and Chagunda *et al.* (2009) later introduced its use for ruminant enteric CH_4 emission measurement. Compared to the RC and SF_6 tracer technique, the LMD is relatively inexpensive, convenient and practical (Grobler *et al.*, 2014; Troy *et al.*, 2016). The LMD collects real-time measurements, is sensitive and has a fast response time, allowing frequent measurements to be taken from ruminants and assisting with on-farm GHG mitigation decision support (Chagunda, 2013; Chagunda *et al.*, 2013; Hammond *et al.*, 2016). Roessler *et al.* (2018) found the LMD to be reliable for estimating ruminant enteric CH_4 emissions. However, as the LMD is hand-held, it is labour-intensive and can introduce variation (Hammond *et al.*, 2016). Ambient conditions significantly affect the accuracy and precision of the readings obtained, limiting the LMD's application (Chagunda, 2013). As it is still a relatively new measurement technique and does not yet have a measuring protocol for different livestock species, comparing different studies that have used this technique is difficult (Roessler *et al.*, 2018).

The emergence of the fourth industrial revolution (4IR), a term coined by Klaus Schwab (2017), is leading the transformation of many sectors, among which also the agriculture industry. The agriculture industry has entered into Agriculture 4.0, characterised by smart farming, an increasingly automated and data-driven agriculture (Charania & Li, 2020). Combining of the various technologies available, characteristic of the 4IR, can be used to the advantage of the agriculture industry to assist with decision-making and enable livestock production to progress toward improved sustainability. One of the main aspects of the 4IR is the internet of things (IoT). The IoT structure has three layers, namely the perception layer (consisting of sensors), the network layer (through which data is transferred), and the application layer (where data is stored and manipulated) (Tzounis *et al.*, 2017; Villa-Henriksen *et al.*, 2020). LoRa (Long Range) is a sub-technology of the IoT and consists of wide-area network (WAN) technology with a star-of-star topology that can relay data between devices and a central network over large geographical distances using low-power wireless networks (Charania & Li, 2020). LoRa was developed in 2009 by Cycleo (Grenoble, France) and bought in 2012 by Semtech Corporation (USA) (Miles *et al.*, 2020). LoRa is the physical layer, and LoRaWAN (Long Range wide area network) is the communication protocol, standardised by the LoRa Alliance, allowing the transmission of small amounts of data over long distances (Peña Queralta *et al.*, 2019; Saban *et al.*, 2021). The LoRa represents a low-power, wireless technology that transmits small amounts of data to a gateway over long geographical distances. The gateway forwards data to a network server. The network server collects messages from all gateways, filters out duplicate data and determines the gateway with the best reception. The LoRa technology can be used in areas with no network coverage and can relay data between 5km and 20km in urban and rural areas, respectively (Miles *et al.*, 2020). The technology has been applied in various aspects of livestock production. In the poultry industry, it has provided real-time measurements of the bird's environment and increased hatching results by monitoring egg incubator conditions. Swine farming has applied it in monitoring pig health, production, environmental conditions and supply chain tracking and monitoring. The cattle sector uses LoRa for herd monitoring and management, environmental monitoring on dairy farms, optimising cattle health and wellbeing, animal tracking, and internal body condition tracking (LoRa Alliance, 2020). A recent study by Mohteshamuddin *et al.* (2022), tested the use of an orally ingested bio-capsule to monitor cattle health, using a customised LoRa network and gateway server installed on the farm for communication of animal condition to a cloud server and mobile device. As the agriculture industry increasingly embraces the application of technology in the industry through Smart Farming and Agriculture 4.0, there is an opportunity to use the technology available to assist as a decision support tool (Bahlo *et al.*, 2019).

The combination of LoRa technology with a CH_4 sensor has the potential to provide a hands-free measurement technique for extensive production systems. This technology is ideal for extensive farming systems as it has low energy consumption and subsequent low economic cost,

can transmit data over long distances, can be used in areas with no network coverage, and uses an open spectral band without license fees (Miles *et al.*, 2020; Moysiadis *et al.*, 2021).

The successful assessment of mitigation strategies and the compilation and validation of national CH_4 inventories requires a measurement technique that is simple, reliable, accurate, robust, low-cost, and can be used under commercial production conditions to measure emissions from a large number of animals (Berends *et al.*, 2014; Hammond *et al.*, 2015; Huhtanen *et al.*, 2015; Huhtanen *et al.*, 2019; Coppa *et al.*, 2021). A technique that meets this description would enable producers to determine the success of applied mitigation strategies, and assist with compiling on-farm, local and national CH_4 inventories. This system could assist producers in determining farm-specific net emissions and identifying the most efficient and cost-effective mitigation strategies ideal for their region and production system, thereby allowing them to remain economically viable while reducing enteric CH_4 emissions. It would also assist researchers in determining farm and region-specific emission inventories and mitigation techniques, ultimately leading to significant and sustainable mitigation of enteric CH_4 emissions. The accumulated database would help producers make decisions toward increased sustainability, improved production, and good animal health and welfare.

This study therefore aimed to determine the potential of LoRa technology to be used in combination with an infrared CH_4 sensor to measure enteric CH_4 emissions in sheep under South African grazing conditions.

3.2 Materials and methods

This study was approved by the Research Ethics Committee: Animal Care and Use of Stellenbosch University (ACU-2021-11233).

3.2.1 Experimental location

This study was conducted on the Welgevallen Experimental Farm of the University of Stellenbosch, Western Cape, South Africa, in a small camp (-33.94458, 18.86628), with an average size of 0.07 ha. The average recorded temperature of this area in December and January varies between 20ºC and 21.4ºC, with the minimum temperature ranging between 14.6ºC and 15.9ºC and the maximum temperature between 26ºC and 27.8ºC (Climate-data.org, 2021). The average rainfall is between 23mm and 27mm, and the average recorded relative humidity is between 60% and 61% (Climate-data.org, 2021).

3.2.2 Experimental animals and husbandry

Ten intact Dohne Merino rams were randomly selected from the Welgevallen Sheep research flock and used for the duration of the study. The rams were three and a half years old at the time of the study. They had *ad libitum* access to fresh water and kikuyu (*Pennisetum clandestinum*) grazing for the entire experimental period, and were fed oat hay supplement at 07:00 each morning. The

husbandry of the rams was according to normal farm protocols, and rams were checked daily for the duration of the study to record any abnormal behaviour or discomfort.

3.2.3 Device design and components

The functional LoRa Methane detection unit consisted of the following components: a TTGO T-Beam ESP32 LoRa 868MHz, Infrared CH_4 Sensor NDIR Gas Sensor MH-440D, DFRobot ADS1115 16-BIT ADC Module, solar panel, battery, charge controller, and electrical cover box (Figure 3.1).

Figure 3.1 An indication of the different components of the LoRa methane measurement units that was located in the camp on the Welgevallen Experimental Farm, throughout the study period.

The TTGO T-Beam consists of an on-board SemTech SX1276 for LoRaWAN communication in the 868/915 MHz band and a U-Blox Neo-6M GPS receiver, which provides location tracking (RIOT, 2022). The TTGO T-Beam has a built-in LoRa chip that operates at 868MHz.The function of the TTGO T-Beam is to send data to the LoRa gateway. Visual Studio (VS) Code was used as the integrated development environment (IDE) with the platformio plug-in to write the code and upload it to the TTGO, enabling it to send and receive data. The infrared CH_4 sensor detects the concentration of CH_4 in the air using the non-dispersive infrared (NDIR) theory (Zhengzhou Winsen Electronics Technology Co., Ltd., 2022). This sensor had a 500 ppm error. A more sensitive one

was not chosen as none were available that were sensitive enough to measure the fluctuations in sheep enteric CH_4 emissions. The more sensitive sensors were also costly and more susceptible to environmental noise, where a slight change in ambient conditions such as temperature and humidity could significantly affect the output. The function of the ADC Module was to enable higher resolution measurements to be taken, and the charge controller charged the battery and ensured an output of a constant voltage.

3.2.4 Testing and calibration of the device

A prototype device was assembled and tested for one month, on-farm, from the 13th of September to the 20th of October 2021. The prototype was calibrated using the 2-point calibration method, as the sensor is expected to follow a relatively linear pattern of CH_4 detection over the full range. The device was calibrated in a fume hood by comparing the readings obtained using the LoRa CH_4 measurement device and a laser methane detector (LMD – laser methane mini, SA3C06A, Tokyo Gas Engineering) as the reference device. The LoRa CH_4 device was placed inside the fume hood for the duration of the calibration period. The TTGO T-Beam of the device was connected to a laptop using a USB to micro-USB cable. The laptop, with Arduino installed, was used to monitor, and record the CH_4 readings. The LMD was held outside the fume hood, and the laser pointed next to the CH_4 sensor of the LoRa device. The concentration was recorded every 0.5s with the LMD (in ppm-m) and every 1s with the LoRa CH_4 measurement device (in mV). A minimum ppm value was obtained by recording the CH_4 concentration for 60 seconds while no gases were released into the fume hood. A maximum ppm value was obtained by recording the CH_4 concentration for 60 seconds while CH_4 gas was released into the fume hood. Once the mV values of the LoRa CH_4 measurement device and the ppm-m values of the LMD were recorded, they were related according to time. The minimum and maximum mV values were correlated to the measured LMD ppm values, as seen in Table 3.1.

Table 3.1 The minimum and maximum CH_4 concentration values obtained in the calibration of the LoRa-CH_4 sensor using D_2 (measuring in mv) and the LMD (measuring in ppm-m).

	LoRa (mV)	LMD (ppm-m)
Minimum	405	11
Maximum	456	3758

Once these values were obtained the calibrated (ppm_{cal}) CH_4 concentrations were determined using the following equation:

$$PPM = (value - in_{min}) \times \frac{(out_{max} - out_{min})}{(in_{max} - in_{min})} + out_{min} \qquad \textit{Equation 3.1}$$

Where PPM is the calculated CH_4 concentration in parts per million (ppm), value is the mV output of the sensor, in_{min} is the minimum mV of the device (theoretical = 400 mV), out_{max} is the maximum

ppm output (theoretical = 100000 ppm), out_{min} is the minimum ppm output (theoretical = 0 ppm), in_{max} is the maximum mV of the device (theoretical = 2000 mV). This equation is a standard straight line y = mx + c formula for linear scaling, where $\frac{(out_{max} - out_{min})}{(in_{max} - in_{min})}$ is m, $(value - in_{min})$ is x and out_{min} is c.

Three devices were built and used in this study. The second device (D_2) was calibrated in the fume hood. All values obtained using this device were corrected based on the minimum and maximum values from the calibration and were calculated as follows, using the recorded mV value at any given time:

$$PPMcal = (Value - 405) \times \frac{(3758-11)}{(456-405)} + 11 \qquad \textit{Equation 3.2}$$

Where PPM_{cal} is the CH_4 concentration determined based on the calibration and value is the mV output of the sensor. The other two devices (Device 1, D_1 and Device 3, D_3) used for the study were calibrated during the first measurement period [11 - 13 December 2021] by allowing them to measure ambient CH_4 concentrations. A reading (mV) was taken from each of the three sensors at a specific time (on the 13th of December, before the sheep were introduced into the field). D_1 recorded 440mV, D_2 recorded 432mV and D_3 recorded 373mV. As D_2 was calibrated, its mV value of 432 was inserted into '*Value'* of Equation 3.2 to determine the ppm at that particular time:

$$PPMcal = (432 - 405) \times \frac{(3758-11)}{(456-405)} + 11 = 1994.71ppm \qquad \textit{Equation 3.3}$$

This is done under the assumption that the three sensors are all measuring the same atmospheric CH_4 concentration (in ppm) at a specific time when no sheep are on the pasture. This calculated concentration was then used to determine the minimum mV of D_1 and D_3, using the theoretical in_{max}, out_{max} and out_{min} values specified for the device (Zhengzhou Winsen Electronics Technology Co., Ltd.. 2022):

$$1994.71 = (440 - x) \times \frac{(100000 - 0)}{(2000 - x)} + 0$$

D_1 in_{min}=408 mV

$$1994.71 = (373 - x) \times \frac{(100000 - 0)}{(2000 - x)} + 0$$

D_3 in_{min}= 340 mV

This calculated minimum mV (in_{min}) was then used to determine the CH_4 concentration (PPM_{cal}) based on the calibration for Devices 1 and 3 for the remainder of the trial:

$$PPMcal = (Value - in_{min}) \times \frac{(100000-0)}{(2000-in_{min})} + 0 \qquad \textit{Equation 3.4}$$

3.2.5 Placement of sensors

Three functional LoRa methane detection units (D_1, D_2, and D_3) were installed in the camp on the morning of the 11th of December 2021. Their position in the camp, as mounted on 4m poles, is indicated in Figure 3.2.

Figure 3.2 Illustration of the camp, on the Welgevallen Experimental Farm, in which the experiment was conducted and the position of the three LoRa methane detection units in the camp.

The electrical box containing the battery and charge controller and the solar panel were secured at the top of the pole about 3m above the ground. A PVC pipe, 1.5m long, was fastened on the pole, 50cm above the ground. The TTGO T-Beam, CH_4 sensor and ADC module were placed at the bottom of the PVC pipe, nearest to the ground. A small hole the size of the CH_4 sensor was drilled into the PVC pipe, and the sensor was placed in the hole to be flush with the PVC pipe. The device setup can be seen in Figure 3.3.

Figure 3.3 An indication of the LoRa methane detection units installed in the camp, on the Welgevallen Experimental Farm, on 4m poles for the duration of the study.

A feeding trough was placed at each of the three poles. The three devices were connected to three LoRa gateways located on Bottelary Hill (± 10 km from the experimental location), Kanonkop (± 28 km from the experimental location) and Delaire Graff (± 8 km from the experimental location), respectively.

3.2.6 Parameters recorded

This study consisted of three defined measurement periods using the LoRa CH_4 detection units (Table 3.2). Rams were weighed on the morning of the 13th of December, and a numbered GPS collar was placed on each ram. The live weight of each sheep, sheep number and tracker number were recorded. The sheep were weighed again at the end of the trial on the morning of the 12th of January. Three LoRa CH_4 detection units were installed in period 1 and 2, while only one unit was installed in period 3 to compare the use of three versus one unit in measuring emission in sheep under grazing conditions.

Table 3.2 Dates of the three measurement periods of this trial, whether the sheep were present in the camp for that measurement period and the data recorded in that measurement period.

Measurement period	Dates	Sheep absent/present	Data recorded
Period 1	11 December 14:00 - 13 December 09:00	Absent	Background methane concentrations measured by D_1, D_2 and D_3
			Ambient conditions
Period 2	13 December 09:11 - 20 December 15:00	Present	Ram live weights before trial
			Sheep methane emissions from three devices
			Ram GPS locations
			Ambient conditions
Period 3	29 December 00:00 - 12 January 07:00	Present	Sheep methane emissions from D_2
			Ram GPS locations
			Ram weights after trial
			Ambient conditions

All three LoRa CH_4 measurement devices stopped functioning in Period 2 due to water damage. Methane concentrations measured prior to this were unaffected by the water damage. The ADC module and CH_4 sensor of D_2 were replaced between Periods 2 and 3, and Device 2, used in Period 3, was placed on the middle pole, centred in the camp.

A weather station (CR800 series, Campbell Scientific) installed on the farm was used to collect hourly meteorological data. The maximum wind speed (m/s), mean wind speed (m/s), wind direction (degree), average air temperature (ºC), relative humidity (%), total rainfall (mm), and average solar radiation (W/m^2) were recorded. The data were collected throughout the

experimental period to determine the influence of different ambient conditions on the devices' observed CH_4 measurement.

During the second and third measurement periods, ram movement was recorded using the GPS trackers (Yabby Edge LoRaWAN®, Digital Matter), as seen in Figure 3.4.

Figure 3.4 Image of the Yabby Edge LoRaWAN®, Digital Matter GPS tracker used to monitor ram movement in Periods 2 and 3.

All of the LoRa CH_4 detection unit data were sent via the TTGO T-Beam to the gateways and from the gateways to the cloud where data were logged, using The Things Network, to a Google Sheet. The GPS location data were sent to the gateways, and from the gateways, using The Things Network, to a SQL database. The data were then exported from the SQL database as a .xlsx file.

3.2.7 Statistical analysis

Outliers within each device's mV output were identified using the modified z-score:

$$Modified\ z-score = \frac{0.6745(x_i - \tilde{x})}{MAD} \qquad \textit{Equation 3.5}$$

Where x_i is a single data value, $\tilde{x}$ is the median of the dataset, and MAD is the median absolute deviation of the dataset. Data associated with modified z-scores greater than 3.5 or smaller than -3.5 were considered outliers and removed from the dataset.

Ram live weights were recorded at the start and end of the study and considered to calculate the change in weight per ram over the measurement period.

A correlation analysis approach was used to determine the effect of ambient conditions on each of the devices. Hourly device data were paired with the hourly ambient conditions for each measurement period. Pearson correlation analyses of device CH_4 concentration observation and corresponding ambient conditions were carried out to determine the relationship between ambient conditions and CH_4 readings from each of the three devices.

Descriptive statistics were obtained for each measurement period and each device within the three measurement periods. Bonferroni analysis of means was used to determine whether there was significant between-device variation by comparing the means of the three devices in the first and second measurement periods. The Bonferroni post hoc test was used as each device had

a different number of observations. One-way ANOVA and Bonferroni pairwise comparisons were used to determine, for each device, whether there was a significant difference between the CH_4 measurements recorded in the first, second and third measurement periods.

The distance of the sheep from each LoRa device was determined for each recorded GPS coordinate. The data pre-processing was done in the Geopandas python library by converting the recorded latitude and longitude values into a spatial geometry value. The data were then outputted to Geopackage format for further processing. The QGIS (Quantum Geographic Information System) 3.16.2 was then used to reproject the sensor locations as well as the sheep logger data into the relevant SACRS (South African coordinate reference systems) local projection (ESRI: 102562). The 'Distance Matrix' tool was used to calculate the linear distance from each ram logged distance to each of the three sensor sites in meters. The output was then exported as a .csv file. All recorded distances greater than 30m were removed from the dataset, as these distances exceeded the size of the camp and were considered unreliable.

The data were further organised into 1- and 5-minute time intervals. The number of animals within each time interval was counted. Within each time interval, the average CH_4 concentration measured and the average distance of the rams from the device were also determined. Analyses were performed on this data to determine whether there was a relationship between the distance of the sheep from the device and the CH_4 concentration measured. A Pearson correlation was used to determine whether there was a significant correlation between the distance of the rams from the device and the CH_4 concentration measured by each device in Periods 2 and 3.

Further analyses were done to determine the effect of the distance of the rams from the devices, when organised into categories, on the observed CH_4 measurement. The distances of the rams from each device were organised into four categories. Namely, 0-4.99 m, 5-9.99 m, 10-19.99 m and 20-30 m from the device. Descriptive statistics of the CH_4 concentration measured by each device when the distance of the sheep from the device was within one of these four distance categories were obtained. A Pearson correlation was used to determine whether the CH_4 concentration measured by each device was significantly correlated with the distance of the sheep from the device, as organised into the four categories. Both a One-way ANOVA and Bonferroni pairwise comparison were used to determine whether there was a significant difference between observed CH_4 concentrations based on the defined distance categories.

The mean CH_4 concentration measured by each device in Period 1 was used as the background CH_4 concentration and subtracted from each device's observed concentrations (ppm) in Periods 2 and 3. The resulting values were used to determine the overall daily CH_4 emissions from the sheep under grazing conditions. Descriptive statistics were obtained for the CH_4 emission values measured by each device in Periods 1 and 2.

Data were analysed using XLSTAT, SAS, and Python. Descriptive statistics, correlations, one-way ANOVA, and Bonferroni pair-wise comparisons were computed using XLSTAT. Graphs

were created using Python. The distance and CH_4 concentration datasets were joined, based on the time of observation, using a SAS work query.

3.3 Results

Nine of the ten rams used in the study were recorded to have lost weight over the month of the trial period. However, the rams were all still at a normal, healthy weight despite the weight loss.

3.3.1 Correlation between ambient conditions and device CH_4 measurements

The Pearson's correlation coefficients for the correlation between the recorded meteorological parameters and methane levels measured are presented in Table 3.3.

Table 3.3 Pearson correlation coefficients between the CH_4 concentrations measured by the three functional LoRa CH_4 detection units and ambient conditions in Period 1.

Device	Ambient condition	r
Device 1	RH (%)	0.414[a]
	Air temperature (ºC)	-0.170
	Wind speed (m/s)	-0.222
	Solar radiation (W)	-0.181
Device 2	RH (%)	0.342[a]
	Air temperature (ºC)	-0.112
	Wind speed (m/s)	-0.173
	Solar radiation (W)	-0.087
Device 3	RH (%)	0.692[a]
	Air temperature (ºC)	-0.480[a]
	Wind speed (m/s)	-0.420[a]
	Solar radiation (W)	-0.497[a]

[a] Pearson correlations with a superscript are statistically significant ($p \leq 0.05$)

In the first measurement period, the CH_4 concentrations measured by all three devices were significantly positively correlated with relative humidity (RH). While the CH_4 concentrations measured by D_3 were also significantly negatively correlated with air temperature, wind speed and solar radiation. The CH_4 concentrations measured by D_1 and D_2 also have a moderately weak correlation with air temperature, wind speed and solar radiation. However, these correlations were not significant. The correlation between RH and CH_4 measurements, for D_1, D_2 and D_3, is presented in Figure 3.5.

3.3.2 Between-device variation

Descriptive statistics of the CH_4 concentrations measured with each device in measurement periods 1 and 2 are presented in Table 3.4.

The respective LoRa devices differed in the average methane levels measured during the study ($p \leq 0.05$, Table 3.4). The mean CH_4 concentrations measured by D_2 differed from D_1 and D_3 ($p \leq 0.0001$) in measurement Period 1. While the mean CH_4 concentrations measured by all three

devices were significantly different in Period 2 ($p \leq 0.0001$). Device 2 measured an overall higher mean CH_4 concentration in both Periods 1 and 2.

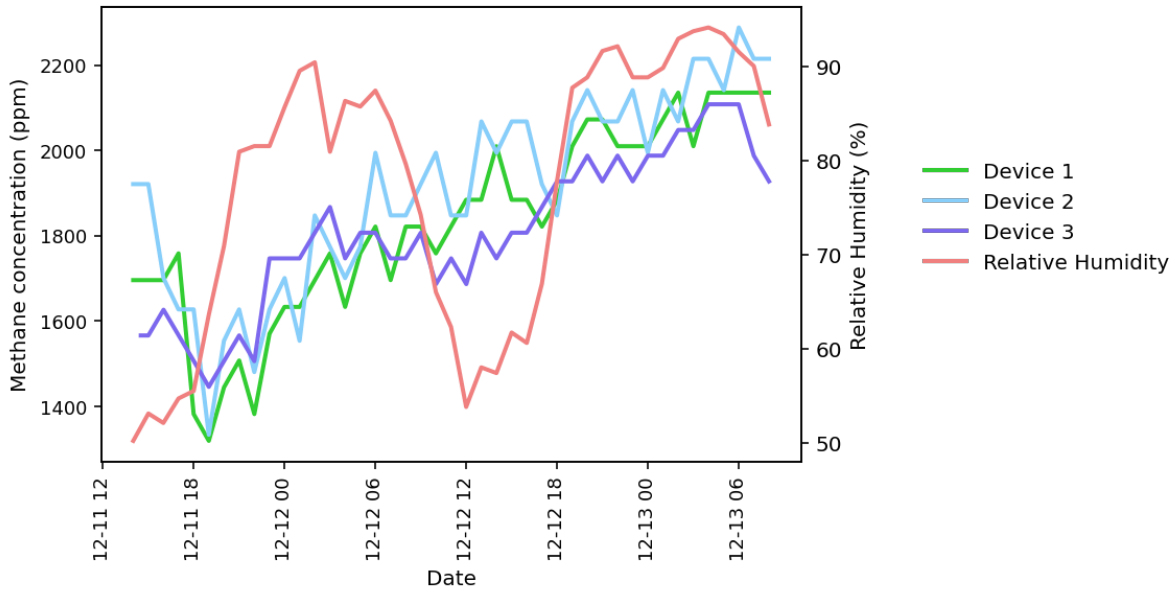

Figure 3.5 Fluctuations in the CH_4 concentrations (ppm) measured by the three functional LoRa CH_4 detection units in Period 1 and the changes in the relative humidity (%).

Table 3.4 Descriptive statistics for methane levels (mean ± SD) recorded in Period 1 (from the 11th to the 13th of December), Period 2 (from the 13th to the 20th of December), and Period 3 (from the 29th of December to the 12th of January) on the Welgevallen Experimental Farm.

Measurement period	Device	n	Methane level (ppm)	Minimum (ppm)	Maximum (ppm)	CV
Period 1	Device 1	3021	1830.6[a] ± 215.3	1256.3	2261.3	11.8
	Device 2	3081	1907.2[b] ± 214.9	1333.5	2435.5	11.3
	Device 3	3002	1822.1[a] ± 176.8	1325.30	2228.9	9.7
Period 2	Device 1	6159	2069.7[c] ± 115.9	1758.8	2386.9	5.6
	Device 2	6774	2376.1[d] ± 297.0	1700.82	2949.8	12.5
	Device 3	11337	1965.1[e] ± 636.9	1024.1	3373.5	32.4
Period 3	Device 2	19100	3326.1[f] ± 214.9	2582.5	4051.9	6.5

[a-f] Different superscripts indicate significant differences ($p \leq 0.05$)

The coefficient of variation (CV) of the CH_4 levels measured by D_1 decreased between Periods 1 and 2. In comparison, the CV of the CH_4 levels measured by D_2 increased slightly, and the CV of the CH_4 levels measured by D_3 increased substantially between Periods 1 and 2. The CV of the CH_4 levels measured by D_2 in Period 3 was half that of the device in Period 2.

The fluctuation in methane levels detected during the three measurement periods, are presented in Figure 3.6, Figure 3.7, and Figure 3.8, respectively. The three devices followed a similar pattern of CH_4 concentration detection in Period 1, steadily increasing over the measurement period and levelling out over the last few hours (Figure 3.6).

In Period 2, the three devices followed a similar pattern of CH_4 concentration detection until early morning on the 15th of December. Thereafter, the mean CH_4 levels detected by D_3 varied

greatly, increasing from about 1800 ppm in the first two days to about 2800 ppm for the next two days and then declining to about 1400 ppm for the remainder of Period 2 (Figure 3.7). These differences could result from a fault in the program, or the code installed on the LoRa TTGO crashing.

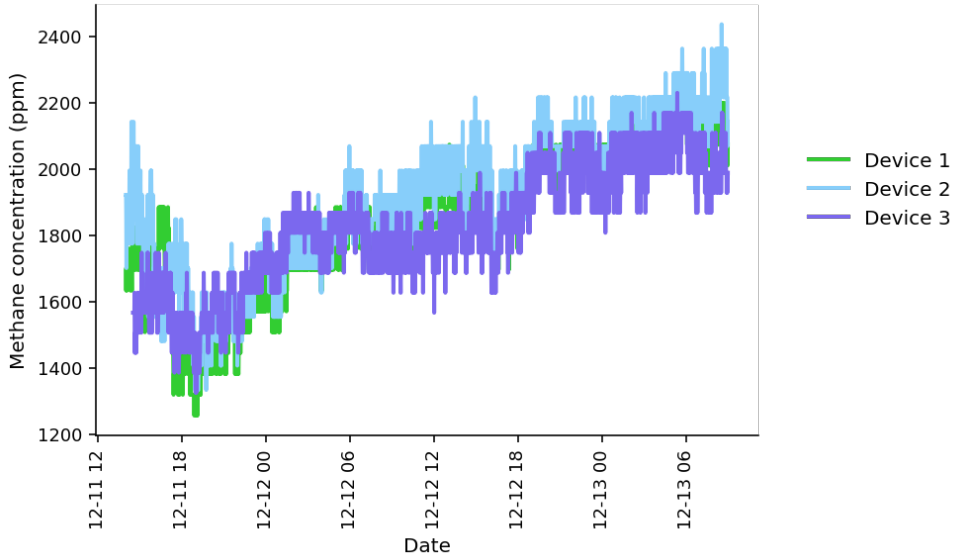

Figure 3.6 Fluctuations in the CH_4 concentrations (ppm) measured by the three functional LoRa CH_4 detection units in Period 1.

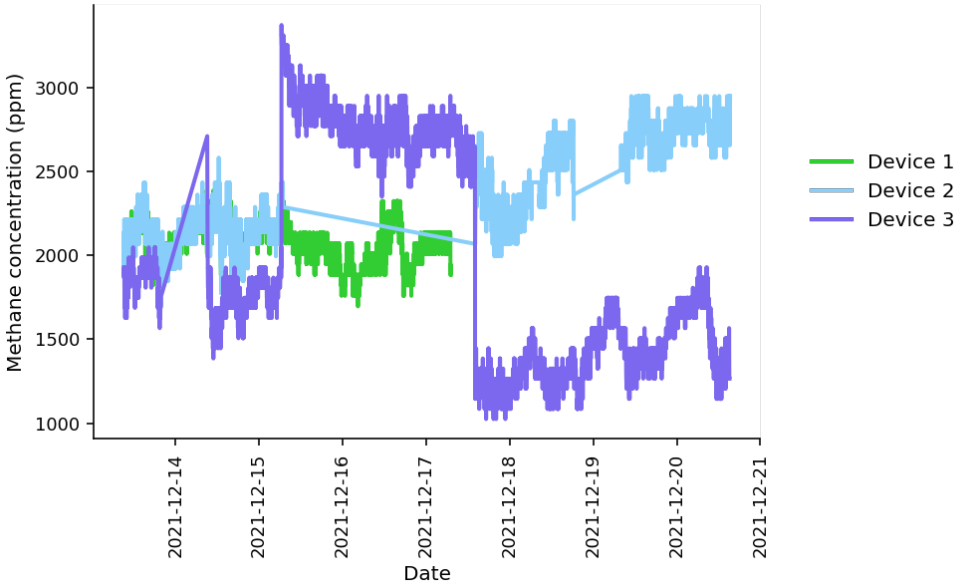

Figure 3.7 Fluctuations in the CH_4 concentrations (ppm) measured by the three functional LoRa CH_4 detection units in Period 2.

Device 1 stopped recording CH_4 levels on the 17th of December due to device malfunction caused by water damage. Despite this, the CH_4 concentrations measured by D_1 in Period 2 were much more consistent than in Period 1, following a relatively flat pattern compared to the increasing pattern in Period 1. Device 2 followed a similar pattern of CH_4 level detection seen in Period 1 (Figure 3.6). However, the device stopped recording CH_4 concentrations at two points in Period 2, seen in Figure 3.7, potentially due to a lost connection between the LoRa TTGO and the

gateways, code crashing or a fault in the program. D_2 stopped sending data simultaneously as D_3's mean CH_4 level detection jumped to 2800 ppm, and D_2 started sending data again simultaneously as D_3's mean CH_4 level detection dropped to 1400 ppm. The CH_4 levels measured by D_2 in Period 3 followed a relatively consistent pattern with values ranging from about 2600 to 3800 ppm. However, an unusual increase in the range of CH_4 levels measured was seen on the 7th and 8th of January, where the values range from 2000 to 4600 ppm (Figure 3.8). Furthermore, the mean CH_4 concentration measured by D_2 in Period 3 was much higher than the device's previous measurements in Periods 1 and 2.

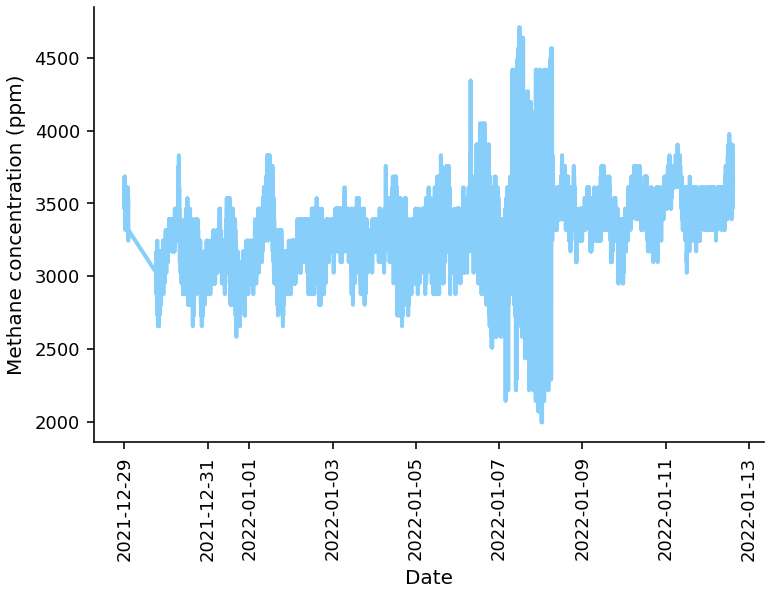

Figure 3.8 Fluctuation in the CH_4 concentrations (ppm) measured by Device 2 in Period 3.

3.3.3 Average and daily methane levels measured by the LoRa devices

The mean CH_4 concentration measured by each device differed between the measurement periods ($p \leq 0.05$, Table 3.3). The mean CH_4 levels determined in Period 2 were significantly greater, for each device, than those determined in Period 1. Furthermore, the mean CH_4 levels determined by D_2 in Period 3 were significantly greater than in Periods 1 and 2.

Descriptive statistics of the daily CH_4 emissions measured from the sheep under grazing conditions are presented in Table 3.5. The values presented in Table 3.5 were collected in periods 2 and 3 and were corrected for the ambient CH_4 concentrations measured in period 1.

Table 3.5 Descriptive statistics for the daily CH_4 levels (mean ± SD), corrected for background CH_4 concentrations, recorded in Period 2 (from the 13th to the 20th of December) and Period 3 (from the 29th of December to the 12th of January) on the Welgevallen Experimental Farm.

Device	Methane level (ppm)	Minimum (ppm)	Maximum (ppm)	CV
Device 1[a]	239.7 ± 116.8	-71.5	556.6	48.7
Device 2[a]	475.3 ± 294.7	-206.3	1042.7	62.0
Device 3[a]	158.6 ± 645.5	-798.0	1551.4	407.1
Device 2[b]	1414.2 ± 208.2	675.3	2144.7	14.7

[a] Devices in Period 2 [13 December – 20 December]
[b] Device 2 in Period 3 [29 December -12 January]

3.3.4 The effect of the distance of rams from the device on measured CH_4 concentrations

Descriptive statistics of the mean CH_4 levels determined by each device in Period 2 and by D_2 in Period 3, when the sheep were either 0-4.99, 5-9.99, 10-19.99 or 20-30 m away from the device, are presented in Table 3.6.

Table 3.6 Descriptive statistics for CH_4 levels (mean ± SD) recorded in Period 2 (from the 13th to the 20th of December) and Period 3 (from the 29th of December to the 12th of January), depending on the distance of the rams from each device, on the Welgevallen Experimental Farm.

	Distance (m)	n	Methane level (ppm)	Minimum (ppm)	Maximum (ppm)	CV
Device 1[a]	0-4.99	821	2068.7[c] ± 116.4	1758.8	2324.1	5.6
	5-9.99	2596	2062.6[c] ± 112.8	1758.8	2324.1	5.5
	10-19.99	5087	2068.5[c] ± 119.0	1758.8	2386.9	5.8
	20-30	1690	2086.9[d] ± 115.1	1758.8	2386.9	5.5
Device 2[a]	0-4.99	612	2414.6[c] ± 283.7	1774.3	2949.8	11.8
	5-9.99	1916	2403.4[c] ± 289.6	1774.3	2949.8	12.1
	10-19.99	5928	2371.6[d] ± 290.4	1700.8	2949.8	12.2
	20-30	2416	2384.3[cd] ± 310.1	1774.3	2949.8	13.0
Device 3[a]	0-4.99	180	2030.6[c] ± 671.1	1024.1	3313.3	33.1
	5-9.99	1083	1938.4[c] ± 660.9	1024.1	3253.0	34.1
	10-19.99	7403	1998.9[c] ± 656.5	1024.1	3313.3	32.9
	20-30	7246	1967.1[c] ± 630.4	1024.1	3373.5	32.0
Device 2[b]	0-4.99	507	3306.5[c] ± 217.0	2729.4	3978.4	6.6
	5-9.99	1623	3326.1[c] ± 217.0	2729.4	4051.9	6.5
	10-19.99	4713	3317.3[c] ± 206.1	2582.5	3978.4	6.2
	20-30	2573	3328.6[c] ± 204.1	2582.5	4051.9	6.1

[a] Devices in Period 2 [13 December – 20 December]
[b] Device 2 in Period 3 [29 December -12 January]
[c, d] Means within a device with different superscripts are significantly different ($p \leq 0.05$)

The CH_4 concentration measured by D_1 and the distance of the sheep from the device had a weak positive correlation (r = 0.066, $p \leq 0.0001$). Methane concentration measured by D_2 had a weak negative correlation with the distance of the sheep from the device (r = -0.034, p = 0.0004). Although not significant, a weak negative correlation was also found between CH_4 concentration measured by D_3 and the distance of the sheep from the device (r = -0.012, p = 0.137).

Furthermore, the correlation between each defined distance category and the CH_4 concentration measured by each device was weak. The CH_4 concentrations measured by D_1 when sheep were 20-30 m away from the device were significantly greater than those measured when the distances of the sheep from the device were within the other three categories. When sheep were 0-4.99 m away from D_2 or D_3, these devices recorded an overall higher mean CH_4 concentration. However, the differences in the CH_4 concentrations measured between the four distance categories were not significant for D_3. The CH_4 concentrations measured by D_2 in Period 2 when sheep were 20-30 m away from the device were significantly lower than those measured when the sheep were 0-4.99 and 5 -9.99 m away from the device. The differences in CH_4 concentrations measured between the four distance categories were not significant for D_2 in Period 3.

3.4 Discussion

3.4.1 Correlation between ambient conditions and device CH_4 measurements

The positive correlation between the CH_4 levels measured by each device and RH could result from two factors. The correlation suggests either that one of the components of the functional LoRa CH_4 detection unit is sensitive to changes in relative humidity or that the stability of CH_4 in the air increased due to increased density of the air resulting in larger CH_4 concentration measurements. The negative correlations observed between the CH_4 levels measured by each device and wind speed indicate that the wind blew the CH_4 gas emitted by the sheep away from the area where the sheep were grazing, possible causing a quicker dissipation of methane, thereby decreasing the CH_4 concentrations measured. The negative correlation of the CH_4 levels measured by the LoRa devices and solar radiation could result from the interaction of the solar radiation with the infrared light emitted or detected by the CH_4 sensors, decreasing the determined CH_4 concentration. Lastly, the negative correlation between the CH_4 levels measured by the LoRa devices and air temperature is potentially due to the effect of increased air temperatures on the functionality of the LoRa CH_4 detection unit, decreasing its performance as temperatures increase. The negative correlation observed between air temperature and the methane concentrations measured could also be an indication of reduce animal activity caused by increasing temperatures, resulting in lower metabolic rates and a subsequent decrease in CH_4 production.

3.4.2 Variation within the CH_4 concentrations measured by the LoRa devices

The CH_4 sensors used in each LoRa CH_4 detection functional unit had a 500ppm error. Therefore, the large differences between minimum and maximum CH_4 concentrations measured by each device in all three measurement periods can be ascribed to this error. Due to this large error, these devices are limited in determining sheep CH_4 emissions. Compared to individual sheep CH_4

emissions, the error recorded for the device is substantial, making it difficult to precisely distinguish between sheep contribution and error contribution to the observed changes in CH_4 concentration.

During Period 1, no sheep were present in the camp, and the LoRa devices were presumed to measure only the ambient CH_4 levels. Therefore, significant differences in the mean CH_4 concentrations measured by the LoRa CH_4 measurement devices were not expected. However, the CH_4 concentrations measured by D_2 in Period 1 were significantly greater than D_1 and D_3. These results indicate that there was between-device variation before any external factors, expected to cause differences in the CH_4 concentrations measured by each device, were present. The higher concentrations measured by D_2 were maintained through Period 2. This variation was potentially a result of the CH_4 sensor of D_2 being more susceptible to environmental noise or simply having a larger error than those of D_1 and D_3. This between-device variation could also result from the ambient conditions' effects on the components of the functional unit.

The coefficients of variation of CH_4 levels measured by the three devices were similar in Period 1. An increase in the CV values was expected when the sheep were added to the camp, with their emissions subsequently increasing the variation in CH_4 levels measured. This increase was true for D_2 and D_3 in Period 2. However, the CV of CH_4 levels measured by D_1 decreased in Period 2, and the CV of CH_4 levels measured by D_2 decreased in Period 3. The steady increase in the CH_4 concentration measured by the three devices in Period 1 may result from the device warming up, only reaching a relatively level measurement pattern 24 hours after installation. The CH_4 sensor used has a 3-minute warm-up time, but overall the functional unit could have a much longer warm-up time while it accounts for the environmental conditions. The lower variation in CH_4 levels measured by D_1 in Period 2 compared to Period 1 can be seen in the different patterns of CH_4 level observation between the two periods. The difference is potentially a result of an increase in the stability of the functional unit following its adjustments for ambient conditions made in the first 24 hours after installation. The substantial change in the CV of D_3 between periods 1 and 2 is due to the two drastic increases in CH_4 concentration measurement. Numerous factors could be responsible for the drastic increase and following decrease in CH_4 levels measured by D_3 and the lack of data from D_2 (Figure 3.7). These factors include the environmental conditions affecting the functional unit and data transfer, the code installed on the LoRa TTGO crashing, lost connection, or a fault in the program. These factors could also have caused the software code to stop operating properly or to terminate completely over that period. The unusual increase in the range of CH_4 concentrations measured by D_2 in Period 3 over the 7^{th} and 8^{th} of January could also be a result of these factors. The CH_4 levels measured by D_2 in Period 3 were significantly larger than in Periods 1 and 2. This increase can be attributed to replacing the ADC Module and CH_4 sensor between Periods 2 and 3, indicating the drastic effect the components of the LoRa CH_4 detection units have on the measured CH_4 concentrations.

Cattani *et al.* (2017) evaluated the effect of ambient conditions on LoRa reliability. They found that both temperature and humidity were significantly correlated to received signal strength

indication (RSSI) and packet reception ratio, affecting the sending, and receiving of data. They found that signal strength decreased as temperature and humidity increased. Several studies evaluating the use and scalability of LoRa technology found that when end-devices are close to each other, sending data at the same time, frequency and spreading factor, a data burst can occur, causing package collision and reduced package reception by the gateways (Aftab *et al.*, 2020; Mroue *et al.*, 2020; Saban *et al.*, 2021). Aftab *et al.* (2020) also found that multiple gateways in the same geographical region can reduce the performance of gateways. These studies indicate that it is likely that the effect of temperature and humidity on sending and receiving of data caused D_2 to have no logged data for a short time in Period 2 and the drastic increases in the CH_4 measurements of D_3.

3.4.3 Methane levels measured by the LoRa devices

The overall increase in CH_4 concentrations measured in Period 2 compared to Period 1 reveal that at least one factor in the second measurement period caused an overall increase in surrounding CH_4 concentration. However, as this study was performed in an uncontrolled environment, numerous factors may have contributed to the observed variation. These factors include the ambient conditions, the components from which the device is made up, ram enteric CH_4 emissions, and the CH_4 sensor error.

The mean daily CH_4 concentrations measured by the three devices in Period 2, ranging from 158.6 to 475.3 ppm, are relatively high. These values may result from correcting the observed CH_4 measurements using background CH_4 concentrations lower than the actual background CH_4 concentrations. The steady increase in the CH_4 concentrations measured by the three devices in the first 24 hours of period 1 potentially lowered the mean background CH_4 concentrations obtained. Therefore, the CH_4 concentrations measured when the sheep were in the camp could be slightly greater than the actual CH_4 emissions of the sheep. The high CH_4 concentration values measured by the three devices in Period 2 could also be attributed to the sensor error or the ambient conditions affecting the functionality and, subsequently, the readings obtained by each device. The CH_4 concentrations measured by D_2 in Period 3 were much larger than in Period 2. The replacement of the ADC module and CH_4 sensor of D_2 resulted in a device that was no longer calibrated. Therefore, the background concentrations used to correct the CH_4 concentrations obtained in Period 3 for atmospheric CH_4 concentrations were no longer relevant for the device, and the CH_4 levels measured by D_2 in Period 3 do not give any information on the contribution of the sheep to the increased CH_4 concentrations observed.

Various studies quantifying sheep enteric CH_4 emissions have found a range between 18 and 40 g/d (Savian *et al.*, 2014; Bhatt *et al.*, 2021; Malik *et al.*, 2022). Ricci *et al.* (2014) and Chagunda *et al.* (2013) used the LMD to determine sheep enteric CH_4 emissions. They measured between 17.3 ± 1.48 µL/L and 28.6 ± 2.73 µL/L and 68.9 ± 32.6 ppm, respectively. The CH_4

concentrations measured by each device in Period 2 were higher than those reported in previous studies. However, with further analyses and refinement of the CH_4 detection units the values recorded could be more similar to those of precious studies. Two studies analysed the CH_4 concentrations in naturally ventilated cattle buildings. Hempel *et al.* (2020) measured the CH_4 concentration in two different buildings, the first with 355 lactating cows and the second with 50 lactating cows. They recorded CH_4 concentrations of 16.4 ± 10.2 ppm and 18.8 ± 6.7 ppm inside the building, and 3.1 ± 2.1 ppm and 3.3 ± 2.1 ppm outside the building for the two groups, respectively (Hempel *et al.*, 2020). Bjerg *et al.* (2011) recorded various CH_4 concentrations, depending on the sensor's location, ranging from 3.9 ± 7.4 ppm to 21.2 ± 10.1 ppm inside the building, housing 150 cows, and 0.9 ± 1.2 ppm outside the building. These results indicate that the presence of the ruminants raises the area's overall CH_4 concentration. However, compared to these results, the CH_4 concentrations obtained in this study are much higher, potentially resulting from inaccurate background CH_4 concentrations.

3.4.4 The effect of the distance of rams from the device on measured CH_4 concentrations

The proximity of the sheep to the sensors was expected to influence the observed CH_4 concentrations measured by the device. However, contrary to the hypothesised association, no correlation was found between sheep distance and device measurements. Nonetheless, the higher average CH_4 concentrations for D_2 and D_3 in the 0-4.99m distance category could be a result of the proximity of the rams to the device, raising the CH_4 concentration surrounding the device. However, the reliability of this data was impacted by the inconsistency with which the tracker data was received. Each animal's GPS locations were sent at different time intervals. Therefore, the distances of each ram were not always known, and subsequently their distances from each device were occasionally unknown. The trackers used had a sleep mode where stationary devices sleep until movement occurs to conserve battery life (Digital Matter, Yabby Edge LoRaWAN®) which potentially caused the sporadic sending of sheep locations.

3.4.5 The design and placement of the LoRa devices for CH_4 emission measurement

When comparing the use of three versus one LoRa CH_4 detection functional unit to determine CH_4 emissions from sheep under grazing conditions, three is more optimal. Using more than one sensor is beneficial in the event that one malfunctions, as seen in this study when D_2 stopped sending data and D_3 had two big changes in CH_4 emissions measured. Sensor placement could be optimised by placing one at each corner of the camp and one in the middle for better coverage of the grazing area. Another sensor can be placed near the camp to determine the background CH_4 concentrations, enabling the system to determine sheep CH_4 emissions better.

Hammond *et al.* (2016) list the characteristics necessary for an enteric CH_4 measurement technique to be acceptable. This “ideal” method should be non-invasive, non-intrusive, cost-

effective, rapid, automated, reduce estimating errors, and enable measurement in commercial production environments (Hammond *et al.*, 2016). Through further refinement, the LoRa-CH_4 measurement device could meet the standards for the ideal measurement technology described by Hammond *et al.* (2016). It is both a non-invasive and non-intrusive method. It is cost-effective compared to other methods available, and it takes automatic, real-time measurements at a time interval determined by the user. It requires no human intervention and no technical skills and training to operate, thereby removing variation caused by human operation. Lastly, it does not disturb the animal's normal activity and could be used under certain commercial production conditions, limited to use in smaller, controlled camps. Further advantages of this technique are its ability to add numerous sensors to the device, enabling the measurement of various parameters simultaneously. It has low labour requirements and costs, with a quick and simple installation and easy maintenance. This system can easily be incorporated into a smart farming system along with other technologies to assist producers in decision-making and help the shift toward automation, keeping the farm profitable through increased production efficiency and improved resource management (Charania & Li, 2020). However, this system requires refinement through determining the effects of ambient conditions and the distance of the rams from the device on the CH_4 concentrations measured and creating a model to account for these effects. Furthermore, determining the optimal placement and design of the device to reduce the chance of the devices malfunctioning or losing connection with the gateways and resulting in a loss of data. The successful refinement of this system could benefit producers and the industry in sustainability by decreasing the environmental impact of production, improving production efficiency and traceability, and decreasing production costs.

3.5 Conclusion

The results of this study reveal that the use of more than one measurement device is necessary in the case that one malfunctions or loses connection with the gateway. It also highlights the importance of the placement of the sensors ensuring that they are not too close, to avoid a data burst and package collision, but also to ensure their placement accounts for the entire experimental area. In its current form, the device is not able to measure sheep enteric emissions accurately, as well as quantify the contribution of other factors that may have affected the CH_4 concentrations measured. However, with further development and refinement, the LoRa CH_4 detection device potentially will be able to determine enteric CH_4 emissions in sheep under grazing conditions.

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Chapter 4

A comparison of LoRa and LMD approaches to determine diurnal methane emissions in sheep under grazing conditions

Abstract

This study compares the laser methane detector (LMD) and LoRa (Long Range) technology in combination with an infrared methane sensor, to determine the diurnal methane emissions in sheep under grazing conditions. Ten intact Dohne Merino rams were placed in a small camp on Welgevallen Experimental Farm, Stellenbosch University, South Africa. The sheep were allowed to graze freely on kikuyu pasture, and were fed oat hay supplement at 07:00 each morning. Three LoRa devices were installed in the camp at equidistance from each other and the fenced boundaries of the camp. Methane emissions from the rams were recorded using the LMD and LoRa techniques, respectively, for seven consecutive days in December 2021. The LMD was used each day, either in the morning (05:00-06:00), mid-day (12:00-13:00) or afternoon (17:00-18:00), for four minutes per ram. The LoRa devices collected readings every 50 seconds throughout the seven days. Using linear regression, the comparison of these techniques showed a weak relationship between the methane concentration measured with the LMD and LoRa devices. However, the average methane level measured by the LMD (13.9 ppm) and LoRa Device 3 (11.9 ppm) were not significantly different. Furthermore, the LMD and Device 3 values were slightly lower than those measured in previous studies. The mean methane concentration measured by Device 1 (24.0 ppm) and Device 2 (52.9 ppm) were similar to previous studies, but significantly different from that measured by the LMD in this study. The results indicate that the LoRa technique has the potential to be used as an enteric methane measurement technique for animals under grazing conditions, limited to use in smaller, controlled camps.

Keywords: Laser methane detector, CH_4, LoRa, methane measurement, sheep, grazing

4.1 Introduction

The significant contribution of ruminants to global methane (CH_4) emissions and their resultant contribution to climate change make them a relevant component of climate change mitigation. Ruminants produce large amounts of CH_4 through fibre digestion as a by-product of enteric fermentation in the rumen (Brouček, 2015). They are responsible for approximately 24-28% of global anthropogenic CH_4 emissions (Yusuf *et al.*, 2012; Glasson *et al.*, 2022). As CH_4 is a potent GHG with a short atmospheric lifespan, its reduction could be an effective strategy for the short-term slowing of climate change (Cain *et al.*, 2019; Glasson *et al.*, 2022).

Approximately 50% of the South African domesticated ruminant population is comprised of sheep (Meissner *et al.*, 2013). The small ruminant population of South Africa contributes an estimated 15.6% of the country's total livestock CH_4 emissions, with sheep said to be responsible for 80% of these emissions (du Toit *et al.*, 2013). Sheep production in South Africa is a vital component of sustainable South African agriculture, due to the fact that sheep can convert fibre resources into products suitable for human consumption as well as use land unsuitable for intensive agricultural production. Sheep can thus be considered as models for the development of CH_4 measurement techniques and mitigation strategies, due to their contribution to global CH_4 emissions (Marino *et al.*, 2016). Furthermore, there is little knowledge regarding enteric CH_4 emissions by ruminants in extensive production conditions (Pérez-Barbería *et al.*, 2020).

The efficient reduction of the ruminant industry's enteric CH_4 emissions requires national GHG inventories, against which mitigation targets can be set and measured, and the verification of on-farm mitigation techniques (Velazco *et al.*, 2016). To validate national GHG inventories and verify mitigation strategies, reliable and low-cost enteric CH_4 measurement techniques that can quantify emissions from large numbers of individual animals in a wide range of production environments are required (Velazco *et al.*, 2016; Huhtanen *et al.*, 2019). Coppa *et al.* (2021) state the importance of accurate techniques to quantify enteric CH_4 emissions to mitigate CH_4 emissions from ruminants significantly and sustainably.

The respiration chamber (RC) and Sulphur Hexafluoride (SF_6) tracer technique are the most commonly used measurement techniques for enteric CH_4 emission quantification. Currently, the respiration chamber is the primary source of data on which ruminant emission estimates are based (Johnson & Johnson, 1995; Gardiner *et al.*, 2015). These two techniques, however, can only be utilised at the controlled experimental level, while application at a commercial, applied research level is not feasible (Chagunda *et al.*, 2013). The RC confines animals for the duration of the measurement period, restricting their movement, diet selection, and their interaction with peers and the environment (Pinares-Patiño *et al.*, 2011). Consequently, the emission values obtained using the RC cannot be used to obtain reliable estimates for grazing animals (Johnson & Johnson, 1995). The SF_6 tracer technique was designed for CH_4 emission measurement from individual free-ranging animals. Both these techniques are expensive, labour intensive and therefore restricted in their application for a high throughput of animals.

Chagunda *et al.* (2009) identified the laser methane detector (LMD) as an alternative measurement technique that could meet the need for an inexpensive, practical measurement technique which can be used in applied research on commercial farms to quantify ruminant enteric CH_4 emissions. Due to its remote nature and portability, this technique could have the capacity to screen a large number of animals for genetic selection and analyse mitigation techniques (Sorg *et al.*, 2018). The LMD is a portable, hand-held device that uses infrared absorption spectroscopy to measure the CH_4 concentration between the device and the target (Tokyo Gas Engineering, 2006). The device is held 1-3m from the animal, with the laser pointed at the animal's mouth and nose

area for about 2 to 5 continuous minutes. Repeated measurements are collected every 0.5s in parts per million per meter (ppm-m). The LMD measures the animal's respiratory cycle represented as a series of peaks (exhalation or eructation) and troughs (inhalation) (Ricci *et al.*, 2014).

The agricultural industry is undergoing a transformation toward automated and data-driven agriculture by integrating various information and communication technologies (Boursianis *et al.*, 2020). This transformation, known as Farming 4.0, is the agricultural industry's response to the fourth industrial revolution (4IR). The Internet of Things (IoT) is one of the most popular technologies driving the 4IR. LoRa (Long Range), a low-power, wide-area technology designed for IoT communication, has the potential to be used in extensive farming applications (Semtech, Co. Camarillo, California, United States). It can be used in areas with no network coverage and transmits small amounts of data over distances of about 5km in urban and 20km in rural areas (Miles *et al.*, 2020). LoRa is the physical layer of the protocol, and LoRaWAN (Long Rand Wide Area Network, LoRa Alliance) is the communication protocol that consists of the application layer and media access control (MAC) layer. It uses a proprietary chirp spread spectrum modulation that enables low-power, long-range data transmission over the unlicensed ISM band. LoRa uses the 868MHz frequency band in South Africa. Combined with a CH_4 sensor, this technology could measure enteric CH_4 emissions in sheep under extensive grazing conditions. This technique consists of wireless sensor nodes placed in the field that transmit data every 50s. The LoRa node is connected to a CH_4 sensor and runs on batteries charged by solar power and, depending on the data transmission rate, is expected to last several years. In the LoRaWAN protocol, nodes are arranged in a star-of-star topology, with each node having bidirectional communication with multiple gateways. The LoRa nodes send observed CH_4 emission information to the LoRa gateway, which sends the data to the network server. The network server removes all duplicate data, and The Things Network logs the remaining data to a Google Sheet.

This study aims to compare the LMD, using the operating protocol described in van Wyngaard (2018), and the LoRa-CH_4 measurement techniques for determining diurnal CH_4 emissions from sheep under South African grazing conditions.

4.2 Materials and methods

This study was approved by the Research Ethics Committee: Animal Care and Use of Stellenbosch University (ACU-2021-11233).

4.2.1 Experimental location

This study was completed on the Welgevallen Experimental Farm (-33.94458, 18.86628) of Stellenbosch University, located in the Western Cape Province of South Africa. The trial ran in the summer from the 11th to the 23rd of December 2021. The average historical temperature during this period is 20°C, with a minimum of 14.6°C and a maximum of 26°C (Climate-data.org, 2021). The

average historical rainfall is 27mm, and the average historical humidity is 61% (Climate-data.org, 2021).

4.2.2 Experimental animal and husbandry

Ten intact Dohne Merino rams, with an average body weight of 96.6 ± 6.1 kg, were randomly selected from the Welgevallen Research Flock, and were used for the duration of this study. The rams were three and a half years old at the time of the study. All rams had *ad libitum* access to fresh water and kikuyu (*Pennisetum clandestinum*) grazing for the entire experimental period, and were fed oat hay supplement at 07:00 each morning. The husbandry of the rams was according to standard farm protocols, and rams were checked daily during the study duration to monitor behaviour and wellbeing.

4.2.3 Methane measurement

In this experiment, CH_4 emissions were measured from rams using the proprietary hand-held LMD and LoRa technology in combination with an infrared CH_4 sensor to compare these devices in determining diurnal CH_4 emissions from sheep under grazing conditions. Methane measurements were recorded in two periods (Table 4.1).

Table 4.1 Dates of the two measurement periods of this trial, whether the sheep were present in the camp for that measurement period and the data recorded in that measurement period.

Measurement period	Dates	Sheep absent/present	Data recorded
Period 1	11 December 14:00 - 13 December 09:00	Absent	Background methane concentrations measured by LoRa D_1, D_2 and D_3
			Ambient conditions
Period 2	13 December 09:11 - 23 December 13:00	Present	Ram live weights before trial
			Sheep methane emissions measured by LoRa D_1, D_2 and D_3 (13 to 20 December)
			Ram GPS locations
			Sheep methane emissions measured daily using the LMD (14 to 23 December)
			Ambient conditions

4.2.3.1 LoRa technology

The LoRa measurement devices were used for CH_4 measurement from the 11th to the 20th of December 2021. The functional unit of the LoRa technology technique consisted of a TTGO T-Beam ESP32 LoRa 868MHz, Infrared CH_4 Sensor NDIR Gas Sensor MH-440D, DFRobot ADS1115 16-BIT ADC Module, solar panel, battery, charge controller, and electrical cover box. The infrared CH_4 sensor detects the concentration of CH_4 in the air using the non-dispersive

infrared (NDIR) theory (Zhengzhou Winsen Electronics Technology Co., Ltd., 2022). The TTGO T-Beam has a built-in LoRa chip that operates at 868MHz, which enables it to send the data collected by the CH_4 sensor to the LoRa gateways. Visual Studio (VS) Code was used as the integrated development environment (IDE) with the platformio plug-in to write the code and upload it to the TTGO, enabling it to send and receive data. The sensor used had a 500 ppm error, as none were available with the sensitivity required to measure sheep CH_4 emissions and the more sensitive ones were more susceptible to environmental noise.

Three complete functional units (namely device 1 (D_1), device 2 (D_2) and device 3 (D_3)) were assembled. Device 2 was calibrated using a 2-point calibration method as described in Chapter 3, section 3.2.4. Devices 1 and 3 were calibrated during the first measurement period when all three devices were set up in the field to measure ambient CH_4 concentrations, described in Chapter 3, section 3.2.4. This calibration method assumed that the three sensors all measure the same atmospheric CH_4 concentration (ppm) at a specific time when no sheep are on the pasture. The calibration of the three devices used the following equation:

$$PPM = (value - in_{min}) \times \frac{(out_{max} - out_{min})}{(in_{max} - in_{min})} + out_{min} \qquad \textit{Equation 4.1}$$

Where PPM is the calculated CH_4 concentration in parts per million (ppm), value is the mV output of the CH_4 sensor, in_{min} is the minimum mV of the device (theoretical = 400 mV), out_{max} is the maximum ppm output (theoretical = 100000 ppm), out_{min} is the minimum ppm output (theoretical = 0 ppm), in_{max} is the maximum mV output of the device (theoretical = 2000 mV).

On the morning of the 11th of December, following the calibration of D_2, the three LoRa functional units were installed, as described in Chapter 3, section 3.2.5, on one of three poles set up in the camp, at equidistance from one another and the fenced boundaries of the camp, illustrated in Figure 3.2, Chapter 3. A feeding trough was placed at each of the three poles. Each LoRa device was set to take and send measurements every 50s.

The three devices were connected to three LoRa gateways located on Bottelary Hill (± 10 km from the experimental location), Kanonkop (± 28 km from the experimental location) and Delaire Graff (± 8 km from the experimental location). Rams were weighed on the morning of the 13th of December, and a numbered GPS collar was placed on each sheep. Ram movement was recorded using GPS trackers (Yabby Edge LoRaWAN®, Digital Matter). All LoRa CH_4 detection unit data was sent via the TTGO T-Beam to the gateways and from the gateways to the cloud, where data was logged, using The Things Network, to a Google Sheet. The GPS location data was sent to the gateways and from the gateways, using The Things Network, to a SQL database. The data was exported from the SQL database as a .xlsx file. The mean CH_4 concentration measured by each device in the first measurement period was used as the background CH_4 concentration and subtracted from each device's observed concentrations (ppm) in the second measurement period.

4.2.3.2 The laser methane detector

The hand-held Laser Methane Mini™ (model SA3C2A; Tokyo Gas Engineering Solutions, Co. Ltd., Otaku, Tokyo, Japan) was used to measure CH_4 concentration in parts-per-million-meter (ppm-m) in the expired air of Dohne Merino rams. A modified version of the operating protocol described in van Wyngaard (2018) was used. Measurements were taken from each ram once a day for ten consecutive days, from the 14th to the 23rd of December 2021. The time of the measurement alternated between the morning (05:00-06:00), mid-day (12:00-13:00) and late afternoon (17:00-18:00) as described by van Wyngaard (2018). This measurement protocol was used to prevent bias caused by the diurnal fluctuations in enteric CH_4 production in grazing animals and to reduce animal variation (van Wyngaard, 2018). Measurements from each sheep were taken for 4 minutes daily, with the LMD set to measure the CH_4 concentration every 0.5s. The LMD was held 1m from the animal's mouth and nose. Therefore, values did not need to be corrected for distance before further analysis (Roessler *et al.*, 2018). All measurements were taken while the sheep were on pasture, without restraining the animal, to ensure the animal's behaviour was not disturbed during the sampling period. Their position (lying or standing) was recorded for each measurement period. The LMD screen was video recorded during each sampling period to capture the LMD CH_4 measurement output. Ten emission profiles were obtained per ram, with approximately 480 observations per daily sampling period. Due to the high sensitivity of the LMD, the minimum CH_4 concentration recorded in each sampling period was set as the background CH_4 concentration. This value was subtracted from the rest of the sample to adjust for the background CH_4 concentration (Ricci *et al.*, 2014).

4.2.4 Data analysis

Errors in laser beam reflectance were automatically identified by the LMD and manually removed from the dataset. All data obtained from the LMD when the laser moved off the mouth and nose area of the sheep due to sheep movement was manually removed. Outliers in the raw LoRa measurement dataset were identified using the modified z-score.

Raw data (mV) of LoRa D_1 and D_3 were converted to CH_4 concentration (ppm) using the following equation, where in_{min} was calculated in the first measurement period:

$$PPMcal = (Value - in_{min}) \times \frac{(100000-0)}{(2000-in_{min})} + 0 \qquad \textit{Equation 4.2}$$

Raw data (mV) of LoRa D_2 were converted to CH_4 concentration (ppm) using the following equation:

$$PPMcal = (Value - 405) \times \frac{(3758-11)}{(456-405)} + 11 \qquad \textit{Equation 4.3}$$

Where PPM_{cal} in both Equation 4.2 and Equation 4.3 is the CH_4 concentration corrected based on the calibration and in_{min} in Equation 4.2 is the minimum mV output of the sensor, which differs for each device.

The converted CH_4 concentration (ppm), corrected for background CH_4 concentration, was divided by the number of sheep to analyse data on a per sheep basis (ppm per sheep). The distance of the sheep from the LoRa device was determined for each recorded GPS coordinate. The data pre-processing was done in the Geopandas python library and outputted to Geopackage format for further processing. Further processing was completed in the QGIS (Quantum Geographic Information System) 3.16.2 as described in Chapter 3, section 3.2.7. All recorded distances greater than 30m were removed from the dataset, as these distances exceeded the size of the camp and were considered unreliable. Based on time, the distance and converted LoRa CH_4 concentration (ppm) datasets were joined using a SAS work query. The data were then grouped according to tracker number, time of day (morning, mid-day, afternoon, and night), date and distance (0-4.99 m, 5-9.99 m, 10-19.99 m and 20-30 m). Data were also grouped into the time periods that correlate with those in which the LMD measurements were taken, i.e. (05:00-10:59), mid-day (11:00-14:59), and afternoon (15:00-19:59), and a fourth period was added (evening) to account for measurements taken between 20:00 and 04:59.

As the LMD emission measurements were taken at a 1m distance from the ram mouth and nose area, no correction needed to be made for the observed CH_4 concentrations to account for the distance between the ram and LMD. Each emission profile obtained each day for each ram was plotted on a graph. For each sampling period, a threshold was calculated and plotted on the same graph to distinguish between respired and eructated CH_4 emissions (Sorg *et al.*, 2018). The following calculation was used to determine the threshold Sorg *et al.* (2018):

$$T = Q3 + (1.5\ x\ IQR) \qquad \textit{Equation 4.4}$$

Where IQR is the interquartile range (IQR = Q3 - Q1) and Q1 and Q3 are the first and third quartile of the distribution of all the CH_4 concentration (ppm-m) values in a single profile. Three phenotypes were produced as described by Sorg *et al.* (2018). Namely, the MEAN, P_MEAN and ERP_MEAN that were used for further analyses. The MEAN is the arithmetic mean of all the values recorded in one sampling period. P_MEAN is the arithmetic mean of all the peaks of the sampling period. ERP_MEAN is the arithmetic mean of all eructation peaks, separated from respiration peaks using the boxplot method described in Sorg *et al.* (2018). Further analyses used only the peak values as done by Chagunda *et al.* (2009) and Ricci *et al.* (2014). The spot-sampled CH_4 concentrations observed for each sheep were converted to g/d (grams/day) to determine the daily CH_4 production per sheep, using the following equation as specified by van Wyngaard (2018):

$$Y = d\ x\ (5.76\ x\ m) \qquad \textit{Equation 4.5}$$

Where Y is the CH_4 production (g/d), d is 0.31 (if the animal is lying down) or 0.38 (if the animal is standing), and m is the average CH_4 concentration (ppm-m).

Descriptive statistics were calculated for both the LMD and LoRa device datasets to determine the distribution of the data. Pearson correlation was used to determine the correlation between the distance of the ram from the LoRa device and the observed CH_4 concentrations.

The CH_4 concentration (ppm-m) emitted by the rams measured by the LMD was compared with the average daily CH_4 concentration (ppm per sheep) measured by the LoRa devices. A linear regression analysis with the observed CH_4 concentration of the LMD and LoRa devices as the response variable and date as the constant variable was used to determine the strength of the linear relationship between the LMD and LoRa devices. A two-sample t-test was performed to compare the CH_4 concentration measured by the LMD and the three LoRa devices. One-way ANOVA and Bonferroni pairwise comparisons were used to determine whether there was a significant difference in the CH_4 concentration observed by the LMD and LoRa devices at different times of the day. The measurements were divided into morning, mid-day, and afternoon for the LMD, correlating to the specified sampling periods. The CH_4 measurements of the LoRa devices were divided into morning, mid-day, afternoon, and evening. The LMD and LoRa data were only compared with respects to their ppm readings as there is not yet a method available that can be used to convert the LoRa measurements in ppm to g/day as done with the LMD measurements.

All data were analysed using XLSTAT.

4.3 Results

No correlation was found between the distance of the rams from the LoRa devices and the CH_4 concentrations measured by each device. In addition, no correlation was observed between the distance categories and the subsequent CH_4 measurement output.

The LMD-measured CH_4 emission profile of one randomly selected ram can be seen in Figure 4.1. This figure represents a typical CH_4 dataset obtained during the 4-minute sampling period. The threshold indicated by the horizontal dashed line separates the respiratory and eructation CH_4 emissions. Each peak below the threshold represents an exhalation of CH_4, and each peak above the threshold represents eructation of CH_4.

The MEAN, P_MEAN, and ERP_MEAN phenotypes calculated from the profiles of the CH_4 concentration (ppm-m) measured in the expired air of the rams are reported in Table 4.2. The average number of spot samples per sampling period and the maximum respired and eructated CH_4 concentration, measured by the LMD, are also presented in Table 4.2. Further analyses were done using only the peak values, as they represent the respiratory or eructation events that make up the respiratory tidal cycle (Chagunda *et al.*, 2013).

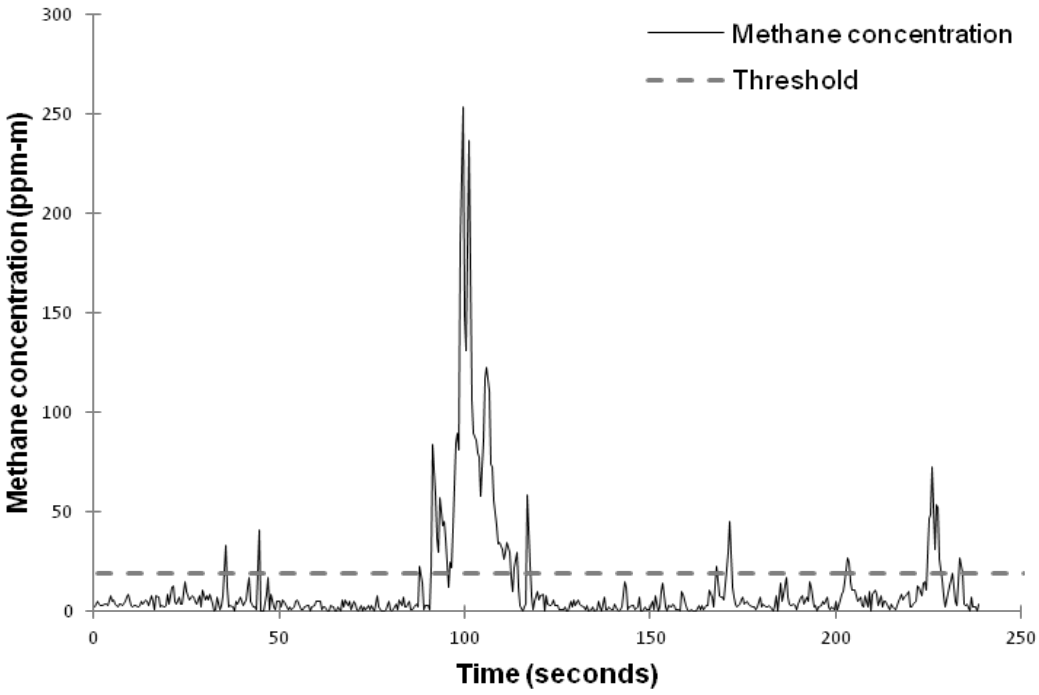

Figure 4.1 The profile of the CH_4 concentration (ppm-m) in the breath of a randomly selected ram measured with the laser methane detector (LMD). The threshold calculated as T = Q3 + (1.5 x IQR).

Table 4.2 The average number of spot-samples obtained per sampling period, the mean ± standard deviation (ppm-m) of the phenotypes, described by Sorg *et al.* (2018), calculated from the 4 min sampling period of the CH_4 concentration measured as spot-samples with the laser methane detector (LMD), as well as the maximum respired and eructated CH_4 concentration (ppm-m) from Dohne Merino rams on Kikuyu pasture.

Parameter	**Methane concentration (mean ± SD)**
Spot samples per sampling period	470.8 ± 31.5
MEAN[a]	9.3 ± 3.8
P_MEAN[b]	13.9 ± 5.2
ERP_MEAN[c]	43.0 ± 20.4
Maximum respired CH_4	50 ± 9.6
Maximum eructated CH_4	468.0 ± 71.4

[a] The arithmetic mean (ppm-m) of all the CH_4 values measured by the LMD
[b] The arithmetic mean (ppm-m) of all the peak CH_4 values
[c] The arithmetic mean (ppm-m) of all the eructation peak CH_4 values

Descriptive statistics for the CH_4 concentrations measured by the three LoRa CH_4 measurement devices throughout the second measurement period (13th to 20th December) are listed in Table 4.3. The values are presented in ppm per sheep. These values were obtained by subtracting each device's measured background concentration from the observed value and dividing the total CH_4 concentration by ten to determine the CH_4 production per sheep. Descriptive

statistics of the LMD-measured CH_4 concentrations from the 14th to the 23rd of December are also presented in Table 4.3.

Table 4.3 Descriptive statistics for the CH_4 concentration (ppm/sheep) measured by the three LoRa devices from the 13th of December to the 20th of December, and the LMD measured CH_4 concentration (ppm-m) from the 14th of December to the 23rd of December.

	Mean ± SD	Minimum	Maximum	Skewness	Kurtosis	CV[d]	CV[e]
D_1[a]	24.0 ± 0.3	15.5	31.5	-0.162	-0.437	1.37	21.0
D_2[a]	52.9 ± 0.8	22.2	89.5	0.271	-1.291	1.43	47.3
D_3[a]	11.9 ± 1.0	-51.8[f]	90.4	0.365	-1.463	8.41	441.2
LMD[b]	13.9 ± 2.1	5.4	29.2	0.908	0.406	15.0	37.5
LMD[c]	27.6 ± 3.6	9.7	54.3	0.592	-0.089	13.0	36.6

[a] mean, standard deviation, minimum and maximum measured as parts per million (ppm) per sheep (obtained by dividing the total measured CH_4 concentration by the number of sheep)
[b] mean, standard deviation, minimum and maximum measured as parts per million per meter (ppm-m)
[c] mean, standard deviation, minimum and maximum in g/d
[d] between-ram coefficient of variation, as a percentage (%)
[e] coefficient of variation (%) of CH_4 measurements between-days and –rams
[f] The negative value is caused by the measured CH_4 concentration being lower than the recorded background CH_4 concentration

The LoRa devices D_1 and D_2 measured numerically higher mean CH_4 concentration values than the LMD. In contrast, D_3 measured a mean CH_4 concentration slightly lower than the LMD. The LMD had higher variation than the LoRa devices in its measurements, shown by the between-ram coefficient of variation. Table 4.3 also presents the coefficients of variation of the LMD and three LoRa devices, including between-day and between-ram variations. Device 3 (CV = 441.2%) had a much higher variation in its CH_4 measurements than D_1, D_2 and the LMD.

This trial intended to collect CH_4 concentration measurements from the LMD and all three LoRa devices for seven consecutive days. However, only three non-consecutive days of the seven days had observations recorded from all devices. Device 1 of the LoRa devices got water damage and stopped working the morning of the 17th of December. The water damage was a result of the PVC pipe (in which the ADC module and CH_4 sensor was placed) not being completely waterproof. No data was received from D_2 from 06:50 on the 15th of December until 14:06 on the 17th of December. Device 3 also logged no data from 19:46 on the 13th of December to 09:07 on the 14th of December. The mean CH_4 concentrations measured by D_2 increased significantly after the device stopped sending data. The device's mean CH_4 concentration measured before the device stopped sending data was 26.0 ppm, and after the period when the device stopped sending data, the average CH_4 concentration was 66.3 ppm. When observing only the results obtained in the first two days before the device stopped sending data, the measured CH_4 concentration agrees with that of D_1 (24.0 ppm). Therefore, the high average observed for D_2 is a result of this substantial increase in measured CH_4 concentration, indicating a potential fault in the TTGO T-Beam or ADC module.

No linear relationship was observed between the CH_4 concentrations measured using the LMD and three LoRa devices. The relationship between the CH_4 concentrations measured by the LMD and LoRa devices can be seen in Figures 4.2 to 4.4.

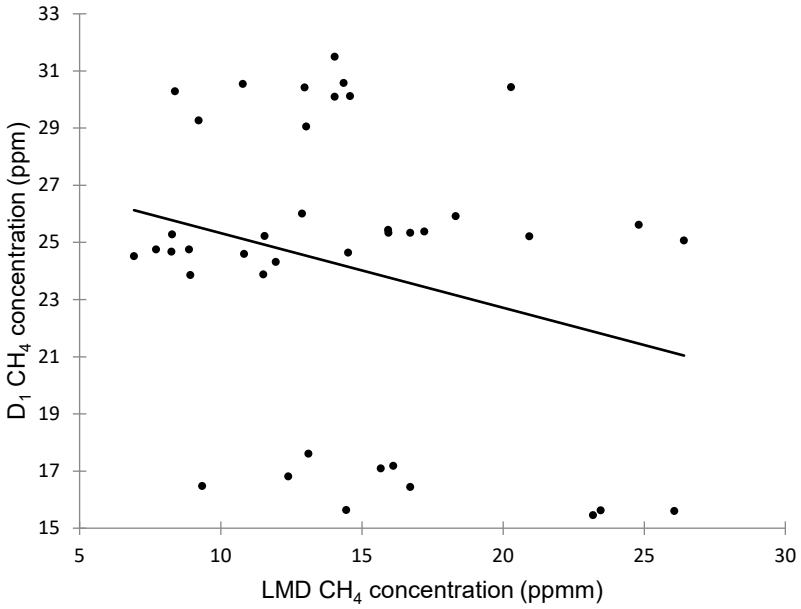

Figure 4.2 The relationship between CH_4 concentrations measured using the LMD and LoRa Device 1.

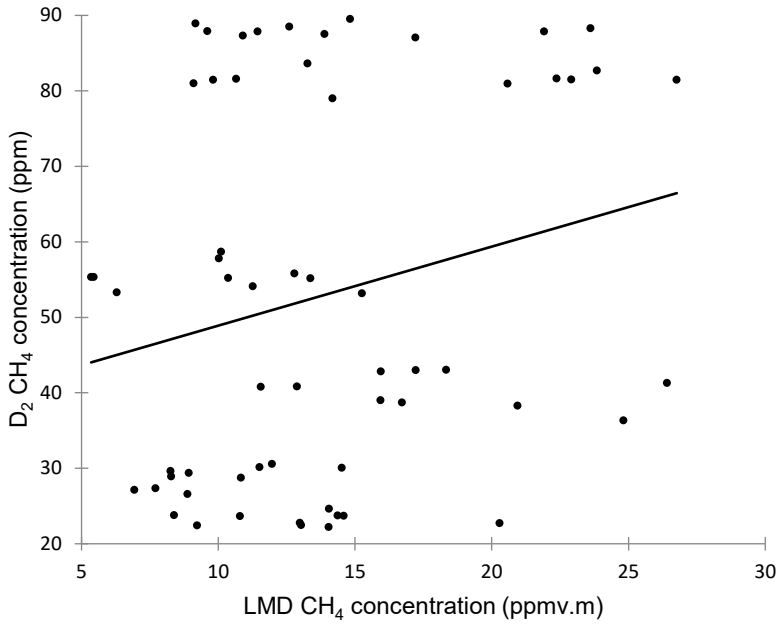

Figure 4.3 The relationship between CH_4 concentrations measured using the LMD and LoRa Device 2.

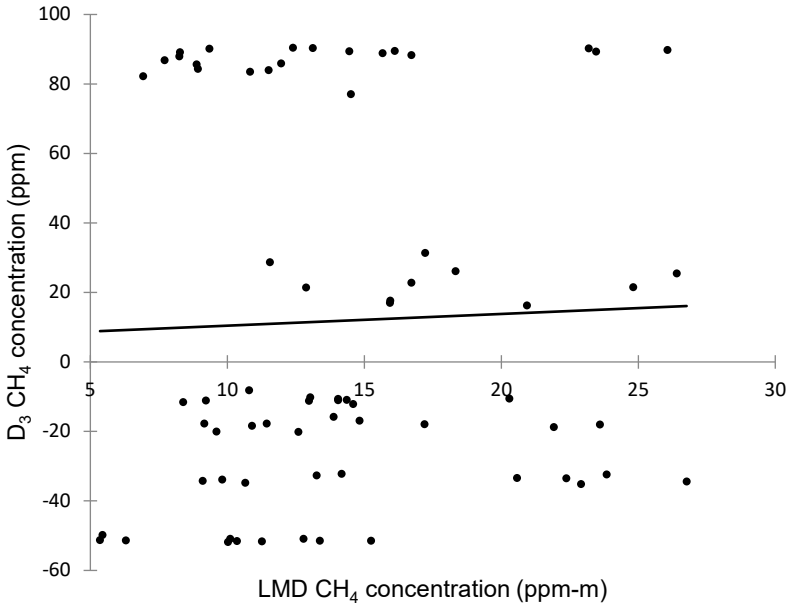

Figure 4.4 The relationship between CH_4 concentrations measured using the LMD and LoRa Device 3.

The mean CH_4 concentrations measured by the LMD were significantly different from those measured by D_1 ($p \leq 0.0001$) and D_2 ($p \leq 0.0001$). In contrast, a significant difference was not found between the mean CH_4 concentrations measured by the LMD and D_3 ($p = 0.704$).

The mean CH_4 concentrations measured by the LMD and LoRa devices differed in terms of the time of the day the measurements were obtained. The mean CH_4 concentrations measured by the LMD in the morning and mid-day higher than those measured in the afternoon ($p \leq 0.0001$ and $p = 0.0002$, respectively, Table 4.4). The mean CH_4 concentrations measured by D_1 and D_2 in the four times of day periods specified for the LoRa measurements were significantly different from each other, where mid-day CH_4 concentration readings were the highest followed by the morning, afternoon and evening readings. For D_3 the mean CH_4 concentration measured in the morning measurement period was significantly higher than the mid-day, afternoon and evening measurements. The CH_4 concentrations measured by D_3 in the mid-day and evening measurements did not differ, and the afternoon mean CH_4 measurement was significantly lower than the morning, mid-day and evening measurements.

The means and standard deviations of the CH_4 concentration measured by the LMD and LoRa devices for the different times of day are presented in Table 4.4. The morning and mid-day periods had higher CH_4 concentration measurements than the afternoon and evening in all three LoRa devices and the LMD. A visualisation of the differences in the CH_4 concentrations measured by the LMD and three LoRa devices at these different times of the day is presented in Figure 4.5.

Table 4.4 The CH_4 concentration (mean ± SD) measured by the LMD and three LoRa devices in the morning, mid-day, afternoon and evening.

Time of day	LMD[a]	D_1[b]	D_2[b]	D_3[b]
Morning	17.2[c] ± 5.9	26.2[c] ± 11.5	50.6[c] ± 27.7	30.6[c] ± 63.9
Mid-day	15.3[c] ± 4.8	30.3[d] ± 10.5	58.2[d] ± 27.6	13.0[d] ± 65.2
Afternoon	9.9[d] ± 2.9	21.9[e] ± 12.4	46.3[e] ± 29.6	6.2[e] ± 65.4
Evening	N/A	20.8[f] ± 10.4	39.9[f] ± 29.3	11.9[d] ± 61.8

[a] measurements in parts per million per meter (ppm-m)
[b] measurements in parts per million (ppm)
[c-e] Means with different superscripts within each CH_4 measurement device are significantly different ($p \leq 0.05$)

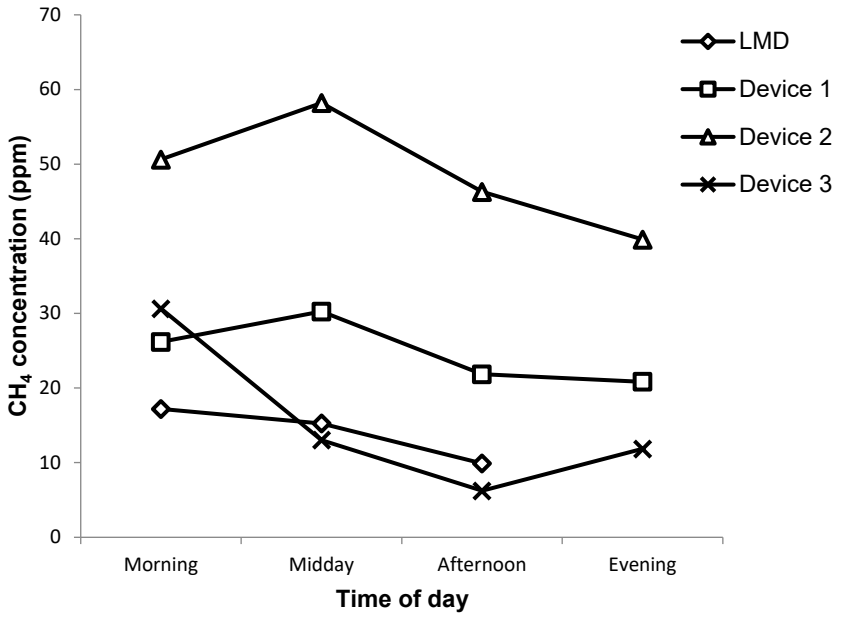

Figure 4.5 A visualisation of the changes in observed CH_4 concentration measured by the LMD and LoRa devices at different times of the day.

4.4 Discussion

The aim of the study was to compare the LMD and LoRa CH_4 measurement techniques in determining diurnal CH_4 emissions from sheep under grazing conditions.

The proximity of the sheep to the LoRa devices was expected to influence the CH_4 concentration measured by the device, hypothesising that as the sheep moved closer to the device, the device would be more likely to detect the changes in CH_4 produced by the sheep. Contrary to the hypothesised association, the low correlation between distance and measured CH_4 concentration indicates a lack of association between these two variables. However, the reliability of the GPS location data is impacted by the inconsistency through which the distance data was received. Each tracker sent the GPS locations at varying time intervals, making it impossible to

determine the exact distance of all animals from each device for each observed CH_4 concentration measurement and the size and distance of a group of animals from the device.

Various methods have been investigated to analyse the LMD data, separating it into peaks and troughs for further analysis (Chagunda *et al.*, 2013; Ricci *et al.*, 2014; Sorg *et al.*, 2018). Chagunda *et al.* (2013) used 2 standard deviations of the mean as the threshold value. Sorg *et al.* (2018) used the boxplot method (T = Q3 + (1.5 * IQR)). Ricci *et al.* (2014) described a more robust process. It involves fitting a double normal distribution to natural logarithmic transformed CH_4 concentrations, where one distribution represents the low values of respiration and the other the high values associated with eructation. The method of Ricci *et al.* (2014) is however labour intensive and is best suited when analysing the difference between treatment means. Each of these studies separated each LMD output into peaks and troughs using different methods, assuming that the peaks and troughs follow the cyclic nature of respiration. Sorg *et al.* (2018) compared the peaks to the observed respiration rate of cows and found that each peak corresponded to one exhalation. While troughs are linked with periods of inhalation, and as inhalation is independent of CH_4 production, it is not included in emission analysis.

This study used the method described by Sorg *et al.* (2018) as it is associated with the actual respiratory cycle of the animal and is sufficient for analysing the aim of the study. The MEAN, P_MEAN, and ERP_MEAN phenotypes calculated for each sampling period reveal the difference in observed CH_4 concentration when looking at all collected spot samples, only at the peaks and only at peaks above the described threshold, respectively. Sorg *et al.* (2018) found the P_MEAN value to have higher repeatability than the other phenotypes. The MEAN value is lower than the P_MEAN value as it includes the inhalation events (troughs) that are hypothesised to be artificially diluted by background CH_4 concentrations (Sorg *et al.*, 2018).

It was not always possible to obtain the 480 observations in each sampling period resulting from the movement of the rams during grazing and the occasional movement of the ram's head while standing or lying. Due to the lack of a reflecting surface for CH_4 measurement, when the rams head moved away from the laser, the LMD measured inaccurate CH_4 concentration values removed from each sampling period dataset. The study of van Wyngaard (2018) had a drastically reduced average sample size resulting from cow movement while grazing and ambient conditions. This is a limitation in the application of the LMD on animals under grazing conditions.

The mean CH_4 concentration measured with the LMD in this study (13.9 ppm-m) is lower than that measured by Chagunda *et al.* (2013) and Ricci *et al.* (2014). Chagunda *et al.* (2013) used the LMD and RC to measure CH_4 production from four yearling ewes, measuring 68.9 ppm and 15.6 ppm, respectively. The mean CH_4 concentration measured with the LMD in this study is more similar to the CH_4 concentration measured by the RC (15.6 ppm) in Chagunda *et al.* (2013). Ricci *et al.* (2014) used the LMD to measure the CH_4 concentration from 24 lactating ewes at different times after feeding and for ad libitum and restricted feeding. Their measurements ranged from 17.3 to 28.6 ppm depending on the time of measurement and 21.7 ppm for the ad libitum feeding

treatment. The LMD was held 2.75 m from the ewes in Chagunda *et al.* (2013), the values were converted to ppm by correcting for the distance, and 1 m from the ewes in Ricci *et al.* (2014). The distance of the LMD from the ewes and the correction for distance by Chagunda *et al.* (2013) is potentially the reason for the much higher CH_4 concentration measured by Chagunda *et al.* (2013) compared to this study (68.9 ppm vs 13.9 ppm) and Ricci *et al.* (2014). The mean CH_4 concentration measured by LoRa D_3 (11.9 ppm) agrees with this study's LMD measurement but is also lower than reported in previous studies. However, LoRa D_1 measured a mean CH_4 concentration of 24.0 ppm, similar to Ricci *et al.* (2014) but much lower than that reported by Chagunda *et al.* (2013). Device 2 (26.0 ppm) agrees with D_1 and Ricci *et al.* (2014) when looking only at the measurements before the device stopped sending data. However, the mean CH_4 concentration measured by D_2 (52.9 ppm) is similar to that of Chagunda *et al.* (2013) measured with the LMD. Both D_1 and D_2 measured CH_4 concentrations larger than that measured by the RC (15.6 ppm) in Chagunda *et al.* (2013).

Despite the observed difference in this study's mean LMD-measured CH_4 concentration (ppm) compared to previous studies, the estimated mean daily CH_4 production calculated in this study (27.6 g/d) is similar to previous studies using the SF_6 tracer technique to determine sheep enteric CH_4 production. Malik *et al.* (2022) measured the CH_4 emissions from nine adult *Mandya* sheep in India measuring a mean CH_4 of 19.7 g/d. Bhatt *et al.* (2021) used the SF_6 tracer technique to measure CH_4 production from 36 adult ewes split into four treatment groups. They measured 18.9 g/d, 26.3 g/d, 31.7 g/d and 40.8 g/d for the four different groups. Savian *et al.* (2014) measured CH_4 emissions from sheep as 22.7 g/d in summer and autumn and 39.9 g/d in winter and spring. Pinares-Patiño *et al.* (2008) measured the CH_4 concentration in ten sheep in two trials using the SF_6 tracer technique and the RC. The first trial measured CH_4 production in the spring, and the mean CH_4 concentrations measured were 24 g/d by the SF_6 tracer technique and 17.8 g/d by the RC. The second trial measured emissions in autumn, and the mean CH_4 concentrations measured were 18.8 g/d by the SF_6 tracer technique and 19.5 g/d by the RC.

The small between-ram coefficient of variation (CV) seen for the three LoRa devices compared to the LMD is likely due to the method used to determine the CH_4 concentration per ram, where the methane production was calculated per ram by dividing the difference between the first and second measurement period by the number of rams. A higher coefficient of variation (CV) is expected for both the LoRa and LMD approaches when including the between-day variation. These results build on the evidence Roessler *et al.* (2018) found that the particular day influenced the mean CH_4 concentration measured by the LMD, which could be a result of changes in ambient conditions, such as humidity and air pressure. However, the extremely high between-day CV for the CH_4 concentrations measured by D_3 was unexpected but can be attributed to the large increase in the range of CH_4 levels measured.

The weak linear relationship between the LMD and the three LoRa devices indicates a potential lack of sensitivity to measure the small changes in CH_4 emissions, such as specific

eructation and expiration emission events like the LMD does. The LoRa devices do, however, still have the potential as a measurement technique that can be used to determine the overall daily emission in sheep under grazing conditions. These weak relationships could also result from the difference in the spot-sample measurements collected by the LMD versus the 24-hour measurements taken by the LoRa devices. Despite the large between-day variation in the CH_4 concentration data measured by D_3 and its weak relationship with the LMD, it is the only LoRa device that did not have a significantly different mean compared to the LMD. Furthermore, despite the lack of agreement between the LMD and D_1 and D_2, these two devices measured CH_4 concentrations similar to previous studies.

The analysis of the mean CH_4 concentration measured at different times of the day identified a diurnal pattern of CH_4 production. Ricci *et al.* (2014) used the LMD to measure the CH_4 concentration from ewes housed in pens at different times (ranging from 10:00 to 15:00) and subsequently different hours after feeding. They reported an overall decrease in the mean CH_4 concentration as the time after feeding increased. Roessler *et al.* (2018) evaluated the diurnal pattern of CH_4 emission from goats fed at 08:00 each morning. The highest concentration was observed about one to two hours after feeding, similar to the results obtained by Ricci *et al.* (2014). A similar pattern was found in this study, with higher CH_4 concentrations measured after the 07:00 feeding of the supplement and slowly decreasing throughout the day. The morning CH_4 concentrations measured by the LMD and D_3 were higher, followed by the mid-day readings and, lastly, by the afternoon readings. At the same time, D_1 and D_2 observed the highest concentration mid-day, followed by the morning and the afternoon. The LMD was used before the supplemental feed was given to the sheep taking measurements at dawn, between 05:00 and 06:00. The slightly higher mean CH_4 concentration measured in the morning compared to the mid-day reading may be a result of better reflectance of the laser.

The results indicate that LoRa technology in combination with a CH_4 sensor has the potential to measure CH_4 emissions from sheep under grazing conditions, however limited to smaller, controlled camps.

The difference in the CH_4 concentration measured by D_2 before and after the device stopped sending data is a limitation of this study, affecting the reliability of D_2's results. Device 3 also had a short period where it stopped sending data which could have caused the extremely high variability in the measurements. These devices were likely affected by changes in the ambient conditions, the proximity of the LoRa nodes to each other and the frequency with which the data was transferred. A study by Cattani *et al.* (2017), evaluating the reliability of LoRa, found that temperature and humidity were significantly correlated with the received signal strength indication (RSSI) and packet reception ratio resulting in decreased signal strength with an increase in temperature and humidity. Various studies also found that end-devices close to each other, sending data at the same time, frequency and spreading factor can cause a data burst and collision of packages,

reducing the package reception by gateways, causing data to be lost (Aftab *et al.*, 2020; Mroue *et al.*, 2020; Saban *et al.*, 2021).

Further studies should investigate the influence of distance on the measured CH_4 concentration with more consistent data to determine whether this method has the potential to measure individual animal emissions. Future research should also focus on comparing the LoRa measurement technique to the RC or SF_6 techniques, testing the devices under more controlled environments to determine whether the devices are sensitive enough to detect the changes in individual animal CH_4 emissions and to validate the LoRa technique.

4.5 Conclusion

There is still a need for a practical and low-cost enteric CH_4 measurement technique that can be used under grazing conditions. The evaluation of the association between the LMD and LoRa measurement techniques provides insight into the potential of these techniques to determine diurnal CH_4 emissions in sheep under grazing conditions. Although no association was found between the LMD and LoRa devices measured CH_4 concentration, the obtained values agree sufficiently with values reported in previous studies. The LoRa device has the potential to measure CH_4 emissions from animals under grazing conditions, although limited to smaller camps, and to be used as a practical and low-cost measurement technique. Further research can assist with refining the device and measurement protocol, respectively.

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Chapter 5

A comparison of LoRa, LMD and IPCC approaches to determine methane emissions in sheep under grazing conditions

Abstract

Climate change has become a prominent global issue and is one of the main driving factors that are moving industries toward more sustainable production. There has been increased attention on the ruminant livestock industry to lower its contribution to climate change by reducing enteric methane emissions. Mitigating ruminant livestock methane emissions requires accurate knowledge of their emissions under various production conditions. However, there is little information regarding the methane production of ruminant animals under extensive conditions, as most of the available measurement techniques cannot be used under these conditions, and their results cannot be extrapolated to animals under these conditions. This study compared the LoRa technology, laser methane detector and IPCC Tier 2 approach to determine methane emissions from sheep under extensive grazing conditions to identify an accurate approach for methane emission determination under these production conditions. Methane emissions of ten intact Dohne Merino rams were estimated using these three approaches. The LoRa technology and laser methane detector approaches were found to agree with each other and previous studies. In contrast, the IPCC Tier 2 approach underestimated methane emissions in this study. The results indicate that the LoRa and LMD approaches have the potential to collect sheep methane emission information under grazing conditions, however limited to smaller, controlled camps in the case of the LoRa technology approach.

Keywords: methane, sheep, LoRa, Laser methane detector, IPCC Tier 2, South Africa, grazing

5.1 Introduction

The focus on developing methane (CH_4) emission reduction strategies has increased due to the significant rise in global CH_4 emissions caused by changing human activities. The ruminant livestock industry has considerably contributed to this increase through enteric fermentation and manure management. This has led to increased analysis of the ruminant livestock sector as researchers investigate various mitigation strategies and attempt to accurately quantify CH_4 emissions from this sector.

Methane emissions in ruminants originate as a non-utilisable product of enteric fermentation and from manure. Herrero *et al.* (2008) state that of these emissions, 97% are eructated or

expired, and 3% are from manure. The livestock sector produces 3.1 gigatonnes (Gt) of carbon dioxide-equivalent (CO_2-eq), equating to 44% of anthropogenic CH_4 emissions (Gerber *et al.*, 2013). Of the livestock supply chain's greenhouse gas (GHG) emissions, 39.1% are attributed to enteric CH_4 emissions and 4.3% to manure management. Small ruminants are responsible for 6.7% of the livestock sector's global CH_4 emissions, producing approximately 0.475 Gt CO_2-eq (Gerber *et al.*, 2013).

There are over 18 million sheep in South Africa, making up about 50% of the South African domesticated ruminant population (Abstract of Agricultural Statistics, 2022; Meissner *et al.*, 2013). Du Toit *et al.* (2013) quantified the CH_4 emissions from the South African small ruminant industry. They found that the South African small ruminant population contributes 15.6% to the country's total livestock CH_4 emissions, with sheep being responsible for 80% of those emissions. In South Africa, sheep production occurs mainly under extensive grazing conditions, as sheep can use land unsuitable for intensive crop production. Therefore, sheep production forms an important part of South African agriculture as 83% of the land available for farming is suitable only for extensive livestock production, as this land is found in arid and semi-arid zones, and sheep production contributes to sustainable production in these areas (Schoeman *et al.*, 2010; Abstract of Agricultural Statistics, 2018).

Producers are under increasing pressure to reduce CH_4 emissions from domesticated ruminants to decrease the environmental impact of production and thus increase sustainability. However, progress in this regard is limited by the uncertainties in the available emission data, making the setting and measuring of emission reduction goals and the verification of mitigation strategies difficult. Furthermore, there is little knowledge regarding enteric CH_4 emissions by ruminants in extensive production conditions (Pérez-Barbería *et al.*, 2020). This lack of knowledge results from insufficient data regarding ruminant livestock CH_4 emissions, their diets and seasonal variation (Pérez-Barbería, 2017).

Obtaining data to fill these gaps in information requires accurate CH_4 measurement techniques that can quantify ruminant CH_4 emissions under extensive production conditions. Establishing national emission inventories, validating mitigation strategies and developing CH_4 quantification protocols also require accurate CH_4 measurement techniques (Hammond *et al.*, 2016). Methane measurement techniques that are accurate, reliable, robust, low-cost and that can be used to determine emissions from large numbers of animals under various production conditions are needed to assess CH_4 mitigation strategies, and determine which of these strategies to apply (Huhtanen *et al.*, 2015; Huhtanen *et al.*, 2019; Coppa *et al.*, 2021).

There are various methods available for determining CH_4 emission in ruminants. These methods range from direct approaches such as the respiration chamber (RC) and sulphur hexafluoride (SF_6) tracer technique, to short-term techniques such as the laser methane detector (LMD) and GreenFeed (GF) system, and include indirect methods such as the *in vitro* gas production technique and prediction equations and models.

Due to its reliability, the RC is the main measurement technique used and is currently the primary source of data on which ruminant CH_4 emissions are based (Gardiner *et al.*, 2015). However, the RC confines the animal, limiting movement, diet selection, and normal interaction with peers and the environment (Pinares-Patiño *et al.*, 2011). The chamber also decreases dry matter intake, and as intake is directly linked to CH_4 production, total CH_4 production decreases (Storm *et al.*, 2012). Therefore, the results obtained using the RC cannot be applied to grazing animals, as grazing animals select their diets and have a higher intake and energy expenditure than animals in the RC (Li *et al.*, 2014; Hammond *et al.*, 2016).

The SF_6 tracer technique was developed for use under grazing conditions (Zimmerman, 1993). It is, however, a labour-intensive technique with high among- and within-animal variation (Grainger *et al.*, 2007). The RC and SF_6 tracer technique are both limited in their application at the applied research level on commercial farms (Chagunda & Yan, 2011).

The LMD was identified as a technique that could measure CH_4 emissions from cattle in their natural environment (Chagunda *et al.*, 2009). It has since been used by Chagunda *et al.* (2013) and Ricci *et al.* (2014) to measure CH_4 emissions from sheep. The LMD is a portable, hand-held device that uses infrared absorption spectroscopy to measure the CH_4 concentration between the device and the target (Tokyo Gas Engineering, 2006). A laser from the device is pointed at the nose and mouth area of the animal for about 2 to 5 continuous minutes. Repeated measurements are collected every 0.5s in parts per million per meter (ppm-m). The LMD measures the animal's respiratory cycle, represented as a series of peaks (exhalation or eructation) and troughs (inhalation) (Ricci *et al.*, 2014). Chagunda *et al.* (2013) and Ricci *et al.* (2014) reported weak but positive correlations between the LMD and RC CH_4 measurements from sheep ($r = 0.18$, $p \leq 0.01$ and $r = 0.12$, $p=0.362$, respectively). Due to the remote nature of the device, the LMD could have the capacity to screen a large number of animals for genetic selection and analysing mitigation techniques (Sorg *et al.*, 2018).

The Intergovernmental Panel for Climate Change (IPCC) developed the Tier (1, 2 and 3) methodology intended for quantifying national and global livestock emissions (IPCC, 2006; IPCC, 2019). The choice of which method to use depends on the amount of data available and the area for which estimates are required (Aljaloud *et al.*, 2011). The IPCC Tier 1 and 2 models are widely used for compiling GHG emissions inventories for various livestock production systems in different regions and countries (Aljaloud *et al.*, 2011). These models are essential tools for predicting national CH_4 emissions data for domesticated ruminant populations, and can be used to compile national and global GHG inventories that assist with developing environmental policies and strategies to reduce overall emissions (IPCC, 2006; Storm *et al.*, 2012). However, compared to direct methods, the estimations of these models generally have lower accuracy and are associated with greater uncertainties (Hristov *et al.*, 2018). These inaccuracies have been partly linked to the small datasets used for model development and parameterisation, which do not accurately take into account the diversity between different regions and diets (Moraes *et al.*, 2014; Hristov *et al.*,

2018). Hristov *et al.* (2018) state that databases should require more than 1000 individual observations or treatment means, including dietary animal factors that affect CH_4 emission production. Many countries have only recently started measuring enteric CH_4 emissions due to the high cost of equipment and the specialised methodologies required to obtain measurements from live animals (Benaouda *et al.*, 2020). Therefore, a limited amount of data specific to these countries is available to be used in prediction models. When these models are applied to areas where little research has been done, it could lead to inaccurate CH_4 emission estimates that are not suited for the region, which can differ in genetic potential, diet, climate, and management practices (Benaouda *et al.*, 2020). Du Toit (2017) determined the enteric and manure CH_4 emissions from sheep in South Africa using equations defined for the Australian National Inventory Report (ANIR), as no values are available that are specific to South Africa. The ANIR equations were chosen because Australian methodology is based on Australian conditions that are more representative of South African conditions.

To reduce the uncertainties of CH_4 emission estimates an accurate measurement technique is needed that can be used under grazing conditions to better quantify sheep CH_4 emissions in South Africa. Emissions data needs to be collected from large groups of animals under various production conditions allowed to exhibit normal behaviour.

Sheep are essential in developing CH_4 measurement techniques and mitigation strategies due to their contribution to CH_4 emissions (Marino *et al.*, 2016). Having accurate baseline emission figures and inventory data of sheep production in South Africa is important to effectively set reduction targets and measure the progress toward achieving these targets. The availability of measurement techniques that can be used under grazing conditions is also crucial for developing and verifying mitigation strategies while the sheep are in their natural environment and able to express their normal behaviour.

The agricultural industry is transforming toward automated and data-driven agriculture by integrating various information and communication technologies (Boursianis *et al.*, 2020). This transformation, known as Farming 4.0, is the agricultural industry's response to the fourth industrial revolution (4IR). The Internet of Things (IoT) is one of the most popular technologies driving the 4IR. LoRa (Long Range), a low-power, wide-area technology designed for IoT communication, has the potential to be used in extensive farming applications (Semtech, Co. Camarillo, California, United States). It can be used in areas with no network coverage and transmits small amounts of data over distances of about 5km in urban and 20km in rural areas (Miles *et al.*, 2020). LoRa is the physical layer of the protocol, and LoRaWAN (Long Rand Wide Area Network, LoRa Alliance) is the communication protocol that consists of the application layer and media access control (MAC) layer. It uses a proprietary chirp spread spectrum modulation that enables low-power, long-range data transmission over the unlicensed ISM band. LoRa uses the 868MHz frequency band in South Africa. Combined with a CH_4 sensor, this technology could measure enteric CH_4 emissions in sheep under extensive grazing conditions. This technique consists of wireless sensor nodes

placed in the field that transmit data every 50s. The LoRa node is connected to a CH_4 sensor and runs on batteries charged by solar power and, depending on the data transmission rate, is expected to last several years. In the LoRaWAN protocol, nodes are arranged in a star-of-star topology, with each node having bidirectional communication with multiple gateways. The LoRa nodes send observed CH_4 emission information to the LoRa gateway, which sends the data to the network server. The network server removes all duplicate data, and The Things Network logs the remaining data to a Google Sheet.

The aim of this study is to compare the LoRa, LMD and IPPC Tier 2 approaches in determining CH_4 emissions in sheep under grazing conditions.

5.2 Materials and methods

This study was approved by the Research Ethics Committee: Animal Care and Use of Stellenbosch University (ACU-2021-11233).

5.2.1 Experimental location

This study was conducted on the Welgevallen Experimental Farm of the University of Stellenbosch, Western Cape, South Africa, in a 0.07 ha camp (-33.94458, 18.86628). The trial was completed in summer, with an average recorded temperature of 20°C, a minimum of 14.6°C and a maximum of 26°C (Climate-data.org, 2021). The average rainfall is 27mm, and the average recorded relative humidity is 61% (Climate-data.org, 2021).

5.2.2 Experimental animals and husbandry

Ten intact Dohne Merino rams, with an average body weight of 96.6 ± 6.1 kg, were randomly selected from the Welgevallen Sheep Research flock and used for the duration of the study. The rams were three and a half years old at the time of the study. They had *ad libitum* access to fresh water and kikuyu (*Pennisetum clandestinum*) grazing for the entire experimental period, and were fed oat hay supplement at 07:00 each morning. The husbandry of the rams was according to normal farm protocols, and rams were checked daily for the duration of the study to ensure their welfare was maintained.

5.2.3 Methane measurement by LoRa technology

The LoRa devices measured methane levels in two separate measurement periods (Table 5.1). The functional LoRa CH_4 detection units used in this trial are described in Chapter 3, section 3.2.3. Three complete units (device 1 (D_1), device 2 (D_2) and device 3 (D_3)) were assembled. The three devices were calibrated as described in Chapter 3, section 3.2.4. On the morning of the 11th of December, the three functional LoRa CH_4 detection units were installed on one of three poles set

up in the camp, at equidistance from one another and the sides of the camp, as described in Chapter 3, section 3.2.5. There placement in the camp is illustrated in Figure 3.2, Chapter 3, section 3.2.5.

Table 5.1 Dates of the two measurement periods of this trial, whether the sheep were present in the camp for that measurement period and the data recorded in that measurement period.

Measurement period	Dates	Sheep absent/present	Data recorded
Period 1	11 December 14:00 - 13 December 09:00	Absent	Background methane concentrations measured by LoRa D_1, D_2 and D_3
			Ambient conditions
Period 2	13 December 09:11 - 20 December 15:00	Present	Ram live weights before trial
			Sheep methane emissions measured by LoRa D_1, D_2 and D_3 (13 to 20 December)
			Ram GPS locations
			Ambient conditions

Each LoRa device was set to take and send measurements every 50s. The three devices were connected to three LoRa gateways located on Bottelary Hill (± 10 km from the experimental location), Kanonkop (± 28 km from the experimental location) and Delaire Graff (± 8 km from the experimental location). All LoRa CH_4 concentration data measured by each device was sent via the TTGO T-Beam to the gateways and from the gateways to the cloud, where data was logged, using The Things Network, to a Google Sheet.

A numbered GPS collar was placed on each ram before moving into the camp where the sensors were installed. Their movement relative to the sensors was recorded using these GPS trackers (Yabby Edge, LoRaWAN®, Digital Matter). The GPS location data was sent to the gateways and from the gateways, using The Things Network, to a SQL database. The data was exported from the SQL database as a .xlsx file. This data was used to determine the distance of sheep from the device and the relative CH_4 concentration measured. The CH_4 concentration measured per sheep was determined using the distance of the sheep from the device and the CH_4 concentration measured by the device.

The raw data (mV) of D_1, D_2 and D_3 were converted to CH_4 concentrations (ppm) using Equations 4.2 and 4.3 presented in Chapter 4, section 4.2.4,

The mean CH_4 concentration measured by each device in the first measurement period was used as the background CH_4 concentration and subtracted from each device's observed concentrations (ppm) in the second measurement period. The CH_4 concentrations (ppm), corrected for background CH_4 concentration, were divided by the number of sheep to determine individual sheep emissions.

5.2.4 Methane measurement and quantification by the LMD

The hand-held Laser Methane Mini™ (model SA3C2A; Tokyo Gas Engineering Solutions, Co. Ltd., Otaku, Tokyo, Japan) was used to measure CH_4 concentration (ppm-m) in the expired air of Dohne Merino rams, as described in Chapter 4, section 4.2.3.2.

As the LMD was held 1m from the animal, the values were not corrected for distance (Roessler *et al.*, 2018). Ten emission profiles were obtained per ram, with approximately 480 observations per daily sampling period. Due to the high sensitivity of the LMD, the minimum CH_4 concentration was set as the background CH_4 concentration. This value was subtracted from the rest of the sample to adjust for background CH_4 concentration (Ricci *et al.*, 2014). For each sampling period, a threshold was calculated to distinguish between respired and eructated CH_4 emissions using Equation 4.4 reported in Chapter 4, section 4.2.4 (Sorg *et al.*, 2018).

Each emissions profile was analysed to detect the peaks (of expiration and eructation) using the method described by Sorg *et al.* (2018). The difference between a data point (x_i) and the preceding value ($x_i - 1$) was determined for each profile. If the difference of $x_i - x_{i-1}$ was < 0, and the difference between the two data points before x_i ($x_{i-1} - x_{i-2}$) was ≥ 0, the data point x_{i-1} was classified as a peak (Sorg *et al.*, 2018). Further analyses used only the peak values as done by Chagunda *et al.* (2009) and Ricci *et al.* (2014).

The spot-sampled CH_4 concentrations observed for each sheep were converted to g/d (grams/day) to determine the daily CH_4 production per sheep using Equation 4.5 in Chapter 4, section 4.2.4.

5.2.5 Methane quantification using Tier 2 methodology

Methane emissions for the sheep were calculated using a Tier 2 approach based on the IPCC 2019 Refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories (IPCC, 2019) and the Australian national greenhouse accounts, National Inventory Report 2019 (ANIR, 2021). The Australian methodology is based on the IPCC methodology, but is adapted for Australian conditions, containing both Australian-specific and IPCC default methodologies and emission factors. Du Toit *et al.* (2013) employed this method to compile national inventories for the South African small stock population. The Australian methodology was used as it is adapted to Australian conditions, which are more representative of South African conditions (du Toit *et al.*, 2013).

5.2.5.1 Enteric CH_4

Rams were weighed on the morning of the 13th of December, and their live weights were used for further calculations.

The potential intake (PI, kg/head/day) is given by AFRC (1990) as:

$$PI = (104.7q_m + 0.307W - 15.0)W^{0.75}/1000 \quad \textit{Equation 5.1}$$

Where W is the live weight (kg, Table 5.2), and q_m is the metabolisability of the diet (ME/GE). The metabolisability of the diet is calculated using the equation of Minson and McDonald (1987), q_m = 0.00795DMD – 0.0014, where the dry matter digestibility (DMD) is expressed as a percentage. A DMD percentage of 63.1% was used in this study (García *et al.*, 2014).

Table 5.2 The live weights of each Dohne Merino ram measured on the morning of the 13th of December.

Ram	Live weight (kg)
1	89
2	93
3	110.4
4	102.4
5	94.4
6	94
7	97.2
8	94
9	99.2
10	92.4

Howden & Reyenga (1987) reported a strong relationship between dry matter intake and methane production, stating 87% of the variation in CH_4 production is explained by feed intake. The daily CH_4 production (M, kg/head/day) was calculated using the daily intake figures determine from Equation 5.1, based on the relationship described by Howden & Reyenga (1987):

$$M = I \times 0.0188 + 0.00158 \quad \textit{Equation 5.2}$$

5.2.5.2 Manure management

As the sheep were kept in the camp for the duration of the study, under extensive conditions, the manure was deposited onto the veld with no active manure management. Methane emissions from sheep manure (M, kg/head/day) were therefore calculated as:

$$M = I \times (1 - DMD) \times MEF \quad \textit{Equation 5.3}$$

Where I is the daily intake calculated from Equation 5.1, DMD is the digestibility of the feed expressed as a percentage, and MEF is the emission factor (kg CH_4/kg DM manure). An emission factor of 1.4 x 10^{-5} was used in this study (Gonzalez-Avalos & Ruiz-Suarez, 2001).

5.2.4 Data analysis

Errors in laser beam reflectance were automatically identified by the LMD and manually removed from the dataset. All data obtained from the LMD when the laser moved off the mouth and nose area of the sheep due to sheep movement were manually removed. Outliers in the raw LoRa measurement dataset were identified using the modified z-score.

Descriptive statistics were calculated for the LoRa, LMD and Tier 2 data sets. The CH_4 concentrations measured by the LMD (ppm) were compared with the CH_4 concentrations measured by the LoRa devices (ppm). The CH_4 emissions quantified for each ram using the LMD (g/day) were compared to the CH_4 emissions determined using the Tier 2 approach (g/day). It was not possible to compare the CH_4 concentrations measured by the LoRa detection units and the CH_4 emissions estimated by the Tier 2 approach as their estimations were in different units (ppm versus g/day, respectively). A two-sample t-test was performed to compare the mean CH_4 concentration measured by the LMD (ppm) and each of the three LoRa devices (ppm). Another two-sample t-test was performed to compare the CH_4 emission estimates determined using the Tier 2 approach and the LMD when its ppm values were converted to g/day.

The correlation between CH_4 concentrations measured by the LMD (ppm) and three LoRa devices (ppm) and between the CH_4 emissions estimated using the LMD (g/day) and Tier 2 approach were calculated. Correlation coefficients were calculated using the average emissions obtained per ram. All data were analysed using XLSTAT.

5.3 Results

The CH_4 emissions for each ram, resulting from enteric emissions and from the manure, calculated using the Tier 2 approach, are presented in Table 5.3. The average daily CH_4 emission of the 10 rams was estimated as 3.77288 kg/head/day.

Table 5.3 Estimated methane emissions of Dohne Merino rams for enteric and manure management under grazing conditions.

Ram	Enteric methane production (kg/head/year)	Manure methane production (kg/head/year)	Total (kg/head/year)
1	3.10	0.00069	3.10069
2	3.44	0.00079	3.44079
3	5.08	0.00124	5.08124
4	4.29	0.00102	4.29102
5	3.56	0.00082	3.56082
6	3.53	0.00081	3.53081
7	3.81	0.00089	3.81089
8	3.53	0.00081	3.53081
9	3.99	0.00094	3.99094
10	3.39	0.00077	3.39077
Average	3.77	0.00088	3.77288

Descriptive statistics of the CH_4 levels measured by each LoRa device and the LMD are presented in Table 5.4. When comparing the mean CH_4 concentrations measured by these four devices, the CH_4 concentrations measured by D_2 differed from those measured by D_1, D_3 and the LMD, while the CH_4 concentrations measured by D_1, D_3 and the LMD were similar.

Table 5.4 Descriptive statistics for the methane levels (mean ± SD) recorded by the three functional LoRa methane detection units and the laser methane detector.

Technique	Methane level (ppm)	Minimum (ppm)	Maximum (ppm)	CV
LoRa D_1	24.0^a ± 0.3	15.5	31.5	1.37
LoRa D_2	52.9^b ± 0.8	22.2	89.5	1.43
LoRa D_3	11.9^a ± 1.0	-51.8	90.4	8.41
LMD	13.9^a ± 2.1	5.4	29.2	15.0

$^{a-b}$ Methane levels with different superscripts are significantly different ($p \leq 0.05$)

The negative CH_4 concentration measured by D_3 observed in Table 5.4 is a result of the background concentration being higher than the concentration recorded by the device at a specific time. When comparing the CH_4 concentration measured by the four devices separately using two-sample t-tests, the mean CH_4 concentration measured by D_1 and D_2 were significantly higher than those measured by the LMD ($t(78) = -8.384$, $p \leq 0.0001$ and $t(118) = -11.837$, $p \leq 0.0001$, respectively). While the CH_4 concentrations measured by D_3 were similar to those measured by the LMD, $t(138) = 0.381$, $p = 0.704$. The CH_4 concentration measured by D_2 was higher than those measured by D_1 and D_3 ($t(58) = 2.002$, $p = 0.006$ and $t(118) = 1.980$, $p \leq 0.0001$, respectively). The CH_4 concentration measured by D_1 were higher than those measured by D_3, $t(78) = 1.991$, $p = 0.050$.

Descriptive statistics of the CH_4 emissions estimated using the LMD and Tier 2 approach are presented in Table 5.5.

Table 5.5 Descriptive statistics for the methane production estimated (mean ± SD) by the three the laser methane detector and Tier 2 approach.

Technique	Methane production (kg/head/year)	Methane production (g/head/day)	Minimum (g/day)	Maximum (g/day)	CV
LMD	10.1 ± 1.3	27.6 ± 3.6	9.7	54.3	13.0
Tier 2 enteric	3.8 ± 0.6	10.3 ± 1.6	8.499	13.920	15.1
Tier 2 manure	0.0008 ± 0.0001	0.0024 ± 0.0004	0.0019	0.0034	17.8

The LMD estimated higher daily CH_4 production in sheep under grazing conditions than the Tier 2 technique ($p \leq 0.0001$). The coefficient of variation (CV) of the CH_4 concentrations estimated for each ram by the LMD was lower than the CV of the manure and enteric CH_4 emissions estimated by the Tier 2 approach.

The CH_4 levels measured by the three functional LoRa CH_4 detection units for each ram had moderately strong positive correlations (Table 5.6). The CH_4 levels measured for each ram by D_2 and D_3 had a weak correlation with those measured by the LMD and the CH_4 levels measured for each ram by D_1 had almost no correlation with those measured by the LMD.

Table 5.6 Correlation coefficients between the CH_4 levels measured for each ram by the three LoRa devices and the LMD.

Measurement technique	LoRa D_1	LoRa D_2	LoRa D_3	LMD	IPCC
LoRa D_1	1				
LoRa D_2	0.674[a]	1			
LoRa D_3	0.688[a]	0.750[a]	1		
LMD	-0.017	0.223	0.251	1	
Tier 2	-0.175	-	-	-	1

[a] Correlations with a superscript are statistically significant ($p \leq 0.05$)

The estimated CH_4 emissions determined using the LMD had a weak correlation with those estimated using the Tier 2 approach.

5.4 Discussion

Sheep CH_4 emissions determined using the LoRa technology, LMD and Tier 2 approaches were compared in this study.

5.4.1 Methane concentrations measured using LoRa technology

The average CH_4 concentrations measured for individual sheep by the three functional LoRa CH_4 detection units ranged from 11.9 to 52.9 ppm. This variation between the CH_4 concentrations detected by each device was observed in the first and second measurement periods. The CH_4 concentrations measured by D_2 were higher in both periods, as reported in Chapter 3, section 3.3.2. This significant variation between the emissions measured by the three devices reduce the reliability of the measurements and is potentially caused by reduced device functionality caused by fluctuations in the ambient conditions or faults in the equipment or program of the LoRa equipment.

Device 3 measured a negative minimum CH_4 concentration in period 2 after the values were corrected for the background concentration measured in the first period (Table 5.3). This could be due to the sheep moving too far away from the device for the device to detect the emissions released by the sheep. Therefore, resulting in the CH_4 concentrations recorded by the device being similar to or lower than the background concentration. However, D_3 also had two big changes in the CH_4 concentrations detected, as mentioned in Chapter 3, section 3.3.2 (Figure 3.7). The significant decrease in CH_4 concentration measurement at the end of the second measurement period could have caused this negative value. It is likely also the reason that the mean CH_4

concentration measured by D_3 is lower than the LMD, while both D_1 and D_2 were larger than the LMD.

The moderately strong positive correlations between the three LoRa devices indicate that they followed a similar pattern of CH_4 measurement, ranking the rams similarly according to CH_4 production. This correlation could also be because of changing environmental factors affecting the concentrations detected by the devices. Atmospheric CH_4 concentrations have been found to be negatively correlated with humidity (r = -0.48, p = 0.14) and positively correlated with air temperature (r = 0.49, p =0.24) (Javadinejad *et al.*, 2019). These relationships could cause changes in the atmospheric CH_4 concentration in the area and result in a similar pattern of CH_4 changes observed by the three LoRa devices.

5.4.2 Methane concentration measured using the LMD

The mean daily CH_4 production measured and estimated by LMD were 13.9 ppm and 27.6 g/day, respectively. The measurements of the LMD were based only on enteric CH_4 emissions, while the LoRa and IPCC approaches determined emissions from both enteric and manure emissions. Despite this, the emissions determined by the LMD were greater than the IPCC calculations and lower than only two of the LoRa devices (D_1 and D_2). Furthermore, the emissions estimated using the LMD have better agreement with previous studies than the Tier 2 approach.

5.4.3 Determining methane concentrations using a Tier 2 approach

The annual enteric and manure management CH_4 emissions for each Dohne Merino ram were calculated using an Australian methodology, based on the IPCC guidelines but adapted for Australian conditions (ANIR, 2021). As presented in Table 5.3, the average enteric CH_4 emissions from these ten rams were estimated at about 3.8 kg/head/year. While the average manure management emission factor calculated in this study was 0.88 g/head/year. These emission factors are lower than the default enteric CH_4 emission factor of 5 kg/head/year and manure CH_4 emission factor of 1.3 g CH_4 kg/VS reported by the IPCC for sheep in Africa (IPCC, 2019). The methane emission factors calculated in this study were also much lower than those calculated for wool sheep by du Toit *et al.* (2013) and by the Australian National Greenhouse accounts (ANIR, 2021). Du Toit *et al.* (2013) calculated a 10.6 kg/head/year emission factor for enteric CH_4 and 0.007 kg/head/year for manure CH_4 emissions. The Australian national inventory report reported enteric emission factors of 6.8 kg/head/year (ANIR, 2021). This underestimation compared to these previous studies could simply be because the Australian methodology is not well-suited to South African conditions and therefore the underestimation is the result of a lack of information regarding South African production conditions and the effect of these conditions on CH_4 emission in sheep under these conditions.

5.4.4 Comparison of the LoRa technology, LMD and Tier 2 approach to determine methane emissions

The values measured by the three LoRa devices and the LMD are similar to other studies' CH_4 concentrations measured using the LMD. Chagunda *et al.* (2013) and Ricci *et al.* (2014) used the LMD to measure CH_4 produced by sheep. They both measured higher CH_4 concentrations than measured by the LMD in this study (13.9 ppm). Chagunda *et al.* (2013) measured an average CH_4 concentration of 68.9 ppm, and Ricci *et al.* (2014) measured concentrations between 17.3 and 28.6 ppm, depending on the time of measurement. The mean CH_4 concentration measured by LoRa D_3 (11.9 ppm) did not differ from that measured by the LMD in this study. However, the CH_4 concentrations measured by D_3 are also lower than those measured by Chagunda *et al.* (2013) and Ricci *et al.* (2014). Chagunda *et al.* (2013) also measured CH_4 concentrations of emissions from sheep of 15.6 ppm using the RC to compare to the LMD. This value has a better agreement with the CH_4 concentrations measured using the LMD and D_3 in this study. In comparison, LoRa D_1 measured a mean CH_4 concentration of 24.0 ppm, similar to Ricci *et al.* (2014) but lower than that of Chagunda *et al.* (2013). The CH_4 concentration measured by D_2 is similar to the CH_4 concentration measured using the LMD in Chagunda *et al.* (2013). This demonstrates the potential of the LoRa devices to measure individual or flock-level CH_4 emissions in sheep under grazing conditions.

The CH_4 emissions (g/day) estimated using the LMD, and Tier 2 approach had a low agreement. Therefore, these two approaches ranked the rams differently according to CH_4 production. The Tier 2 approach considers the animal's weight as the only differing factor between the animals due to the relationship between intake and CH_4 production. Therefore, the animal's live weight is directly linked to the estimated CH_4 emissions. The Tier 2 approach used in this study considers animal species, animal liveweight, potential intake and the digestibility of the feed. This method does not consider the effects of changing ambient conditions and genetic influence on daily CH_4 production. Various studies quantifying sheep enteric CH_4 emissions using the SF_6 tracer technique have found they range between 18 and 40 g/d (Savian *et al.*, 2014; Bhatt *et al.*, 2021; Malik *et al.*, 2022). The mean daily CH_4 production determined using the LMD (27.6 g/day) agrees with these studies. In contrast, the Tier 2 (10.1 g/day) approach significantly underestimated the daily CH_4 production per sheep, based on the observations of these previous studies. This is potentially due to the small number of factors that are taken into account when using this approach to determine sheep CH_4 production. Although the CV in the CH_4 estimates of the Tier 2 approach was slightly higher than the CV in the CH_4 emission estimates of the LMD, the variation in the Tier 2 approach is caused only by the differences in the live weights of the sheep. In contrast, the variation in CH_4 emissions estimated using the LMD approach could have resulted from different genetic profiles of the sheep, changing ambient conditions and sheep movement during the

sampling period. The variation in the CH_4 emissions measured per ram by the LoRa CH_4 detection units, although low (Table 5.4), can also be affected by these factors.

Of these three approaches, the LMD is the most labour-intensive, requiring daily measurements from each ram, amounting to about an hour of measurements each day. As observed in van Wyngaard (2018), it is not always possible to obtain a full dataset from the animals under grazing conditions using the LMD as the laser loses contact with the animal as it moves its head. The LMD also only considers the enteric CH_4 emissions of the animal and not the CH_4 emissions from the manure. Even though the contribution of manure management per ram to CH_4 emissions is small compared to enteric emissions, the lack of that data from the LMD could lead to lower national inventories due to these emissions being unaccounted for.

Compared to the LMD, the LoRa technology approach is much less labour-intensive as this technique requires only assembling, calibration and installation of the detection units. The LoRa technology approach also enables measurements to be collected throughout the day, presenting the opportunity to easily analyse the changes in CH_4 production based on season and time of day. The LoRa technology approach is more affordable compared to the LMD, and is much more practical when considering the labour-intensity of each technique.

While the Tier 2 approach has the least labour requirements and is ideal for determining emissions at the national level, the emission factors estimated using this method vary significantly and were seen in this study to underestimate sheep CH_4 emissions. The accuracy of the Tier 2 estimates can be improved through further collection of emission data under various production conditions and analysis of the numerous factors that influence CH_4 production using methods such as the LMD and LoRa technology.

5.5 Conclusion

While the Tier 2 approach is the most cost-effective and least labour-intensive of the three approaches it is not suitable for accurate determination of on-farm CH_4 emissions data due to a lack of South African relevant data. The LMD and LoRa technology approaches are more suited for determining emission production of sheep under extensive production conditions, and the results of this study indicate that the LoRa technique could have the potential as a practical and low-cost measurement technique that can be used under grazing conditions. With further development, the LoRa technology and LMD approaches could potentially collect emissions data from animals under grazing conditions to compile national inventories, evaluate mitigation strategies, and improve prediction equations and models.

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Chapter 6
General conclusions and recommendations

Climate change is a growing global issue, and its effect on the livestock sector makes it imperative for immediate action to be taken to increase the sustainability of farming by developing and applying enteric methane (CH_4) emission mitigation strategies. Due to the rapidly growing population, increased affluence in developing countries, and the subsequent increase in demand for livestock products, the option of reducing the global ruminant population is not viable. The global ruminant industry instead needs to focus on increasing the sustainability of production, partly by lowering enteric CH_4 production. However, for this to be accomplished, valid emission inventories must be compiled, and mitigation strategies developed and evaluated. This requires an accurate and affordable technique that can take constant measurements, has low labour requirements, requires no specialised training and is widely available to take measurements in various production conditions.

A technique of this calibre would allow producers to quantify CH_4 emissions from their ruminant animals and test various mitigation strategies based on the collected data. Such a technique would also allow producers to choose the most effective mitigation strategy for their production conditions, reducing emissions while maintaining or increasing production efficiency. Based on literature, it is evident that the current CH_4 measurement techniques available are not sufficient for assisting with creating global and national GHG inventories and accurately determining CH_4 emissions from animals under grazing conditions. A measurement technique does not yet exist that is affordable and simple to use, which can be applied on commercial farms, and which can be used by producers to measure GHG emissions on-farm. Long-range (LoRa) technology has the potential to meet the abovementioned requirements as a CH_4 measurement technique that researchers and producers can use under field conditions to measure real-time GHG levels.

The aim of this study was to determine the potential of LoRa technology to be used in combination with an infrared CH_4 sensor to measure CH_4 emissions from sheep under South African grazing conditions. A novel LoRa CH_4 detection unit was conceptualized, designed, and produced for testing under extensive grazing conditions. Methane levels recorded were compared with methane readings obtained with a hand-held laser methane detector (LMD), and the Australian-adapted Tier 2 approach based on the Intergovernmental Panel on Climate Change (IPCC) default values of ruminant CH_4 emission estimation.

The design and placement of the LoRa devices

Three functional LoRa detection units were installed in a 0.07 ha camp, at equidistance from each other and the fenced boundaries of the camp. The LoRa devices were used to collect CH_4 readings before the rams were introduced to the camp to determine the background CH_4 levels, and to determine the effect of ambient conditions on the observed CH_4 levels detected by each device. Ten Dohne Merino rams, each fitted with a GPS collar, were placed in the camp and allowed to freely graze the kikuyu pasture. The live weight of the rams was recorded at the beginning and end of the trial. Once the rams were moved into the camp, the three LoRa devices collected CH_4 concentrations for eight days. These values were corrected for background CH_4 concentrations and divided by the number of rams to determine the CH_4 production per ram.

The three LoRa devices were set to take measurements every 50s. Before the sheep were moved into the camp, the effect of ambient conditions on the CH_4 concentrations recorded by the devices was investigated. A significant positive correlation was observed between the relative humidity and the CH_4 concentrations measured by each device, while the CH_4 concentrations measured by Device 3 had a significant negative correlation with air temperature, wind speed and solar radiation. The CH_4 concentrations measured by Devices 1 and 2 were also negatively correlated with air temperature, wind speed and solar radiation; however, these correlations were not significant.

The distance of each ram from the devices and the CH_4 measured by each device, was determined using the data from the GPS collars and the locations of each device, respectively. The proximity of the sheep to each device was expected to influence the CH_4 concentrations measured by the device. However, no correlation was found between the CH_4 concentrations measured by each device, and the distance of the rams from the devices.

The potential of LoRa technology to measure CH_4 emissions in sheep under grazing conditions

The CH_4 emissions in sheep measured by each LoRa device in this trial differed significantly. However, the similarity in the CH_4 emissions measured by the LoRa devices and those measured in previous studies reveal that this approach could potentially measure CH_4 emissions from animals under grazing conditions, although limited to use in small, controlled camps. The similarity indicates that the LoRa devices show potential as a practical and low-cost measurement technique that can be used under grazing conditions. Therefore, providing information to quantify ruminant CH_4 emissions, and thus consequently contribute to the improvement of emission inventories and verifying mitigation strategies.

Comparison of the LoRa technology, the LMD technique, and the IPCC Tier 2 approach in determining CH_4 emissions from sheep under grazing conditions

While the LoRa devices were installed in the camp, the hand-held Laser Methane Mini™ (model SA3C2A; Tokyo Gas Engineering Solutions, Co. Ltd., Otaku, Tokyo, Japan) methane detection device (LMD) was used for ten days to collect daily 4-minute emission samples for each ram. The time of measurements alternated each day between morning (05:00-06:00), mid-day (12:00-13:00) and afternoon (17:00-18:00) to reduce bias caused by daily fluctuations in enteric CH_4 production. The LMD was held 1m from each animal's nose and mouth area, and the device was set to take measurements every 0.5s. The resulting emission profiles obtained per ram for each day were analysed to identify the peaks associated with expiration and eructation, and these peaks were used for further analysis.

Total daily emissions measured for each sheep using the LMD were compared to those measured by the LoRa devices. Despite weak linear relationships observed between the LMD and the three LoRa devices, these two techniques measured CH_4 emission levels similar to those reported in previous studies.

The live weight of each ram recorded at the start of the trial and the digestibility of the kikuyu pasture was used in equations specified by the Australian National Inventory Report to estimate the CH_4 emissions of each ram. The CH_4 emissions of each ram estimated using the Australian-adapted Tier 2 approach were compared to those estimated using the LMD. The CH_4 emissions estimated using the Tier 2 approach underestimated emissions compared to previous studies, and the LMD emission estimates in this study.

Limitations of the study

As the LoRa technique is intended for use by producers and researchers, the system's practicality and ease of use are essential. This study showed that the system is easy to use, requiring only installation and occasional calibration. The system is, however, limited in practicality by the need for a small measurement area. During the study, the available grazing ran low, and as a result, nine of the ten rams lost weight. The rams were, however, still at a healthy, normal weight and thus their wellbeing was not affected. Since enteric CH_4 production is determined by the amount and type of feed ingested, with higher intakes resulting in significantly higher emissions, a reduced feed intake potentially may have resulted in less CH_4 being produced and thus detected or measured. Despite the grazing running low, the sheep were not given extra supplemental feed as the intention of the study was to determine the potential of the LoRa-CH_4 measurement device to measure enteric CH_4 emissions under grazing conditions. The sheep were also not moved to a different camp as no others were available at the time of the study.

Devices 2 and 3 encountered problems gathering data in the second measurement period, where both devices at different times, did not recorded any measurements. On the 13th of

December from 19:46 until 09:07 on the 14th of December D_3 had no logged mV data, and on the 15th of December from 06:50 until 14:06 on the 17th of December D_2 had no logged mV data. The inability of the devices to recorded data resulted from environmental factors affecting the functional units and data transfer, animal interference, code crashing, lost connection, or a fault in the software associated with the methane detection and logging of the data. These factors affected the efficiency of the devices in gathering CH_4 emission data from the rams.

The study was also limited by inconsistent GPS location data, with an unequal number and sporadic timing of separate animals' GPS coordinates. Data from each of the ten trackers should have been recorded every minute; however, at each minute interval only one to four of the GPS trackers sent the location of the ram in that minute. The inconsistent GPS location data made it challenging to determine the location of each ram relative to each device for each CH_4 level measurement. These inconsistencies also made it challenging to determine the potential effect of the distance of the rams from the LoRa device on the observed CH_4 measurement. Ensuring consistent sending and receiving of every animal's location simultaneously will assist with analyses.

As the LoRa technology and Tier 2 approaches determined the daily CH_4 production per sheep in different units (ppm and g/day, respectively), the comparison between their emission estimates was not possible.

Recommendations

Future studies should focus on refining the LoRa detection units. Firstly, by increasing the sensitivity of the units, thereby creating a device that can accurately measure ruminant CH_4 emissions and the diurnal fluctuations in emissions. Secondly, by analysing the components of the units and identifying the components that will improve the robustness, stability, accuracy, and reliability of the device. This study revealed how changing components of the LoRa detection unit can affect the sensitivity of the units and alter the CH_4 levels measured. The infrared CH_4 sensor and the DFRobot ADS1115 16-BIT ADC Module, which enables higher resolution readings, were replaced in D_2 between Periods 2 and 3. This caused a significant difference in the mean CH_4 levels measured by this device in Periods 2 and 3. Therefore, analysing the impact of each of the components (namely the TTGO T-Beam ESP32 LoRa 868MHz, Infrared CH_4 Sensor NDIR Gas Sensor MH-440D and DFRobot ADS1115 16-BIT ADC Module) on the observed CH_4 concentration will provide valuable insight into the optimal design and functioning of the device. Thirdly, determining which factors affect the validity of the device's measurements, such as determining the temperature and humidity range under which the device will function optimally and determining the effect of various ambient conditions on observed CH_4 measurements, as well as testing the device under a controlled and fixed environment kept at standard temperature and pressure to observe the consistency of the readings. The device could further be tested under a

controlled environment where certain conditions are changed in order to determine the effect of various ambient conditions on the CH_4 concentrations measured. Lastly, testing and improving the device's durability will enable it to withstand harsh environmental conditions and avoid equipment damage.

Once the LoRa detection unit is optimised, further studies can test it against conventional techniques, such as the RC, to determine whether the LoRa detection unit is accurate and reliable in its enteric CH_4 emission measurements.

If, in future studies, the LoRa technique was found to accurately determine CH_4 emissions of sheep under grazing conditions, it is recommended that numerous small camps be set up with poles and waterproof boxes throughout the year to allow measurement equipment to be moved between camps as the sheep are moved between camps. Studies investigating the placement of the LoRa detection units in the camp could place a unit at each corner of the fenced camp and in the middle of the camp to ensure that at least one device can measure the CH_4 emissions from the sheep while they move around the camp.

Because of the nature of CH_4, the development of a small device that can be carried by the animal, with the measurement apparatus placed near the mouth and nose of the animal, could be better suited for ruminant CH_4 emission measurement. Zero Emissions Livestock Project (ZELP) have developed a mask that measures and oxidises eructated and respired CH_4 from cattle, converting about 60% of methane. However, a mask such as the ZELP face mask is not yet available for sheep. Future studies could focus on the development of a face mask CH_4 measurement device, that utilises LoRa technology, for sheep.

Due to the small amount of data available regarding the CH_4 production of ruminant animals under extensive conditions and consequently the uncertainties in the IPCC estimates and national emission inventories of South African sheep production, future studies can use the LoRa CH_4 detection unit under extensive conditions to collect sheep CH_4 emission data and production data. Thereby, generating values that can be used in an IPCC model to predict CH_4 emission values for sheep under South African production conditions.

To compare the LoRa technology and Tier 2 approaches of CH_4 emission estimation for sheep accurately future studies should focus on generating information to correlate these two approaches.